The BTER Foundation's

Biotherapy Journal Review

Vol 2, No 4; Fall 2016

Table of Contents

Publication Credits p. ii

Letter from the Editor p. 3

Section I: Journal Reviews p. 5

SAMPLE FORMATTING:
Brundage AL, Crippen TL, Tomberlin JK. **Methods for external disinfection of blow fly (Diptera: Calliphoridae) eggs prior to use in wound debridement therapy.** Wound Repair Regen. 2016;24(2):384-93.

Section II: Biotherapy Citations for Spring 2016 p. ?

A. Apitherapy/Bee Venom Therapy p. ?

B. Helminthic Therapy p. ?

C. Ichthyotherapy p. ?

D. Leech Therapy p. ?

E. Mammals- Service & Therapy Animals p. ?

F. Maggot Debridement Therapy p. ?

G. Olfactory Detection and Diagnosis p. ?

H. Phage Therapy p. ?

I. Probiotics and Bacteriotherapy p. ?

From the Editor's Desk:

This month, we have selected to review pivotal studies from the literature on maggot therapy, helminthic therapy, phage therapy, and probiotics. While most of these studies offer important findings that will move their respective fields forward, the helminthic therapy study did not identify any statistically significant benefits in multiple sclerosis patients treated with *Trichuris suis* ova therapy. This "negative study" hit many in the helminthic therapy community very hard, and stifled much of the enthusiasm of helminthic therapy researchers and patients alike.

We are reviewing the study here because we believe that protocols that do not work can be as educational – and can potentially advance the field – as much as protocols that do work. In the words of Johnny Cash, ". . . build on failure. . . . use it as a stepping stone." The authors discussed several explanations for their findings; the reviewer notes a few possible explanations as well. Perhaps the reader has additional thoughts . . . about this article, or about the Biotherapy Journal Review in general. If so, visit our website and comment. Or send us your comments and submissions in a letter to the editor at editor@bterfoundation.org. We are here for you, so tell us what you think, and what you want.

Indeed, while I have your attention, let me share my excitement over our upcoming issue dedicated to biotherapy with mammals! Not only will we be reviewing some interesting work in the fields of service animals, hippotherapy and olfactory detection, but we will also be featuring some original artwork between the sections. So be sure to take a close look at the coming issues, and to share with us YOUR reviews, manuscripts, artwork, or announcements.

As always, this issue is brought to you by the generous support of BTER Foundation members and donors, along with the contributions of its authors and reviewers. Thank you, all.



Ronald A. Sherman, MD, MSc, DTM&H
Director, BTER Foundation

P.S. Don't miss an issue of the Biotherapy Journal Review! To ensure you recieve each quarterly issue via email, please sign up here!



Looking to delve deeper into Biotherapy? The BTER Foundation maintains a presence at a variety of biotherapy conferences, and periodically offers webinars and workshops. Learn more at http://www.bterfoundation.org/education

Section I: Journal Reviews

The BTER Foundation's

Biotherapy Journal Review

Volume 2, Number 3; Summer 2016

Title: Methods for external disinfection of blow fly (Diptera: Calliphoridae) eggs prior to use in wound debridement therapy.
Authors: Brundage AL, Crippen TL, Tomberlin JK.
Reviewer: Mr. Frank Stadler, BSc Hon, doctoral candidate
To view the article review, visit page 6

Title: Effect of the probiotic strain *Bifidobacterium animalis subsp. lactis*, BB-12®, on defecation frequency in healthy subjects with low defecation frequency and abdominal discomfort: a randomised, double-blind, placebo-controlled, parallel-group trial.
Authors: Eskesen D, Jespersen L, Michelsen B, Whorwell PJ, Müller-Lissner S, Morberg CM.
Reviewer: Ronald Sherman, MD
To view the article review, visit page 10

Title: Bacteriophage treatment of intransigent diabetic toe ulcers: a case series.
Authors: Fish R, Kutter E, Wheat G, Blasdel B, Kutateladze M, Kuhl S.
Reviewer: Ronald Sherman, MD
To view the article review, visit page 14

Title: Trichuris suis ova therapy in relapsing multiple sclerosis is safe but without signals of beneficial effect.
Authors: Voldsgaard A, Bager P, Garde E, Åkeson P, Leffers AM, Madsen CG, Kapel C, Roepstorff A, Thamsborg SM, Melbye M, Siebner H, Søndergaard HB, Sellebjerg F, Sørensen PS.
Reviewer: Ronald Sherman, MD
To view the article review, visit page 18

Maggot Debridement Therapy Reviews

Journal Citation

Brundage AL, Crippen TL, Tomberlin JK. **Methods for external disinfection of blow fly (Diptera: Calliphoridae) eggs prior to use in wound debridement therapy.** Wound Repair Regen. 2016;24(2):384-93.

Published Abstract

Maggot debridement therapy (MDT) is the use of the larval stage of flies (i.e., Calliphoridae) to remove necrotic tissue and disinfect wounds. Effective MDT requires aseptic technique to prevent the unintentional introduction of pathogenic bacteria into a wound to be debrided; yet the external surface of Calliphoridae eggs is often heavily contaminated with bacteria. Studies of external disinfection of dipteran eggs have been reported, but neither their efficacy nor effect on egg viability has been adequately assessed. The present study evaluated the efficacy of ten disinfection techniques involving immersion, rinse, or a combination of both in formalin, Lysol, formaldehyde, bleach, ethanol, Sporgon, or benzalkonium chloride. All techniques resulted in significant decreases in culturable, aerobic bacterial load on *Lucilia cuprina* eggs. For *L. cuprina*, a 10 minute 3% Lysol immersion was the most efficacious, disinfecting 96.67% of egg samples, while resulting in 31.84% egg mortality. The 5% formalin immersion was least efficacious, disinfecting only 3.33% of *L. cuprina* egg samples, while resulting in 33.51% egg mortality. A formaldehyde immersion, one of the most commonly used disinfection techniques, was moderately effective, disinfecting 66.7% of egg samples, while resulting in 40.16% egg mortality. For *Chrysomya rufifacies* and *Cochliomyia macellaria* egg samples, the 10 minute 3% Lysol immersion disinfected 100% of the samples, and for *Lucilia sericata*, 80% of egg samples, while resulting in 33.97%, 7.34%, and 36.96% egg mortality, respectively. H_2CO disinfected 16.67% of *Ch. rufifacies*, 26.67% of *C. macellaria*, and 56.67% of *L. sericata* egg samples, while resulting in 21.98%, 10.18%, and 32.19% egg mortality, respectively. Due to its high disinfection efficacy and relatively low egg mortality, a 10 minutes 3% Lysol immersion is recommended for sterilizing Calliphoridae eggs prior to rearing of larvae for use in MDT.

Context (briefly, why and how the research was carried out)

The authors point out that while a number of papers outline various sterilization protocols and reagents for fly eggs in medical maggot production, there has been insufficient evaluation or validation of the efficacy of sterilization methods, not only regarding the sterility of the eggs but also regarding egg survival. In this study ten sterilization treatments were tested on eggs from *Lucilia cuprina*. *Cochliomyia macellaria, Chrysomya rufifacies* and *Lucilia sericata* were exposed to two treatments, H2CO and 10 minutes soak in Lysol.

Main experimental treatments to determine sterilization efficacy and eclosion rate included:
Immersion (for 5 minutes unless otherwise stated) in

a) 5% formalin,
b) 10% formalin,
c) 3% Lysol,
d) 3% Lysol for 10 minutes
e) 5% H_2CO
f) 5% NaOCl followed by 5% H_2CO.

Rinsed in

g) 10 cc 70% EtOH
h) 30 cc 1% NaOCl

Combination of

i) 10 minutes immersion in ADBAC followed by rinse in 10 cc 70% EtOH
j) Immersion in 95% EtOH then evaporation for 5 minutes followed by immersion in SporGon®.

Other experiments included

- Effect of agitation on sterilization efficiency and eclosion
- The testing of immersion tolerance for medical fly eggs,
- Identification of baseline contamination of unsterilized eggs,
- Chorion visualization to determine chorion damage

It is important to introduce here the polycarbonate filter system that was used to hold the eggs during sterilization treatments (Figure 1) because it is relevant to the discussion below.



EW-29550-40
Polycarbonate Filter Holder, 13 mm
http://www.coleparmer.com/Product/Polycarbonate_Filter_Holder_13_mm_12_pk/EW-29550-40 (Accessed, 21 August 2016)

Findings

With regard to the sterilization efficacy experiments, please refer to the abstract for detailed results. Overall, the authors conclude from their findings that a 10 minute 3%

Lysol immersion provides the best sterilization and eclosion results for Calliphorid eggs. Immersion tolerance results are somewhat intriguing and will be discussed below in more detail. In summary, *Ch. Rufifacies* and *C. macellaria* exhibited high eclosion rates of above 88%. *L. cuprina* was the most sensitive to length of immersion with a statistically significant drop in eclosion rate for longer immersion times. However, even at 10 minutes eclosion was still high (76.3% +- 7.1). The most surprising result was that for *L. sericata*. Eclosion was below 50% and not significantly different between the treatments. Even eggs that were not immersed had very low eclosion of (47.3%).

Discussion (looking at author's conclusions and your own thoughts)

First up, the authors are to be congratulated for contributing experimental data to the discussion of sterilization efficacy in maggot debridement therapy. Confidence in the quality and sterility of medical maggots is key to making this treatment more popular and more widely available.

This brief review will mainly focus on 1) validity and comparability of test outcomes of this study with the wider literature and medical maggot production practice, 2) the unusually low eclosion rate of *L. sericata* in the immersion experiment compared with the additional sterilization experiment conducted with *L. sericata*, and 3) general relevance of findings for medical maggot production and maggot debridement therapy.

Validity and comparability of sterilization efficacy testing

The authors conducted sterility testing using reagents that have been used by researchers and medial maggot producers as published in the literature as far back as the early 20th century. It is important to note that sterility testing was not conducted using the same methodology as described in the respective literature or practiced in commercial laboratories. To the reviewer's knowledge, there has been no report of the use of a polycarbonate filter holder (Fig 1) for egg sterilization in MDT.

Independent of the chemical reagents used there is a clear difference in the protocols, procedures for sterilization described in this paper compared to the literature and practice. First, in many labs sterilization is conducted in open vessels such as beakers or larger test tubes and greater volumes of sterilization reagent are used to treat and rinse the eggs. The polycarbonate filter holder appears to only hold a volume of 2.5 mL for immersion. This does not allow for sufficiently generous washing of eggs. Moreover, when the sterilization reagent and rinse are pushed through the filter, the eggs may be pressed against the filter paper preventing proper circumferential removal of bacteria from the chorion. Second, deagglutination in practice involves usually both mechanical separation of eggs and surfactant treatment with NaOCl or sodium sulfite. Only treatments f) and h) used this approach with NaOCl and with higher sterilization rates. Third, the most used species for MDT is *L. sericata* and to a lesser degree *L. cuprina*. It should therefore be a priority to thoroughly test sterilization methods on these two species. The authors argue for preferential use of *L. cuprina* on the basis of its sensitivity to immersion. It appears to the reviewer that for the study to have practical use, both *L.*

sericata and *L. cuprina* should have been tested in equal measure or preference should have been given to *L. sericata*.

In short, while this study compares the respective sterilization options on a level playing field with the same albeit new methodology, it does not replicate and evaluate sterilization procedures reported in the literature. This is somewhat problematic because the poor sterilization outcomes reported in this study for many of the commonly used reagents implies inefficient and perhaps even inadequate sterilization of medial maggots used for MDT. However, this is unlikely because any commercial producer faced with such poor sterilization outcomes would rapidly improve the methodology in order to avoid costly waste of resources due to rejection of production batches. Moreover, it must be stressed and made explicit to the uninitiated reader that best practice medical maggot production does include sterility testing of maggots before application in MDT. This ensures that contaminated batches if they should occur are discarded and not used in treatment.

To conclude, this study is invaluable as it establishes the superiority of Lysol as a disinfection reagent under given experimental conditions. The problem, however, is the widely differing experimental methods employed here and elsewhere and the limitations this imposes on generalizability of findings. In fact, it appears to the reviewer that the objectives of this research have been achieved only in part.

Low eclosion rates for L. sericata

The authors report very poor eclosion rates (<50%) for *L. sericata* in the immersion experiment compared to the other species tested, even for those maggots not immersed in water. This appears highly unusual particularly when considering that in the sterilization experiment, eclosion rates of around 70% were noted for *L. sericata*.

Relevance and implication of this study

The reviewer would like to emphasize that quality control in best practice medical production ensures that no contaminated medical maggots reach the patient. The outcomes of this study are likely the consequence of methodological differences and the different equipment used compared to sterilization protocols described in the literature. Nevertheless, the authors have clearly established that under their experimental conditions, Lysol is the best sterilization reagent.

With regard to egg mortality due to sterilization, it should be noted that there is probably an overproduction and surplus of eggs in commercial production meaning that higher mortality rates can probably be tolerated in favor of high sterility. After all, only successfully eclosed larvae are packaged and used for MDT. Higher egg mortality would only be a problem if eggs instead of larvae were packaged and dispatched for MDT treatment.

-Reviewed by Mr. Frank Stadler, BSc Hon, doctoral candidate

Probiotics and Bacteriotherapy Reviews

Journal Citation:

Eskesen D, Jespersen L, Michelsen B, Whorwell PJ, Müller-Lissner S, Morberg CM. **Effect of the probiotic strain *Bifidobacterium animalis subsp. lactis*, BB-12®, on defecation frequency in healthy subjects with low defecation frequency and abdominal discomfort: a randomised, double-blind, placebo-controlled, parallel-group trial.** *Br J Nutr*. 2015;114(10):1638-46.

Published Abstract

The aim of the present study was to investigate the effect of Bifidobacterium animalis subsp. lactis, BB-12®, on two primary end points - defecation frequency and gastrointestinal (GI) well-being - in healthy adults with low defecation frequency and abdominal discomfort. A total of 1248 subjects were included in a randomised, double-blind, placebo-controlled trial. After a 2-week run-in period, subjects were randomised to 1 or 10 billion colony-forming units/d of the probiotic strain BB-12® or a matching placebo capsule once daily for 4 weeks. Subjects completed a diary on bowel habits, relief of abdominal discomfort and symptoms. GI well-being, defined as global relief of abdominal discomfort, did not show significant differences. The OR for having a defecation frequency above baseline for ≥50% of the time was 1·31 (95% CI 0·98, 1·75), P=0·071, for probiotic treatment overall. Tightening the criteria for being a responder to an increase of ≥1 d/week for ≥50 % of the time resulted in an OR of 1·55 (95% CI 1·22, 1·96), P=0·0003, for treatment overall. A treatment effect on average defecation frequency was found (P=0·0065), with the frequency being significantly higher compared with placebo at all weeks for probiotic treatment overall (all P<0·05). Effects on defecation frequency were similar for the two doses tested, suggesting that a ceiling effect was reached with the one billion dose. Overall, 4 weeks' supplementation with the probiotic strain BB-12® resulted in a clinically relevant benefit on defecation frequency. The results suggest that consumption of BB-12® improves the GI health of individuals whose symptoms are not sufficiently severe to consult a doctor (ISRCTN18128385).

Context

Gastrointestinal (GI) microbiota are now recognized to play an integral role in such functional bowel disorders as irritable bowel syndrome (IBS) and constipation. Therefore, it is reasonable to investigate whether administered microbes (probiotics) could alleviate the symptoms of functional bowel disorders or even the mild symptoms in otherwise “normal” individuals.

In prior studies, the administration of *Bifidobacterium animalis* subsp. *lactis* (BB-12®) has already been shown to improve abdominal distress and defecation irregularity in certain patient groups.

In this study, the authors "set out with the primary objective to investigate the effect of the probiotic strain BB-12® in different dosages on defecation frequency and global relief in healthy subjects with low defecation frequency and abdominal discomfort. The secondary objective was to investigate the effect on abdominal pain and bloating. As little is known about healthy populations with minor GI complaints, we also wanted to explore whether certain subgroups were more likely to benefit from treatment with the probiotic strain BB-12®

The study was designed as a randomized, double-blind, placebo-controlled, multicenter clinical trial, conducted in two parts: a 2-week baseline ("run-in period") followed by a 4 week intervention period. The intervention consisted of administering 1 capsule of BB-12® to subjects each day, in doses of either 1 or 10 billion colony-forming units (CFU) per capsule. The control group received identical placebo capsules. Subjects were allowed to continue all other medications and their normal activities, except excessive physical exercise or the consumption of probiotics or fermented dairy products.

Subjects were recruited voluntarily if they were between 18 – 70 years old, with a low defecation frequency (defined as 2 - 4 days/week) and complaints of general abdominal discomfort. Subjects were excluded if they had a history or diagnosis of specific GI disease such as IBS, prior complicated GI surgery, depressive disorder, or the use of oral antibiotics within the preceding 4 weeks. The use of drugs or large doses of vitamins and minerals or food or herbal supplements, even for digestive symptoms, was permitted as long as the substance and dose was stable.

After enrollment and randomization, subjects began the 6 week study, completing a daily and weekly diary assess study outcomes, their symptoms, their self-assessment of any changes over the past week, and their compliance with the study regimen. Subjects also completed questionnaires regarding their physical activity and adverse events.

Primary end points were defecation frequency and GI well-being. Responders were defined as subjects with an average weekly defecation frequency above baseline for at least 50% of the time (at least 2 weeks of the 4-week intervention period); GI well-being responders were defined as subjects who achieved "relief" for at least 50% of the time. Secondary end points were symptom severity scores for abdominal pain and bloating. Data was analyzed with rigorous standard statistical methodology. Before completion of the study, data was submitted for two interim analyses.

Findings

A total of 1248 subjects were randomized into the study and analyzed in the intent-to-treat analysis. Attrition was less than 1%. The characteristics of the three study groups

were similar at Baseline. Subject compliance with study protocol was reported as 100%. The low dose (1 million CFU) arm of the study was closed early, as the result of the interim analysis.

The odds ratio (OR) – which can be likened to statistical probability – for having a defecation frequency above baseline for ≥50% of the time was 1·31 (95% CI 0·98, 1·75), P=0·071, for probiotic treatment overall. However, when looking only at those subjects who increased their bowel frequency by at least ≥1 day/week for ≥50% of the time, there was a significant difference between subjects who received the probiotic supplements and those who did not: OR of 1·55 (95% CI 1·22, 1·96), P=0·0003. Defecation frequency increased for subjects in all three treatment groups, though it increased more for those in the probiotics arms than in the control arm (P=0·0065). Defecation frequency increased most for those subjects whose baseline frequency was ≥3 day/week. No dosage-related difference was seen with defecation frequency nor with any other finding of benefit or harm.

There were no statistically significant differences in symptoms of GI well-being between the three treatment groups. Abdominal pain and bloating decreased by about 40% for subjects in all treatment groups during the study period (no significant difference between different interventions). Stool consistency became softer for subjects in all study groups, though maybe more so for those in the one billion group (P= 0·056).

While 337 adverse events were reported in 233 subjects (18·7 %), all but 17 events in 14 subjects were considered unrelated to the study. Most of these were GI related, and there was no difference between the number or severity of adverse event and the treatment group.

Discussion

The authors are to be commended for executing a rigorous scientific study, complete with randomization and controls, a baseline observational period, extensive statistical analyses (including intent-to-treat analysis and intermediate analysis), and a large enrollment of over 1,200 subjects. Their study demonstrated a clear increase in defecation frequency in patients with baseline stooling of only 3 – 4 days per week, if they received the daily BB-12® probiotic supplement (either 1 billion or 10 billion CFU dose). Although the probiotic was not definitely associated with increased defecation frequency for those with baseline stooling of ≤3 days / week (since most patients increased their defecation frequency during the study, regardless of treatment group), those who increased their frequency by at least one more day per week, for at least 2 weeks out of the 4-week intervention period, were more likely to have received the probiotic than not. Why were the benefits less visible in subjects with lower defecation frequency? This could be a result of a difference in severity, or even a difference in etiology. Clearly the subject population was not a uniform group, even though the distribution among the three treatment arms appears to have been homogeneous. Then again, one can always blame probability – a reality whose effects are minimized only by studying larger and larger sample sizes.

It is interesting that there was no dose effect seen with the BB-12®. This suggests that the benefits of this organisms are reached at some minimal level of consumption (presumably ≤ 1 million CFU / day). Higher doses were associated with no greater benefits and no greater harms.

The authors claim that "the results suggest that consumption of BB-12® improves the GI health of individuals whose symptoms are not sufficiently severe to consult a doctor." But I would temper that statement because – while there were clearly benefits in defecation frequency for many of the subjects – increased defecation frequency should not be interpreted as improved GI health overall. Indeed, all other measurements of GI health, such as symptoms of abdominal discomfort, stool consistency, etc., improved in subjects from all intervention groups, with no clear statistically significant association with the probiotics intervention arms.

Still, the study demonstrated BB-12® to be safe, with no more adverse events than those seen in the placebo-controlled arm. The results suggest that it is reasonable for individuals with infrequent defecation and GI symptoms who have no specific etiology identified and/or who have not felt their symptoms warranted consulting a doctor might benefit from a trial of BB-12®, and have little to lose from doing so.

-Reviewed by Ronald Sherman, MD



Phage Therapy Reviews

Journal Citation

Fish R, Kutter E, Wheat G, Blasdel B, Kutateladze M, Kuhl S. **Bacteriophage treatment of intransigent diabetic toe ulcers: a case series. *J Wound Care*.** 2016;25 Suppl 7:S27-33.

Published Abstract

Objective: Diabetic foot ulcer (DFU) infections are a growing public health problem, with increasing prevalence, poor response to antibiotics and bacterial resistance to traditional antimicrobials leading to increased morbidity and mortality. Bacteriophages (phages), the viruses that target specific bacteria, are one option for addressing bacterial infections, especially where antibiotics fail. Of particular value is a class of virulent staphylococcal phages that hit almost all Staphylococcus aureus, including most methicillin-resistant Staphylococcus aureus (MRSA) strains. Here we report a continuous case series assessing the effectiveness of treating infected and poorly vascularised toe ulcers with exposed bone, after failure of recommended antibiotic therapy, using topically applied Staphylococcus aureus-specific phage. ***Method***: This was a compassionate-use case series of nine patients with diabetes and poorly perfused toe ulcers containing culture-proven Staphylococcus aureus infected bone and soft tissue, who had responded poorly to recommended antibiotic therapy. Six representative cases are presented here. The only generally accepted other option in each case was toe amputation. Exposed portions of the infected phalanges were removed in three cases and left in place in two cases. One case presented as a micro-clot induced gangrene following vascular stenting. In this case, phage were used to prevent infection. The phage used was a commercially available fully sequenced preparation of staphylococcal phage Sb-1. Phage solution was applied topically to the ulcerations once weekly, following standard good wound care. The amount of phage solution applied varied from 0.1 to 0.5 cc depending on volume and area of the ulceration. ***Results***: All infections responded to the phage applications and the ulcers healed in an average of seven weeks with infected bone debridement. One ulcer, where vascularity was extremely poor and bone was not removed to preserve hallux function, required 18 weeks of treatment. In the case of the toe with the micro-clot gangrene, the toe was salvaged and healed in seven weeks without complications. ***Conclusion***: Topical application of a staph mono-phage preparation can be used successfully to treat infected toe ulcerations with bone involvement, despite very poor vascularity and failure of antibiotic treatment. The success within this small series provides the groundwork for controlled clinical trials of staph phage for diabetic foot infections.

Context

This research – an exploration of the utility of bacteriophage to treat infected diabetic foot ulcers – was carried out to address several significant problems currently plaguing our society. Diabetes is on the rise, and so are the complications, not the least of which are foot ulcers that do not heal, and often result in amputations. While antibiotics are not a simple fix for infected diabetic foot ulcers, even under the best of circumstances, the fact that more and more bacteria are becoming resistant to our antibiotics have made the antibiotics less useful. Some organisms, such as methicillin-resistant *Staphylococcus* aureus (MRSA) are now susceptible only to the most toxic of antibiotics, if they are susceptible to any at all.

Bacteriophages are viruses that parasitize only specific bacteria. They do not infect other cells or other animals, and they die out when their host bacteria have been destroyed. The therapeutic use of bacteriophage died out, too, in most of the world, with the antibiotic era. Use and research continued only in a few centers, predominantly in Eastern Europe.

The past couple decades have seen a renewed interest in using bacteriophage to control pathogenic bacteria, but large-scale, controlled, prospective trials are still quite rare.

It is in this context that this project was undertaken: an analysis of the experience of treating diabetic foot ulcers at a hospital-based wound clinic with a 10^7 - 10^8 phage/ml solution of a commercial preparation of staphylococcal phage Sb-1 (Eliava Sb-1). Staphylococcal phage Sb-1 was first isolated in 1977 at the Eliava Institute (Tbilisi, Republic of Georgia) and is a relative of the phages already approved by the U.S. Food and Drug Administration (FDA) for eliminating *Listeria monocytogenes* in some prepared foods. In fact, a closely related phage can be found in another phage product (Staphage Lysate produced by Delmont Laboratories in Philadelphia) which is already legally marketed for use in animals in the U.S. as an immune stimulant against *Staphylococcus* infections.

Patients were considered eligible for the compassionate use of this investigational product if they failed to respond to conventional wound care and antibiotic therapy, with amputation being the only remaining routine option. The authors briefly described their treatment protocol as follows:

> "Treatment involved standard wound care, including weekly soft tissue debridement if necessary. The softened, exposed and obviously infected bone was debrided in three cases and left in place in two cases where bone debridement would have led to a less favorable functional and cosmetic result. For experimental treatment, the phage preparation was dripped into the wound cavity; gauze packing was placed over the wound and soaked with the phage preparation (0.1 to 0.5cc, depending on ulcer volume), covered with petroleum gauze and wrapped with dry

gauze. The patient was instructed to leave the treatment in place for 48 hours before removing it and replacing the treatment with moist dressings. This protocol was repeated weekly until the ulcer became too small to pack."

Findings

A total of nine patients were treated over two years, all with vascular insufficiency, four with osteomyelitis and bone. MRSA was found in one of the wounds, methicillin-susceptible *S aureus* (MSSA) in all of the others. Eight of the wounds were clinically infected; one was gangrenous at considered to be "at high risk" of infection. All 9 infections responded to treatment; all nine wounds healed in an average of seven weeks (range: 4 - 18 weeks). There were no adverse events observed during treatment or follow-up.

Discussion

The authors concluded that "Topical application of a staph mono-phage preparation can be used successfully to treat infected toe ulcerations with bone involvement, despite very poor vascularity and failure of antibiotic treatment. The success within this small series provides the groundwork for controlled clinical trials of staph phage for diabetic foot infections." Both conclusions are valid.

It is worth noting that phage Sb-1 was not the only treatment administered. Subjects received the gamut of good wound care, including frequent debridement and antibiotics, where indicated. But standard of care treatments like these had been attempted all along, and these subjects failed to respond, as defined by the compassionate use protocol. We must assume that they would have gone on to receive an amputation, had they not been allowed to try phage therapy.

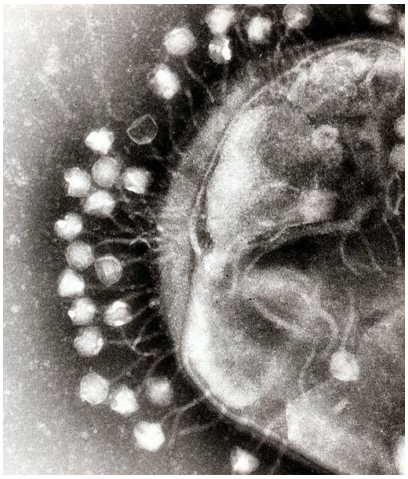
The authors have contributed not only to the literature of phage therapy, but also to our understanding of wound infections in general. This study has demonstrated that even though most wounds are colonized and often infected by multiple organisms, a treatment aimed at a single microbe was still associated with successful treatment of the infected wound. This gives us much food for thought, not only as we begin to design the much-needed and highly justified clinical trials of phage therapy, but also as we re-consider how we currently treat diabetic foot ulcers with broad-spectrum antibiotics in order to kill multiple organisms, despite the commonly seen risks of killing beneficial microbes in the gut (the gastrointestinal biome) and further selecting for microbes with even greater antimicrobial resistance.

Some readers may ask: why did the subjects not receive maggot therapy? How can they be described as having no commercially available treatment options if they had not been considered for maggot therapy? At least in the biotherapy community, this is a valid question. The answer is quite simple: medicinal maggots are NOT currently approved for managing infection, even if they are approved for debridement in

general. Could maggot therapy have been used successfully, either with or instead of phage therapy? While there is data to suggest that this is the case, and although many people may use maggot therapy in tough cases like this, there are no clinical trials to support it. And here we get to the crux of the matter: it is only through well-designed controlled studies – preferably randomized prospective studies, but at least controlled – that we can say with confidence that intervention A is significantly responsible for outcome B. The authors know that they have not proven that phage therapy was the deciding factor, but they hope (and I hope) that their experience and analysis contributes significantly to the body of evidence that we now have to support some good prospective clinical trials of phage therapy.

The authors have contributed not only by providing additional justification for clinical trials, but they have also identified some of the impediments that have blocked clinical trials from having already been done, and they have provided guidance to help overcome some of those impediments. Clinical trials are expensive, and are generally sponsored only by those organizations that believe they can adequately benefit from such a large investment. The authors propose that public money could kick-start some studies, or be combined with corporate money to sponsor such studies. Another obstacle to moving forwards with clinical trials is that there are many different phages and many different ways to combine them into a useful product; and many different infections that could likely benefit. Where do we start? By demonstrating the benefits of a pure strain of naturally occurring phage, they convincingly argue that perhaps we should start with clinical trials of pure, naturally occurring, inexpensive phage, and let the results of those trials generate private support to develop the more expensive – but more lucrative – modified and mixed phage products. This is another approach that may help move us forwards with conducting the clinical trials that are needed before phage therapy can become a reality for the rest of us.

-Reviewed by Ronald Sherman, MD



Helminthic Therapy Reviews

Journal Citation

Voldsgaard A, Bager P, Garde E, Åkeson P, Leffers AM, Madsen CG, Kapel C, Roepstorff A, Thamsborg SM, Melbye M, Siebner H, Søndergaard HB, Sellebjerg F, Sørensen PS. ***Trichuris suis* ova therapy in relapsing multiple sclerosis is safe but without signals of beneficial effect.** Mult Scler. 2015;21(13):1723-9.

Published Abstract

BACKGROUND: An observational study has suggested that relapsing-remitting multiple sclerosis patients with helminth infections have lower disease activity and progression than uninfected multiple sclerosis patients. **OBJECTIVE:** To evaluate the safety and efficacy on MRI activity of treatment with TSO in relapsing MS. **METHODS:** The study was an open-label, magnetic resonance imaging assessor-blinded, baseline-to-treatment study including ten patients with relapsing forms of multiple sclerosis. Median (range) age was 41 (24-55) years, disease duration 9 (4-34) years, Expanded Disability Status Scale score 2.5 (1-5.0), and number of relapses within the last two years 3 (2-5). Four patients received no disease modifying therapy, while six patients received IFN-β. After an observational period of 8 weeks, patients received 2500 ova from the helminth *Trichuris suis* orally every second week for 12 weeks. Patients were followed with serial magnetic resonance imaging, neurological examinations, laboratory safety tests and expression of immunological biomarker genes. **RESULTS:** Treatment with *Trichuris suis* orally was well-tolerated apart from some gastrointestinal symptoms. Magnetic resonance imaging revealed 6 new or enlarged T2 lesions in the run-in period, 7 lesions in the early period and 21 lesions in the late treatment period. Two patients suffered a relapse before treatment and two during treatment. Eight patients developed eosinophilia. The expression of cytokines and transcription factors did not change. **CONCLUSIONS:** In a small group of relapsing multiple sclerosis patients, oral therapy was well tolerated but without beneficial effect.

Context

Multiple sclerosis (MS) is a debilitating neurological disease which progressively leads to the disappearance of myelin around motor nerve cells, thereby slowing the conduction along those pathways. The result is a progressive weakness or paralysis which may wax and wane over time, but ultimately progresses in severity. MS is much more prevalent in regions of the world with well-developed sanitation, but rare in countries with high rates of helminth carriage. This has led many to believe that helminth exposure may protect against the development of MS, and may even be able to slow the progression of this disease.

T. suis, the porcine whipworm, is a helminth naturally infecting pigs. It can transiently colonize humans, but generally cannot survive long in the human gut, and has not been observed to cause human disease. In small, unblinded trials, treatment with *T. suis* ova (TSO) was found to be safe and effective in active in treating inflammatory bowel disease.

While there are some anecdotal reports of MS patients whose symptoms improved with the administration of helminths (helminthic therapy), there has been little in the way of published studies. Correale and colleagues followed 12 patients with MS and various helminth infections for an average of 4.5 years and noted fewer exacerbations, less disease progression and less disease activity on magnetic resonance imaging (MRI) than 12 matched uninfected controls (1). Helminth-infected individuals also had less pro-inflammatory cytokines than non-infected subjects (2).

In 2021, Fleming and colleagues (3) published a Phase 1 trial of 5 treatment-naive relapsing-remitting multiple sclerosis subjects who were given 2500 TSO orally every 2 weeks for 3 months in a baseline versus treatment control exploratory trial. The treatments were well tolerated, MRI lesions decreased, and favorable changes is cytokine levels (increases in IL-4 and IL-10) were measured. The same year, Benzel and colleagues (4) published their studies of 4 MS patients treated with TSO for 6 months. These researchers demonstrated a slight decrease in interleukin (IL)-2, a temporary increase of IL-4, and a mild eosinophily, all suggesting that TSO therapy had a moderate beneficial immunomodulatory impact in MS.

Given these encouraging results, the authors of the present study "conducted a 20-week phase IIa baseline-to-treatment trial to evaluate the safety and efficacy of treatment with TSO in relapsing MS." Their study design was a prospective randomized pilot study of safety and efficacy, with baseline-to-treatment comparisons of clinical findings and blindly assessed MRIs. The 6-week baseline or run-in period was followed by an early (6-week) and late (6 week) treatment periods, with assessments at the beginning and end of each 6-week interval. The intervention (treatment) consisted of an aqueous suspension 2500 TSO per treatment (15 ml) administered in the outpatient clinic every two weeks (total of 6 doses over the 12 weeks of treatment).

Subjects were recruited from among those with relapsing forms of MS for more than a year, at the outpatient clinic at the Danish Multiple Sclerosis Center in Copenhagen. Subjects needed to be between 19 and 55 years old, with an Expanded Disability Status Scale score $\leqslant$ 5.5, no or unchanged IFN-β therapy for the previous three months, at least two documented relapses during the prior 2 years (at least one of which was within the prior 1 year), and MRI results meeting the Barkhof criteria for MS. Subjects were excluded if they received steroids in the previous month, received previous treatment with cyclophosphamide, mitoxantrone or other intensive immunosuppression, received total body irradiation or any experimental therapy, received treatment with azathioprine or any immunomodulatory drug apart from IFN-β within 6 months prior to enrolment, stayed in tropical areas during the preceding 3 months or had a history of current pregnancy or breastfeeding, cardiac diseases, malignant hypertension, diabetes mellitus, renal insufficiency, concurrent systemic infections with hepatitis B or C

virus or HIV, or any medical or psychiatric condition that could interfere with the subject's participation in the study protocol.

Primary endpoints of efficacy were the number of new or enlarging lesions on T2-weighted MRI scans performed at baseline and every 6 weeks. Secondary endpoints were the number of gadolinium enhancing lesions in T1-weighted MRI scans and the number of documented relapses. Standard safety parameters were monitored to assess overall safety. Blood samples were obtained at baseline and after 6 and 12 weeks of therapy for the analysis of cytokine gene expression. MRI and immunological results were analyzed using the non-parametrical chi-square contingency table and Friedman tests with post hoc Kruskal–Wallis tests corrected for multiple comparisons yielding a level of statistical significance at $p = 0.025$. Safety data was assessed using descriptive statistics.

Findings

Ten subjects were enrolled in the study.

The total number of new and/or enlarged T2 lesions was 6 lesions in the pre-treatment scans, 7 lesions in the early treatment period, and 21 lesions in the later treatment period ($p = 0.034$). The number of patients with new and/or enlarged T2 lesions was three in the pre-treatment period, two in the early, and eight in the later treatment period. Two patients experienced a relapse during the baseline period; two patients experienced relapses in the late treatment period.

It should be noted that, of the 10 subjects enrolled, Six were treated with IFN-β during the study; four patients did not receive any immunomodulating drugs. Four patients missed one MRI scan (no. 2) and one patient missed two scans (no. 2 and 4) due to renovation of the MRI scanner. Due to technical problems with a too-short interval between injection of gadolinium contrast and the T1-weighted MRI scan, the count of the number of contrast-enhancing lesions was unreliable and had to be omitted.

No serious adverse events were reported during the study. Four subjects complained of abdominal discomfort or pain, mostly mild, lasting from days to several weeks. Two patients reported flatulence and one of these patients also had diarrhea. Except for eosinophilia, all other blood parameters showed no significant changes.

For the whole patient group, the blood eosinophil count was significantly higher at 6 weeks ($p = 0.02$) and twelve weeks ($p = 0.01$) than at baseline. No significant difference was found in the level of gene expression for any of the cytokines tested: Th1 cytokines and transcription factors *IFNG, TBX21, HLX1* or *EOMES*; the Th2 transcription factor *GATA3*; the Th17 transcription factor *RORC*; regulatory cytokines and the transcription factor *FOXP3, IL10* and *TGFB1*; the antigen-presenting cell cytokines *IL1B, IL12A, IL12B, IL23A, IL27* and *EBI3*; the tumor necrosis factor superfamily molecules *TNF, LTA, LTB* and *TRAIL*; or the matrix metalloproteinase *MMP9, TIMP1* or *SPP1* molecule.

Discussion

This study is not new (it was published in 2015), but it is the latest helminthic therapy clinical trial to be published, and it is an important contribution on several levels.

The authors successfully demonstrated with this small but rigorous study that TSO is a relatively safe modality, having found no serious adverse events after 12 weeks of use. This is consistent with earlier studies of TSO – in MS patients and non-MS patients – where mild GI complaints are the only side effects attributable to TSO.

However, the authors did not demonstrate any clinical benefit of TSO for these 10 MS subjects. Therefore, the authors concluded that "treatment with TSO is safe and well tolerated, but was without any trend towards a positive effect on disease activity when administered to patients with relapsing MS. Hence, we found it unjustified to embark on the planned double-blind, placebo-controlled phase II study."

The authors should be congratulated for having published their work, and the publisher congratulated for printing it, since often studies that fail to prove the desired hypothesis do not see the light of day. The reason? As a result of these findings, some have described this study as a "failure," or worse yet, they have described helminthic therapy as a failure, at least as a possible treatment for MS. I prefer the interpretation of Thomas Edison, who is reported to have said: "I have not failed. I've just found 10,000 ways that won't work." Voldsgaard and colleagues may have proved that this treatment did not work, but what treatment will work? What comes next? A different dose or frequency of administration? A different helminth? A different subset of MS patients, or a different target illness altogether?

The authors go through this very process of examination when reviewing their findings. They clearly gave it the same careful thought when designing the project. Prior research with TSO for MS and other disorders used the same dose. Perhaps there was a difference in the method of preparation of the reagents. Perhaps some of the TSO was less viable than normal, or prepared in a different way (which would explain not only why there was no clinical benefit, but also why there was no significant change in immunological cytokines). While MS is probably a heterogeneous disease, and as such, patients should not all be expected to respond to any treatment in the same way, reviewing the subject profiles did not allow me to identify any characteristic that might select a subset of potential responders for the next clinical trial.

What? Another clinical trial? Didn't the authors state that they found it "unjustified to embark on the planned double-blind, placebo-controlled phase II study." Yes, they did, but that was a large, expensive trial. But I hope they or someone else will be willing to try another small clinical trial of helminthic therapy for MS again. After all, if these researchers identified a significant improvement, we would not accept that as the final verdict, but rather would want to repeat the study, likely with additional twists, to make sure that the results were really valid and reproducible. So why not follow this study with another small, relatively inexpensive study – but with a different helminthic product, or a

different treatment regimen – to test whether or not helminthic therapy really does not benefit MS. After all, the more we learn what does not work, the closer we get to finding out what does work.

-Reviewed by Ronald Sherman, MD

ADDITIONAL REFERENCES

1) Correale J and Farez M. Association between parasite infections and immune responses in multiple sclerosis. *Ann Neurol* 2007; 61: 97–108.

2) Correale J and Farez M. Helminth-infections associated with multiple sclerosis induce regulatory B cells. *Ann Neurol* 2008; 64: 187–199.

3) Fleming JO, Isaak A, Fabry Z, et al. Probiotic helminth administration in relapsing–remitting
multiple sclerosis: A phase 1 study. *Multiple Sclerosis* 2011; 17(6): 743–754.

4) Benzel F, Erdur H, Rosche B, et al. Immune monitoring of *Trichuris suis* egg therapy in multiple
sclerosis patients. *J Helminthology* 2012; 86(3): 339–347.



Your Ad Could Be Here!

Full page for $40, half page for $25, quarter page for $15 - ad will be located within the first 15 pages, for maximum readership.

Contact
editor@BTERFoundation.org
for more information

Section II: Biotherapy Citations for Summer 2016

A. Apitherapy and Bee Venom Therapy

Fratini, F., Cilia, G., Mancini, S., and Felicioli, A. Royal Jelly: An ancient remedy with remarkable antibacterial properties. Microbiol.Res. 2016; 192:130-141; PM: 27664731

ABSTRACT: Royal Jelly (RJ), a honeybee hypopharyngeal gland secretion of young nurse and an exclusive nourishment for bee queen, has been used since ancient times for care and human health and it is still very important in traditional and folkloristic medicine, especially in Asia within the apitherapy. Recently, RJ and its protein and lipid components have been subjected to several investigations on their antimicrobial activity due to extensive traditional uses and for a future application in medicine. Antimicrobial activities of crude Royal Jelly, Royalisin, 10-hydroxy-2-decenoic acid, Jelleines, Major Royal Jelly Proteins against different bacteria have been reported. All these beehive products showed antimicrobial activities that lead their potential employment in several fields as natural additives. RJ and its derived compounds show a highest activity especially against Gram positive bacteria. The purpose of this Review is to summarize the results of antimicrobial studies of Royal Jelly following the timescale of the researches. From the first scientific applications to the isolation of the single components in order to better understand its application in the past years and propose an employment in future studies as a natural antimicrobial agent.

Lee, G. and Bae, H. Bee Venom Phospholipase A2: Yesterday's Enemy Becomes Today's Friend. Toxins.(Basel). 2-22-2016; 8(2):48; PM: 26907347

ABSTRACT: Bee venom therapy has been used to treat immune-related diseases such as arthritis for a long time. Recently, it has revealed that group III secretory phospholipase A2 from bee venom (bee venom group III sPLA2) has in vitro and in vivo immunomodulatory effects. A growing number of reports have demonstrated the therapeutic effects of bee venom group III sPLA2. Notably, new experimental data have shown protective immune responses of bee venom group III sPLA2 against a wide range of diseases including asthma, Parkinson's disease, and drug-induced organ inflammation. It is critical to evaluate the beneficial and adverse effects of bee venom group III sPLA2 because this enzyme is known to be the major allergen of bee venom that can cause anaphylactic shock. For many decades, efforts have been made to avoid its adverse effects. At high concentrations, exposure to bee venom group III sPLA2 can result in damage to cellular membranes and necrotic cell death. In this review, we summarized the current knowledge about the therapeutic effects of bee venom group III sPLA2 on several immunological diseases and described the detailed mechanisms of bee venom group III sPLA2 in regulating various immune responses and physiopathological changes.

Roshan, N., Rippers, T., Locher, C., and Hammer, K. A. Antibacterial activity and chemical characteristics of several Western Australian honeys compared to manuka honey and pasture honey. Arch.Microbiol. 10-26-2016; PM: 27785532

ABSTRACT: The physicochemical parameters and antibacterial activity of 10 Western Australian (WA) and two comparator honeys were determined. Honeys showed a pH range of 4.0-4.7, colour range of 41.3-470.7 mAU, methylglyoxal levels ranging from 82.2 to 325.9 mg kg-1 and hydrogen peroxide levels after 2 h of 22.7-295.5 microM. Antibacterial activity was assessed by the disc diffusion assay, phenol equivalence assay, determination of minimum inhibitory and bactericidal concentrations and a time-kill assay. Activity was shown for all honeys by one or more method, however, activity varied according to which assay was used. Minimum inhibitory concentrations for WA honeys against 10 organisms ranged from 4.0 to >32.0% (w/v). Removal of hydrogen peroxide activity by catalase resulted in decreased activity for several honeys. Overall, the data showed that honeys in addition to those derived from Leptospermum spp. have antimicrobial activity and should not be overlooked as potential sources of clinically useful honey.

Saral, O., Yildiz, O., Aliyazicioglu, R., Yulug, E., Canpolat, S., Ozturk, F., and Kolayli, S. Apitherapy products enhance the recovery of CCL4-induced hepatic damages in rats. Turk.J.Med.Sci. 1-5-2016; 46(1):194-202; PM: 27511354

ABSTRACT: ***BACKGROUND/AIM***: Our objective was to identify the antioxidant properties of honeybee products from Turkey, chestnut honey, pollen, propolis, and royal jelly, and their hepatoprotective activity against CCl4-induced hepatic damage in rats. ***MATERIALS AND METHODS***: Animals were fed with honeybee products for 7 days following CCl4 injection. Development of liver damage and oxidative stress were monitored by measuring the activities of the enzymes alanine transaminase, aspartate transaminase, malondialdehyde, superoxide dismutase, and catalase. Antioxidant capacities of the bee products were identified using FRAP and DPPH assays, as well as by measuring total phenolic and flavonoid contents. ***RESULTS***: The antioxidant activities of the honeybee products were highest in propolis, followed, in order, by pollen, honey, and royal jelly. Despite their different levels of antioxidant capacity, their roles in the prevention of liver damage induced by CCl4 were very similar, which can be explained through their bioavailability to the treated animals. ***CONCLUSIONS***: Our results suggest that honey, propolis, pollen, and royal jelly significantly enhanced the healing of CCl4-induced liver damage, partially due to their antioxidant properties and bioavailability.

B. Helminthic Therapy

Dige, A., Rasmussen, T. K., Nejsum, P., Hagemann-Madsen, R., Williams, A. R., Agnholt, J., Dahlerup, J. F., and Hvas, C. L. Mucosal and systemic immune modulation by *Trichuris trichiura* in a self-infected individual. Parasite Immunol. 10-15-2016; PM: 27743501

ABSTRACT: Helminthic therapy of immune-mediated diseases has gained attention in recent years, but we know little of how helminths modulate human immunity. In this study, we investigated how self-infection with Trichuris (T.) trichiura in an adult man without intestinal disease affected mucosal and systemic immunity. Colonic mucosal biopsies were obtained at baseline, during T. trichiura infection, and after its clearance following mebendazole treatment. Unexpectedly, the volunteer experienced a Campylobacter colitis following T. trichiura clearance, and this served as a positive infectious control. T. trichiura colonisation induced equally increased expressions of T-helper (h)1-, Th2-, Th17-, and Treg- associated cytokines and transcription factors, measured by quantitative polymerase chain reaction. We observed several indicators of modulation of systemic immunity during the T. trichiura infection. Plasma eosinophils and anti-Trichuris antibodies rose markedly during the inoculation phase, and a shift towards a Th2-dominated T cell response at the expense of the Th1-response was observed in circulating T cells. Taken together, our findings corroborate that helminths modulate regional and systemic human immunity. This article is protected by copyright. All rights reserved.

Laan, L. C., Williams, A. R., Stavenhagen, K., Giera, M., Kooij, G., Vlasakov, I., Kalay, H., Kringel, H., Nejsum, P., Thamsborg, S. M., Wuhrer, M., Dijkstra, C. D., Cummings, R. D., and van, Die, I. The whipworm (*Trichuris suis*) secretes prostaglandin E2 to suppress proinflammatory properties in human dendritic cells. FASEB J. 11-2-2016; PM: 27806992

ABSTRACT: Clinical trials have shown that administration of the nematode Trichuris suis can be beneficial in treating various immune disorders. To provide insight in the mechanisms by which this worm suppresses inflammatory responses, an active component was purified from T. suis-soluble products (TsSPs) that suppress---- TNF and IL-12 secretion from LPS-activated human dendritic cells (DCs). Analysis by liquid chromatography tandem mass spectrometry identified this compound as prostaglandin (PG)E2. The purified compound showed similar properties compared with TsSPs and commercial PGE2 in modulating LPS-induced expression of many cytokines and chemokines and in modulating Rab7B and P2RX7 expression in human DCs. Furthermore, the TsSP-induced reduction of TNF secretion from DCs is reversed by receptor antagonists for EP2 and EP4, indicating PGE2 action. T. suis secretes extremely high amounts of PGE2 (45-90 ng/mg protein) within their excretory/secretory products but few related lipid mediators as established by metabololipidomic analysis. Culture of T. suis with several cyclooxygenase (COX) inhibitors that inhibit mammalian prostaglandin synthesis affected the worm's motility but did not inhibit PGE2 secretion, suggesting that the worms can synthesize PGE2 via a COX-independent pathway. We conclude that T. suis secretes PGE2 to suppress proinflammatory responses in human DCs, thereby modulating the host's immune response.-Laan, L. C., Williams, A. R., Stavenhagen, K., Giera, M., Kooij, G., Vlasakov, I., Kalay, H., Kringel, H., Nejsum, P., Thamsborg, S. M., Wuhrer, M., Dijkstra, C. D., Cummings, R. D., van Die, I. The whipworm (Trichuris suis) secretes prostaglandin E2 to suppress proinflammatory properties in human dendritic cells.

Lopes, F., Matisz, C., Reyes, J. L., Jijon, H., Al-Darmaki, A., Kaplan, G. G., and McKay, D. M. Helminth Regulation of Immunity: A Three-pronged Approach to Treat Colitis. Inflamm.Bowel.Dis. 2016; 22(10):2499-2512; PM: 27575495

ABSTRACT: By reputation, the parasite is a pariah, an unwelcome guest. Infection with helminth parasites evokes stereotypic immune responses in humans and mice that are dominated by T helper (Th)-2 responses; thus, a hypothesis arises that infection with helminths would limit immunopathology in concomitant inflammatory disease. Although infection with some species of helminths can cause devastating disease and affect the course of microbial infections, analyses of rodent models of inflammatory disease reveal that infection with helminth parasites, or treatment with helminth extracts, can limit the severity of autoinflammatory disease, including colitis. Intriguing, but fewer, studies show that adoptive transfer of myeloid immune cells treated with helminth products/extracts in vitro can suppress inflammation. Herein, 3 facets of helminth therapy are reviewed and critiqued: treatment with viable ova or larvae, treatment with crude extracts of the worm or purified molecules, and cellular immunotherapy. The beneficial effect of helminth therapy often converges on the mobilization of IL-10 and regulatory/alternatively activated macrophages, while there are reports on transforming growth factor (TGF)-beta, regulatory T cells and dendritic cells, and recent data suggest that helminth-evoked changes in the microbiota should be considered when defining anticolitic mechanisms. We speculate that if the data from animal models translate to humans, noting the heterogeneity therein, then the choice between use of viable helminth ova, helminth extracts/molecules or antigen-pulsed immune cells could be matched to disease management in defined cohorts of patients with inflammatory bowel disease.

C. Ichthyotherapy

No New Publications for Fall 2016.



Do you shop at Amazon.com as much as we do? Amazon has made it easy for anyone to support their favorite nonprofit just by beginning their shopping at smile.amazon.com - all you do is select your nonprofit and the rest of your shopping experience will be just like it is on the standard Amazon website, with the added bonus of 0.5% of your purchase going to the BTER Foundation! Please bookmark smile.amazon.com so that your eligible shopping will benefit Biotherapeutics Education And Research Foundation.

To sign up to support BTER Foundation, visit: https://smile.amazon.com/ch/83-0351746

To find out more about how you can advertise in the Biotherapy Journal review, contact editor@bterfoundation.org

D. Leech Therapy

Emtiazi, M., Jokar, S. Z., and Sadeghmanesh, R. Nasal Polyp in Iranian Traditional Medicine & Conventional Medicine. Iran J.Med.Sci. 2016; 41(3 Suppl):S44; PM: 27840510

ABSTRACT: ***BACKGROUND***: The prevalence of nasal polyp in the population has been estimated to be 0.5-4%. It is noticed that nasal polyp could have a negative impact on the patient's quality of life and indubitably exert a substantial cost on the society. It is observed that, in some cases, complete cure may not be achieved and relapse can occur. In Iranian traditional medicine (ITM), nasal polyp is called "bawaseer e anff" and ITM offers several treatments for it. ***METHODS***: In this review article the definition, signs, symptoms, and treatments of nasal polyp in conventional and traditional medicine are investigated and compared. The Canon of Medicine of Avicenna, liber Medicinalis Almansor of Rhazez, Tohfe Khani, and Makhazen Alta'lim were studied for ITM. Additionally, Google Scholar and PubMed were searched for conventional medicine. ***RESULTS***: The current common treatment for nasal polyp is by applying corticosteroids (locally or orally) and surgery. Definition, signs, symptoms, and prognosis (considering color, shape, consistency) of nasal polyp are described in ITM and its treatments (depending on the kind of polyp and the time from its incidence) are oral or local administration of drugs and using physical manipulations such as phlebotomy, leech therapy, and surgery. ***CONCLUSION***: According to the same definition of nasal polyp and "bawaseer e anff" in the texts, a combination of traditional and conventional treatments could be helpful in order to reduce relapse, surgery indications, and usage of corticosteroids.

Isik, M., Ugur, M., Yakisan, R. S., Sari, T., and Yilmaz, N. Comparison of the effectiveness of medicinal leech and TENS therapy in the treatment of primary osteoarthritis of the knee : A randomized controlled trial. Z.Rheumatol. 8-17-2016; PM: 27535276

ABSTRACT: ***OBJECTIVES***: The aim of this study was to evaluate the effects of leech therapy in the treatment of knee osteoarthritis in terms of duration of effectiveness and symptom relief and to compare these results with transcutaneous electrical nerve stimulation (TENS) therapy. ***MATERIAL AND METHODS***: This study was designed as a prospective, single center, randomized, single-blind and parallel group study. A total of 90 patients were included in the study, 46 in the leech group and 44 in the TENS group. Primary outcome measures were changes of the pain scores in visual analogue scale (VAS) and Western Ontario and McMaster Universities Osteoarthritis Index (WOMAC) on the measurements day 0, 21 and 180. Secondary outcome measures were the changes in the sub-groups of the WOMAC scores. Five leeches were applied to the affected knee, once every week for 3 weeks. ***RESULTS***: The VAS pain score showed a similar decrease in both groups in the evaluation on day 21 ($p < 0.001$). The course of the change of the VAS pain score in both

groups was similar in the comparisons between groups. Long-term benefits of the TENS therapy group were slightly more than the leech therapy group. All the sub-scores of WOMAC in both therapy groups showed a similar decrease ($p = 0.819$). Throughout the study this decrease was statistically significant in both groups ($p < 0.001$). ***CONCLUSION***: Leech therapy relieves symptoms in patients with osteoarthritis of the knee and is as effective as TENS therapy in the management of osteoarthritis of the knee. This treatment has the potential of being an additional or alternative therapy for the non-surgical management of osteoarthritis of the knee.

Marden, J. N., McClure, E. A., Beka, L., and Graf, J. Host Matters: Medicinal Leech Digestive-Tract Symbionts and Their Pathogenic Potential. Front Microbiol. 2016; 7:1569; PM: 27790190

ABSTRACT: Digestive-tract microbiota exert tremendous influence over host health. Host-symbiont model systems are studied to investigate how symbioses are initiated and maintained, as well as to identify host processes affected by resident microbiota. The medicinal leech, Hirudo verbana, is an excellent model to address such questions owing to a microbiome that is consistently dominated by two species, Aeromonas veronii and Mucinivorans hirudinis, both of which are cultivable and have sequenced genomes. This review outlines current knowledge about the dynamics of the H. verbana microbiome. We discuss in depth the factors required for A. veronii colonization and proliferation in the leech crop and summarize the current understanding of interactions between A. veronii and its annelid host. Lastly, we discuss leech usage in modern medicine and highlight how leech-therapy associated infections, often attributable to Aeromonas spp., are of growing clinical concern due in part to an increased prevalence of fluoroquinolone resistant strains.

Rothenberger, J., Petersen, W., Schaller, H. E., and Held, M. Determining the appropriate number and duration of leech therapy in congested tissues using tissue spectrophotometry and laser Doppler flowmetry. Wound.Repair Regen. 10-12-2016; PM: 27733014

ABSTRACT: A universal protocol determining the number of leeches and their application time does not exist. The aim of this study, therefore, is to quantify perfusion dynamics in venous congested tissues after leech application to get more detailed information about changes due to leech-induced skin microcirculation and to evaluate the usability of the O2C device in terms of determining the appropriate number of leeches and the duration of therapy. Twelve patients with the need for leech therapy participated in the study. Perfusion dynamics of the congested tissue was assessed using the O2C device, which determines blood flow (BF), the relative amount of hemoglobin (rHB), and the oxygen saturation (SO2). Measurements were carried out before leech application and on various intervals like 10 minutes, one hour, and three hours after leech application. The leech application effectuated after 10 minutes a non-significant perfusion improvement,

which further increased after one hour with a significant reduction of the relative amount of hemoglobin and a significant increase of blood flow and oxygen saturation (BF= +56.7%; rHB= - 25.5%; SO2= +53.7%). After three hours, the values returned to the levels before leech administration. In two cases, in which further administration of leeches within the measurement period was necessary, no substantial perfusion changes were obtained. The results of this study forms a more precise pattern of microcirculatory changes of leech therapy in congested tissues. According to our measurements a venous drainage improvement can be expected in congested tissue one hour after leech administration. The O2C seems to be a useful method to determine the appropriate number and duration of leech therapy. This article is protected by copyright. All rights reserved.

Spear, M. Medicinal Leech Therapy: Friend or Foe. Plast.Surg.Nurs. 2016; 36(3):121-125; PM: 27606587

ABSTRACT: Leech therapy is the ancient practice of bloodletting or bloodsucking and is considered one of the oldest medical practices known. Its use has spanned centuries and is still used in plastic surgery practices today when performing free flaps and replants. The purpose of this article is to discuss the history of this ancient therapy and to describe the leech and its present indications, complications, contraindications, and techniques.



E. Mammals- Service & Therapeutic Animals

Antunes, F. N., Pinho, A. S., Kleiner, A. F., Salazar, A. P., Eltz, G. D., de Oliveira Junior, A. A., Cechetti, F., Galli, M., and Pagnussat, A. S. Different horse's paces during hippotherapy on spatio-temporal parameters of gait in children with bilateral spastic cerebral palsy: A feasibility study. Res.Dev.Disabil. 8-9-2016; 59:65-72; PM: 27518920

ABSTRACT: Hippotherapy is often carried out for the rehabilitation of children with Cerebral Palsy (CP), with the horse riding at a walking pace. This study aimed to explore the immediate effects of a hippotherapy protocol using a walk-trot pace on spatio-temporal gait parameters and muscle tone in children with Bilateral Spastic CP (BS-CP). Ten children diagnosed with BS-CP and 10 healthy aged-matched children (reference group) took part in this study. The children with BS-CP underwent two sessions of hippotherapy for one week of washout between them. Two protocols (lasting 30min) were applied on separate days: Protocol 1: the horse's pace was a walking pace; and Protocol 2: the horse's pace was a walk-trot pace. Children from the reference group were not subjected to treatment. A wireless inertial measurement unit measured gait spatio-temporal parameters before and after each session. The Modified Ashworth Scale was applied for muscle tone measurement of hip adductors. The participants underwent the gait assessment on a path with surface irregularities (ecological context). The comparisons between BS-CP and the reference group found differences in all spatio-temporal parameters, except for gait velocity. Within-group analysis of children with BS-CP showed that the swing phase did not change after the walk pace and after the walk-trot pace. The percentage of rolling phase and double support improved after the walk-trot. The spasticity of the hip adductors was significantly reduced as an immediate result of both protocols, but this decrease was more evident after the walk-trot. The walk-trot protocol is feasible and is able to induce an immediate effect that improves the gait spatio-temporal parameters and the hip adductors spasticity.

Maenpaa, H., Kela, K., and Satila, H. [Riding therapy in the rehabilitation of mobility-impaired children]. Duodecim. 2016; 132(13-14):1279-1285; PM: 27522837

ABSTRACT: Riding therapy is a comprehensive and functional form of rehabilitation, in which the rehabilitee, the horse and the riding therapist collaborate in order to achieve individually assigned goals that support rehabilitation. In Finland, riding therapy is therapeutic rehabilitation carried out by riding therapists who have undergone approved training. The therapy is mainly implemented in an individual form, but small group working is also applied, e.g. in the form of pair therapy and therapeutic vaulting. In Europe, this form of rehabilitation has been divided into hippotherapy supporting motor functions and heilpedagogical riding therapy functioning in support of upbringing.

Mani, I. and Weese, J. S. Pet Therapy: Enhancing Patient Care Through Time with Animals. Am.Fam.Physician. 11-1-2016; 94(9):737-740; PM: 27929251

No Abstract Available.

Moraes, A. G., Copetti, F., Angelo, V. R., Chiavoloni, L. L., and David, A. C. The effects of hippotherapy on postural balance and functional ability in children with cerebral palsy. J.Phys.Ther.Sci. 2016; 28(8):2220-2226; PM: 27630401

ABSTRACT: ***Purpose***: This study evaluated the effects of hippotherapy on seated postural balance, dynamic balance, and functional performance in children with cerebral palsy and compared the effects of 12 and 24 sessions on seated postural balance. ***Subjects and Methods***: This study included 15 children with cerebral palsy aged between 5 and 10 years. ***Interventions***: A hippotherapy protocol was performed for 30 minutes, twice a week, for 12 weeks. Postural balance in a sitting position was measured using an AMTI AccuSway Plus force platform 1 week before initiating the hippotherapy program and after 12 and 24 weeks. The Berg Balance Scale (BBS) and Pediatric Evaluation of Disability Inventory (PEDI) were used before and after 24 sessions. ***Results***: Significant differences were observed for center of pressure (COP) variables, including medio-lateral (COPml), anteroposterior displacement (COPap), and velocity of displacement (VelCOP), particularly after 24 sessions. There were also significant differences in BBS scores and PEDI score increases associated with functional skills (self-care, social function, and mobility), caregiver assistance (self-care), social function, and mobility. ***Conclusion***: Hippotherapy resulted in improvement in postural balance in the sitting position, dynamic balance, and functionality in children with cerebral palsy, an effect particularly significant after 24 hippotherapy sessions.

Mutoh, T., Mutoh, T., Takada, M., Doumura, M., Ihara, M., Taki, Y., Tsubone, H., and Ihara, M. Application of a tri-axial accelerometry-based portable motion recorder for the quantitative assessment of hippotherapy in children and adolescents with cerebral palsy. J.Phys.Ther.Sci. 2016; 28(10):2970-2974; PM: 27821971

ABSTRACT: ***Purpose***: This case series aims to evaluate the effects of hippotherapy on gait and balance ability of children and adolescents with cerebral palsy using quantitative parameters for physical activity. ***Subjects and Methods***: Three patients with gait disability as a sequela of cerebral palsy (one female and two males; age 5, 12, and 25 years old) were recruited. Participants received hippotherapy for 30 min once a week for 2 years. Gait parameters (step rate, step length, gait speed, mean acceleration, and horizontal/vertical displacement ratio) were measured using a portable motion recorder

equipped with a tri-axial accelerometer attached to the waist before and after a 10-m walking test. **Results**: There was a significant increase in step length between before and after a single hippotherapy session. Over the course of 2 year intervention, there was a significant increase in step rate, gait speed, step length, and mean acceleration and a significant improvement in horizontal/vertical displacement ratio. ***Conclusion***: The data suggest that quantitative parameters derived from a portable motion recorder can track both immediate and long-term changes in the walking ability of children and adolescents with cerebral palsy undergoing hippotherapy.

Petersen, S., Houston, S., Qin, H., Tague, C., and Studley, J. The Utilization of Robotic Pets in Dementia Care. J.Alzheimers.Dis. 11-19-2016; 55(2):569-574; PM: 27716673

ABSTRACT: ***BACKGROUND***: Behavioral problems may affect individuals with dementia, increasing the cost and burden of care. Pet therapy has been known to be emotionally beneficial for many years. Robotic pets have been shown to have similar positive effects without the negative aspects of traditional pets. Robotic pet therapy offers an alternative to traditional pet therapy. ***OBJECTIVE***: The study rigorously assesses the effectiveness of the PARO robotic pet, an FDA approved biofeedback device, in treating dementia-related symptoms. ***METHODS***: A randomized block design with repeated measurements guided the study. Before and after measures included reliable, valid tools such as: RAID, CSDD, GDS, pulse rate, pulse oximetry, and GSR. Participants interacted with the PARO robotic pet, and the control group received standard activity programs. Five urban secure dementia units comprised the setting. ***RESULTS***: 61 patients, with 77% females, average 83.4 years in age, were randomized into control and treatment groups. Compared to the control group, RAID, CSDD, GSR, and pulse oximetry were increased in the treatment group, while pulse rate, pain medication, and psychoactive medication use were decreased. The changes in GSR, pulse oximetry, and pulse rate over time were plotted for both groups. The difference between groups was consistent throughout the 12-week study for pulse oximetry and pulse rate, while GSR had several weeks when changes were similar between groups. ***CONCLUSIONS***: Treatment with the PARO robot decreased stress and anxiety in the treatment group and resulted in reductions in the use of psychoactive medications and pain medications in elderly clients with dementia.

Schoenfeld-Tacher, R. and Kogan, L. R. Professional Veterinary Programs' Perceptions and Experiences Pertaining to Emotional Support Animals and Service Animals, and Recommendations for Policy Development. J.Vet.Med.Educ. 9-30-2016; 1-13; PM: 27689948

ABSTRACT: Given the unique nature of programs in professional veterinary medicine (PVM), the increasing numbers of students requesting accommodations for emotional support animals (ESAs) in higher education settings is of growing interest to student affairs and administrative staff in PVM settings. Since the legislation pertaining to this type of

support animal differs from the laws governing disability service animals, colleges and universities now need to develop new policies and guidelines. Representatives from a sample of 28 PVM programs completed a survey about the prevalence of student requests for ESAs and service animals. PVM associate deans for academic affairs also reported their perceptions of this issue and the challenges these requests might pose within veterinary teaching laboratories and patient treatment areas. Responses indicated that approximately one third of PVM programs have received requests for ESAs (32.1%) in the last 2 years, 17.9% have had requests for psychiatric service animals, and 17.9% for other types of service animals. Despite this, most associate deans reported not having or not being aware of university or college policies pertaining to these issues. Most associate deans are interested in learning more about this topic. This paper provides general recommendations for establishing university or PVM program policies.

Strashko, E. Y., Kapustianska, capital A., and Bobyreva, L. E. Experience of using hippotherapy in complex effects on muscle spirals in children with spastic forms of cerebral palsy. Wiad.Lek. 2016; 69(3 pt 2):527-529; PM: 27717938

ABSTRACT: Matters of physical and medical rehabilitation of children with organic lesions of the nervous system, in particular, with cerebral palsy, are actual in countries around the world. Hippotherapy is neurophysiologically oriented therapy using horses. Determine whether a combination of hippotherapy as a method of rehabilitation in the aftermath of outpatient comprehensive impact on MS on a stationary phase; Study of the effect of hippotherapy as securing and preparation method for learning new postures and movements in children with spastic cerebral palsy forms; The study of the possible optimization of psychophysical state, activation motivations of patients; Determination of the optimal timing of hippotherapy sessions, the number of procedures, the study of possible fatigue factor children. HT classes were conducted at the Ippotsentra "Wind of Change" in the period 2010-2013 the main group of children surveyed (36 people) with spastic forms of cerebral palsy. HT procedure took place twice a day - morning and evening - 30 minutes during 10-12 days. Thus, the proposed integration of the HT program of complex effects on muscle spirals children with spastic cerebral palsy forms is physiologically and anthropologically based on 4-5 day training children adequately transferred the full amount of lessons learned new postures and movements, HT does not cause complications in the somatic and psycho-emotional state of the children, HT enables sensorimotor and psychomotor effects, save and normalize muscle tone for a longer period (up to three months), compared with traditional methods of physiotherapy. HT can serve as a method of learning a new "postures and movements", the preparation of the locomotor apparatus to learn walking.

Tew, S. and Taicher, B. M. A Dog Is a Doctor's Best Friend: The Use of a Service Dog as a Perioperative Assistant. Case.Rep.Pediatr. 2016; 2016:9013520; PM: 27843665

ABSTRACT: Service dogs are beneficial in providing assistance to people with multiple types of disabilities and medical disorders including visual impairment, physical disabilities, seizure disorders, diabetes, and mental illness. Some service animals have been trained as a screening tool for cancer. We review a case involving a 6-year-old female with a history of mast cell mediator release and immediate hypersensitivity due to the urticaria pigmentosa variant of cutaneous mastocytosis who underwent a cystourethroscopy. Her service dog, JJ, who would alert to mast cell mediator release, was used throughout the perioperative course as a means of anxiolysis and comfort and to monitor for mast cell mediator release. This case presents an example of a service dog used in a family-care model in the field of anesthesiology and provides a unique example of using a service dog as an additional monitor to alert the care team for impending mast cell mediator release.

Your Ad Here!

Publicize your business or company with the BTER Foundation's Biotherapy Research Journal! For more information, email editor@bterfoundation.org

F. Maggot Debridement Therapy

Contreras-Ruiz, J., Fuentes-Suarez, A., rroyo-Escalante, S., Moncada-Barron, D., Sosa-de-Martinez, M. C., Maravilla-Franco, E., and Dominguez-Cherit, J. G. [Comparative study of the efficacy of larva therapy for debridement and control of bacterial burden compared to surgical debridement and topical application of an antimicrobial]. Gac.Med.Mex. 2016; 152(Suppl 2):78-87; PM: 27792719

ABSTRACT: Maggot debridement therapy (MDT) is the use of medical grade maggots of the fly Lucilia sericata for wound debridement. Recent observations show that MDT decreases bacterial burden as well. Venous ulcers are the most commonly seen in wound clinics and require, besides adequate treatment of venous hypertension, proper wound bed preparation with debri dement of necrotic tissue and control of potential infections. To evaluate the efficacy of MDT in venous ulcers a randomized controlled trial was designed to compare MDT to surgical debridement and topical application of silver sulfadiazine (SSD) in 19 patients for 4 weeks. The study variables were area reduction, wound bed characteristics, pain, odor, anxiety and bacterial burden using quantitative tissue biopsies. MDT was effective as surgical debridement associated with topical SDD in the debridement of the wound and in reducing its size. A significant difference was observed in the reduction of bacterial burden in favor of the MDT group. Odor and anxiety increased in the MDT group without any difference in the pain intensity between groups. In conclusion, this study suggests that MDT is as effective as surgical debridement for the debridement of necrotic tissue and promote wound healing in venous ulcers and better at reducing bacterial burden.

Cruz-Saavedra, L., az-Roa, A., Gaona, M. A., Cruz, M. L., Ayala, M., Cortes-Vecino, J. A., Patarroyo, M. A., and Bello, F. J. The effect of Lucilia sericata- and Sarconesiopsis magellanica-derived larval therapy on Leishmania panamensis. Acta Trop. 2016; 164:280-289; PM: 27686957

ABSTRACT: This study's main objective was to evaluate the action of larval therapy derived from Lucilia sericata and Sarconesiopsis magellanica (blowflies) regarding Leishmania panamensis using an in vivo model. Eighteen golden hamsters (Mesocricetus auratus) were used; they were divided into 6 groups. The first three groups consisted of 4 animals each; these, in turn, were internally distributed into subgroups consisting of 2 hamsters to be used separately in treatments derived from each blowfly species. Group 1 was used in treating leishmanial lesions with larval therapy (LT), whilst the other two groups were used for evaluating the used of larval excretions and secretions (ES) after the ulcers had formed (group 2) and before they appeared (group 3). The three remaining groups (4, 5 and 6), consisting of two animals, were used as controls in the experiments. Biopsies were taken for histopathological and molecular analysis before,

during and after the treatments; biopsies and smears were taken for assessing parasite presence and bacterial co-infection. LT and larval ES proved effective in treating the ulcers caused by the parasite. There were no statistically significant differences between the blowfly species regarding the ulcer cicatrisation parameters. There were granulomas in samples taken from lesions at the end of the treatments. The antibacterial action of larval treatment regarding co-infection in lesions caused by the parasite was also verified. These results potentially validate effective LT treatment against cutaneous leishmaniasis aimed at using it with humans in the future.

Dauros, Singorenko P., Rosario, R., Windsor, J. A., Phillips, A. R., and Blenkiron, C. The transcriptional responses of cultured wound cells to the excretions and secretions of medicinal Lucilia sericata larvae. Wound.Repair Regen. 11-21-2016; PM: 27868332

ABSTRACT: Maggots, through their excretions and secretions (ES), promote wound healing by removing necrotic tissue, counter bacterial infection, and activate wound associated cells. We investigated the effects of a physiological dose of maggot ES on four wound-associated cell types in vitro with Affymetrix gene expression arrays; keratinocytes, endothelial cells, fibroblasts and monocytes. Keratinocytes showed the fewest (n=5; p<0.05, fold-change +/-2) and smallest fold-changes (up to 2.32x) in gene expression and conversely THP1 monocytes had the most (n=233) and greatest magnitude (up to 44.3x). There were no genes that were altered in all four cell-lines. Gene pathway analysis identified an enrichment of immune response pathways in three of the treated cell-lines. Analyses by quantitative RT-PCR found many genes dynamically expressed in ES dose dependent manner during the three day treatments. Phenotype analyses however found no effects of ES on cell viability, proliferation, migration and angiogenesis. ES was 100x less potent at triggering IL-8 secretion than fibroblasts treated with purified bacterial lipopolysaccharide (LPS; in equivalent amounts to that found in ES; approximately 40EU/mL). Furthermore, co-treatment with LPS and ES decreased the LPS-alone triggered IL-8 secretion by 13%. Although ES had no direct effect on wound cell phenotypes it did partially reduce the immune response to bacterial LPS exposure. These observations were consistent with the profile of transcriptional responses that were dominated by modulation of immune response genes. Maggot therapy may therefore improve wound healing through the secondary effects of these gene changes in the wound cells. This article is protected by copyright. All rights reserved.

Masiero, F. S., Aquino, M. F., Nassu, M. P., Pereira, D. I., Leite, D. S., and Thyssen, P. J. First Record of Larval Secretions of Cochliomyia macellaria (Fabricius, 1775) (Diptera: Calliphoridae) Inhibiting the Growth of Staphylococcus aureus and Pseudomonas aeruginosa. Neotrop.Entomol. 10-1-2016; PM: 27696133

ABSTRACT: Maggot debridement therapy (MDT) consists on the intentional and

controlled application of sterilized larvae of the order Diptera on necrotic skin lesions with the purpose of cleaning necrotic tissue and removing pathogenic bacteria. During MDT, a marked antimicrobial activity has been reported in literature specially associated with antibacterial substances from Lucilia sericata (Meigen); however, regarding Cochliomyia macellaria (Fabricius), little is known. This study aimed to evaluate in vitro inhibition of bacterial growth of Pseudomonas aeruginosa and Staphylococcus aureus in contact with excretions and secretions (ES) from C. macellaria larvae. Larval ES were extracted in sterile distilled water and divided in three groups: ES, containing 400 muL of autoclaved ES; ES+BAC, containing 400 muL of autoclaved ES+0.5-muL bacterial inoculum; and CONT-BAC, containing 400 muL of sterile distilled water +0.5 muL of bacterial inoculum. Aliquots of each experimental group were plated by spreading onto Petri dishes. Seedings were made at 0, 1, 2, 4, and 12 h after the extraction of ES. In ES+BAC groups, inhibition of S. aureus was verified between times 1 and 2 h and P. aeruginosa was inhibited between 0 and 4 h. There was no growth observed in any ES group. In the CONT-BAC groups, the number of colonies from time 4 h became countless for S. aureus and decreased for P. aeruginosa. As reported in the literature, we note here that ES have excellent bactericidal activity for both gram-positive and gram-negative bacteria, and this study shows for the first time the action of the bactericidal activity of exosecretions of C. macellaria against S. aureus and P. aeruginosa.

G. Olfactory Detection and Diagnosis

Clark, J. T. and Ray, A. Olfactory Mechanisms for Discovery of Odorants to Reduce Insect-Host Contact. J.Chem.Ecol. 2016; 42(9):919-930; PM: 27628342

ABSTRACT: Insects have developed highly sophisticated and sensitive olfactory systems to find animal or plant hosts for feeding. Some insects vector pathogens that cause diseases in hundreds of millions of people and destroy billions of dollars of food products every year. There is great interest, therefore, in understanding how the insect olfactory system can be manipulated to reduce their contact with hosts. Here, we review recent advances in our understanding of insect olfactory detection mechanisms, which may serve as a foundation for designing insect control programs based on manipulation of their behaviors by using odorants. Because every insect species has a unique set of olfactory receptors and olfactory-mediated behaviors, we focus primarily on general principles of odor detection that potentially apply to most insects. While these mechanisms have emerged from studies on model systems for study of insect olfaction, such as Drosophila melanogaster, they provide a foundation for discovery of odorants to repel vector insects or reduce their host-seeking behavior.

Pinnelli, G. R., Singh, N. K., Soroker, V., and Plettner, E. Synthesis of Enantiopure Alicyclic Ethers and Their Activity on the Chemosensory Organ of the Ectoparasite of Honey Bees, Varroa destructor. J.Agric.Food Chem. 11-16-2016; 64(45):8653-8658; PM: 27792333

ABSTRACT: The preparation of enantiopure conformationally restricted alicyclic ethers and their inhibitory activities on the chemosensory organ of the Varroa destructor, a parasite of honey bees, are reported in this article. We tested the effect of enantiopure ethers of cis-5-(2'-hydroxyethyl)cyclopent-2-en-1-ol on the Varroa chemosensory organ by electrophysiology, for their ability to inhibit the responses to two honey bee-produced odors that are important for the mite to locate its host: nurse bee head space odor and (E)-beta-ocimene, a honey bee brood pheromone. Previous work with the racemic compounds showed that they suppress the mite's olfactory response to its bee host, which led to incorrect host choice. Based on a structure-activity relationship, we predicted that the two most active compounds-cis-1-butoxy-5-(2'-methoxyethyl)cyclopent-2-ene, cy{4,1}, and (cis-1-ethoxy-5-(2'ethoxyethyl)cyclopent-2-ene, cy{2,2}-could have opposite active enantiomers. Here we studied the enantiomers of both ethers, whose preparation involved enzymatic resolution of racemic diol cis-5-(2'-hydroxyethyl)cyclopent-2-en-1-ol using Lipase AK with vinyl acetate. The racemic diol was prepared from commercially available 2,5-norbornadiene. We observed that the responses of the chemosensory organ to honey bee head space volatiles were significantly decreased by both enantiomers of cy{4,1} and cy{2,2}, but that responses to (E)-beta-ocimene were decreased significantly only by (+)-cy{4,1} (1R,5S) and (-)-cy{2,2} (1S,5R) and not by their respective enantiomers. The importance of this result is that the

racemates could be used to inhibit olfactory detection of bee odors by mites, without a loss in activity relative to the more expensive enantiopure compounds.

Willis, C. M., Britton, L. E., Swindells, M. A., Jones, E. M., Kemp, A. E., Muirhead, N. L., Gul, A., Matin, R. N., Knutsson, L., and Ali, M. Invasive melanoma in-vivo can be distinguished from basal cell carcinoma, benign naevi and healthy skin by canine olfaction: a proof of principle study of differential volatile organic compound emission. Br.J.Dermatol. 7-25-2016; PM: 27454583

ABSTRACT: ***BACKGROUND***: Volatile organic compounds (VOCs) are continuously released by the body during normal metabolic processes, but their profiles change in the presence of cancer, offering an objective, non-invasive approach to diagnosis. Robust evidence that invasive melanoma in-vivo emits a characteristic VOC signature is currently lacking. ***OBJECTIVE***: To conduct a canine olfactory, proof of principle study, to investigate whether VOCs captured from the surface of invasive melanoma are distinguishable from those of basal cell carcinoma (BCC), benign naevi and healthy skin in-vivo. ***METHODS***: After a 13 month training period, the dog's ability to discriminate melanoma was evaluated in a series of 20 double-blind tests, each requiring the selection of one melanoma sample from amongst nine controls (three each of BCC, naevi and healthy skin; all samples being new to the dog), assuring a power of 88% with respect to the analysis. ***RESULTS***: The dog correctly selected the melanoma sample on nine of the 20 occasions, a success rate of 45% (exact 95% CI [0.23, 0.68]), compared with the 10% expected by chance alone. A one-sided exact binomial test gave a p-value of <0.001, supporting the hypothesis that samples were not chosen at random, but that some degree of VOC signal from the melanoma samples significantly increased the probability of their detection. Use of a discrete choice model confirmed melanoma as the most influential of the recorded medical/personal covariates in determining the dog's choice of sample. Notably, accuracy rates based on familiar samples during training were not a reliable indicator of the dog's ability to distinguish melanoma, when confronted with new, unknown samples. ***CONCLUSIONS***: Our study demonstrates that invasive melanoma in-vivo releases odorous VOCs which are distinct from those of BCC, benign naevi and healthy skin, adding to the body of evidence that the volatile metabolome of melanoma contains diag nostically useful biomarkers. This article is protected by copyright. All rights reserved.

H. Phage Therapy

[Anonymous]. Silk route to the acceptance and re-implementation of bacteriophage therapy. Biotechnol.J. 2016; 11(5):595-600; PM: 27008250

ABSTRACT: This multidisciplinary expert panel opinion on bacteriophage therapy has been written in the context of a society that is confronted with an ever-increasing number of antibiotic resistant bacteria. To avoid the return to a pre-antibiotic era, alternative treatments are urgently needed. The authors aim to contribute to the opinion formation of relevant stakeholders on how to potentially develop an infrastructure and legislation that paves the way for the acceptance and re-implementation of bacteriophage therapy.

Aanei, I. L., ElSohly, A. M., Farkas, M. E., Netirojjanakul, C., Regan, M., Taylor, Murphy S., O'Neil, J. P., Seo, Y., and Francis, M. B. Biodistribution of Antibody-MS2 Viral Capsid Conjugates in Breast Cancer Models. Mol.Pharm. 9-30-2016; PM: 27611245

ABSTRACT: A variety of nanoscale scaffolds, including virus-like particles (VLPs), are being developed for biomedical applications; however, little information is available about their in vivo behavior. Targeted nanoparticles are particularly valuable as diagnostic and therapeutic carriers because they can increase the signal-to-background ratio of imaging agents, improve the efficacy of drugs, and reduce adverse effects by concentrating the therapeutic molecule in the region of interest. The genome-free capsid of bacteriophage MS2 has several features that make it well-suited for use in delivery applications, such as facile production and modification, the ability to display multiple copies of targeting ligands, and the capacity to deliver large payloads. Anti-EGFR antibodies were conjugated to MS2 capsids to construct nanoparticles targeted toward receptors overexpressed on breast cancer cells. The MS2 agents showed good stability in physiological conditions up to 2 days and specific binding to the targeted receptors in in vitro experiments. Capsids radiolabeled with 64Cu isotopes were injected into mice possessing tumor xenografts, and both positron emission tomography-computed tomography (PET/CT) and scintillation counting of the organs ex vivo were used to determine the localization of the agents. The capsids exhibit surprisingly long circulation times (10-15% ID/g in blood at 24 h) and moderate tumor uptake (2-5% ID/g). However, the targeting antibodies did not lead to increased uptake in vivo despite in vitro enhancements, suggesting that extravasation is a limiting factor for delivery to tumors by these particles.

Abdallah, K., Hartman, K., Pletzer, D., Zhurina, D., and Ullrich, M. S. The bacteriophage-derived transcriptional regulator, LscR, activates the expression of levansucrase genes in Pseudomonas syringae. Mol.Microbiol. 9-24-2016; PM: 27664099

ABSTRACT: Synthesis of the exopolysaccharide levan occurs in the bacterial blight pathogen of soybean, Pseudomonas syringae pv. glycinea PG4180, when this bacterium encounters moderate to high concentrations of sucrose inside its host plant. The process is mediated by the temperature-dependent expression and secretion of two levansucrases, LscB and LscC. Previous studies showed the importance of a prophage-

associated promoter element in driving the expression of levansucrase genes. Herein, heterologous screening for transcriptional activators revealed that the prophage-borne transcriptional regulator, LscR, from P. syringae mediates expression of levansucrase. A lscR-deficient mutant was generated and exhibited a levan-negative phenotype when grown on a sucrose-rich medium. This phenotype was confirmed by zymographic analysis and Western blots which demonstrated absence of levansucrase in the supernatant and total cell lysates. Transcriptional analysis showed a down-regulation of expression levels of levansucrase and glycosyl hydrolase genes in the lscR-deficient mutant. Ultimately, a direct binding of LscR to the promoter region of levansucrase was demonstrated using electrophoretic mobility shift assays allowing to conclude that a bacteriophage-derived regulator dictates expression of bacterial genes involved in in planta fitness.

Abedon, S. T. Commentary: Phage Therapy of Staphylococcal Chronic Osteomyelitis in Experimental Animal Model. Front Microbiol. 2016; 7:1251; PM: 27558214

No Abstract Available.

Abedon, S. T. Phage therapy dosing: The problem(s) with multiplicity of infection (MOI). Bacteriophage. 2016; 6(3):e1220348. PM: 27738558

ABSTRACT: The concept of bacteriophage multiplicity of infection (MOI) - ratios of phages to bacteria - historically has been less easily applied than many phage workers would prefer or, perhaps, may be aware. Here, toward clarification of the concept, I discuss multiplicity of infection in terms of semantics, history, mathematics, pharmacology, and actual practice. For phage therapy and other biocontrol purposes it is desirable, especially, not to solely employ MOI to describe what phage quantities have been applied during dosing. Why? Bacterial densities can change between bacterial challenge and phage application, may not be easily determined immediately prior to phage dosing, and/or target bacterial populations may not be homogeneous with regard to phage access and thereby inconsistent in terms of what MOI individual bacteria experience. Toward experiment reproducibility and as practiced generally for antibacterial application, phage dosing instead should be described in terms of concentrations of formulations (phage titers) as well as volumes applied and, in many cases, absolute numbers of phages delivered. Such an approach typically will be far more desirable from a pharmacological perspective than solely indicating ratios of agents to bacteria. This essay was adapted, with permission, from an appendix of the 2011 monograph, Bacteriophages and Biofilms, Nova Science Publishers.

Adabi, E., Saebi, F., Moradi Hasan-Abad, A., Teimoori-Toolabi, L., and Kardar, G. A. Evaluation of an Albumin-Binding Domain Protein Fused to Recombinant Human IL-2 and Its Effects on the Bioactivity and Serum Half-Life of the Cytokine. Iran Biomed.J. 11-2-2016; **PM:** 27805072

ABSTRACT: ***BACKGROUND***: Cancer immunotherapy is a promising strategy for cancer treatment. In this strategy, the immune system is triggered to destroy cancer cells. IL-2 is an important factor in passive cancer immunotherapy that helps modulating some important immune functions. One of the IL-2 limitations is low serum half-life; therefore, repetitive high doses of the injections are required to maintain effective concentrations.

High-dose IL-2 therapy results in severe side effects; thus, improvement of its serum half-life would provide therapeutic benefits. ***METHODS***: We have investigated a strategy that is able to utilize an albumin-binding domain (ABD) from streptococcal protein G. In this strategy, the fusion protein ABD-rIL-2 binds to serum albumin, which results in improvement of the IL-2 serum half-life. PET26b+ plasmid was used as an expression vector, which encoded rIL-2 and ABD-rIL-2 both fused to pelB secretion signal under the control of the strong bacteriophage T7 promoter. The constructs were expressed in E. coli Rosetta (DE3) and secreted into the periplasm. ***RESULTS***: The analysis of in vitro bioactivity proved that the fusion of ABD to rIL-2 does not interfere with its bioactivity. ABD-rIL-2 fusion protein indicated higher serum half-life compared to rIL-2, when it was tested in the BALB/c mice. ***CONCLUSION***: The current study provides an alternative strategy to extend the half-life and improve pharmacokinetic properties of rIL-2 without reducing its bioactivity in vitro.

Ahlgren, N. A., Ren, J., Lu, Y. Y., Fuhrman, J. A., and Sun, F. Alignment-free d_2^* oligonucleotide frequency dissimilarity measure improves prediction of hosts from metagenomically-derived viral sequences. Nucleic Acids Res. 11-28-2016; PM: 27899557

ABSTRACT: Viruses and their host genomes often share similar oligonucleotide frequency (ONF) patterns, which can be used to predict the host of a given virus by finding the host with the greatest ONF similarity. We comprehensively compared 11 ONF metrics using several k-mer lengths for predicting host taxonomy from among approximately 32 000 prokaryotic genomes for 1427 virus isolate genomes whose true hosts are known. The background-subtracting measure [Formula: see text] at k = 6 gave the highest host prediction accuracy (33%, genus level) with reasonable computational times. Requiring a maximum dissimilarity score for making predictions (thresholding) and taking the consensus of the 30 most similar hosts further improved accuracy. Using a previous dataset of 820 bacteriophage and 2699 bacterial genomes, [Formula: see text] host prediction accuracies with thresholding and consensus methods (genus-level: 64%) exceeded previous Euclidian distance ONF (32%) or homology-based (22-62%) methods. When applied to metagenomically-assembled marine SUP05 viruses and the human gut virus crAssphage, [Formula: see text]-based predictions overlapped (i.e. some same, some different) with the previously inferred hosts of these viruses. The extent of overlap improved when only using host genomes or metagenomic contigs from the same habitat or samples as the query viruses. The [Formula: see text] ONF method will greatly improve the characterization of novel, metagenomic viruses.

Ahmadi, M., Karimi Torshizi, M. A., Rahimi, S., and Dennehy, J. J. Prophylactic Bacteriophage Administration More Effective than Post-infection Administration in Reducing Salmonella enterica serovar Enteritidis Shedding in Quail. Front Microbiol. 2016; 7:1253; PM: 27555842

ABSTRACT: Infections caused by Salmonella bacteria, often through poultry products, are a serious public health issue. Because of drawbacks associated with antibiotic prophylaxis, alternative treatments are sought. Bacterial viruses (bacteriophages) may provide an effective alternative, but concerns remain with respect to bacteriophage stability and effectiveness. To this end, we assessed the stability of a novel bacteriophage isolated from poultry excreta, siphovirus PSE, and its effectiveness in reducing Salmonella enterica serovar Enteritidis colonization in vitro and in vivo. Moreover, we sought to determine how the timing (prophylactic or therapeutic) and route (oral gavage or vent lip) of PSE administration impacted its effectiveness. Here we

report that significant quantities of viable PSE bacteriophages were recovered following exposure to high and low pH, high temperatures, and bile salts, testifying to its ability to survive extreme conditions. In addition, we found that ileal lactic acid bacteria and Streptococcus spp. counts increased, but colibacilli and total aerobe counts decreased, in quail receiving phage PSE through both oral gavage and vent lip routes. In other experiments, we assessed the efficiency of PSE administration, in both prophylactic and therapeutic contexts, via either oral gavage or vent lip administration, on S. Enteritidis colonization of quail cecal tonsils. Our results demonstrate that administration of PSE as a preventive agent could reduce the S. Enteritidis colonization more effectively than post-challenge administration. Furthermore, oral administration of PSE phage is a more effective prophylactic tool for reduction of S. Enteritidis shedding in poultry than is vent lip administration.

Alhmidi, H., Koganti, S., Tomas, M. E., Cadnum, J. L., Jencson, A., and Donskey, C. J. A pilot study to assess use of fluorescent lotion in patient care simulations to illustrate pathogen dissemination and train personnel in correct use of personal protective equipment. Antimicrob.Resist.Infect.Control. 2016; 5:40; PM: 27777761

ABSTRACT: ***BACKGROUND***: Simulations using fluorescent tracers can be useful in understanding the spread of pathogens and in devising effective infection control strategies. ***METHODS***: During simulated patient care interactions in which providers wore gloves and gowns, we evaluated environmental and personnel dissemination of fluorescent lotion and bacteriophage MS2 from a contaminated mannequin. The frequency of skin and clothing contamination after removal of personal protective equipment (PPE) was compared before versus after an intervention that included education and practice in PPE donning and doffing. ***RESULTS***: Ten healthcare personnel participated in 30 pre-intervention and 30 post-intervention patient care simulations. Fluorescent lotion and bacteriophage MS2 were rapidly disseminated to touched surfaces throughout the room; there was no difference in the frequency of contamination before versus after the PPE training intervention. After the intervention, there was a decrease in skin and/or clothing contamination with fluorescent lotion (9/30, 30 % versus 1/30, 3 %; P = 0.01) and bacteriophage MS2 (8/30, 27 % versus 2/30, 7 %; P = 0.08) and there was a significant reduction in the concentration of bacteriophage MS2 recovered from hands (0.31 versus 0.07 log10plaque-forming units; P < 0.01). ***CONCLUSIONS***: Our findings suggest that simulations with fluorescent lotion can be a useful teaching tool to illustrate the spread of pathogens and provide further evidence that simple PPE training interventions can be effective in reducing contamination of personnel.

Amarillas, L., Chaidez, C., Gonzalez-Robles, A., Lugo-Melchor, Y., and Leon-Felix, J. Characterization of novel bacteriophage phiC119 capable of lysing multidrug-resistant Shiga toxin-producing Escherichia coli O157:H7. PeerJ. 2016; 4:e2423; PM: 27672499

ABSTRACT: ***BACKGROUND***: Shiga toxin-producing Escherichia coli (STEC) is one of the most common and widely distributed foodborne pathogens that has been frequently implicated in gastrointestinal and urinary tract infections. Moreover, high rates of multiple antibiotic-resistant E. coli strains have been reported worldwide. Due to the emergence of antibiotic-resistant strains, bacteriophages are considered an attractive alternative to biocontrol pathogenic bacteria. Characterization is a preliminary step towards designing a phage for biocontrol. ***METHODS***: In this study, we describe the characterization of a bacteriophage designated phiC119, which can infect and lyse several multidrug-

resistant STEC strains and some Salmonella strains. The phage genome was screened to detect the stx-genes using PCR, morphological analysis, host range was determined, and genome sequencing were carried out, as well as an analysis of the cohesive ends and identification of the type of genetic material through enzymatic digestion of the genome. ***RESULTS***: Analysis of the bacteriophage particles by transmission electron microscopy showed that it had an icosahedral head and a long tail, characteristic of the family Siphoviridae. The phage exhibits broad host range against multidrug-resistant and highly virulent E. coli isolates. One-step growth experiments revealed that the phiC119 phage presented a large burst size (210 PFU/cell) and a latent period of 20 min. Based on genomic analysis, the phage contains a linear double-stranded DNA genome with a size of 47,319 bp. The phage encodes 75 putative proteins, but lysogeny and virulence genes were not found in the phiC119 genome. ***CONCLUSION***: These results suggest that phage phiC119 may be a good biological control agent. However, further studies are required to ensure its control of STEC and to confirm the safety of phage use.

Amin, M. G., Pedersen, C. O., Forslund, A., Veith, T. L., and Laegdsmand, M. Influence of soil structure on contaminant leaching from injected slurry. J.Environ.Manage. 12-15-2016; 184(Pt 2):289-296; PM: 27729178

ABSTRACT: Animal manure application to agricultural land provides beneficial organic matter and nutrients but can spread harmful contaminants to the environment. Contamination of fresh produce, surface water and shallow groundwater with the manure-borne pollutants can be a critical concern. Leaching and persistence of nitrogen, microorganisms (bacteriophage, E. coli, and Enterococcus) and a group of steroid hormone (estrogens) were investigated after injection of swine slurry into either intact (structured) or disturbed (homogeneous repacked) soil. The slurry was injected into hexaplicate soil columns at a rate of 50 t ha-1 and followed with four irrigation events: 3.5-h period at 10 mm h-1 after 1, 2, 3, and 4 weeks. The disturbed columns delayed the leaching of a conservative tracer and microorganisms in the first irrigation event compared to the intact columns due to the effect of disturbed macropore flow paths. The slurry constituents that ended up in or near the macropore flow paths of the intact soil were presumably washed out relatively quickly in the first event. For the last three events the intact soil leached fewer microorganisms than the disturbed soil due to the bypassing effect of water through the macropore flow path in the intact soil. Estrogen leached from the intact soil in the first event only, but for the disturbed soil it was detected in the leachates of last two events also. Leaching from the later events was attributed to higher colloid transport from the disturbed soils. In contrast, NO3-N leaching from the intact soil was higher for all events except the first event, probably due to a lower nitrification rate in the disturbed soil. A week after the last irrigation event, the redistribution of all slurry constituents except NO3-N in most of the sections of the soil column was higher for the disturbed soil. Total recovery of E. coli was significantly higher from the disturbed soil and total leaching of mineral nitrogen was significantly lower from the disturbed soil. Results demonstrate how manure-borne constituents injected into undisturbed soil columns respond more as expected in the field, in terms of leaching and persistence, than do the same constituents injected into typically constructed columns of disturbed soil.

Andriolo, J. M., Rossi, R. J., McConnell, C. A., Connors, B. I., Trout, K., Hailer, M. K., Pedulla, M. L., and Skinner, J. L. Influence of iron-doped apatite nanoparticles on viral infection examined in bacterial versus algal systems. IEEE Trans.Nanobioscience. 10-19-2016; PM: 27775532

ABSTRACT: The Centers for Disease Control and Prevention have estimated that each year, two million people in the United States become infected with antibiotic-resistant bacteria, of which, approximately 23 000 die as a direct result of these infections. Phage therapy, or the treatment of bacterial infection by specific, antagonistic viruses, provides one alternative to traditional antibiotics. Bacteriophages, or phages, are bacteriaspecific viruses that possess biological traits that allow for not only the removal of bacterial infection, but also the evasion of bacterial resistance which renders antibiotics ineffective. Previous research has shown the addition of iron-doped apatite nanoparticles (IDANPs) to bacteria prior to phage exposure results in increased bacterial plaques in vitro. Coupled with the biocompatible nature of apatite, these results provide promise for future use of IDANPs as adjuvants to phage therapy along with anti-bacterial applications yet to be explored. Although IDANP enhancement of phage infection has been replicated many times in gram-positive and gram-negative prokaryotic hosts as well as with the utilization of both RNA and DNA viruses, the specific mechanisms involved remain elusive. To further understand increased phage infections in a prokaryotic system, and to evaluate the safety of IDANPs as a treatment used in a eukaryotic system, we have replicated plaque assay experiments in an algal system using Chlorella variabilis NC64A and its virus, Paramecium bursaria chlorella virus 1 (PBCV-1). Statistical modeling was used to evaluate alteration in numbers of plaques observed after viral introduction in IDANP-exposed versus non- IDANP-exposed bacterial and algal cell cultures. While IDANPs synthesized between 25-45 degrees C and doped with 30 % iron have been shown to influence dramatic increases in phage-induced bacterial death, experiments replicated in an algal system indicated viral infections do not increase when C. variabilis cells are pre-exposed to IDANPs. It is essential to potential use of IDANPs as an antibacterial adjuvant that IDANPs do not increase viral infection of eukaryotic host cells during treatment.

Arnold, J. W., Spacht, D., and Koudelka, G. B. Determinants that govern the recognition and uptake of E. coli O157:H7 by Acanthamoeba castellanii. Cell Microbiol. 3-15-2016; PM: 26990156

ABSTRACT: Predation by phagocytic predators is a major source of bacterial mortality. The first steps in protozoan predation are recognition and consumption of their bacterial prey. However, the precise mechanisms governing prey recognition and phagocytosis by protists and the identities of the molecular and cellular factors involved in these processes are, as yet, ill-characterized. Here we show that that the ability of the phagocytic bacterivorous amoebae, Acanthamoeba castellanii to recognize and internalize E. coli, a bacterial prey, varies with LPS structure and composition. The presence of an O-antigen carbohydrate is not required for uptake of E. coli by A castellanii. However O1, but not, O157 O-antigen types inhibit recognition and uptake of bacteria by amoeba. This finding implies O-antigen may function as an anti-predator defense molecule. Recognition and uptake of E. coli by A castellanii is mediated by the interaction of mannose binding protein located on amoebae's surface (aMBP) with LPS carbohydrate. Phagocytic mammalian cells also use mannose-binding lectins to recognize and/or mediate phagocytosis of E. coli. Nonetheless, aMBP apparently displays no sequence similarity with any known metazoan mannose binding protein. Hence the similarity in bacterial recognition mechanisms of amoebae and mammalian phagocytes may be a result of convergent evolution.

Arthur, T. M., Kalchayanand, N., Agga, G. E., Wheeler, T. L., and Koohmaraie, M. Evaluation of Bacteriophage Application to Cattle in Lairage at Beef Processing Plants to Reduce Escherichia coli O157:H7 Prevalence on Hides and Carcasses. Foodborne.Pathog.Dis. 11-21-2016; PM: 27870549

ABSTRACT: Escherichia coli O157:H7 is a major food safety concern for the beef industry. Several studies have provided evidence that cattle hides are the main source of beef carcass contamination during processing and that reductions in the E. coli O157:H7 load on the hides of cattle entering processing facilities will lead to reductions in carcass contamination. Bacteriophages have been proposed as a novel preharvest antimicrobial intervention to reduce the levels of E. coli O157:H7 on cattle hides. The objective of this study was to evaluate a commercialized phage application administered in the lairage area of commercial beef processing plants for the ability to reduce E. coli O157:H7 contamination of cattle hides and carcasses. Cattle lots either received phage spray treatment (n = 289) or did not (n = 301), as they entered the lairage environments in two separate experiments at two different commercial beef processing plants. Hide and carcass samples were collected and analyzed for E. coli O157:H7 prevalence and concentration. Cattle hides receiving phage treatment had an E. coli O157:H7 prevalence of 51.8%, whereas untreated hides had a prevalence of 57.6%. For carcass samples, the E. coli O157 prevalence in treated and untreated samples was 17.1% and 17.6%, respectively. The results obtained from these experiments demonstrated that the treatment of cattle hides with bacteriophages before processing did not produce a significant reduction of E. coli O157:H7 on cattle hides or beef carcasses during processing.

Asensio, M. A., Morella, N. M., Jakobson, C. M., Hartman, E. C., Glasgow, J. E., Sankaran, B., Zwart, P. H., and Tullman-Ercek, D. A Selection for Assembly Reveals That a Single Amino Acid Mutant of the Bacteriophage MS2 Coat Protein Forms a Smaller Virus-like Particle. Nano.Lett. 9-14-2016; 16(9):5944-5950; PM: 27549001

ABSTRACT: Virus-like particles are used to encapsulate drugs, imaging agents, enzymes, and other biologically active molecules in order to enhance their function. However, the size of most virus-like particles is inflexible, precluding the design of appropriately sized containers for different applications. Here, we describe a chromatographic selection for virus-like particle assembly. Using this selection, we identified a single amino acid substitution to the coat protein of bacteriophage MS2 that mediates a uniform switch in particle geometry from T = 3 to T = 1 icosahedral symmetry. The resulting smaller particle retains the ability to be disassembled and reassembled in vitro and to be chemically modified to load cargo into its interior cavity. The pair of 27 and 17 nm MS2 particles will allow direct examination of the effect of size on function in established applications of virus-like particles, including drug delivery and imaging.

Benjamin, C. J., Wright, K. J., Bolton, S. C., Hyun, S. H., Krynski, K., Grover, M., Yu, G., Guo, F., Kinzer-Ursem, T. L., Jiang, W., and Thompson, D. H. Selective Capture of Histidine-tagged Proteins from Cell Lysates Using TEM grids Modified with NTA-Graphene Oxide. Sci.Rep. 10-17-2016; 6:32500; PM: 27748364

ABSTRACT: We report the fabrication of transmission electron microscopy (TEM) grids bearing graphene oxide (GO) sheets that have been modified with Nalpha, Nalpha-dicarboxymethyllysine (NTA) and deactivating agents to block non-selective binding between GO-NTA sheets and non-target proteins. The resulting GO-NTA-coated grids

with these improved antifouling properties were then used to isolate His6-T7 bacteriophage and His6-GroEL directly from cell lysates. To demonstrate the utility and simplified workflow enabled by these grids, we performed cryo-electron microscopy (cryo-EM) of His6-GroEL obtained from clarified E. coli lysates. Single particle analysis produced a 3D map with a gold standard resolution of 8.1 A. We infer from these findings that TEM grids modified with GO-NTA are a useful tool that reduces background and improves both the speed and simplicity of biological sample preparation for high-resolution structure elucidation by cryo-EM.

Bhowmick, T. S., Das, M., Heinz, K. M., Krauter, P. C., and Gonzalez, C. F. Transmission of phage by glassy-winged sharpshooters, a vector of Xylella fastidiosa. Bacteriophage. 2016; 6(3):e1218411; PM: 27738554

ABSTRACT: Xylella fastidiosa subsp. fastidiosa (Xff) is the causal agent of Pierce's Disease (PD) of grapevines and is vectored by the glassy-winged sharpshooter (GWSS, Homalodisca vitripennis). Previously we have reported the development of a bacteriophage (phage) based biocontrol system for PD, but no information on insect transmission of phages has been reported. Here we communicate that laboratory reared GWSSs fed on cowpea plants (Vigna unguiculata subsp. unguiculata) harboring the virulent phage Paz were able to uptake of phage efficiently when the phage was present in high concentration, but were inefficient in transfer to plants.

Billiau, A. At the Centennial of the Bacteriophage: Reviving the Overlooked Contribution of a Forgotten Pioneer, Richard Bruynoghe (1881-1957). J.Hist Biol. 2016; 49(3):559-580; PM: 26515105

ABSTRACT: The year 2015 marks the 100th anniversary of a publication by William Twort, in which he first described lysis of bacterial cultures by a filterable, self-replicating agent. In 1917, Felix d'Herelle, coined the name "bacteriophage" for the proposed agent. Two Belgian teams of microbiologists were among the few to critically examine the nature of the bacteriophage at that time. Although their experimental results agreed, their interpretations did not. Richard Bruynoghe (University of Louvain/Leuven) interpreted them as supportive of d'Herelle's notion of an ultramicroscopic microorganism. Jules Bordet (University of Brussels) found the proposal of a complex organism unnecessary and saw the bacteriophage as a simple endogenous bacterial enzyme endowed with capacity to induce its own secretion as well as ability to cause lysis of the bacteria. Two decades would elapse before bacteriophages were visualized and confirmed to be organized particles. However, by that time, Bruynoghe's work, that had only been published in short notes in society proceedings, was virtually forgotten. The present paper revives his original observations and arguments, while also recognizing that Bordet's alternative hypothesis had scientific merit.

Binzel, D. W., Khisamutdinov, E., Vieweger, M., Ortega, J., Li, J., and Guo, P. Mechanism of three-component collision to produce ultrastable pRNA three-way junction of Phi29 DNA-packaging motor by kinetic assessment. RNA. 2016; 22(11):1710-1718; PM: 27672132

ABSTRACT: RNA nanotechnology is rapidly emerging. Due to advantageous pharmacokinetics and favorable in vivo biodistribution, RNA nanoparticles have shown promise in targeted delivery of therapeutics. RNA nanotechnology applies bottom-up assembly, thus elucidation of the mechanism of interaction between multiple components is of fundamental importance. The tendency of diminishing concern about RNA instability has accelerated by the finding of the novel thermostable three-way junction (3WJ) motif of the phi29 DNA-packaging motor. The kinetics of these three components, each averaging 18 nucleotides (nt), was investigated to elucidate the mechanism for producing the stable 3WJ. The three fragments coassembled into the 3WJ with extraordinary speed and affinity via a two-step reaction mechanism, 3WJb + 3WJc <--> 3WJbc + 3WJa <--> 3WJabc The first step of reaction between 3WJb and 3WJc is highly dynamic since these two fragments only contain 8 nt for complementation. In the second step, the 3WJa, which contains 17 nt complementary to the 3WJbc complex, locks the unstable 3WJbc complex into a highly stable 3WJ. The resulting pRNA-3WJ is more stable than any of the dimer species as shown in the much more rapid association rates and slowest dissociation rate constant. The second step occurs at a very high association rate that is difficult to quantify, resulting in a rapid formation of a stable 3WJ. Elucidation of the mechanism of three-component collision in producing the ultrastable 3WJ proves a promising platform for bottom-up assembly of RNA nanoparticles as a new class of anion polymers for material science, electronic elements, or therapeutic reagents.

Blazquez, B., Fresco-Taboada, A., Iglesias-Bexiga, M., Menendez, M., and Garcia, P. PL3 Amidase, a Tailor-made Lysin Constructed by Domain Shu ffl ing with Potent Killing Activity against Pneumococci and Related Species. Front Microbiol. 2016; 7:1156; PM: 27516758

ABSTRACT: The emergence and spread of antibiotic-resistant bacteria is pushing the need of alternative treatments. In this context, phage therapy is already a reality to successfully fight certain multiresistant bacteria. Among different phage gene products, murein hydrolases responsible of phage progeny liberation (also called lysins or endolysins) are weapons that target specific peptidoglycan bonds, leading to lysis and death of susceptible bacteria when added from the outside. In the pneumococcal system, all but one phage murein hydrolases reported to date share a choline-binding domain that recognizes cell walls containing choline residues in the (lipo)teichoic acids. Some purified pneumococcal or phage murein hydrolases, as well as several chimeric proteins combining natural catalytic and cell wall-binding domains (CBDs) have been used as effective antimicrobials. In this work we have constructed a novel chimeric N-acetylmuramoyl-L-alanine amidase (PL3) by fusing the catalytic domain of the Pal amidase (a phage-coded endolysin) to the CBD of the LytA amidase, the major pneumococcal autolysin. The physicochemical properties of PL3 and the bacteriolytic effect against several pneumococci (including 48 multiresistant representative strain) and related species, like Streptococcus pseudopneumoniae, Streptococcus mitis, and Streptococcus oralis, have been studied. Results have shown that low doses of PL3, in the range of 0.5-5 mug/ml, are enough to practically sterilize all choline-containing strains tested. Moreover, a single 20-mug dose of PL3 fully protected zebrafish embryos from infection by S. pneumoniae D39 strain. Importantly, PL3 keeps 95% enzymatic activity after 4 weeks at 37 degrees C and can be lyophilized without losing activity, demonstrating a remarkable robustness. Such stability, together with a prominent efficacy against a narrow spectrum of human pathogens, confers to PL3 the

characteristic to be an effective therapeutic. In addition, our results demonstrate that the structure/function-based domain shu ffl ing approach is a successful method to construct tailor-made endolysins with higher bactericidal activities than their parental enzymes.

Bocian, K., Borysowski, J., Zarzycki, M., Pacek, M., Weber-Dabrowska, B., Machcinska, M., Korczak-Kowalska, G., and Gorski, A. The Effects of T4 and A3/R Bacteriophages on Differentiation of Human Myeloid Dendritic Cells. Front Microbiol. 2016; 7:1267; PM: 27582733

ABSTRACT: Bacteriophages (phages) are viruses of bacteria. Here we evaluated the effects of T4 and A3/R bacteriophages, as well as phage-generated bacterial lysates, on differentiation of human myeloid dendritic cells (DCs) from monocytes. Neither of the phages significantly reduced the expression of markers associated with differentiation of DCs and their role in the activation of T cells (CD40, CD80, CD83, CD86, CD1c, CD11c, MHC II, PD-L1, PD-L2, TLR2, TLR4, and CCR7) and phagocytosis receptors (CD64 and DEC-205). By contrast, bacterial lysate of T4 phage significantly decreased the percentages of DEC-205- and CD1c-positive cells. The percentage of DEC-205-positive cells was also significantly reduced in DCs differentiated in the presence of lysate of A3/R phage. Thus while bacteriophages do not substantially affect differentiation of DCs, some products of phage-induced lysis of bacterial cells may influence the differentiation and potentially also some functions of DCs. Our results have important implications for phage therapy of bacterial infections because during infections monocytes recruited to the site of inflammation are an important source of inflammatory DCs.

Bocian, K., Borysowski, J., Zarzycki, M., Wierzbicki, P., Klosowska, D., Weber-Dabrowska, B., Korczak-Kowalska, G., and Gorski, A. LPS-Activated Monocytes Are Unresponsive to T4 Phage and T4-Generated Escherichia coli Lysate. Front Microbiol. 2016; 7:1356; PM: 27630621

ABSTRACT: A growing body of data shows that bacteriophages can interact with different kinds of immune cells. The objective of this study was to investigate whether T4 bacteriophage and T4-generated Escherichia coli lysate affect functions of monocytes, the key population of immune cells involved in antibacterial immunity. To that end, we evaluated how T4 and E. coli lysate influence the expression of main costimulatory molecules including CD40, CD80 and CD86, TLR2, TLR4 on monocytes, as well as the production of IL-6 and IL-12 in cultures of peripheral blood mononuclear cells (PBMCs). Separate experiments were performed on unactivated and LPS-activated PBMCs cultures. Both studied preparations significantly increased the percentage of CD14(+)CD16(-)CD40(+) and CD14(+)CD16(-)CD80(+) monocytes in unactivated PBMCs cultures, as well as the concentration of IL-6 and IL-12 in culture supernates. However, neither purified T4 nor E. coli lysate had any significant effect on monocytes in LPS-activated PBMCs cultures. We conclude that LPS-activated monocytes are unresponsive to phages and products of phage-induced lysis of bacteria. This study is highly relevant to phage therapy because it suggests that in patients with infections caused by Gram-negative bacteria the administration of phage preparations to patients and lysis of bacteria by phages are not likely to overly stimulate monocytes.

Bodier-Montagutelli, E., Morello, E., L'Hostis, G., Guillon, A., Dalloneau, E., Respaud, R., Pallaoro, N., Blois, H., Vecellio, L., Gabard, J., and Heuze-Vourc'h, N. Inhaled phage therapy: a promising and challenging approach to treat bacterial respiratory infections. Expert.Opin.Drug Deliv. 11-10-2016; 1-14; PM: 27776446

ABSTRACT: ***INTRODUCTION***: Bacterial respiratory tract infections (RTIs) are increasingly difficult to treat due to evolving antibiotic resistance. In this context, bacteriophages (or phages) are part of the foreseen alternatives or combination therapies. Delivering phages through the airways seems more relevant to accumulate these natural antibacterial viruses in proximity to their bacterial host, within the infectious site. ***Areas covered***: This review addresses the potential of phage therapy to treat RTIs and discusses preclinical and clinical results of phages administration in this context. Recent phage formulation and aerosolization attempts are also reviewed, raising technical challenges to achieve efficient pulmonary deposition via inhalation. ***Expert opinion***: Overall, the inhalation of phages as antibacterial treatment seems both clinically relevant and technically feasible. Several crucial points still need to be investigated, such as phage product pharmacokinetics and immunogenicity. Furthermore, given phage-specific features, appropriate regulatory and manufacturing guidelines will need to be defined. Finally, randomized controlled clinical trials should be carried out to establish phage therapy's clinical positioning in the antimicrobial arsenal against RTIs.

Bojanowski, C. L., Crookes-Goodson, W. J., and Robinson, J. B. Use of bacteriophage to prevent Pseudomonas aeruginosa contamination and fouling in Jet A aviation fuel. Biofouling. 2016; 32(10):1163-1170; PM: 27718644

ABSTRACT: In the present study, the use of bacteriophages to prevent growth and/or biofouling by Pseudomonas aeruginosa PAO1 was investigated in microcosms containing Jet A aviation fuel as the carbon source. Bacteriophages were found to be effective at preventing biofilm formation but did not always prevent planktonic growth in the microcosms. This result was at odds with experiments conducted in nutrient-rich medium, demonstrating the necessity to test antimicrobial and antifouling strategies under conditions as near as possible to the 'real world'. The success of the bacteriophages at preventing biofilm formation makes them potential candidates as antifouling agents for fuel systems.

Bonilla, N., Rojas, M. I., Netto Flores, Cruz G., Hung, S. H., Rohwer, F., and Barr, J. J. Phage on tap-a quick and efficient protocol for the preparation of bacteriophage laboratory stocks. PeerJ. 2016; 4:e2261; PM: 27547567

ABSTRACT: A major limitation with traditional phage preparations is the variability in titer, salts, and bacterial contaminants between successive propagations. Here we introduce the Phage On Tap (PoT) protocol for the quick and efficient preparation of homogenous bacteriophage (phage) stocks. This method produces homogenous, laboratory-scale, high titer (up to 10(10-11) PFU.ml(-1)), endotoxin reduced phage banks that can be used to eliminate the variability between phage propagations and improve the molecular characterizations of phage. The method consists of five major parts, including phage propagation, phage clean up by 0.22 mum filtering and chloroform treatment, phage

concentration by ultrafiltration, endotoxin removal, and the preparation and storage of phage banks for continuous laboratory use. From a starting liquid lysate of > 100 mL, the PoT protocol generated a clean, homogenous, laboratory phage bank with a phage recovery efficiency of 85% within just two days. In contrast, the traditional method took upwards of five days to produce a high titer, but lower volume phage stock with a recovery efficiency of only 4%. Phage banks can be further purified for the removal of bacterial endotoxins, reducing endotoxin concentrations by over 3,000-fold while maintaining phage titer. The PoT protocol focused on T-like phages, but is broadly applicable to a variety of phages that can be propagated to sufficient titer, producing homogenous, high titer phage banks that are applicable for molecular and cellular assays.

Bordenstein, S. R. and Bordenstein, S. R. Eukaryotic association module in phage WO genomes from Wolbachia. Nat.Commun. 10-11-2016; 7:13155; PM: 27727237

ABSTRACT: Viruses are trifurcated into eukaryotic, archaeal and bacterial categories. This domain-specific ecology underscores why eukaryotic viruses typically co-opt eukaryotic genes and bacteriophages commonly harbour bacterial genes. However, the presence of bacteriophages in obligate intracellular bacteria of eukaryotes may promote DNA transfers between eukaryotes and bacteriophages. Here we report a metagenomic analysis of purified bacteriophage WO particles of Wolbachia and uncover a eukaryotic association module in the complete WO genome. It harbours predicted domains, such as the black widow latrotoxin C-terminal domain, that are uninterrupted in bacteriophage genomes, enriched with eukaryotic protease cleavage sites and combined with additional domains to forge one of the largest bacteriophage genes to date (14,256 bp). To the best of our knowledge, these eukaryotic-like domains have never before been reported in packaged bacteriophages and their phylogeny, distribution and sequence diversity imply lateral transfers between bacteriophage/prophage and animal genomes. Finally, the WO genome sequences and identification of attachment sites will potentially advance genetic manipulation of Wolbachia.

Breslin, E. F., Cornell, J., Schuhmacher, Z., Himelright, M., Andos, A., Childs, A., Clem, A., Gerber, M., Gordillo, A., Harb, L., Hossain, R., Hutchinson, T., Miller, I., Morton, E., Walters, R., Webb, D., and Temple, L. Complete Genome Sequence of Bacillus Phage Belinda from Grand Cayman Island. Genome Announc. 10-13-2016; 4(5); PM: 27738022

ABSTRACT: Soil from George Town, Grand Cayman Island, yielded the bacteriophage Belinda, isolated on Bacillus thuringiensis DSM 350. We present here the analysis of the complete genome sequence of 162,308 bp, with 298 predicted genes. The genome also contains three tRNA genes. Belinda belongs to the C1 cluster of Bacillus phages.

Brown, D. R., Sheppard, C. M., Burchell, L., Matthews, S., and Wigneshweraraj, S. The Xp10 Bacteriophage Protein P7 Inhibits Transcription by the Major and Major Variant Forms of the Host RNA Polymerase via a Common Mechanism. J.Mol.Biol. 10-9-2016; 428(20):3911-3919; PM: 27515396

ABSTRACT: The sigma factor is a functionally obligatory subunit of the bacterial transcription machinery, the RNA polymerase. Bacteriophage-encoded small proteins that either modulate or inhibit the bacterial RNAP to allow the temporal regulation of

bacteriophage gene expression often target the activity of the major bacterial sigma factor, sigma70. Previously, we showed that during Xanthomonas oryzae phage Xp10 infection, the phage protein P7 inhibits the host RNAP by preventing the productive engagement with the promoter and simultaneously displaces the sigma70 factor from the RNAP. In this study, we demonstrate that P7 also inhibits the productive engagement of the bacterial RNAP containing the major variant bacterial sigma factor, sigma54, with its cognate promoter. The results suggest for the first time that the major variant form of the host RNAP can also be targeted by bacteriophage-encoded transcription regulatory proteins. Since the major and major variant sigma factor interacting surfaces in the RNAP substantially overlap, but different regions of sigma70 and sigma54 are used for binding to the RNAP, our results further underscore the importance of the sigma-RNAP interface in bacterial RNAP function and regulation and potentially for intervention by antibacterials.

Brown, T. W., Chen, W., and Casanova, L. M. Survival and disinfection of an enveloped surrogate virus on Tyvek suits used for health care personal protective equipment. Am.J.Infect.Control. 12-1-2016; 44(12):1734-1735; PM: 27597391

ABSTRACT: The survival and disinfection of bacteriophage Phi6, an enveloped surrogate virus, was evaluated on Tyvek suits used as health care personal protective equipment. After 6 hours there was 2-log10 inactivation of virus on Tyvek suits at both 40% and 60% relative humidity. Both hypochlorite and quaternary ammonium produced a >3.21- and >4.33-log10 reduction of the virus, respectively, after 1-minute contact time. Enveloped viruses can survive on Tyvek suits beyond the length of a single patient care encounter, but they can be inactivated by chemical disinfectants.

Bryan, D., El-Shibiny, A., Hobbs, Z., Porter, J., and Kutter, E. M. Bacteriophage T4 Infection of Stationary Phase E. coli: Life after Log from a Phage Perspective. Front Microbiol. 2016; 7:1391; PM: 27660625

ABSTRACT: Virtually all studies of phage infections investigate bacteria growing exponentially in rich media. In nature, however, phages largely encounter non-growing cells. Bacteria entering stationary phase often activate well-studied stress defense mechanisms that drastically alter the cell, facilitating its long-term survival. An understanding of phage-host interactions in such conditions is of major importance from both an ecological and therapeutic standpoint. Here, we show that bacteriophage T4 can efficiently bind to, infect and kill E. coli in stationary phase, both in the presence and absence of a functional stationary-phase sigma factor, and explore the response of T4-infected stationary phase cells to the addition of fresh nutrients 5 or 24 h after that infection. An unexpected new mode of response has been identified. "Hibernation" mode is a persistent but reversible dormant state in which the infected cells make at least some phage enzymes, but halt phage development until appropriate nutrients become available before producing phage particles. Our evidence indicates that the block in hibernation mode occurs after the middle-mode stage of phage development; host DNA breakdown and the incorporation of the released nucleotides into phage DNA indicate that the enzymes of the nucleotide synthesizing complex, under middle-mode control, have been made and assembled into a functional state. Once fresh glucose and amino acids become available, the standard lytic infection process rapidly resumes and concentrations of up to 10(11) progeny phage (an average of about 40 phage per initially present cell) are produced. All evidence is consistent with the hibernation-mode control point lying between middle mode and late mode T4 gene expression. We have

also observed a "scavenger" response, where the infecting phage takes advantage of whatever few nutrients are available to produce small quantities of progeny within 2 to 5 h after infection. The scavenger response seems able to produce no more than an average of one phage per originally available cell, and few if any further progeny are produced by cells in this mode even if fresh nutrients are made available later.

Burmeister, A. R., Lenski, R. E., and Meyer, J. R. Host coevolution alters the adaptive landscape of a virus. Proc.Biol.Sci. 9-28-2016; 283(1839); PM: 27683370

ABSTRACT: The origin of new and complex structures and functions is fundamental for shaping the diversity of life. Such key innovations are rare because they require multiple interacting changes. We sought to understand how the adaptive landscape led to an innovation whereby bacteriophage lambda evolved the new ability to exploit a receptor, OmpF, on Escherichia coli cells. Previous work showed that this ability evolved repeatedly, despite requiring four mutations in one virus gene. Here, we examine how this innovation evolved by studying six intermediate genotypes of lambda isolated during independent transitions to exploit OmpF and comparing them to their ancestor. All six intermediates showed large increases in their adsorption rates on the ancestral host. Improvements in adsorption were offset, in large part, by the evolution of host resistance, which occurred by reduced expression of LamB, the usual receptor for lambda. As a consequence of host coevolution, the adaptive landscape of the virus changed such that selection favouring four of the six virus intermediates became stronger after the host evolved resistance, thereby accelerating virus populations along the path to using the new OmpF receptor. This dependency of viral fitness on host genotype thus shows an important role for coevolution in the origin of the new viral function.

Buttimer, C., O'Sullivan, L., Elbreki, M., Neve, H., McAuliffe, O., Ross, R. P., Hill, C., O'Mahony, J., and Coffey, A. Genome Sequence of Jumbo Phage vB_AbaM_ME3 of Acinetobacter baumanni. Genome Announc. 8-25-2016; 4(4); PM: 27563033

ABSTRACT: Bacteriophage (phage) vB_AbaM_ME3 was previously isolated from wastewater effluent using the propagating host Acinetobacter baumannii DSM 30007. The full genome was sequenced, revealing it to be the largest Acinetobacter bacteriophage sequenced to date with a size of 234,900 bp and containing 326 open reading frames (ORFs).

Cardoso, M. E., Fernandez, L., Tejeria, E., Esperon, P., and Teran, M. Evaluation of a Labelled Bacteriophage with 99mTc as a Potential Agent for Infection Diagnosis. Curr.Radiopharm. 2016; 9(2):137-142; PM: 26876366

ABSTRACT: ***BACKGROUND AND OBJECTIVE***: The design of target-specific molecular imaging probes to determine infection sites are mainly based on the biochemistry of the inflammatory response that may lead to an ideal agent for infection imaging. Infectious diseases timely and specifically diagnosed can be clinically challenging but essential for the patient's recovery. Laboratory tests can detect the responsible microorganism but cannot discriminate between sterile inflammatory disease and truly infectious disease. On the other hand, scintigraphic images, can pinpoint the infection in the body. ***METHODS***: Bacteriophages (phages) are viruses that infect specific bacterial strains. Given the composition of the protein capsid, they could be used as

radiopharmaceuticals to diagnose bacterial infection. In this case, PP7 phage was labelled and evaluated as a specific tracer for Pseudomonas aeruginosa infections. 99mTc-Phage synthesis used HYNIC as a bifunctional agent. Physicochemical evaluation included studies such as stability in time, ligand exchange, lipophilicity and bacterial binding assay. Three groups of animals namely; healthy, infected with Pseudomonas aeruginosa and induced sterile inflammation were used to conduct biological evaluation ***RESULTS***: The radiolabelling process required size exclusion purification of the 99mTc-Phage, which was obtained with a radiochemical purity higher than 90%, during 18 hours post labelling. The collective accumulation in the stomach, small intestine and large intestine and thyroid of 99mTc-Phage was negligible, indicating no in vivo reoxidation. The complex presented urinary elimination. Target/ non-target ratio (T/NT) was determined both for sterile inflammation and for infection. Values were 2.5 +/- 0.4 and 4.2 +/- 0.3 respectively. These values indicate significant differences between sterile inflammation and infection by Pseudomonas aeruginosa ($p<0.05$ unpaired two sided t-test). ***CONCLUSION***: Targeted biodistribution profile and good T/NT ratios, indicate that this complex presents enough specificity to discriminate between infection caused by Pseudomonas aeruginosa and sterile inflammation.

Carpena, N., Manning, K. A., Dokland, T., Marina, A., and Penades, J. R. Convergent evolution of pathogenicity islands in helper cos phage interference. Philos.Trans.R.Soc.Lond B Biol.Sci. 11-5-2016; 371(1707); PM: 27672154

ABSTRACT: Staphylococcus aureus pathogenicity islands (SaPIs) are phage satellites that exploit the life cycle of their helper phages for their own benefit. Most SaPIs are packaged by their helper phages using a headful (pac) packaging mechanism. These SaPIs interfere with pac phage reproduction through a variety of strategies, including the redirection of phage capsid assembly to form small capsids, a process that depends on the expression of the SaPI-encoded cpmA and cpmB genes. Another SaPI subfamily is induced and packaged by cos-type phages, and although these cos SaPIs also block the life cycle of their inducing phages, the basis for this mechanism of interference remains to be deciphered. Here we have identified and characterized one mechanism by which the SaPIs interfere with cos phage reproduction. This mechanism depends on a SaPI-encoded gene, ccm, which encodes a protein involved in the production of small isometric capsids, compared with the prolate helper phage capsids. As the Ccm and CpmAB proteins are completely unrelated in sequence, this strategy represents a fascinating example of convergent evolution. Moreover, this result also indicates that the production of SaPI-sized particles is a widespread strategy of phage interference conserved during SaPI evolution.This article is part of the themed issue 'The new bacteriology'.

Castillo, D. and Middelboe, M. Genomic diversity of bacteriophages infecting the fish pathogen Flavobacterium psychrophilum. FEMS Microbiol.Lett. 12-2-2016; PM: 27915247

ABSTRACT: Bacteriophages infecting the fish pathogen Flavobacterium psychrophilum can potentially be used to prevent and control outbreaks of this bacterium in salmonid aquaculture. However, application of bacteriophages in disease control requires detailed knowledge on their genetic composition. To explore the diversity of F. pyschrophilum bacteriophages we have analyzed the complete genome sequences of 17 phages isolated from two distant geographic areas (Denmark and Chile), including the previously characterized temperate bacteriophage 6H. Phage genome size ranged

from 39302 to 89010 bp with a G+C content of 27 to 32%. None of the bacteriophages isolated in Denmark contained genes associated with lysogeny, whereas the Chilean isolates were all putative temperate phages and similar to bacteriophage 6H. Comparative genome analysis showed that phages grouped in three different genetic clusters based on genetic composition and gene content, indicating a limited genetic diversity of F. psychrophilum-specific bacteriophages. However, amino acid sequence dissimilarity (25%) was found in putative structural proteins, which could be related to the host specificity determinants. This study represents the first analysis of genomic diversity and composition among bacteriophages infecting the fish pathogen F. psychrophilum and discusses the implications for the application of phages in disease control.

Cech, G. M., Szalewska-Palasz, A., Kubiak, K., Malabirade, A., Grange, W., Arluison, V., and Wegrzyn, G. The Escherichia Coli Hfq Protein: An Unattended DNA-Transactions Regulator. Front Mol.Biosci. 2016; 3:36; PM: 27517037

ABSTRACT: The Hfq protein was discovered in Escherichia coli as a host factor for bacteriophage Qbeta RNA replication. Subsequent studies indicated that Hfq is a pleiotropic regulator of bacterial gene expression. The regulatory role of Hfq is ascribed mainly to its function as an RNA-chaperone, facilitating interactions between bacterial non-coding RNA and its mRNA target. Thus, it modulates mRNA translation and stability. Nevertheless, Hfq is able to interact with DNA as well. Its role in the regulation of DNA-related processes has been demonstrated. In this mini-review, it is discussed how Hfq interacts with DNA and what is the role of this protein in regulation of DNA transactions. Particularly, Hfq has been demonstrated to be involved in the control of ColE1 plasmid DNA replication, transposition, and possibly also transcription. Possible mechanisms of these Hfq-mediated regulations are described and discussed.

Chan, B. K., Brown, K., Kortright, K. E., Mao, S., and Turner, P. E. Extending the lifetime of antibiotics: how can phage therapy help? Future.Microbiol. 2016; 11:1105-1107; PM: 27545690

No Abstract Available.

Cheng, R., Li, G., Shi, L., Xue, X., Kang, M., and Zheng, X. The mechanism for bacteriophage f2 removal by nanoscale zero-valent iron. Water Res. 11-15-2016; 105:429-435; PM: 27665430

ABSTRACT: Nanoscale zero-valent iron (NZVI) has shown excellent performance for pathogenic microorganism removal but the inactivation mechanism has not been understood clearly enough. In this study, the bacteriophage f2 removal by NZVI under aerobic and anaerobic conditions was investigated, and various factors involved in f2 removal were analyzed in detail, including the ion products of NZVI (Fe(II), Fe(III)), solid phase products, the reactive oxygen species (ROS), O2 and H+. In addition, the morphologies of bacteriophage f2 during reaction were observed. The results showed that the removal efficiency of bacteriophage f2 was much higher under aerobic conditions than that in anaerobic systems, and oxygen and pH were determinants for f2 removal. The oxidation of Fe(II) was a fundamental step and played a significant role in bacteriophage f2 removal, especially in the aerobic systems. In the presence of oxygen, the virus removal was attributed to the generation of ROS (namely .OH and .O2-) and the

oxidized iron, in which the ROS (.OH and .O2-) made a predominant contribution. And the adsorption of iron oxide was responsible for the removal in oxygen depleted circumstance. In the anaerobic system, the virus removal was mainly attributed to the interaction between NZVI and bacteriophage f2. Besides, from the perspective of TEM images, the virus removal was mainly attributed to the damage of infective ability by NZVI at the initial stage of reaction, and later the virus was inactivated by the ROS generated.

Cheng, S., Xing, S., Zhang, X., Pei, G., An, X., Mi, Z., Huang, Y., and Tong, Y. Complete Genome Sequence of a New Enterococcus faecalis Bacteriophage, vB_EfaS_IME197. Genome Announc. 9-15-2016; 4(5); PM: 27634987

ABSTRACT: We report here the whole-genome sequence of a new Enterococcus faecalis phage, vB_EfaS_IME197, which has a linear double-stranded DNA genome of 41,307 bp with 34% G+C content. We describe the main features of the genome of vB_EfaS_IME197.

Cinek, O., Kramna, L., Lin, J., Oikarinen, S., Kolarova, K., Ilonen, J., Simell, O., Veijola, R., Autio, R., and Hyoty, H. Imbalance of bacteriome profiles within the Finnish Diabetes Prediction and Prevention study: Parallel use of 16S profiling and virome sequencing in stool samples from children with islet autoimmunity and matched controls. Pediatr.Diabetes. 11-17-2016; PM: 27860030

ABSTRACT: ***BACKGROUND***: We set out to explore associations between the stool bacteriome profiles and early-onset islet autoimmunity, taking into account the interactions with the virus component of the microbiome. ***METHODS***: Serial stool samples were longitudinally collected from 18 infants and toddlers with early-onset islet autoimmunity (median age 17.4 months) followed by type 1 diabetes, and 18 tightly matched controls from the Finnish Diabetes Prediction and Prevention (DIPP) cohort. Three stool samples were analyzed, taken 3, 6, and 9 months before the first detection of serum autoantibodies in the case child. The risk of islet autoimmunity was evaluated in relation to the composition of the bacteriome 16S rDNA profiles assessed by mass sequencing, and to the composition of DNA and RNA viromes. ***RESULTS***: Four operational taxonomic units were significantly less abundant in children who later on developed islet autoimmunity as compared to controls-most markedly the species of Bacteroides vulgatus and Bifidobacterium bifidum. The alpha or beta diversity, or the taxonomic levels of bacterial phyla, classes or genera, showed no differences between cases and controls. A correlation analysis suggested a possible relation between CrAssphage signals and quantities of Bacteroides dorei. No apparent associations were seen between development of islet autoimmunity and sequences of yet unknown origin. ***CONCLUSIONS***: The results confirm previous findings that an imbalance within the prevalent Bacteroides genus is associated with islet autoimmunity. The detected quantitative relation of the novel "orphan" bacteriophage CrAssphage with a prevalent species of the Bacteroides genus may exemplify possible modifiers of the bacteriome.

Cisek, A. A., Dabrowska, I., Gregorczyk, K. P., and Wyzewski, Z. Phage Therapy in Bacterial Infections Treatment: One Hundred Years After the Discovery of Bacteriophages. Curr.Microbiol. 11-28-2016; PM: 27896482

ABSTRACT: The therapeutic use of bacteriophages has seen a renewal of interest blossom in the last few years. This reversion is due to increased difficulties in the treatment

of antibiotic-resistant strains of bacteria. Bacterial resistance to antibiotics, a serious problem in contemporary medicine, does not implicate resistance to phage lysis mechanisms. Lytic bacteriophages are able to kill antibiotic-resistant bacteria at the end of the phage infection cycle. Thus, the development of phage therapy is potentially a way to improve the treatment of bacterial infections. However, there are antibacterial phage therapy difficulties specified by broadening the knowledge of the phage nature and influence on the host. It has been shown during experiments that both innate and adaptive immunity are involved in the clearance of phages from the body. Immunological reactions against phages are related to the route of administration and may vary depending on the type of bacterial viruses. For that reason, it is very important to test the immunological response of every single phage, particularly if intravenous therapy is being considered. The lack of these data in previous years was one of the reasons for phage therapy abandonment despite its century-long study. Promising results of recent research led us to look forward to a phage therapy that can be applied on a larger scale and subsequently put it into practice.

Citi, S. and Berg, D. E. Grete Kellenberger-Gujer: Molecular biology research pioneer. Bacteriophage. 2016; 6(2):e1148805; PM: 27607140

ABSTRACT: Grete Kellenberger-Gujer was a Swiss molecular biologist who pioneered fundamental studies of bacteriophage in the mid-20(th) century at the University of Geneva. Her life and career stories are reviewed here, focusing on her fundamental contributions to our early understanding of phage biology via her insightful analyses of phenomena such as the lysogenic state of a temperate phage (lambda), genetic recombination, radiation's in vivo consequences, and DNA restriction-modification; on her creative personality and interactions with peers; and how her academic advancement was affected by gender, societal conditions and cultural attitudes of the time. Her story is important scientifically, putting into perspective features of the scientific community from just before the molecular biology era started through its early years, and also sociologically, in illustrating the numerous "glass ceilings" that, especially then, often hampered the advancement of creative women.

Connolly, J. P. and Roe, A. J. Intracellular d-Serine Accumulation Promotes Genetic Diversity via Modulated Induction of RecA in Enterohemorrhagic Escherichia coli. J.Bacteriol. 12-15-2016; 198(24):3318-3328; PM: 27698085

ABSTRACT: We recently discovered that exposure of enterohemorrhagic Escherichia coli (EHEC) to d-serine resulted in accumulation of this unusual amino acid, induction of the SOS regulon, and downregulation of the type III secretion system that is essential for efficient colonization of the host. Here, we have investigated the physiological relevance of this elevated SOS response, which is of particular interest given the presence of Stx toxin-carrying lysogenic prophages on the EHEC chromosome that are activated during the SOS response. We found that RecA elevation in response to d-serine, while being significant, was heterogeneous and not capable of activating stx expression or stx phage transduction to a nonlysogenic recipient. This "SOS-like response" was, however, capable of increasing the mutation frequency associated with low-level RecA activity, thus promoting genetic diversity. Furthermore, this response was entirely dependent on RecA and enhanced in the presence of a DNA-damaging agent, indicating a functional SOS response, but did not result in observable cleavage of the LexA repressor alone, indicating a controlled mechanism of induction. This work demonstrates that

environmental factors not usually associated with DNA damage are capable of promoting an SOS-like response. We propose that this modulated induction of RecA allows EHEC to adapt to environmental insults such as d-serine while avoiding unwanted phage-induced lysis. *IMPORTANCE*: The SOS response is a global stress network that is triggered by the presence of DNA damage due to breakage or stalled replication forks. Activation of the SOS response can trigger the replication of lytic bacteriophages and promote genetic diversification through error-prone polymerases. We have demonstrated that the host-associated metabolite d-serine contributes to Escherichia coli niche specification and accumulates inside cells that cannot catabolize it. This results in a modulated activation of the SOS antirepressor RecA that is insufficient to trigger lytic bacteriophage but capable of increasing the SOS-associated mutation frequency. These findings describe how relevant signals not normally associated with DNA damage can hijack the SOS response, promoting diversity as E. coli strains adapt while avoiding unwanted phage lysis.

Cooper, C. J., Khan, Mirzaei M., and Nilsson, A. S. Adapting Drug Approval Pathways for Bacteriophage-Based Therapeutics. Front Microbiol. 2016; 7:1209; PM: 27536293

ABSTRACT: The global rise of multi-drug resistant bacteria has resulted in the notion that an "antibiotic apocalypse" is fast approaching. This has led to a number of well publicized calls for global funding initiatives to develop new antibacterial agents. The long clinical history of phage therapy in Eastern Europe, combined with more recent in vitro and in vivo success, demonstrates the potential for whole phage or phage based antibacterial agents. To date, no whole phage or phage derived products are approved for human therapeutic use in the EU or USA. There are at least three reasons for this: (i) phages possess different biological, physical, and pharmacological properties compared to conventional antibiotics. Phages need to replicate in order to achieve a viable antibacterial effect, resulting in complex pharmacodynamics/pharmacokinetics. (ii) The specificity of individual phages requires multiple phages to treat single species infections, often as part of complex cocktails. (iii) The current approval process for antibacterial agents has evolved with the development of chemically based drugs at its core, and is not suitable for phages. Due to similarities with conventional antibiotics, phage derived products such as endolysins are suitable for approval under current processes as biological therapeutic proteins. These criteria render the approval of phages for clinical use theoretically possible but not economically viable. In this review, pitfalls of the current approval process will be discussed for whole phage and phage derived products, in addition to the utilization of alternative approval pathways including adaptive licensing and "Right to try" legislation.

Cornell, J. L., Breslin, E., Schuhmacher, Z., Himelright, M., Berluti, C., Boyd, C., Carson, R., Del, Gallo E., Giessler, C., Gilliam, B., Heatherly, C., Nevin, J., Nguyen, B., Nguyen, J., Parada, J., Sutterfield, B., Tukruni, M., and Temple, L. Complete Genome Sequence of Bacillus thuringiensis Bacteriophage Smudge. Genome Announc. 8-18-2016; 4(4); PM: 27540049

ABSTRACT: Smudge, a bacteriophage enriched from soil using Bacillus thuringiensis DSM-350 as the host, had its complete genome sequenced. Smudge is a myovirus with a genome consisting of 292 genes and was identified as belonging to the C1 cluster of Bacillus phages.

Cruz-Flores, R., Caceres-Martinez, J., Munoz-Flores, M., Vasquez-Yeomans, R., Hernandez, Rodriguez M., Angel, Del Rio-Portilla, Rocha-Olivares, A., and Castro-Longoria, E. Hyperparasitism by the bacteriophage (Caudovirales) infecting Candidatus Xenohaliotis californiensis (Rickettsiales-like prokaryote) parasite of wild abalone Haliotis fulgens and Haliotis corrugata from the Peninsula of Baja California, Mexico. J.Invertebr.Pathol. 2016; 140:58-67; PM: 27623402

ABSTRACT: Candidatus Xenohaliotis californiensis (CXc) is a Rickettsiales-like prokaryote that is considered the causal agent of Withering Syndrome (WS), a chronic disease of abalone, from the west coast of North America and it is listed by the International Organization for Animal Health (OIE) as a reportable agent due to its pathogenicity. This bacterium in red abalone Haliotis rufescens, black abalone Haliotis cracherodii, and yellow abalone Haliotis corrugata from California, US and Baja California, Mexico has been found to be infected by a bacteriophage. To date, there is no information on the epizootiology of CXc and its bacteriophage in natural populations of abalone; furthermore, it is unknown if the bacteriophage was also present in CXc infecting blue abalone Haliotis fulgens. The objective of this study was to determine the distribution, prevalence and intensity of CXc, as well as to determine the distribution and prevalence of the bacteriophage and to study interactions between host sex and hyperparasitism in blue abalone and yellow abalone. Tissue samples were obtained from seven localities where the commercial capture of wild abalone is carried out. Samplings were conducted throughout the 2012-2013 capture seasons and a total of 182 blue abalone and 170 yellow abalone were obtained. The prevalence and intensity of CXc and the prevalence of the bacteriophage were determined by histology. The identity of CXc was confirmed by PCR, product sequence analysis and in situ hybridization while the identity of the bacteriophage was corroborated by TEM. The prevalence of CXc infected and uninfected by the bacteriophage was 80% in blue abalone and 62% in yellow abalone. Low infection intensities were found in 86% of blue abalone and 82% of yellow abalone. Infection intensity was significantly higher in undifferentiated yellow abalone. The bacteriophage in CXc showed a prevalence of 22% and 31% in blue abalone and yellow abalone respectively. These results show that CXc and its bacteriophage are widely distributed in the peninsula of Baja California and that they are well established in natural populations of blue abalone and yellow abalone. Additionally, this data constitutes the first record of a bacteriophage in blue abalone.

Das, A. and Biswas, M. Changes in the Functional Activity of Phi11 Cro Protein is Mediated by Various Ions. Protein J. 2016; 35(6):407-415; PM: 27812887

ABSTRACT: Phi11, a temperate bacteriophage of Staphylococcus aureus, has been found to harbor a cro repressor gene which facilitates Phi11 to adopt the lytic mode of development. The Cro protein has been found to bind very specifically to a 15-bp operator DNA, located in the Phi11 cI-cro intergenic region [1]. To investigate the effects exerted by different ions upon the interaction between Cro and its cognate operator DNA, we have employed gel shift assays as well as circular dichroism spectral analysis. In this communication, we have shown that NH4+ and acetate- ions better facilitated the binding of Cro with its cognate operator as compared to Na+, K+ and Li+. Interestingly, Mg2+, carbonate2- and Citrate3- have an inhibitory effect upon the binding. The effect of the said ions upon the structure of Cro was also investigated by circular dichroism and it was found that other than Citrate3- ions, none of the other ions destabilised the protein. On the other hand, Mg2+ and carbonate2- ions maintained the structure of the protein but severely hampered its functional activity. Citrate3- ions severely unfolded Cro and

also inhibited its function. Considering all the data, NH4+ and acetate- ions appeared to be more suitable in maintaining the biological activity of Cro.

Davison, M., Treangen, T. J., Koren, S., Pop, M., and Bhaya, D. Diversity in a Polymicrobial Community Revealed by Analysis of Viromes, Endolysins and CRISPR Spacers. PLoS.One. 2016; 11(9):e0160574; PM: 27611571

ABSTRACT: The polymicrobial biofilm communities in Mushroom and Octopus Spring in Yellowstone National Park (YNP) are well characterized, yet little is known about the phage populations. Dominant species, Synechococcus sp. JA-2-3B'a(2-13), Synechococcus sp. JA-3-3Ab, Chloroflexus sp. Y-400-fl, and Roseiflexus sp. RS-1, contain multiple CRISPR-Cas arrays, suggesting complex interactions with phage predators. To analyze phage populations from Octopus Spring biofilms, we sequenced a viral enriched fraction. To assemble and analyze phage metagenomic data, we developed a custom module, VIRITAS, implemented within the MetAMOS framework. This module bins contigs into groups based on tetranucleotide frequencies and CRISPR spacer-protospacer matching and ORF calling. Using this pipeline we were able to assemble phage sequences into contigs and bin them into three clusters that corroborated with their potential host range. The virome contained 52,348 predicted ORFs; some were clearly phage-like; 9319 ORFs had a recognizable Pfam domain while the rest were hypothetical. Of the recognized domains with CRISPR spacer matches, was the phage endolysin used by lytic phage to disrupt cells. Analysis of the endolysins present in the thermophilic cyanophage contigs revealed a subset of characterized endolysins as well as a Glyco_hydro_108 (PF05838) domain not previously associated with sequenced cyanophages. A search for CRISPR spacer matches to all identified phage endolysins demonstrated that a majority of endolysin domains were targets. This strategy provides a general way to link host and phage as endolysins are known to be widely distributed in bacteriophage. Endolysins can also provide information about host cell wall composition and have the additional potential to be used as targets for novel therapeutics.

De, S., Bhattacharyya, S., and Gopmandal, P. P. Importance of core electrostatic properties on the electrophoresis of a soft particle. Phys.Rev.E. 2016; 94(2-1):022611; PM: 27627364

ABSTRACT: The impact of the volumetric charged density of the dielectric rigid core on the electrophoresis of a soft particle is analyzed numerically. The volume charge density of the inner core of a soft particle can arise for a dendrimer structure or bacteriophage MS2. We consider the electrokinetic model based on the conservation principles, thus no conditions for Debye length or applied electric field is imposed. The fluid flow equations are coupled with the ion transport equations and the equation for the electric field. The occurrence of the induced nonuniform surface charge density on the outer surface of the inner core leads to a situation different from the existing analysis of a soft particle electrophoresis. The impact of this induced surface charge density together with the double-layer polarization and relaxation due to ion convection and electromigration is analyzed. The dielectric permittivity and the charge density of the core have a significant impact on the particle electrophoresis when the Debye length is in the order of the particle size. We find that by varying the ionic concentration of the electrolyte, the particle can exhibit reversal in its electrophoretic velocity. The role of the polymer layer softness parameter is addressed in the present analysis.

Descamps, E. C., Meunier, D., Brutesco, C., Preveral, S., Franche, N., Bazin, I., Miclot, B., Larosa, P., Escoffier, C., Fantino, J. R., Garcia, D., Ansaldi, M., Rodrigue, A., Pignol, D., Cholat, P., and Ginet, N. Semi-autonomous inline water analyzer: design of a common light detector for bacterial, phage, and immunological biosensors. Environ.Sci.Pollut.Res.Int. 11-12-2016; PM: 27838908

ABSTRACT: The use of biosensors as sensitive and rapid alert systems is a promising perspective to monitor accidental or intentional environmental pollution, but their implementation in the field is limited by the lack of adapted inline water monitoring devices. We describe here the design and initial qualification of an analyzer prototype able to accommodate three types of biosensors based on entirely different methodologies (immunological, whole-cell, and bacteriophage biosensors), but whose responses rely on the emission of light. We developed a custom light detector and a reaction chamber compatible with the specificities of the three systems and resulting in statutory detection limits. The water analyzer prototype resulting from the COMBITOX project can be situated at level 4 on the Technology Readiness Level (TRL) scale and this technical advance paves the way to the use of biosensors on-site.

Dobroff, A. S., D'Angelo, S., Eckhardt, B. L., Ferrara, F., Staquicini, D. I., Cardo-Vila, M., Staquicini, F. I., Nunes, D. N., Kim, K., Driessen, W. H., Hajitou, A., Lomo, L. C., Barry, M., Krishnamurthy, S., Sahin, A., Woodward, W. A., Prossnitz, E. R., Anderson, R. L., as-Neto, E., Brown-Glaberman, U. A., Royce, M. E., Ueno, N. T., Cristofanilli, M., Hortobagyi, G. N., Marchio, S., Gelovani, J. G., Sidman, R. L., Arap, W., and Pasqualini, R. Towards a transcriptome-based theranostic platform for unfavorable breast cancer phenotypes. Proc.Natl.Acad.Sci.U.S.A. 10-24-2016; PM: 27791177

ABSTRACT: Inflammatory breast carcinoma (IBC) is one of the most lethal forms of human breast cancer, and effective treatment for IBC is an unmet clinical need in contemporary oncology. Tumor-targeted theranostic approaches are emerging in precision medicine, but only a few specific biomarkers are available. Here we report up-regulation of the 78-kDa glucose-regulated protein (GRP78) in two independent discovery and validation sets of specimens derived from IBC patients, suggesting translational promise for clinical applications. We show that a GRP78-binding motif displayed on either bacteriophage or adeno-associated virus/phage (AAVP) particles or loop-grafted onto a human antibody fragment specifically targets orthotopic IBC and other aggressive breast cancer models in vivo. To evaluate the theranostic value, we used GRP78-targeting AAVP particles to deliver the human Herpes simplex virus thymidine kinase type-1 (HSVtk) transgene, obtaining simultaneous in vivo diagnosis through PET imaging and tumor treatment by selective activation of the prodrug ganciclovir at tumor sites. Translation of this AAVP system is expected simultaneously to image, monitor, and treat the IBC phenotype and possibly other aggressive (e.g., invasive and/or metastatic) subtypes of breast cancer, based on the inducible cell-surface expression of the stress-response chaperone GRP78, and possibly other cell-surface receptors in human tumors.

Dong, S., Shi, H., Cao, D., Wang, Y., Zhang, X., Li, Y., Gao, X., and Wang, L. Novel nanoscale bacteriophage-based single-domain antibodies for the therapy of systemic infection caused by Candida albicans. Sci.Rep. 8-25-2016; 6:32256; PM: 27558409

ABSTRACT: Candida albicans (C. albicans) is an important human commensal and opportunistic fungal pathogen. Secreted aspartyl proteinases (Saps) are a major virulence trait of C. albicans, and among these proteases Sap2 has the highest expression levels. It is possible that antibodies against Sap2 could provide an antifungal effect. In this study, two phages displaying anti-rSap2 single chain variable fragments (scFvs) were screened from human single fold scFv libraries, and their potential therapeutic roles were evaluated using a murine model infected by C. albicans. The in vivo efficacies were assessed by mortality rates, fungal burden and histological examination. Overall survival rates were significantly increased while the colony counts and infectious foci were significantly decreased after treatment with the scFv-phages relative to the control groups. In order to investigate the immune response provoked by scFv-phages, three kinds of cytokines (Th1, Th2 and Th17 types) were measured and a clear immune response was observed. These findings suggest that anti-rSap2 scFv-phages have potential in the therapy of systemic infection caused by C. albicans.

Doore, S. M., Schweers, N. J., and Fane, B. A. Elevating fitness after a horizontal gene exchange in bacteriophage phiX174. Virology. 11-14-2016; 501:25-34; PM: 27855283

ABSTRACT: In an earlier study, protein-based barriers to horizontal gene transfer were investigated by placing the bacteriophage G4 G gene, encoding the major spike protein, into the phiX174 genome. The foreign G protein promoted off-pathway assembly reactions, resulting in a lethal phenotype. After three targeted genetic selections, one of two foreign spike proteins was productively integrated into the phiX174 system: the complete G4 or a recombinant G4/phiX174 protein (94% G4:6% phiX174). However, strain fitness was very low. In this study, the chimeras were characterized and experimentally evolved. Inefficient assembly was the primary contributor to low fitness: accordingly, mutations affecting assembly restored fitness. The spike protein preference of the ancestral and evolved strains was determined in competition experiments between the foreign and phiX174G proteins. Before adaptation, both G proteins were incorporated into virions; afterwards, the foreign proteins were strongly preferred. Thus, a previously inhibitory protein became the preferred substrate during assembly.

Duderstadt, K. E., Geertsema, H. J., Stratmann, S. A., Punter, C. M., Kulczyk, A. W., Richardson, C. C., and van Oijen, A. M. Simultaneous Real-Time Imaging of Leading and Lagging Strand Synthesis Reveals the Coordination Dynamics of Single Replisomes. Mol.Cell. 11-15-2016; PM: 27889453

ABSTRACT: The molecular machinery responsible for DNA replication, the replisome, must efficiently coordinate DNA unwinding with priming and synthesis to complete duplication of both strands. Due to the anti-parallel nature of DNA, the leading strand is copied continuously, while the lagging strand is produced by repeated cycles of priming, DNA looping, and Okazaki-fragment synthesis. Here, we report a multidimensional single-molecule approach to visualize this coordination in the bacteriophage T7 replisome by simultaneously monitoring the kinetics of loop growth and leading-strand synthesis. We show that loops in the lagging strand predominantly occur during priming and only infrequently support subsequent Okazaki-fragment synthesis. Fluorescence imaging

reveals polymerases remaining bound to the lagging strand behind the replication fork, consistent with Okazaki-fragment synthesis behind and independent of the replication complex. Individual replisomes display both looping and pausing during priming, reconciling divergent models for the regulation of primer synthesis and revealing an underlying plasticity in replisome operation.

Duerkop, B. A., Huo, W., Bhardwaj, P., Palmer, K. L., and Hooper, L. V. Molecular Basis for Lytic Bacteriophage Resistance in Enterococci. MBio. 8-30-2016; 7(4); PM: 27578757

ABSTRACT: The human intestine harbors diverse communities of bacteria and bacteriophages. Given the specificity of phages for their bacterial hosts, there is growing interest in using phage therapies to combat the rising incidence of multidrug-resistant bacterial infections. A significant barrier to such therapies is the rapid development of phage-resistant bacteria, highlighting the need to understand how bacteria acquire phage resistance in vivo Here we identify novel lytic phages in municipal raw sewage that kill Enterococcus faecalis, a Gram-positive opportunistic pathogen that resides in the human intestine. We show that phage infection of E. faecalis requires a predicted integral membrane protein that we have named PIPEF (for phage infection protein from E. faecalis). We find that PIPEF is conserved in E. faecalis and harbors a 160-amino-acid hypervariable region that determines phage tropism for distinct enterococcal strains. Finally, we use a gnotobiotic mouse model of in vivo phage predation to show that the sewage phages temporarily reduce E. faecalis colonization of the intestine but that E. faecalis acquires phage resistance through mutations in PIPEF Our findings define the molecular basis for an evolutionary arms race between E. faecalis and the lytic phages that prey on them. They also suggest approaches for engineering E. faecalis phages that have altered host specificity and that can subvert phage resistance in the host bacteria. *IMPORTANCE*: Bacteriophage therapy has received renewed attention as a potential solution to the rise in antibiotic-resistant bacterial infections. However, bacteria can acquire phage resistance, posing a major barrier to phage therapy. To overcome this problem, it is necessary to understand phage resistance mechanisms in bacteria. We have unraveled one such resistance mechanism in Enterococcus faecalis, a Gram-positive natural resident of the human intestine that has acquired antibiotic resistance and can cause opportunistic infections. We have identified a cell wall protein hypervariable region that specifies phage tropism in E. faecalis Using a gnotobiotic mouse model of in vivo phage predation, we show that E. faecalis acquires phage resistance through mutations in this cell wall protein. Our findings define the molecular basis for lytic phage resistance in E. faecalis. They also suggest opportunities for engineering E. faecalis phages that circumvent the problem of bacterial phage resistance.

Duncan, A. B., Dusi, E., Jacob, F., Ramsayer, J., Hochberg, M. E., and Kaltz, O. Hot spots become cold spots: coevolution in variable temperature environments. J.Evol.Biol. 10-6-2016; PM: 27711983

ABSTRACT: Antagonistic coevolution between hosts and parasites is a key process in the genesis and maintenance of biological diversity. Whereas coevolutionary dynamics show distinct patterns under favourable environmental conditions, the effects of more realistic, variable conditions are largely unknown. We investigated the impact of a fluctuating environment on antagonistic coevolution in experimental microcosms of Pseudomonas fluorescens SBW25 and lytic phage SBWPhi2. High-frequency temperature

fluctuations caused no deviations from typical coevolutionary arms race dynamics. However, coevolution was stalled during periods of high temperature under intermediate- and low-frequency fluctuations, generating temporary coevolutionary cold spots. Temperature variation affected population density, providing evidence that eco-evolutionary feedbacks act through variable bacteria-phage encounter rates. Our study shows that environmental fluctuations can drive antagonistic species interactions into and out of coevolutionary cold and hot spots. Whether coevolution persists or stalls depends on the frequency of change and the environmental optima of both interacting players.

Eggers, C. H., Gray, C. M., Preisig, A. M., Glenn, D. M., Pereira, J., Ayers, R. W., Alshahrani, M., Acabbo, C., Becker, M. R., Bruenn, K. N., Cheung, T., Jendras, T. M., Shepley, A. B., and Moeller, J. T. Phage-mediated horizontal gene transfer of both prophage and heterologous DNA by varphiBB-1, a bacteriophage of Borrelia burgdorferi. Pathog.Dis. 11-2-2016; PM: 27811049

ABSTRACT: Horizontal gene transfer (HGT) in Borrelia burgdorferi, the Lyme disease agent, is likely mediated by bacteriophage. Studies of the B. burgdorferi phage, varphiBB-1, and its role in HGT have been hindered by the lack of an assay for readily characterizing phage-mediated DNA movement (transduction). Here we describe an in vitro assay in which a clone of B. burgdorferi strain CA-11.2A encoding kanamycin resistance on a varphiBB-1 prophage is co-cultured with different clones encoding gentamicin resistance on a shuttle vector; transduction is monitored by enumerating colonies selected in the presence of both kanamycin and gentamicin. When both clones used in the assay were derived from CA-11.2A, the frequency of transduction was 1.23 x 10-6 transductants per cell, and could be increased fivefold by exposing the phage-producing strain to 5% ethanol. Transduction also was demonstrated between the CA-11.2A clone and clones of both high-passage B. burgdorferi strain B31 and low-passage, virulent B. burgdorferi strain 297, although with lower transduction frequencies. The transductant in the 297 background produced phage capable of transducing another B. burgdorferi clone: this is the first experimental demonstration of transduction from a clone of a virulent strain. In addition to prophage DNA, small E. coli-derived shuttle vectors also were transduced between co-cultured B. burgdorferi strains, suggesting both a broad role for the phage in the HGT of heterologous DNA and a potential use of the phage as a molecular tool. These results enhance our understanding of phage-mediated transduction as a mechanism of HGT in the Lyme disease spirochetes. Furthermore, the reagents and techniques developed herein will facilitate future studies of phage-mediated HGT, especially within the tick vector and vertebrate host.

Eggleston, E. M. and Hewson, I. Abundance of Two Pelagibacter ubique Bacteriophage Genotypes along a Latitudinal Transect in the North and South Atlantic Oceans. Front Microbiol. 2016; 7:1534; PM: 27733846

ABSTRACT: This study characterizes viral and bacterial dynamics along a latitudinal transect in the Atlantic Ocean from approximately 10 N-40 S. Overall viral abundance decreased with depth, on average there were 1.64 +/- 0.71 x 107 virus like particles (VLPs) in surface waters, decreasing to an average of 6.50 +/- 2.26 x 105 VLPs in Antarctic Bottom Water. This decrease was highly correlated to bacterial abundance. There are six major water masses in the Southern Tropical Atlantic Ocean, and inclusion of water mass, temperature and salinity variables explained a majority of the variation in total viral

abundance. Recent discovery of phages infecting bacteria of the SAR11 clade of Alphaproteobacteria (i.e., pelagiphages) leads to intriguing questions about the roles they play in shaping epipelagic communities. Viral-size fraction DNA from epipelagic water was used to quantify the abundance of two pelagiphages, using pelagiphage-specific quantitative PCR primers and probes along the transect. We found that HTVC010P, a member of a podoviridae sub-family, was most abundant in surface waters. Copy numbers ranged from an average of 1.03 +/- 2.38 x 105 copies ml-1 in surface waters, to 5.79 +/- 2.86 x 103 in the deep chlorophyll maximum. HTVC008M, a T4-like myovirus, was present in the deep chlorophyll maximum (5.42 +/- 2.8 x 103 copies ml-1 on average), although it was not as highly abundant as HTVC010P in surface waters (6.05 +/- 3.01 x 103 copies ml-1 on average). Interestingly, HTVC008M was only present at a few of the most southern stations, suggesting latitudinal biogeography of SAR11 phages.

Elena, C. M., Leticia, F., Emilia, T., Patricia, E., and Mariella, T. Evaluation of a Labelled Bacteriophage with 99mTc as a Potential Agent for Infection Diagnosis. Curr.Radiopharm. 2-15-2016; PM: 26876366

ABSTRACT: The design of target-specific molecular imaging probes to determine infection sites are mainly based on the biochemistry of the inflammatory response that may lead to an ideal agent for infection imaging. Infectious diseases timely and specifically diagnosed can be clinically challenging but essential for the patient's recovery. Laboratory tests can detect the responsible microorganism but cannot discriminate between sterile inflammatory disease and truly infectious disease. On the other hand, scintigraphic images, can pinpoint the infection in the body. Bacteriophages (phages) are viruses that infect specific bacterial strains. Given the composition of the protein capsid, they could be used as radiopharmaceuticals to diagnose bacterial infection. In this case, PP7 phage was labelled and evaluated as a specific tracer for Pseudomonas aeruginosa infections.99mTc-Phage synthesis used HYNIC as a bifunctional agent. Physicochemical evaluation included studies such as stability in time, ligand exchange, lipophilicity and bacterial binding assay. The radiolabelling process required size exclusion purification of the 99mTc-Phage, which was obtained with a radiochemical purity higher than 90%, during 18 hours post labelling. Three groups of animals namely; healthy, infected with Pseudomonas aeruginosa and induced sterile inflammation were used to conduct biological evaluation. The collective accumulation in the stomach, small intestine and large intestine and thyroid of 99mTc-Phage was negligible, indicating no in vivo reoxidation. The complex presented urinary elimination. Target/non-target ratio (T/NT) was determined both for sterile inflammation and for infection. Values were 2.5 +/- 0.4 and 4.2 +/- 0.3 respectively. These values indicate significant differences between sterile inflammation and infection by Pseudomonas aeruginosa ($p<0.05$ unpaired two sided t-test). Targeted biodistribution profile and good T/NT ratios, indicate that this complex presents enough specificity to discriminate between infection caused by Pseudomonas aeruginosa and sterile inflammation.

Emmoth, E., Rovira, J., Rajkovic, A., Corcuera, E., Wilches, Perez D., Dergel, I., Ottoson, J. R., and Widen, F. Inactivation of Viruses and Bacteriophages as Models for Swine Hepatitis E Virus in Food Matrices. Food Environ.Virol. 10-25-2016; PM: 27783334

ABSTRACT: Hepatitis E virus has been recognised as a food-borne virus hazard in pork products, due to its zoonotic properties. This risk can be reduced by adequate treatment of the food to inactivate food-borne viruses. We used a spectrum of viruses and

bacteriophages to evaluate the effect of three food treatments: high pressure processing (HPP), lactic acid (LA) and intense light pulse (ILP) treatments. On swine liver at 400 MPa for 10 min, HPP gave log10 reductions of >/=4.2, >/=5.0 and 3.4 for feline calicivirus (FCV) 2280, FCV wildtype (wt) and murine norovirus 1 (MNV 1), respectively. Escherichia coli coliphage varphiX174 displayed a lower reduction of 1.1, while Escherichia coli coliphage MS2 was unaffected. For ham at 600 MPa, the corresponding reductions were 4.1, 4.4, 2.9, 1.7 and 1.3 log10. LA treatment at 2.2 M gave log10 reductions in the viral spectrum of 0.29-2.1 for swine liver and 0.87-3.1 for ham, with varphiX174 and MNV 1, respectively, as the most stable microorganisms. The ILP treatment gave log10 reductions of 1.6-2.8 for swine liver, 0.97-2.2 for ham and 1.3-2.3 for sausage, at 15-60 J cm-2, with MS2 as the most stable microorganism. The HPP treatment gave significantly ($p < 0.05$) greater virus reduction on swine liver than ham for the viruses at equivalent pressure/time combinations. For ILP treatment, reductions on swine liver were significantly ($p < 0.05$) greater than on ham for all microorganisms. The results presented here could be used in assessments of different strategies to protect consumers against virus contamination and in advice to food producers. Conservative model indicators for the pathogenic viruses could be suggested.

Erill, I. and Caruso, S. M. Genome Sequence of Bacillus cereus Group Phage SalinJah. Genome Announc. 9-29-2016; 4(5); PM: 27688335

ABSTRACT: The double-stranded DNA (dsDNA) Myoviridae Bacillus cereus group bacteriophage SalinJah was isolated from soil collected in Gyeonggi-do, South Korea. SalinJah, a cluster C phage with a broad host range, suggests the need to create a new subcluster with SalinJah and Helga as founding members.

Eskelin, K., Lampi, M., Meier, F., Moldenhauer, E., Bamford, D. H., and Oksanen, H. M. Asymmetric flow field flow fractionation methods for virus purification. J.Chromatogr.A. 10-21-2016; 1469:108-119; PM: 27697294

ABSTRACT: Detailed biochemical and biophysical characterization of viruses requires viral preparations of high quantity and purity. The optimization of virus production and purification is an essential, but laborious and time-consuming process. Asymmetric flow field flow fractionation (AF4) is an attractive alternative method for virus purification because it is a rapid and gentle separation method that should preserve viral infectivity. Here we optimized the AF4 conditions to be used for purification of a model virus, bacteriophage PRD1, from various types of starting materials. Our results show that AF4 is well suited for PRD1 purification as monitored by virus recovery and specific infectivity. Short analysis time and high sample loads enabled us to use AF4 for preparative scale purification of PRD1. Furthermore, we show that AF4 enables the rapid real-time analysis of progeny virus production in infected cells.

Estrella, L. A., Quinones, J., Henry, M., Hannah, R. M., Pope, R. K., Hamilton, T., Teneza-Mora, N., Hall, E., and Biswajit, B. Characterization of novel Staphylococcus aureus lytic phage and defining their combinatorial virulence using the OmniLog(R) system. Bacteriophage. 2016; 6(3):e1219440; PM: 27738555

ABSTRACT: Skin and soft tissue infections (SSTI) caused by methicillin resistant Staphylococcus aureus (MRSA) are difficult to treat. Bacteriophage (phage) represent a

potential alternate treatment for antibiotic resistant bacterial infections. In this study, 7 novel phage with broad lytic activity for S. aureus were isolated and identified. Screening of a diverse collection of 170 clinical isolates by efficiency of plating (EOP) assays shows that the novel phage are virulent and effectively prevent growth of 70-91% of MRSA and methicillin sensitive S. aureus (MSSA) isolates. Phage K, which was previously identified as having lytic activity on S. aureus was tested on the S. aureus collection and shown to prevent growth of 82% of the isolates. These novel phage group were examined by electron microscopy, the results of which indicate that the phage belong to the Myoviridae family of viruses. The novel phage group requires beta-N-acetyl glucosamine (GlcNac) moieties on cell wall teichoic acids for infection. The phage were distinct from, but closely related to, phage K as characterized by restriction endonuclease analysis. Furthermore, growth rate analysis via OmniLog(R) microplate assay indicates that a combination of phage K, with phage SA04201, SA04561 or SA04821 have a synergistic phage-mediated lytic effect on MRSA and suppress formation of phage resistance. These results indicate that a broad spectrum lytic phage mixture can suppress the emergence of resistant bacterial populations and hence have great potential for combating S. aureus wound infections.

Fang, P. Y., Gomez Ramos, L. M., Holguin, S. Y., Hsiao, C., Bowman, J. C., Yang, H. W., and Williams, L. D. Functional RNAs: combined assembly and packaging in VLPs. Nucleic Acids Res. 11-29-2016; PM: 27903913

ABSTRACT: We describe here a one pot RNA production, packaging and delivery system based on bacteriophage Qbeta. We demonstrate a method for production of a novel RNAi scaffold, packaged within Qbeta virus-like particles (VLPs). The RNAi scaffold is a general utility chimera that contains a functional RNA duplex with paired silencing and carrier sequences stabilized by a miR-30 stem-loop. The Qbeta hairpin on the 5' end confers affinity for the Qbeta coat protein (CP). Silencing sequences can include mature miRNAs and siRNAs, and can target essentially any desired mRNA. The VLP-RNAi assembles upon co-expression of CP and the RNAi scaffold in E. coli The annealing of the scaffold to form functional RNAs is intramolecular and is therefore robust and concentration independent. We demonstrate dose- and time-dependent inhibition of GFP expression in human cells with VLP-RNAi. In addition, we target the 3'UTR of oncogenic Ras mRNA and suppress Pan-Ras expression, which attenuates cell proliferation and promotes mortality of brain tumor cells. This combination of RNAi scaffold design with Qbeta VLP packaging is demonstrated to be target-specific and efficient.

Feng, Z., Lu, R., Yuan, B., Zhou, Z., Wu, Q., and Nguyen, T. H. Influence of solution chemistry on the inactivation of particle-associated viruses by UV irradiation. Colloids Surf.B Biointerfaces. 12-1-2016; 148:622-628; PM: 27694052

ABSTRACT: MS2 inactivation by UV irradiance was investigated with the focus on how the disinfection efficacy is influenced by bacteriophage MS2 aggregation and adsorption to particles in solutions with different compositions. Kaolinite and Microcystis aeruginosa were used as model inorganic and organic particles, respectively. In the absence of model particles, MS2 aggregates formed in either 1mM NaCl at pH=3 or 50-200mM ionic

strength CaCl2 solutions at pH=7 led to a decrease in the MS2 inactivation efficacy because the virions located inside the aggregate were protected from the UV irradiation. In the presence of kaolinite and Microcystis aeruginosa, MS2 adsorbed onto the particles in either 1mM NaCl at pH=3 or 50-200mM CaCl2 solutions at pH=7. In contrast to MS2 aggregates formed without the presence of particles, more MS2 virions adsorbed on these particles were exposed to UV irradiation to allow an increase in MS2 inactivation. In either 1mM NaCl at pH from 4 to 8 or 2-200mM NaCl solutions at pH=7, the absence of MS2 aggregation and adsorption onto the model particles explained why MS2 inactivation was not influenced by pH, ionic strength, and the presence of model particles in these conditions. The influence of virus adsorption and aggregation on the UV disinfection efficiency found in this research suggests the necessity of accounting for particles and cation composition in virus inactivation for drinking water.

Fernandes, S. and Sao-Jose, C. Probing the function of the two holin-like proteins of bacteriophage SPP1. Virology. 2017; 500:184-189; PM: 27825035

ABSTRACT: Double-stranded DNA bacteriophages employ holin and endolysin functions to lyse host bacteria after virus multiplication. Holins oligomerize in the cytoplasmic membrane and trigger to form holes that cause cell death. For most systems these holes are also required for endolysin release to the cell wall, where it cleaves the peptidoglycan network. Orfs 25 and 26 of Bacillus subtilis phage SPP1 were predicted to encode the endolysin and holin functions, respectively. However, the product of the upstream orf 24.1 exhibits also holin features. We show that production of gp24.1 or gp26 in B. subtilis causes no major impact on cell growth, despite their ability to insert in the cytoplasmic membrane. Instant growth cessation and cell death is observed only upon co-production of the two holin-like proteins. Surprisingly, a constitutive promoter was identified within orf 24.1, which we propose to correspond to the previously described SPP1 early promoter PE5.

Firlieyanti, A. S., Connerton, P. L., and Connerton, I. F. Campylobacters and their bacteriophages from chicken liver: The prospect for phage biocontrol. Int.J.Food Microbiol. 11-21-2016; 237:121-127; PM: 27565524

ABSTRACT: Consumption of foods containing chicken liver has been associated with Campylobacter enteritis. Campylobacters can contaminate the surface of livers post-mortem but can also arise through systemic infection of colonising bacteria in live birds. The use of bacteriophage to reduce levels of Campylobacter entering the food chain is a promising intervention approach but most phages have been isolated from chicken excreta. This study examined the incidence and contamination levels of Campylobacter and their bacteriophage in UK retail chicken liver. Using enrichment procedures, 87% of 109 chicken livers were surface contaminated with Campylobacter and 83% contaminated within internal tissues. Direct plating on selective agar allowed enumeration of viable bacteria from 43% of liver samples with counts ranging from 1.8->3.8log10CFU/cm2 for surface samples, and 3.0->3.8log10CFU/g for internal tissue samples. Three C. jejuni isolates recovered from internal liver tissues were assessed for their ability to colonise the intestines and extra-intestinal organs of broiler chickens following oral infection. All isolates efficiently colonised the chicken intestines but were variable in their abilities to colonise extra-intestinal organs. One isolate, CLB104, could be recovered by enrichment from the livers and kidneys of three of seven chickens. Campylobacter isolates remained viable within fresh livers stored at 4 degrees C over

72h and frozen livers stored at -20 degrees C over 7days in atmospheric oxygen, and therefore constitute a risk to human health. Only three Campylobacter-specific bacteriophages were isolated, and these exhibited a limited host range against the Camplylobacter chicken liver isolates. All were identified as group III virulent bacteriophage based on their genome size of 140kb. The application of broad host range group II virulent phages (8log10PFU/g) to liver homogenates containing C. jejuni strains of diverse origin at 4 degrees C resulted in modest but significant reductions in the viable counts ranging from 0.2 to 0.7log10CFU/g.

Fister, S., Robben, C., Witte, A. K., Schoder, D., Wagner, M., and Rossmanith, P. Influence of Environmental Factors on Phage-Bacteria Interaction and on the Efficacy and Infectivity of Phage P100. Front Microbiol. 2016; 7:1152; PM: 27516757

ABSTRACT: When using bacteriophages to control food-borne bacteria in food production plants and processed food, it is crucial to consider that environmental conditions influence their stability. These conditions can also affect the physiological state of bacteria and consequently host-virus interaction and the effectiveness of the phage ability to reduce bacteria numbers. In this study we investigated the stability, binding, and replication capability of phage P100 and its efficacy to control Listeria monocytogenes under conditions typically encountered in dairy plants. The influences of SDS, Lutensol AO 7, salt, smear water, and different temperatures were investigated. Results indicate that phage P100 is stable and able to bind to the host under most conditions tested. Replication was dependent upon the growth of L. monocytogenes and efficacy was higher when bacterial growth was reduced by certain environmental conditions. In long-term experiments at different temperatures phages were initially able to reduce bacteria up to seven log10 units after 2 weeks at 4 degrees C. However, thereafter, re-growth and development of phage-resistant L. monocytogenes isolates were encountered.

Fokine, A. and Rossmann, M. G. Common Evolutionary Origin of Procapsid Proteases, Phage Tail Tubes, and Tubes of Bacterial Type VI Secretion Systems. Structure. 11-1-2016; 24(11):1928-1935; PM: 27667692

ABSTRACT: Many large viruses, including tailed dsDNA bacteriophages and herpesviruses, assemble their capsids via formation of precursors, called procapsids or proheads. The prohead has an internal core, made of scaffolding proteins, and an outer shell, formed by the major capsid protein. The prohead usually contains a protease, which is activated during capsid maturation to destroy the inner core and liberate space for the genome. Here, we report a 2.0 A resolution structure of the pentameric procapsid protease of bacteriophage T4, gene product (gp)21. The structure corresponds to the enzyme's pre-active state in which its N-terminal region blocks the catalytic center, demonstrating that the activation mechanism involves self-cleavage of nine N-terminal residues. We describe similarities and differences between T4 gp21 and related herpesvirus proteases. We found that gp21 and the herpesvirus proteases have similarity with proteins forming the tubes of phage tails and bacterial type VI secretion systems, suggesting their common evolutionary origin.

Gambelli, L., Cremers, G., Mesman, R., Guerrero, S., Dutilh, B. E., Jetten, M. S., Op den Camp, H. J., and van, Niftrik L. Ultrastructure and Viral Metagenome of Bacteriophages from an Anaerobic Methane Oxidizing Methylomirabilis Bioreactor Enrichment Culture. Front Microbiol. 2016; 7:1740; PM: 27877158

ABSTRACT: With its capacity for anaerobic methane oxidation and denitrification, the bacterium Methylomirabilis oxyfera plays an important role in natural ecosystems. Its unique physiology can be exploited for more sustainable wastewater treatment technologies. However, operational stability of full-scale bioreactors can experience setbacks due to, for example, bacteriophage blooms. By shaping microbial communities through mortality, horizontal gene transfer, and metabolic reprogramming, bacteriophages are important players in most ecosystems. Here, we analyzed an infected Methylomirabilis sp. bioreactor enrichment culture using (advanced) electron microscopy, viral metagenomics and bioinformatics. Electron micrographs revealed four different viral morphotypes, one of which was observed to infect Methylomirabilis cells. The infected cells contained densely packed ~55 nm icosahedral bacteriophage particles with a putative internal membrane. Various stages of virion assembly were observed. Moreover, during the bacteriophage replication, the host cytoplasmic membrane appeared extremely patchy, which suggests that the bacteriophages may use host bacterial lipids to build their own putative internal membrane. The viral metagenome contained 1.87 million base pairs of assembled viral sequences, from which five putative complete viral genomes were assembled and manually annotated. Using bioinformatics analyses, we could not identify which viral genome belonged to the Methylomirabilis- infecting bacteriophage, in part because the obtained viral genome sequences were novel and unique to this reactor system. Taken together these results show that new bacteriophages can be detected in anaerobic cultivation systems and that the effect of bacteriophages on the microbial community in these systems is a topic for further study.

Gao, Y., Meyer, J. R., and Nelson, S. W. A network of allosterically coupled residues in the bacteriophage T4 Mre11-Rad50 complex. Protein Sci. 2016; 25(11):2054-2065; PM: 27571435

ABSTRACT: The Mre11-Rad50 (MR) protein complex, made up of a nuclease and ATPase, respectively, is involved in the processing of double-strand breaks as part of an intricate mechanism for their repair. Although it is clear that the MR complex is subject to allosteric regulation and that there is communication between the nuclease and ATPase active sites, the underlying mechanisms are poorly understood. We performed statistical coupling analysis on Mre11 and Rad50 to predict linked residues based on their evolutionary correlation. This analysis predicted a coevolving sector of six residues that may be allosterically coupled. The prediction was tested using double-mutant cycle analysis of nuclease and ATPase activity. The results indicate that a tyrosine residue located near the active site of Mre11 is allosterically coupled to several Rad50 residues located over 40 A away. This allosteric coupling may be the basis for the reciprocal regulation of the ATPase and nuclease activities of the complex.

Gella, P., Salas, M., and Mencia, M. Engineering Permissive Insertion Sites in the Bacteriophage Phi29 DNA-Linked Terminal Protein. PLoS.One. 2016; 11(10):e0164901; PM: 27780219

ABSTRACT: Many different DNA delivery vehicles have been developed and tested, all

with their advantages and disadvantages. The bacteriophage phi29 terminal protein (TP) is covalently linked to the 5' ends of the phage genome during the DNA replication process. Our approach is to utilize this TP as a platform to incorporate different protein or peptide modules that can target the DNA to the interior of the cell, to the nucleus, or even to subcellular compartments. In order to be able to insert different peptide modules on the TP sequence to endow it with desired functions and/or eliminate unwanted regions of the protein, we have carried out a transposition screening to detect insertion-permissive points on the sequence of the TP. We report the functional characterization of 12 insertion mutants of the TP, and the identification of one site at position 38 that allows the insertion of peptides up to 17 amino acids in length while maintaining the ability of the TP to support DNA amplification in vitro. A protein with one insertion at that position containing a cysteine residue, a linker, and a thrombin recognition site was purified and its amplification activity was optimized.

Ghebati-Maleki, L., Bakhshinejad, B., Baradaran, B., Motallebnezhad, M., ghebati-Maleki, A., Nickho, H., Yousefi, M., and Majidi, J. Phage display as a promising approach for vaccine development. J.Biomed.Sci. 9-29-2016; 23(1):66; PM: 27680328

ABSTRACT: Bacteriophages are specific antagonists to bacterial hosts. These viral entities have attracted growing interest as optimal vaccine delivery vehicles. Phages are well-matched for vaccine design due to being highly stable under harsh environmental conditions, simple and inexpensive large scale production, and potent adjuvant capacities. Phage vaccines have efficient immunostimulatory effects and present a high safety profile because these viruses have made a constant relationship with the mammalian body during a long-standing evolutionary period. The birth of phage display technology has been a turning point in the development of phage-based vaccines. Phage display vaccines are made by expressing multiple copies of an antigen on the surface of immunogenic phage particles, thereby eliciting a powerful and effective immune response. Also, the ability to produce combinatorial peptide libraries with a highly diverse pool of randomized ligands has transformed phage display into a straightforward, versatile and high throughput screening methodology for the identification of potential vaccine candidates against different diseases in particular microbial infections. These libraries can be conveniently screened through an affinity selection-based strategy called biopanning against a wide variety of targets for the selection of mimotopes with high antigenicity and immunogenicity. Also, they can be panned against the antiserum of convalescent individuals to recognize novel peptidomimetics of pathogen-related epitopes. Phage display has represented enormous promise for finding new strategies of vaccine discovery and production and current breakthroughs promise a brilliant future for the development of different phage-based vaccine platforms.

Ghugare, G. S., Nair, A., Nimkande, V., Sarode, P., Rangari, P., and Khairnar, K. Membrane filtration Immobilization technique - A simple and novel method for primary isolation and enrichment of bacteriophages. J.Appl.Microbiol. 11-8-2016; PM: 27862682

ABSTRACT: ***AIM***: To develop a method for isolation and enrichment of bacteriophages selectively against specific bacteria coupled with membrane filtration technique. ***METHODS AND RESULTS***: Rapid isolation and concentration of host specific bacteriophages was achieved by exposure of the sample suspected to contain bacteriophages to specific host immobilized on a 0.45mum membrane in a membrane

filtration unit. The principle behind this method is exploitation of host specific interaction of bacteriophages with their host and maximizing this interaction using classic membrane filtration method. This provides a chance to every bacteriophage in the sample to interact with the specific host on membrane filter under the effect of vacuum with the help of vacuum pump. Specific bacteriophages of the host are retained onto the membrane along with its host cells due to effect of adsorption and these adsorbed bacteriophages (along with their hosts) on filter disc are then amplified and enriched in regular nutritive broth Tryptose Soya Broth (TSB) by incubation. With help of plaque assay method, host specific phages of various bacterial species were isolated, segregated and enriched. ***CONCLUSIONS***: The phage concentration method coupled with membrane filtration immobilization of host bacteria was able to isolate and enrich the host specific bacteriophages by several folds using less quantity of environmental water sample, or other phage suspensions. Enrichment of phages from single plaques too was achieved. *SIGNIFICANCE AND IMPACT OF THE STUDY*: Isolation and detection of host specific bacteriophages from low bacteriophage density water sample in a single step by use of a simple and basic microbiological technique can be achieved. Enrichment of phages from low phage titre suspensions is also achieved very effectively. This article is protected by copyright. All rights reserved.

Girlich, D., Bonnin, R. A., Bogaerts, P., De Laveleye, M., Huang, D. T., Dortet, L., Glaser, P., Glupczynski, Y., and Naas, T. Chromosomal amplification of the blaOXA-58 carbapenemase gene in a Proteus mirabilis clinical isolate. Antimicrob.Agents Chemother. 11-14-2016; PM: 27855079

ABSTRACT: Horizontal gene transfer may occur between distantly related bacteria thus leading to genetic plasticity and in some cases to acquisition of novel resistance traits. Proteus mirabilis is an Enterobacterial species responsible for human infections that may express various acquired ss-lactam resistance genes, including different classes of carbapenemase genes. Here we report a Proteus mirabilis clinical isolate (1091) displaying resistance to penicillin, including temocillin, together with reduced susceptibility to carbapenems and susceptibility to expanded-spectrum cephalosporins. Using biochemical tests, significant carbapenem hydrolysis could be detected in P. mirabilis 1091. Since PCR failed to detect acquired carbapenemase genes commonly found in Enterobactericeae we used a whole genome sequencing approach that revealed the presence of blaOXA-58 class D carbapenemase gene, so far only identified in Acinetobacter sp.. This gene was located on a 3.1-kb element co-harboring a blaAmpC-like gene. Remarkably these two genes were bracketed by putative XerC-XerD binding sites and inserted at a XerC-XerD site located between the terminase-like small and large subunit genes of a bacteriophage. Increased expression of the two bla genes resulted from a 6-time tandem amplification of the element as revealed by Southern blotting. This is the first isolation of a clinical P. mirabilis strain producing OXA-58, a class D carbapenemase and the first description of a XerC-XerD dependent insertion of antibiotic resistance genes within a bacteriophage. This study revealed a new role for the XerC-XerD recombinase in bacteriophage biology.

Glow, D., Kurkowska, M., Czarnecka, J., Szczepaniak, K., Pianka, D., Kappert, V., Bujnicki, J. M., and Skowronek, K. J. Identification of protein structural elements responsible for the diversity of sequence preferences among Mini-III RNases. Sci.Rep. 12-7-2016; 6:38612; PM: 27924926

ABSTRACT: Many known endoribonucleases select their substrates based on the presence of one or a few specific nucleotides at or near the cleavage site. In some cases, selectivity is also determined by the structural features of the substrate. We recently described the sequence-specific cleavage of double-stranded RNA by Mini-III RNase from Bacillus subtilis in vitro. Here, we characterized the sequence specificity of eight other members of the Mini-III RNase family from different bacterial species. High-throughput analysis of the cleavage products of Phi6 bacteriophage dsRNA indicated subtle differences in sequence preference between these RNases, which were confirmed and characterized by systematic analysis of the cleavage kinetics of a set of short dsRNA substrates. We also showed that the sequence specificities of Mini-III RNases are not reflected by different binding affinities for cognate and non-cognate sequences, suggesting that target selection occurs predominantly at the cleavage step. We were able to identify two structural elements, the alpha4 helix and alpha5b-alpha6 loop that were involved in target selection. Characterization of the sequence specificity of the eight Mini-III RNases may provide a basis for better understanding RNA substrate recognition by Mini-III RNases and adopting these enzymes and their engineered derivatives as tools for RNA research.

Gonzalez, S., Fernandez, L., Campelo, A. B., Gutierrez, D., Martinez, B., Rodriguez, A., and Garcia, P. The behavior of Staphylococcus aureus dual-species biofilms treated with bacteriophage phiIPLA-RODI depends on the accompanying microorganism. Appl.Environ.Microbiol. 11-11-2016; PM: 27836851

ABSTRACT: The use of bacteriophages as antimicrobials against pathogenic bacteria offers a promising alternative to traditional antibiotics and disinfectants. Significantly, phages may help to remove biofilms, which are notoriously resistant to commonly used eradication methods. However, the successful development of novel antibiofilm strategies must take into account that real-life biofilms usually consist of mixed-species populations. Within this context, this study aimed to explore the effectiveness of bacteriophage-based sanitation procedures for removing polymicrobial biofilms from food industry surfaces. To do that, dual-species biofilms formed by the food pathogenic bacterium Staphylococcus aureus in combination with Lactobacillus plantarum, Enterococcus faecium or Lactobacillus pentosus were treated with the staphylococcal phage phiIPLA-RODI. Our results suggest that the impact of bacteriophage treatment on S. aureus mixed-species biofilms varies depending on the accompanying species and the infection conditions. For instance, short treatments (4 h) with a phage suspension under nutrient-limiting conditions reduced the number of S. aureus cells in 5 h biofilms by approximately 1 log unit without releasing the non-susceptible species. In contrast, longer infection periods (18 h) with no nutrient limitation resulted in increased killing of S. aureus cells by the phage (decrease of up to 2.9 log units). However, in some cases, these conditions promoted growth of the accompanying species. For example, L. plantarum cell counts were up to 2.3 log units higher in the treated sample compared to the untreated control. Furthermore, phage propagation inside dual-species biofilms was also highly dependent on the accompanying species, with the highest rate detected in biofilms formed by S. aureus-L. pentosus Scanning electron microscopy (SEM) and confocal laser scanning microscopy (CLSM) also showed changes in the three-dimensional structure of mixed-species biofilms after phage treatment. Altogether, the results presented here highlight the need to study the impact of phage therapy on microbial communities that reflect a more realistic setting. *IMPORTANCE*: Biofilms represent a major source of contamination in industrial and hospital settings. Therefore,

the development of efficient strategies to combat bacterial biofilms is of the utmost importance from a medical as well as an economic perspective. Bacteriophages have shown potential as novel antibiofilm agents, but further research is still required to fully understand the interactions between phages and biofilm-embedded bacteria. The results presented in this manuscript contribute to achieving a better understanding of such interactions in a more realistic context, considering that most biofilms in the environment consist of mixed-species populations.

Gorski, A., Miedzybrodzki, R., Weber-Dabrowska, B., Fortuna, W., Letkiewicz, S., Rogoz, P., Jonczyk-Matysiak, E., Dabrowska, K., Majewska, J., and Borysowski, J. Phage Therapy: Combating Infections with Potential for Evolving from Merely a Treatment for Complications to Targeting Diseases. Front Microbiol. 2016; 7:1515; PM: 27725811

ABSTRACT: Antimicrobial resistance is considered to be one of the greatest challenges of medicine and our civilization. Lack of progress in developing new anti-bacterial agents has greatly revived interest in using phage therapy to combat antibiotic-resistant infections. Although a number of clinical trials are underway and more are planned, the realistic perspective of registration of phage preparations and their entering the health market and significantly contributing to the current antimicrobial crisis is rather remote. Therefore, in addition to planning further clinical trials, our present approach of phage treatment carried out as experimental therapy (compassionate use) should be expanded to address the growing and urgent needs of increasing cohorts of patients for whom no alternative treatment is currently available. During the past 11 years of our phage therapy center's operation, we have obtained relevant clinical and laboratory data which not only confirm the safety of the therapy but also provide important information shedding more light on many aspects of the therapy, contributing to its optimization and allowing for construction of the most appropriate clinical trials. New data on phage biology and interactions with the immune system suggest that in the future phage therapy may evolve from dealing with complications to targeting diseases. However, further studies are necessary to confirm this promising trend.

Gregory, A. C., Solonenko, S. A., Ignacio-Espinoza, J. C., LaButti, K., Copeland, A., Sudek, S., Maitland, A., Chittick, L., Dos, Santos F., Weitz, J. S., Worden, A. Z., Woyke, T., and Sullivan, M. B. Genomic differentiation among wild cyanophages despite widespread horizontal gene transfer. BMC.Genomics. 11-16-2016; 17(1):930; PM: 27852226

*ABSTRACT: **BACKGROUND***: Genetic recombination is a driving force in genome evolution. Among viruses it has a dual role. For genomes with higher fitness, it maintains genome integrity in the face of high mutation rates. Conversely, for genomes with lower fitness, it provides immediate access to sequence space that cannot be reached by mutation alone. Understanding how recombination impacts the cohesion and dissolution of individual whole genomes within viral sequence space is poorly understood across double-stranded DNA bacteriophages (a.k.a phages) due to the challenges of obtaining appropriately scaled genomic datasets. ***RESULTS***: Here we explore the role of recombination in both maintaining and differentiating whole genomes of 142 wild double-stranded DNA marine cyanophages. Phylogenomic analysis across the 51 core genes revealed ten lineages, six of which were well represented. These phylogenomic lineages represent discrete genotypic populations based on comparisons of intra- and inter- lineage shared gene content, genome-wide average nucleotide identity, as well as detected gaps in the distribution of pairwise differences between genomes.

McDonald-Kreitman selection tests identified putative niche-differentiating genes under positive selection that differed across the six well-represented genotypic populations and that may have driven initial divergence. Concurrent with patterns of recombination of discrete populations, recombination analyses of both genic and intergenic regions largely revealed decreased genetic exchange across individual genomes between relative to within populations. ***CONCLUSIONS***: These findings suggest that discrete double-stranded DNA marine cyanophage populations occur in nature and are maintained by patterns of recombination akin to those observed in bacteria, archaea and in sexual eukaryotes.

Gu, X., Park, S. Y., Tonelli, M., Cornilescu, G., Xia, T., Zhong, D., and Schroeder, S. J. NMR Structures and Dynamics in a Prohead RNA Loop that Binds Metal Ions. J.Phys.Chem.Lett. 10-6-2016; 7(19):3841-3846; PM: 27631837

ABSTRACT: Metal ions are critical for RNA structure and enzymatic activity. We present the structure of an asymmetric RNA loop that binds metal ions and has an essential function in a bacteriophage packaging motor. Prohead RNA is a noncoding RNA that is required for genome packaging activity in phi29-like bacteriophage. The loops in GA1 and phi29 bacteriophage share a conserved adenine that forms a base triple, although the structural context for the base triple differs. NMR relaxation studies and femtosecond time-resolved fluorescence spectroscopy reveal the dynamic behavior of the loop in the metal ion bound and unbound forms. The mechanism of metal ion binding appears to be an induced conformational change between two dynamic ensembles rather than a conformational capture mechanism. These results provide experimental benchmarks for computational models of RNA-metal ion interactions.

Gundogdu, A., Bolkvadze, D., and Kilic, H. In vitro Effectiveness of Commercial Bacteriophage Cocktails on Diverse Extended-Spectrum Beta-Lactamase Producing Escherichia coli Strains. Front Microbiol. 2016; 7:1761; PM: 27857711

ABSTRACT: The objective of this study is to determine the in vitro susceptibility of Georgian bacteriophage cocktails on multidrug resistant (MDR) extended-spectrum beta-lactamase producing Escherichia coli (ESBL-EC) isolated from patients' blood and urine cultures. A total of 615 E. coli isolates were included in this study. Phene Plate (PhP)-typing and phylogenetic grouping were used for the typing. Antimicrobial resistance profiles and ESBL production of all isolates were confirmed according to Clinical and Laboratory Standards Institute (CLSI) criteria. The activities of four bacteriophage cocktails (Enko-phage, SES-bacteriophage, Pyo-bacteriophage, and Intesti-bacteriophage) were determined against 142 ESBL-EC using in vitro spot tests. According to this, Enko-phage were active against 87.3% of the tested strains while that ratio was 81.7% for Intesti-bacteriophage, 81.7% for Pyo-bacteriophage, and 59.2% for SES-bacteriophage cocktails. Based on the contingency tests, the phage cocktails were observed to be statistically significantly ($p < 0.001$) more effective on ESBL-EC strains belonging to phylogenetic groups D and B2. The employed phage cocktails were found to be affective against all tested resistant types. These results are promising especially for the infections that are caused by MDR pathogens that are difficult to treat. As this is a preliminary step to the potential clinical trials to be designed for the country, in vitro confirmation of their success on a MDR ESBL-EC collection should be accepted as an initial action, which is encouraging to consider clinical trials of phage therapy especially in countries which are not introduce phage therapy.

Guo, Z., Huang, H., Wu, X., Hao, Y., and Sun, Y. Complete Genome Sequence of Lytic Bacteriophage LZ35 Infecting Acinetobacter baumannii Isolates. Genome Announc. 11-17-2016; 4(6); PM: 27856573

ABSTRACT: Acinetobacter baumannii is a Gram-negative opportunistic pathogen that is frequently associated with nosocomial infections. Bacteriophages infecting A. baumannii can be used as effective agents to control these infections. Here, we announce the complete genome sequence of the lytic bacteriophage LZ35 infecting A. baumannii isolates.

Gutierrez, D., Hidalgo-Cantabrana, C., Rodriguez, A., Garcia, P., and Ruas-Madiedo, P. Monitoring in Real Time the Formation and Removal of Biofilms from Clinical Related Pathogens Using an Impedance-Based Technology. PLoS.One. 2016; 11(10):e0163966; PM: 27695058

ABSTRACT: Bacteria found in diverse ecosystems grow in a community of aggregated cells that favors their survival and colonization. Different extracellular polymeric substances are used to entrap this multispecies community forming a biofilm, which can be associated to biotic and abiotic surfaces. This widespread and successful way of bacterial life, however, can lead to negative effects for human activity since many pathogen and spoiling bacteria form biofilms which are not easy to eradicate. Therefore, the search for novel anti-biofilm bio-active molecules is a very active research area for which simple, reliable, and fast screening methods are demanded. In this work we have successfully validated an impedance-based method, initially developed for the study of adherent eukaryotic cells, to monitor the formation of single-species biofilms of three model bacteria in real time. The xCelligence real time cell analyzer (RTCA) equipment uses specific microtiter E-plates coated with gold-microelectrodes that detect the attachment of adherent cells, thus modifying the impedance signal. In the current study, this technology allowed the distinction between biofilm-producers and non-producers of Staphylococcus aureus and Staphylococcus epidermidis, as well as the formation of Streptococcus mutans biofilms only when sucrose was present in the culture medium. Besides, different impedance values permitted discrimination among the biofilm-producing strains tested regardless of the nature of the polymeric biofilm matrix. Finally, we have continuously monitored the inhibition of staphylococcal biofilm formation by the bacteriophage phi-IPLA7 and the bacteriophage-encoded endolysin LysH5, as well as the removal of a preformed biofilm by this last antimicrobial treatment. Results observed with the impedance-based method showed high correlation with those obtained with standard approaches, such as crystal violet staining and bacteria enumeration, as well as with those obtained upon other abiotic surfaces (polystyrene and stainless steel). Therefore, this RTCA technology opens new opportunities in the biofilm research arena and its application could be further explored for other bacterial genera as well as for different bio-active molecules.

Hathaway, H., Ajuebor, J., Stephens, L., Coffey, A., Potter, U., Sutton, J. M., and Jenkins, A. T. Thermally triggered release of the bacteriophage endolysin CHAPK and the bacteriocin lysostaphin for the control of methicillin resistant Staphylococcus aureus (MRSA). J.Control Release. 11-28-2016; 245:108-115; PM: 27908758

ABSTRACT: Staphylococcus aureus infections of the skin and soft tissue pose a major

concern to public health, largely owing to the steadily increasing prevalence of drug resistant isolates. As an alternative mode of treatment both bacteriophage endolysins and bacteriocins have been shown to possess antimicrobial efficacy against multiple species of bacteria including otherwise drug resistant strains. Despite this, the administration and exposure of such antimicrobials should be restricted until required in order to discourage the continued evolution of bacterial resistance, whilst maintaining the activity and stability of such proteinaceous structures. Utilising the increase in skin temperature during infection, the truncated bacteriophage endolysin CHAPK and the staphylococcal bacteriocin lysostaphin have been co-administered in a thermally triggered manner from Poly(N-isopropylacrylamide) (PNIPAM) nanoparticles. The thermoresponsive nature of the PNIPAM polymer has been employed in order to achieve the controlled expulsion of a synergistic enzybiotic cocktail consisting of CHAPK and lysostaphin. The point at which this occurs is modifiable, in this case corresponding to the threshold temperature associated with an infected wound. Consequently, bacterial lysis was observed at 37 degrees C, whilst growth was maintained at the uninfected skin temperature of 32 degrees C.

Henein, A. E., Hanlon, G. W., Cooper, C. J., Denyer, S. P., and Maillard, J. Y. A Partially Purified Acinetobacter baumannii Phage Preparation Exhibits no Cytotoxicity in 3T3 Mouse Fibroblast Cells. Front Microbiol. 2016; 7:1198; PM: 27536286

ABSTRACT: A surge in the level and scale of antibiotic resistance has prompted renewed interest in the application of bacteriophages to treat bacterial infections. However, concerns still exist over their efficacy and safety. Acinetobacter baumannii phage BS46, a member of the family Myoviridae, has previously been shown to be effective in murine models. The cytotoxic effect of this phage was evaluated in mouse fibroblast 3T3 cells using four different assays: trypan blue; staining with Hoechst and propidium iodide; lactate dehydrogenase release; and the MTS assay. The addition of phage concentrations up to 2 x 10(9) pfu/mL showed little to no impact on the viability of 3T3 cells after 24 h exposure using the different assays. This study demonstrates that phage BS46 is non-cytotoxic to 3T3 cells using four different assays and that appropriate quality assurance protocols for phage therapeutics are required.

Heussler, G. E., Miller, J. L., Price, C. E., Collins, A. J., and O'Toole, G. A. Requirements for Pseudomonas aeruginosa Type I-F CRISPR-Cas Adaptation Determined Using a Biofilm Enrichment Assay. J.Bacteriol. 11-15-2016; 198(22):3080-3090; PM: 27573013

ABSTRACT: CRISPR (clustered regularly interspaced short palindromic repeat)-Cas (CRISPR-associated protein) systems are diverse and found in many archaea and bacteria. These systems have mainly been characterized as adaptive immune systems able to protect against invading mobile genetic elements, including viruses. The first step in this protection is acquisition of spacer sequences from the invader DNA and incorporation of those sequences into the CRISPR array, termed CRISPR adaptation. Progress in understanding the mechanisms and requirements of CRISPR adaptation has largely been accomplished using overexpression of cas genes or plasmid loss assays; little work has focused on endogenous CRISPR-acquired immunity from viral predation. Here, we developed a new biofilm-based assay system to enrich for Pseudomonas aeruginosa strains with new spacer acquisition. We used this assay to demonstrate that P. aeruginosa rapidly acquires spacers protective against DMS3vir, an engineered lytic variant of the Mu-like bacteriophage DMS3, through primed CRISPR adaptation from spacers present in

the native CRISPR2 array. We found that for the P. aeruginosa type I-F system, the cas1 gene is required for CRISPR adaptation, recG contributes to (but is not required for) primed CRISPR adaptation, recD is dispensable for primed CRISPR adaptation, and finally, the ability of a putative priming spacer to prime can vary considerably depending on the specific sequences of the spacer. *IMPORTANCE*: Our understanding of CRISPR adaptation has expanded largely through experiments in type I CRISPR systems using plasmid loss assays, mutants of Escherichia coli, or cas1-cas2 overexpression systems, but there has been little focus on studying the adaptation of endogenous systems protecting against a lytic bacteriophage. Here we describe a biofilm system that allows P. aeruginosa to rapidly gain spacers protective against a lytic bacteriophage. This approach has allowed us to probe the requirements for CRISPR adaptation in the endogenous type I-F system of P. aeruginosa Our data suggest that CRISPR-acquired immunity in a biofilm may be one reason that many P. aeruginosa strains maintain a CRISPR-Cas system.

Hille, F. and Charpentier, E. CRISPR-Cas: biology, mechanisms and relevance. Philos.Trans.R.Soc.Lond B Biol.Sci. 11-5-2016; 371(1707); PM: 27672148

ABSTRACT: Prokaryotes have evolved several defence mechanisms to protect themselves from viral predators. Clustered regularly interspaced short palindromic repeats (CRISPR) and their associated proteins (Cas) display a prokaryotic adaptive immune system that memorizes previous infections by integrating short sequences of invading genomes-termed spacers-into the CRISPR locus. The spacers interspaced with repeats are expressed as small guide CRISPR RNAs (crRNAs) that are employed by Cas proteins to target invaders sequence-specifically upon a reoccurring infection. The ability of the minimal CRISPR-Cas9 system to target DNA sequences using programmable RNAs has opened new avenues in genome editing in a broad range of cells and organisms with high potential in therapeutical applications. While numerous scientific studies have shed light on the biochemical processes behind CRISPR-Cas systems, several aspects of the immunity steps, however, still lack sufficient understanding. This review summarizes major discoveries in the CRISPR-Cas field, discusses the role of CRISPR-Cas in prokaryotic immunity and other physiological properties, and describes applications of the system as a DNA editing technology and antimicrobial agent.This article is part of the themed issue 'The new bacteriology'.

Hong, Y., Thimmapuram, J., Zhang, J., Collings, C. K., Bhide, K., Schmidt, K., and Ebner, P. D. The impact of orally administered phages on host immune response and surrounding microbial communities. Bacteriophage. 2016; 6(3):e1211066; PM: 27738553

ABSTRACT: Numerous studies have shown the efficacy of phage therapy in reducing foodborne pathogen carriage in food animals. Fewer studies have focused on host reactions, especially in terms of phage-mediated acute immune responses and effects on the gut microbiome. Here we administered E. coli O157:H7 phages in low (single dose of 105 PFU) or high (single dose of 107 PFU) quantities to mice. While there were time points at which cytokine levels in different treatment groups differed from one another, all cytokine levels remained within normal ranges for mice regardless of treatment. Similarly, the patterns of these differences were not dose related, indicating that phage treatment did not result in a strong acute immune response as measured here. In separate experiments, 3-week-old pigs received a diet containing an in-feed antibiotic or daily phage treatment. After two weeks, microbial DNA of ileal, cecal, and fecal

contents was characterized using 16S rRNA sequencing. There were no statistical differences in performance among the different groups. Compared to control pigs (no antibiotic, no phage), antibiotic treatment significantly altered ileal microbiome composition ($P < 0.05$), with Bacilli being most affected (antibiotic treated: 22%; control: 76%; FDR = 0.0572). No significant differences were observed in cecal and fecal microbiome composition between antibiotic-treated and control pigs, and there were no differences in gut microbiome composition between phage treated and control pigs in any intestinal compartment. Significant abundance differences were observed at the OTU level, with OTUs belonging to genera such as Lactobacillus and Streptococcus being over- or under-represented in either antibiotic or phage treated groups compared to control pigs. Determining whether these changes are deleterious to host, however, requires further study.

Hong, Y., Schmidt, K., Marks, D., Hatter, S., Marshall, A., Albino, L., and Ebner, P. Treatment of Salmonella-Contaminated Eggs and Pork with a Broad-Spectrum, Single Bacteriophage: Assessment of Efficacy and Resistance Development. Foodborne.Pathog.Dis. 10-21-2016; PM: 27768383

ABSTRACT: Numerous studies have assessed the efficacy of phage-based methods to inhibit Salmonella contamination in food products. As with most antibacterials, bacteria can develop resistance to phage in vitro. Here, we applied a single broad-spectrum Salmonella phage, vB_SalS_SJ_2 (SJ2; 108 PFU; MOI = 10), to Salmonella-contaminated meat and eggs to quantify the development of resistance in actual food matrices. Treatment with a single phage significantly reduced Salmonella Typhimurium contamination in both ground pork and liquid egg at various time points. Similarly, the same phage significantly reduced Salmonella Enteritidis in both food matrices. Efficacy was temperature dependent as larger reductions were seen at higher temperatures (21 degrees C) versus lower temperatures (4 degrees C) at 24 h. Following phage treatment, over 10,000 Salmonella isolates were examined for resistance to the treatment phage. The percentages of phage-resistant Salmonella (either serovar) recovered from phage-treated versus untreated pork did not differ. Conversely, significantly ($p < 0.05$) higher percentages of phage-resistant Salmonella Typhimurium (92.50% vs. 0.56% of control) and Salmonella Enteritidis (50.83% vs. 0.56% of control) isolates were observed in phage-treated versus untreated egg samples after incubation at room temperature for 48 h. Taken together, these data indicate that the food matrix may influence the emergence of phage resistance with resistance developing more rapidly in foods with less complex microbial communities. Future studies will focus on the impact the development of resistance in production and processing settings may have on the efficacy of phage treatments for longer term biocontrol of pathogens.

Hook, C. D., Samsonov, V. V., Ublinskaya, A. A., Kuvaeva, T. M., Andreeva, E. V., Gorbacheva, L. Y., and Stoynova, N. V. A novel approach for Escherichia coli genome editing combining in vivo cloning and targeted long-length chromosomal insertion. J.Microbiol.Methods. 2016; 130:83-91; PM: 27567891

ABSTRACT: Despite the abundance of genetic manipulation approaches, particularly for Escherichia coli, new techniques and increased flexibility in the application of existing techniques are required to address novel aims. The most widely used approaches for chromosome editing are based on bacteriophage site-specific and lambdaRed/RecET-mediated homologous recombination. In the present study, these techniques were

combined to develop a novel approach for in vivo cloning and targeted long-length chromosomal insertion. This approach permits direct lambdaRed-mediated cloning of DNA fragment with lengths of 10kb or greater from the E. coli chromosome into the plasmid vector pGL2, which carries the ori of pSC101, the varphi80-attP site of varphi80 phage, and an excisable CmR marker bracketed by lambda-attL/attR sites. In pGL2-based recombinant plasmids, the origin of replication can be eliminated in vitro via hydrolysis by SceI endonuclease and recircularization by DNA ligase. The resulting ori-less circular recombinant DNA can be used for targeted insertion of the cloned sequence into the chromosome at a selected site via varphi80 phage-specific integrase-mediated recombination using the Dual-In/Out approach (Minaeva et al., 2008). At the final stage of chromosomal editing, the CmR-marker can be excised from the chromosome due to expression of the lambdaint/xis genes. Notably, the desired fragment can be inserted as multiple copies in the chromosome by combining insertions at different sites in one strain using the P1 general transduction technique (Moore, 2011). The developed approach is useful for the construction of plasmidless, markerless recombinant strains for fundamental and industrial purposes.

Hoyland-Kroghsbo, N. M., Paczkowski, J., Mukherjee, S., Broniewski, J., Westra, E., Bondy-Denomy, J., and Bassler, B. L. Quorum sensing controls the Pseudomonas aeruginosa CRISPR-Cas adaptive immune system. Proc.Natl.Acad.Sci.U.S.A. 11-14-2016; PM: 27849583

ABSTRACT: CRISPR-Cas are prokaryotic adaptive immune systems that provide protection against bacteriophage (phage) and other parasites. Little is known about how CRISPR-Cas systems are regulated, preventing prediction of phage dynamics in nature and manipulation of phage resistance in clinical settings. Here, we show that the bacterium Pseudomonas aeruginosa PA14 uses the cell-cell communication process, called quorum sensing, to activate cas gene expression, to increase CRISPR-Cas targeting of foreign DNA, and to promote CRISPR adaptation, all at high cell density. This regulatory mechanism ensures maximum CRISPR-Cas function when bacterial populations are at highest risk for phage infection. We demonstrate that CRISPR-Cas activity and acquisition of resistance can be modulated by administration of pro- and antiquorum-sensing compounds. We propose that quorum-sensing inhibitors could be used to suppress the CRISPR-Cas adaptive immune system to enhance medical applications, including phage therapies.

Hung, W. C., Wan, T. W., Kuo, Y. C., Yamamoto, T., Tsai, J. C., Lin, Y. T., Hsueh, P. R., and Teng, L. J. Molecular Evolutionary Pathways toward Two Successful Community-Associated but Multidrug-Resistant ST59 Methicillin-Resistant Staphylococcus aureus Lineages in Taiwan: Dynamic Modes of Mobile Genetic Element Salvages. PLoS.One. 2016; 11(9):e0162526; PM: 27606427

ABSTRACT: Clonal complex 59 (CC59) Staphylococcus aureus in Taiwan includes both methicillin-susceptible S. aureus (MSSA) and methicillin-resistant S. aureus (MRSA). As the most prominent community-associated MRSA (CA-MRSA) in Taiwan, CC59 has two major clones characterized as PVL-negative SCCmec IV (carrying the staphylococcal cassette chromosome mec IV but Panton-Valentine leukocidin-negative) and PVL-positive SCCmec V (5C2&5). We investigated the drug resistance, phylogeny and the distribution and sequence variation of SCCmec, staphylococcal bacteriophage phiSA3, genomic island nuSabeta and MES (an enterococcal mobile genetic element conferring multidrug resistance) in 195 CC59 S. aureus. Sequencing and PCR mapping revealed

that all of the CC59/SCCmec V (5C2&5) MRSA strains had acquired MESPM1 or its segregants, and obtained a phiSA3-related fragment in nuSabeta. In contrast, MES6272-2 and MES4578, which showed gentamicin resistance that was not encoded by MESPM1, were dominant in SCCmec IVg MRSA. Translocation of a whole phiSA3 into nuSabeta instead of only a phiSA3-related fragment was common in SCCmec IVg MRSA. However, the non-subtype-g SCCmec IV MRSA (SCCmec IVa is the major) still carried MES and nuSabeta structures similar to those in SCCmec V (5C2&5) MRSA. A minimum spanning tree constructed by multiple-locus variable-number tandem repeat analysis revealed that SCCmec IVg MRSA and SCCmec V (5C2&5) MRSA grouped respectively in two major clades. The CC59 MSSA was equally distributed among the two clades, while the non-subtype-g SCCmec IV MRSA mostly clustered with SCCmec V (5C2&5) MRSA. Our findings strongly suggest that CC59 MSSA acquired divergent mobile genetic elements and evolved to SCCmec IVg MRSA and SCCmec V (5C2&5) MRSA/non-subtype-g SCCmec IV MRSA independently. The evolutionary history of CC59 S. aureus explains how mobile genetic elements increase the antimicrobial resistance and virulence and contribute to the success of CA-MRSA in Taiwan.

Iacumin, L., Manzano, M., and Comi, G. Phage Inactivation of Listeria monocytogenes on San Daniele Dry-Cured Ham and Elimination of Biofilms from Equipment and Working Environments. Microorganisms. 1-5-2016; 4(1); PM: 27681898

ABSTRACT: The anti-listerial activity of generally recognized as safe (GRAS) bacteriophage Listex P100 (phage P100) was demonstrated in broths and on the surface of slices of dry-cured ham against 5 strains or serotypes (i.e., Scott A, 1/2a, 1/2b, and 4b) of Listeria monocytogenes. In a broth model system, phage P100 at a concentration equal to or greater than 7 log PFU/mL completely inhibited 2 log CFU/cm(2) or 3 log CFU/cm(2) of L. monocytogenes growth at 30 degrees C. The temperature (4, 10, 20 degrees C) seemed to influence P100 activity; the best results were obtained at 4 degrees C. On dry-cured ham slices, a P100 concentration ranging from 5 to 8 log PFU/cm(2) was required to obtain a significant reduction in L. monocytogenes. At 4, 10, and 20 degrees C, an inoculum of 8 log PFU/cm(2) was required to completely eliminate 2 log L. monocytogenes/cm(2) and to reach the absence in 25 g product according to USA food law. Conversely, it was impossible to completely eradicate L. monocytogenes with an inoculum of approximately of 3.0 and 4.0 log CFU/cm(2) and with a P100 inoculum ranging from 1 to 7 log PFU/cm(2). P100 remained stable on dry-cured ham slices over a 14-day storage period, with only a marginal loss of 0.2 log PFU/cm(2) from an initial phage treatment of approximately 8 log PFU/cm(2). Moreover, phage P100 eliminated free L. monocytogenes cells and biofilms on the machinery surfaces used for dry-cured ham production. These findings demonstrate that the GRAS bacteriophage Listex P100 at level of 8 log PFU/cm(2) is listericidal and useful for reducing the L. monocytogenes concentration or eradicating the bacteria from dry-cured ham.

Jeon, J., D'Souza, R., Pinto, N., Ryu, C. M., Park, J., Yong, D., and Lee, K. Characterization and complete genome sequence analysis of two Myoviral bacteriophages infecting clinical carbapenem-resistant Acinetobacter baumannii isolates. J.Appl.Microbiol. 2016; 121(1):68-77; PM: 26991925

ABSTRACT: ***AIMS***: The aim of this study was to characterize phenotypical properties, to analyse whole genomes of novel Acinetobacter baumannii phages infecting carbapenem-resistant Ac. baumannii (CRAB) and to evaluate their potential as

antimicrobial alternatives to control Ac. baumannii in clinical settings. ***METHODS AND RESULTS***: The Ac. baumannii phages, Betavarphi-R1215 and Betavarphi-R2315, were isolated from sewage samples. These phages were characterized by transmission electron microscopy, host spectrum, the thermal/pH stability test, the bacterial lysis assay and the whole genome analysis. Both phages lysed 21 of 45 CRAB hosts, and showed high stability at various pH (pH 4-10) and temperature (25-60 degrees C), and were strongly active against host bacteria in vitro. The genomes of Betavarphi-R1215 and Betavarphi-R2315 are linear double-strands of DNA with 44.866 and 44.846 bp respectively. These two genomes revealed high similarity at the DNA level, but the organization and direction of open reading frames were different. ***CONCLUSIONS***: The Ac. baumannii phages, Betavarphi-R1215 and Betavarphi-R2315, are novel lytic phages lysing CRAB strains which were isolated from respiratory samples of patients. ***SIGNIFICANCE AND IMPACT OF THE STUDY***: In vitro and in silico data showed that these novel Ac. baumannii phages, Betavarphi-R1215 and Betavarphi-R2315, have potential as antimicrobial alternatives to control CRAB in healthcare settings.

Jiao, H., Yang, H., Zhao, D., He, L., Chen, J., and Li, G. The enhanced immune responses induced by Salmonella enteritidis ghosts loaded with Neisseria gonorrhoeae porB against Salmonella in mice. FEMS Microbiol.Lett. 10-18-2016; PM: 27797865

ABSTRACT: Humans health has been seriously endangered by highly prevalent salmonellosis and multidrug resistant Salmonella strains. Current vaccines suffer from variable immune-protective effects, so more effective ones are in need to control Salmonella infection : Bacterial ghosts have been produced by the expression of lysis gene E from bacteriophage PhiX174 and can be filled with considerable exogenous substances such as DNA or drugs as a novel platform. In this study, Salmonella enteritidis (SE) ghosts were developed and loaded with Neisseria gonorrhoeae porin B (porB) to construct a novel inactive vaccine. Our new studies show that SE ghosts loaded with porB displayed increased production of pro-inflammatory cytokines (IL-1beta, IL-6, IL-10 and IL-12 p70) in BMDCs, and elicited significantly higher specific systemic and mucosal immune responses to Salmonella than SE ghosts alone. In addition, the novel porB-loaded ghosts conferred higher protective effects on virulent Salmonella challenge. For the first time, we demonstrate that N. gonorrhoeae porB, as a novel adjuvant, can increase the immunogenicity of SE ghosts. Our studies suggested that Salmonella enteritidis ghosts loaded with Neisseria gonorrhoeae porin B might be a useful mucosal Salmonella vaccine candidate for practical use in future.

Jurtz, V. I., Villarroel, J., Lund, O., Voldby, Larsen M., and Nielsen, M. MetaPhinder-Identifying Bacteriophage Sequences in Metagenomic Data Sets. PLoS.One. 2016; 11(9):e0163111; PM: 27684958

ABSTRACT: Bacteriophages are the most abundant biological entity on the planet, but at the same time do not account for much of the genetic material isolated from most environments due to their small genome sizes. They also show great genetic diversity and mosaic genomes making it challenging to analyze and understand them. Here we present MetaPhinder, a method to identify assembled genomic fragments (i.e.contigs) of phage origin in metagenomic data sets. The method is based on a comparison to a

database of whole genome bacteriophage sequences, integrating hits to multiple genomes to accomodate for the mosaic genome structure of many bacteriophages. The method is demonstrated to out-perform both BLAST methods based on single hits and methods based on k-mer comparisons. MetaPhinder is available as a web service at the Center for Genomic Epidemiology https://cge.cbs.dtu.dk/services/MetaPhinder/, while the source code can be downloaded from: https://bitbucket.org/genomicepidemiology/metaphinder or https://github.com/vanessajurtz/MetaPhinder.

Kaikabo, A. A., AbdulKarim, S. M., and Abas, F. Evaluation of the efficacy of chitosan nanoparticles loaded PhiKAZ14 bacteriophage in the biological control of colibacillosis in chickens. Poult.Sci. 10-4-2016; PM: 27702916

ABSTRACT: Disease inflicted by avian pathogenic Escherichia coli (APEC) causes economic losses and burden to the poultry industry worldwide. In this study, the efficacy of chitosan nanoparticles loaded PhiKAZ14 (C-PhiKAZ14 NPs) as an oral biological therapy for Colibacillosis was evaluated. C-PhiKAZ14 NPs containing 107 PFU/ml of PhiKAZ14 (Myoviridae; T4-like coliphage) bacteriophage were used to treat experimentally APEC-infected COBB 500 broiler chicks. C-PhiKAZ14 NPs and PhiKAZ14 bacteriophage were administered orally in a single dose. The clinical symptoms, mortality, and pathology in the infected birds were recorded and compared with those of control birds that did not receive C-PhiKAZ14 NPs or naked PhiKAZ14 bacteriophage. The results showed that C-PhiKAZ14 NP intervention decreased mortality from 58.33 to 16.7% with an increase in the protection rate from 42.00 to 83.33%. The bacterial colonization of the intestines of infected birds was significantly higher in the untreated control than in the C-PhiKAZ14 NP-treated group (2.30x109 +/- 0.02 and 0.79x103 +/- 0.10 CFU/mL, respectively) (P </= 0.05). Similarly, a significant difference in the fecal shedding of Escherichia coli was observed on d 7 post challenge between the untreated control and the C-PhiKAZ14 NP-treated group (2.35x109 +/- 0.05 and 1.58x103 +/- 0.06 CFU/mL, respectively) (P </= 0.05). Similar trends were observed from d 14 until d 21 when the experiment was terminated. Treatment with C-PhiKAZ14 NPs improved the body weights of the infected chicks. A difference in body weight on d 7 post challenge was observed between the untreated control and the C-PhiKAZ14 NP-treated group (140 +/- 20 g and 160 +/- 20 g, respectively). The increase was significant (P </= 0.05) on d 21 between the 2 groups (240 +/- 30 g and 600 +/- 80 g, respectively). Consequently, the clinical signs and symptoms were ameliorated upon treatment with C-PhiKAZ14 NPs compared with infected untreated birds. In all, based on the results, it can be concluded that the encapsulation of bacteriophage could enhance bacteriophage therapy and is a valuable approach for controlling APEC infections in poultry.

Kaistha, S. D. and Umrao, P. D. Bacteriophage for Mitigation of Multiple Drug Resistant Biofilm Forming Pathogens. Recent Pat Biotechnol. 2016; 10(2):184-194; PM: 27652612

ABSTRACT: ***BACKGROUND***: Microbial communities encased in exopolymeric substances (EPS) attached to suitable substrate as biofilms show heightened resistance to multiple drugs including antibiotics. One promising control strategy in dealing with the ever mounting problem of antibiotic resistance amongst biofilm forming bacterial pathogens

is the use of biological agents. ***OBJECTIVE***: This review focuses on the development of bacteriophages as means of disrupting biofilm forming pathogens and hence mitigation of multiple drug resistant organisms. ***RESULTS***: Bacteriophages are obligatory viral intracellular parasites that can cause lytic infection of their bacterial hosts. Bacteriophage (Phage) therapy is advantageous in being highly host specific, safe and non-toxic to humans and hence environmental friendly. Bacteriophage induced lysis of cells within the biofilm is aided by the production of penetrating enzymes such as endolysins and EPS depolymerases as well quorum sensing inhibitors such as lactonases. ***CONCLUSION***: Phages are a promising alternative therapy for the control of multiple drug resistant (MDR) pathogens. Several phage (indigenous and engineered)/ phage products are currently being patented and developed as commercial biological control agents.

Kasai, Y. and Harayama, S. Construction of Marker-Free Transgenic Strains of Chlamydomonas reinhardtii Using a Cre/loxP-Mediated Recombinase System. PLoS.One. 2016; 11(8):e0161733; PM: 27564988

ABSTRACT: The Escherichia coli bacteriophage P1 encodes a site-specific recombinase called Cre and two 34-bp target sites of Cre recombinase called loxP. The Cre/loxP system has been used to achieve targeted insertion and precise deletion in many animal and plant genomes. The Cre/loxP system has particularly been used for the removal of selectable marker genes to create marker-free transgenic organisms. For the first time, we applied the Cre/loxP-mediated site-specific recombination system to Chlamydomonas reinhardtii to construct marker-free transgenic strains. Specifically, C. reinhardtii strains cc4350 and cc124 carrying an aphVIII expression cassette flanked by two direct repeats of loxP were constructed. Separately, a synthetic Cre recombinase gene (CrCRE), the codons of which were optimized for expression in C. reinhardtii, was synthesized, and a CrCRE expression cassette was introduced into strain cc4350 carrying a single copy of the loxP-flanked aphVIII expression cassette. Among 46 transformants carrying the CrCRE expression cassette stably, the excision of aphVIII by CrCre recombinase was observed only in one transformant. We then constructed an expression cassette of an in-frame fusion of ble to CrCRE via a short linker peptide. The product of ble (Ble) is a bleomycin-binding protein that confers resistance to bleomycin-related antibiotics such as Zeocin and localizes in the nucleus. Therefore, the ble-(linker)-CrCRE fusion protein is expected to localize in the nucleus. When the ble-(linker)-CrCRE expression cassette was integrated into the genome of strain cc4350 carrying a single copy of the loxP-flanked aphVIII expression cassette, CrCre recombinase-mediated excision of the aphVIII expression cassette was observed at a frequency higher than that in stable transformants of the CrCRE expression cassette. Similarly, from strain cc124 carrying a single loxP-flanked aphVIII expression cassette, the aphVIII expression cassette was successfully excised after introduction of the ble-(linker)-CrCRE expression cassette. The ble-(linker)-CrCRE expression cassette remained in the genome after excision of the aphVIII expression cassette, and it was subsequently removed by crossing with the wild-type strain. This precise Cre-mediated deletion method applicable to transgenic C. reinhardtii could further increase the potential of this organism for use in basic and applied research.

Kazi, M. and Annapure, U. S. Bacteriophage biocontrol of foodborne pathogens. J.Food Sci.Technol. 2016; 53(3):1355-1362; PM: 27570260

ABSTRACT: Bacteriophages are viruses that only infect bacterial cells. Phages are

categorized based on the type of their life cycle, the lytic cycle cause lysis of the bacterium with the release of multiple phage particles where as in lysogenic phase the phage DNA is incorporated into the bacterial genome. Lysogeny does not result in lysis of the host. Lytic phages have several potential applications in the food industry as biocontrol agents, biopreservatives and as tools for detecting pathogens. They have also been proposed as alternatives to antibiotics in animal health. Two unique features of phage relevant for food safety are that they are harmless to mammalian cells and high host specificity, keeping the natural microbiota undisturbed. However, the recent approval of bacteriophages as food additives has opened the discussion about 'edible viruses'. This article reviews in detail the application of phages for the control of foodborne pathogens in a process known as "biocontrol".

Khalifa, L., Shlezinger, M., Beyth, S., Houri-Haddad, Y., Coppenhagen-Glazer, S., Beyth, N., and Hazan, R. Phage therapy against Enterococcus faecalis in dental root canals. J.Oral Microbiol. 2016; 8:32157; PM: 27640530

ABSTRACT: Antibiotic resistance is an ever-growing problem faced by all major sectors of health care, including dentistry. Recurrent infections related to multidrug-resistant bacteria such as methicillin-resistant Staphylococcus aureus, carbapenem-resistant Enterobacteriaceae, and vancomycin-resistant enterococci (VRE) in hospitals are untreatable and question the effectiveness of notable drugs. Two major reasons for these recurrent infections are acquired antibiotic resistance genes and biofilm formation. None of the traditionally known effective techniques have been able to efficiently resolve these issues. Hence, development of a highly effective antibacterial practice has become inevitable. One example of a hard-to-eradicate pathogen in dentistry is Enterococcus faecalis, which is one of the most common threats observed in recurrent root canal treatment failures, of which the most problematic to treat are its biofilm-forming VRE strains. An effective response against such infections could be the use of bacteriophages (phages). Phage therapy was found to be highly effective against biofilm and multidrug-resistant bacteria and has other advantages like ease of isolation and possibilities for genetic manipulations. The potential of phage therapy in dentistry, in particular against E. faecalis biofilms in root canals, is almost unexplored. Here we review the efforts to develop phage therapy against biofilms. We also focus on the phages isolated against E. faecalis and discuss the possibility of using phages against E. faecalis biofilm in root canals.

Kick, B., Hensler, S., Praetorius, F., Dietz, H., and Weuster-Botz, D. Specific growth rate and multiplicity of infection affect high-cell-density fermentation with bacteriophage M13 for ssDNA production. Biotechnol.Bioeng. 10-17-2016; PM: 27748519

ABSTRACT: The bacteriophage M13 has found frequent applications in nanobiotechnology due to its chemically and genetically tunable protein surface and its ability to self-assemble into colloidal membranes. Additionally, its single-stranded (ss) genome is commonly used as scaffold for DNA origami. Despite the manifold uses of M13, upstream production methods for phage and scaffold ssDNA are underexamined with respect to future industrial usage. Here, the high-cell-density phage production with Escherichia coli as host organism was studied in respect of medium composition, infection time, multiplicity of infection, and specific growth rate. The specific growth rate and the multiplicity of infection were identified as the crucial state variables that influence phage amplification rate on one hand and the concentration of produced

ssDNA on the other hand. Using a growth rate of 0.15 h-1 and a multiplicity of infection of 0.05 pfu cfu-1 in the fed-batch production process, the concentration of pure isolated M13 ssDNA usable for scaffolded DNA origami could be enhanced by 54% to 590 mg L-1 . Thus, our results help enabling M13 production for industrial uses in nanobiotechnology. Biotechnol. Bioeng. 2016;9999: 1-8. (c) 2016 Wiley Periodicals, Inc.

Kim, E. J., Chang, H. J., Kwak, S., and Park, J. H. Virulence Factors and Stability of Coliphages Specific to Escherichia coli O157:H7 and to Various E. coli Infection. J.Microbiol.Biotechnol. 10-6-2016; PM: 27713209

ABSTRACT: Characteristics of E.coli O157:H7 specific infection bacteriophages (O157 coliphages) and broad-host-range bacteriophages for other E.coli serotypes (broad-host coliphages) were compared. The burst sizes of two groups were ranged from 40 to 176 PFU/infected. Distribution of virulence factors of stx1, stx2, ehxA, and saa between two groups were not differenciated. Broad host coliphages showed a little stable at 70 degrees C than O157 coliphages. However, O157 coliphages showed high acid and ethanol tolerant by reduction of only 22% and 11% phages under pH 3 and 70% ethanol for an hour exposure. Therefore, these results revealed that the O157 coliphages might be more stable under harsh environments, which might be a character to infect effectively the acid tolerant E. coli O157:H7.

Kim, J., Poling-Skutvik, R., Trabuco, J. R., Kourentzi, K., Willson, R. C., and Conrad, J. C. Orientational binding modes of reporters in a viral-nanoparticle lateral flow assay. Analyst. 9-30-2016; PM: 27704069

ABSTRACT: Using microscopy and image analysis, we characterize binding of filamentous viral nanoparticles to a fibrous affinity matrix as models for reporter capture in a lateral flow assay (LFA). M13 bacteriophage (M13) displaying an in vivo-biotinylated peptide (AviTag) genetically fused to the M13 tail protein p3 are functionalized with fluorescent labels. We functionalize glass fiber LFA membranes with antibodies to M13, which primarily capture M13 on the major p8 coat proteins, or with avidin, which captures M13 at the biotin-functionalized tail, and compare orientational modes of reporter capture for the side- versus tip-binding recognition interactions. The number of captured M13 is greater for side-binding than for tip-binding, as expected from the number of recognition groups. Whereas two-thirds of side-bound M13 captured by an anti-M13 antibody bind immediately after colliding with the membrane, tip-bound M13 prominently exhibit three additional orientational modes that require M13 to reorient to enable binding. These results are consistent with the idea that the elongated M13 shape couples with the complex flow field in an open and disordered fibrous LFA membrane to enhance capture.

Koganti, S., Alhmidi, H., Tomas, M. E., Cadnum, J. L., Sass, C., Jencson, A. L., and Donskey, C. J. Evaluation of an Ethanol-Based Spray Disinfectant for Decontamination of Cover Gowns Prior to Removal. Infect.Control Hosp.Epidemiol. 12-5-2016; 1-; PM: 27917733

ABSTRACT: An ethanol-based spray disinfectant significantly reduced bacteriophage MS2 contamination on material from gowns meeting ASTM standard 1671 for resistance to blood and viral penetration and on a cover gown worn by personnel. Effectiveness of

disinfection was affected by the type of gown material and the correctness of fit. Infect Control Hosp Epidemiol 2016;1-3.

Koning, R. I., Gomez-Blanco, J., Akopjana, I., Vargas, J., Kazaks, A., Tars, K., Carazo, J. M., and Koster, A. J. Asymmetric cryo-EM reconstruction of phage MS2 reveals genome structure in situ. Nat.Commun. 8-26-2016; 7:12524; PM: 27561669

ABSTRACT: In single-stranded ribonucleic acid (RNA) viruses, virus capsid assembly and genome packaging are intertwined processes. Using cryo-electron microscopy and single particle analysis we determined the asymmetric virion structure of bacteriophage MS2, which includes 178 copies of the coat protein, a single copy of the A-protein and the RNA genome. This reveals that in situ, the viral RNA genome can adopt a defined conformation. The RNA forms a branched network of stem-loops that almost all allocate near the capsid inner surface, while predominantly binding to coat protein dimers that are located in one-half of the capsid. This suggests that genomic RNA is highly involved in genome packaging and virion assembly.

Koo, C. K., Senecal, K., Senecal, A., and Nugen, S. R. Dehydration of bacteriophages in electrospun nanofibers: effect of excipients in polymeric solutions. Nanotechnology. 12-2-2016; 27(48):485102; PM: 27819799

ABSTRACT: Bacteriophages are viruses capable of infecting and lysing target bacterial cells; as such they have potential applications in agriculture for decontamination of foods, food contact surfaces and food rinse water. Although bacteriophages can retain infectivity long-term using lyophilized storage, the process of freeze-drying can be time consuming and expensive. In this study, electrospinning was used for dehydrating bacteriophages in polyvinylpyrrolidone polymer solutions with addition of excipients (sodium chloride, magnesium sulfate, Tris-HCl, sucrose) in deionized water. The high voltage dehydration reduced the infectivity of bacteriophages following electrospinning, with the damaging effect abated with addition of storage media (SM) buffer and sucrose. SM buffer and sucrose also provided the most protection over extended storage (8 weeks; 20 degrees C; 1% relative humidity) by mitigating environmental effects on the dried bacteriophages. Magnesium sulfate however provided the least protection due to coagulation effects of the ion, which can disrupt the native conformation of the bacteriophage protein coat. Storage temperatures (20 degrees C, 4 degrees C and -20 degrees C; 1% relative humidity) had a minimal effect while relative humidity had substantial effect on the infectivity of bacteriophages. Nanofibers stored in higher relative humidity (33% and 75%) underwent considerable damage due to extensive water absorption and disruption of the fibers. Overall, following storage of nanofiber mats for eight weeks at ambient temperatures, high infective phage concentrations (106-107 PFU ml-1) were retained. Therefore, this study provided valuable insights on preservation and dehydration of bacteriophages by electrospinning in comparison to freeze drying and liquid storage, and the influence of excipients on the viability of bacteriophages.

Korobeinikov, A., Shchepakina, E., and Sobolev, V. Paradox of enrichment and system order reduction: bacteriophages dynamics as case study. Math.Med.Biol. 2016; 33(3):359-369; PM: 26180103

ABSTRACT: The paradox of enrichment in a 3D model for bacteriophage dynamics, with a free infection stage of the phage and a bilinear incident rate, is considered. An

application of the technique of singular perturbation theory allows us to demonstrate why the paradox arises in this 3D model despite the fact that it has a bilinear incident rate (while in 2D predator-prey models it is usually associated with the concavity of the attack rate). Our analysis demonstrates that the commonly applied approach of the model order reduction using the so-called quasi-steady-state approximation can lead to a loss of important properties of an original system.

Kozlowska, K. I., Tymecka-Mulik, J., and Wegrzyn, G. Purified Stx and lambda phage initiator O proteins bind specifically to two different origins of replication in vitro. Protein Expr.Purif. 11-5-2016; 131:16-26; PM: 27826079

ABSTRACT: The O protein is a crucial factor initiating the DNA replication of lambdoid bacteriophage. Efficient DNA replication of Shiga toxin-converting phage is necessary for effective production of Shiga toxin - main virulence factor of STEC strains. We developed an improved protocol for overproduction, bacterial cell lysis and purification of lambdaO protein. With use of this method we have also isolated O proteins of Stx-phage P27 and 933W that were never purified before. Purified proteins were tested for their DNA binding activity and revealed a sequence specific interactions.

Krishnamurthy, M., Moore, R. T., Rajamani, S., and Panchal, R. G. Bacterial genome engineering and synthetic biology: combating pathogens. BMC.Microbiol. 11-4-2016; 16(1):258; PM: 27814687

ABSTRACT: ***BACKGROUND***: The emergence and prevalence of multidrug resistant (MDR) pathogenic bacteria poses a serious threat to human and animal health globally. Nosocomial infections and common ailments such as pneumonia, wound, urinary tract, and bloodstream infections are becoming more challenging to treat due to the rapid spread of MDR pathogenic bacteria. According to recent reports by the World Health Organization (WHO) and Centers for Disease Control and Prevention (CDC), there is an unprecedented increase in the occurrence of MDR infections worldwide. The rise in these infections has generated an economic strain worldwide, prompting the WHO to endorse a global action plan to improve awareness and understanding of antimicrobial resistance. This health crisis necessitates an immediate action to target the underlying mechanisms of drug resistance in bacteria. ***RESEARCH***: The advent of new bacterial genome engineering and synthetic biology (SB) tools is providing promising diagnostic and treatment plans to monitor and treat widespread recalcitrant bacterial infections. Key advances in genetic engineering approaches can successfully aid in targeting and editing pathogenic bacterial genomes for understanding and mitigating drug resistance mechanisms. In this review, we discuss the application of specific genome engineering and SB methods such as recombineering, clustered regularly interspaced short palindromic repeats (CRISPR), and bacterial cell-cell signaling mechanisms for pathogen targeting. The utility of these tools in developing antibacterial strategies such as novel antibiotic production, phage therapy, diagnostics and vaccine production to name a few, are also highlighted. ***CONCLUSIONS***: The prevalent use of antibiotics and the spread of MDR bacteria raise the prospect of a post-antibiotic era, which underscores the need for developing novel therapeutics to target MDR pathogens. The development of enabling SB technologies offers promising solutions to deliver safe and effective antibacterial therapies.

Kruszewski, J. and Golik, P. Pentatricopeptide Motifs in the N-Terminal Extension Domain of Yeast Mitochondrial RNA Polymerase Rpo41p Are Not Essential for Its Function. Biochemistry (Mosc.). 2016; 81(10):1101-1110; PM: 27908235

ABSTRACT: The core mitochondrial RNA polymerase is a single-subunit enzyme that in yeast Saccharomyces cerevisiae is encoded by the nuclear RPO41 gene. It is an evolutionary descendant of the bacteriophage RNA polymerases, but it includes an additional unconserved N-terminal extension (NTE) domain that is unique to the organellar enzymes. This domain mediates interactions between the polymerase and accessory regulatory factors, such as yeast Sls1p and Nam1p. Previous studies demonstrated that deletion of the entire NTE domain results only in a temperature-dependent respiratory deficiency. Several sequences related to the pentatricopeptide (PPR) motifs were identified in silico in Rpo41p, three of which are located in the NTE domain. PPR repeat proteins are a large family of organellar RNA-binding factors, mostly involved in posttranscriptional gene expression mechanisms. To study their function, we analyzed the phenotype of strains bearing Rpo41p variants where each of these motifs was deleted. We found that deletion of any of the three PPR motifs in the NTE domain does not affect respiratory growth at normal temperature, and it results in a moderate decrease in mtDNA stability. Steady-state levels of COX1 and COX2 mRNAs are also moderately affected. Only the deletion of the second motif results in a partial respiratory deficiency, manifested only at elevated temperature. Our results thus indicate that the PPR motifs do not play an essential role in the function of the NTE domain of the mitochondrial RNA polymerase.

Krylov, V., Shaburova, O., Pleteneva, E., Bourkaltseva, M., Krylov, S., Kaplan, A., Chesnokova, E., Kulakov, L., Magill, D., and Polygach, O. Modular Approach to Select Bacteriophages Targeting Pseudomonas aeruginosa for Their Application to Children Suffering With Cystic Fibrosis. Front Microbiol. 2016; 7:1631; PM: 27790211

ABSTRACT: This review discusses the potential application of bacterial viruses (phage therapy) toward the eradication of antibiotic resistant Pseudomonas aeruginosa in children with cystic fibrosis (CF). In this regard, several potential relationships between bacteria and their bacteriophages are considered. The most important aspect that must be addressed with respect to phage therapy of bacterial infections in the lungs of CF patients is in ensuring the continuity of treatment in light of the continual occurrence of resistant bacteria. This depends on the ability to rapidly select phages exhibiting an enhanced spectrum of lytic activity among several well-studied phage groups of proven safety. We propose a modular based approach, utilizing both mono-species and hetero-species phage mixtures. With an approach involving the visual recognition of characteristics exhibited by phages of well-studied phage groups on lawns of the standard P. aeruginosa PAO1 strain, the simple and rapid enhancement of the lytic spectrum of cocktails is permitted, allowing the development of tailored preparations for patients capable of circumventing problems associated with phage resistant bacterial mutants.

Kusradze, I., Karumidze, N., Rigvava, S., Dvalidze, T., Katsitadze, M., Amiranashvili, I., and Goderdzishvili, M. Characterization and Testing the Efficiency of Acinetobacter baumannii Phage vB-GEC_Ab-M-G7 as an Antibacterial Agent. Front Microbiol. 2016; 7:1590; PM: 27757110

ABSTRACT: Acinetobacter baumannii is a gram-negative, non-motile bacterium that, due to its multidrug resistance, has become a major nosocomial pathogen. The increasing number of multidrug resistant (MDR) strains has renewed interest in phage therapy. The aim of our study was to assess the effectiveness of phage administration in Acinetobacter baumannii wound infections in an animal model to demonstrate phage therapy as non-toxic, safe and alternative antibacterial remedy. Using classical methods for the study of bacteriophage properties, we characterized phage vB-GEC_Ab-M-G7 as a dsDNA myovirus with a 90 kb genome size. Important characteristics of vB-GEC_Ab-M-G7include a short latent period and large burst size, wide host range, resistance to chloroform and thermal and pH stability. In a rat wound model, phage application effectively decreased the number of bacteria isolated from the wounds of successfully treated animals. This study highlights the effectiveness of the phage therapy and provides further insight into treating infections caused by MDR strains using phage administration.

Kwiatek, M., Parasion, S., Rutyna, P., Mizak, L., Gryko, R., Niemcewicz, M., Olender, A., and Lobocka, M. Isolation of bacteriophages and their application to control Pseudomonas aeruginosa in planktonic and biofilm models. Res.Microbiol. 11-4-2016; PM: 27818282

ABSTRACT: Pseudomonas aeruginosa is frequently identified as a cause of diverse infections and chronic diseases. It forms biofilms and has natural resistance to several antibiotics. Strains of this pathogen resistant to new-generation beta-lactams have emerged. Due to the difficulties associated with treating chronic P. aeruginosa infections, bacteriophages are amongst the alternative therapeutic options being actively researched. Two obligatorily lytic P. aeruginosa phages, vB_PaeM_MAG1 (MAG1) and vB_PaeP_MAG4 (MAG4), have been isolated and characterized. These phages belong to the PAK_P1likevirus genus of the Myoviridae family and the LIT1virus genus of the Podoviridae family, respectively. They adsorb quickly to their hosts (approximately 90% in 5 min), have a short latent period (15 min), and are stable during storage. Each individual phage propagated in approximately 50% of P. aeruginosa strains tested, which increased to 72.9% when phages were combined into a cocktail. While MAG4 reduced biofilm more effectively after a short time of treatment, MAG1 was more effective after a longer time and selected less for phage-resistant clones. A MAG1-encoded homolog of YefM antitoxin of the bacterial toxin-antitoxin system may contribute to the superiority of MAG1 over MAG4.

Lee, W., Gillies, J. P., Jose, D., Israels, B. A., von Hippel, P. H., and Marcus, A. H. Single-molecule FRET studies of the cooperative and non-cooperative binding kinetics of the bacteriophage T4 single-stranded DNA binding protein (gp32) to ssDNA lattices at replication fork junctions. Nucleic Acids Res. 9-30-2016; PM: 27694621

ABSTRACT: Gene 32 protein (gp32) is the single-stranded (ss) DNA binding protein of the bacteriophage T4. It binds transiently and cooperatively to ssDNA sequences exposed during the DNA replication process and regulates the interactions of the other sub-assemblies of the replication complex during the replication cycle. We here use single-molecule FRET techniques to build on previous thermodynamic studies of gp32 binding to initiate studies of the dynamics of the isolated and cooperative binding of gp32 molecules within the replication complex. DNA primer/template (p/t) constructs are used as models to determine the effects of ssDNA lattice length, gp32 concentration, salt concentration, binding cooperativity and binding polarity at p/t junctions. Hidden

Markov models (HMMs) and transition density plots (TDPs) are used to characterize the dynamics of the multi-step assembly pathway of gp32 at p/t junctions of differing polarity, and show that isolated gp32 molecules bind to their ssDNA targets weakly and dissociate quickly, while cooperatively bound dimeric or trimeric clusters of gp32 bind much more tightly, can 'slide' on ssDNA sequences, and exhibit binding dynamics that depend on p/t junction polarities. The potential relationships of these binding dynamics to interactions with other components of the T4 DNA replication complex are discussed.

Leisi, R., Von, Nordheim M., Ros, C., and Kempf, C. The VP1u Receptor Restricts Parvovirus B19 Uptake to Permissive Erythroid Cells. Viruses. 9-28-2016; 8(10); PM: 27690083

ABSTRACT: Parvovirus B19 (B19V) is a small non-enveloped virus and known as the causative agent for the mild childhood disease erythema infectiosum. B19V has an extraordinary narrow tissue tropism, showing only productive infection in erythroid precursor cells in the bone marrow. We recently found that the viral protein 1 unique region (VP1u) contains an N-terminal receptor-binding domain (RBD), which mediates the uptake of the virus into cells of the erythroid lineage. To further investigate the role of the RBD in connection with a B19V-unrelated capsid, we chemically coupled the VP1u of B19V to the bacteriophage MS2 capsid and tested the internalization capacity of the bioconjugate on permissive cells. In comparison, we studied the cellular uptake and infection of B19V along the erythroid differentiation. The results showed that the MS2-VP1u bioconjugate mimicked the specific internalization of the native B19V into erythroid precursor cells, which further coincides with the restricted infection profile. The successful mimicry of B19V uptake demonstrates that the RBD in the VP1u is sufficient for the endocytosis of the viral capsid. Furthermore, the recombinant VP1u competed with B19V uptake into permissive cells, thus excluding a significant alternative uptake mechanism by other receptors. Strikingly, the VP1u receptor appeared to be expressed only on erythropoietin-dependent erythroid differentiation stages that also provide the necessary intracellular factors for a productive infection. Taken together, these findings suggest that the VP1u binds to a yet-unknown erythroid-specific cellular receptor and thus restricts the virus entry to permissive cells.

Lewis, D. E., Gussin, G. N., and Adhya, S. New Insights into the Phage Genetic Switch: Effects of Bacteriophage Lambda Operator Mutations on DNA Looping and Regulation of PR, PL, and PRM. J.Mol.Biol. 11-6-2016; 428(22):4438-4456; PM: 27670714

ABSTRACT: One of the best understood systems in genetic regulatory biology is the so-called "genetic switch" that determines the choice the phage-encoded CI repressor binds cooperatively to tripartite operators, OL and OR, in a defined pattern, thus blocking the transcription at two lytic promoters, PL and PR, and auto-regulating the promoter, PRM, which directs CI synthesis by the prophage. Fine-tuning of the maintenance of lysogeny is facilitated by interactions between CI dimers bound to OR and OL through the formation of a loop by the intervening DNA segment. By using a purified in vitro transcription system, we have genetically dissected the roles of individual operator sites in the formation of the DNA loop and thus have gained several new and unexpected insights into the system. First, although both OR and OL are tripartite, the presence of only a single active CI binding site in one of the two operators is sufficient for DNA loop formation. Second, in PL, unlike in PR, the promoter distal operator site, OL3, is sufficient to directly repress PL. Third, DNA looping mediated by the formation of CI octamers arising through the interaction of pairs of dimers bound to adjacent operator

sites in OR and OL does not require OR and OL to be aligned "in register", that is, CI bound to "out-of-register" sub-operators, for example, OL1~Ol2 and OR2~OR3, can also mediate loop formation. Finally, based on an examination of the mechanism of activation of PRM when only OR1 or OR2 are wild type, we hypothesize that RNA polymerase bound at PR interferes with DNA loop formation. Thus, the formation of DNA loops involves potential interactions between proteins bound at numerous cis-acting sites, which therefore very subtly contribute to the regulation of the "switch".

Li, E., Yin, Z., Ma, Y., Li, H., Lin, W., Wei, X., Zhao, R., Jiang, A., Yuan, J., and Zhao, X. Identification and molecular characterization of bacteriophage phiAxp-2 of Achromobacter xylosoxidans. Sci.Rep. 9-27-2016; 6:34300; PM: 27669904

ABSTRACT: A novel Achromobacter xylosoxidans bacteriophage, phiAxp-2, was isolated from hospital sewage in China. The phage was morphologically and microbiologically characterized, and its one-step growth curve, host range, genomic sequence, and receptor were determined. Its morphology showed that phiAxp-2 belongs to the family Siphoviridae. Microbiological characterization demonstrated that pH 7 is most suitable for phage phiAxp-2; its titer decreased when the temperature exceeded 50 degrees C; phiAxp-2 is sensitive to ethanol and isopropanol; and the presence of calcium and magnesium ions is necessary to accelerate cell lysis and improve the formation of phiAxp-2 plaques. Genomic sequencing and a bioinformatic analysis showed that phage phiAxp-2 is a novel bacteriophage, consisting of a circular, double-stranded 62,220-bp DNA molecule with a GC content of 60.11% that encodes 86 putative open reading frames (ORFs). The lipopolysaccharide of A. xylosoxidans is involved in the adsorption of phiAxp-2.

Li, J., Shi, H., Zhao, C., Hao, Y., He, Y., and Sun, Y. Complete Genome Sequence of the Siphoviral Bacteriophage Ec-ZZ2, Which Is Capable of Lysing Enterococcus faecium. Genome Announc. 11-17-2016; 4(6); PM: 27856575

ABSTRACT: A virulent bacteriophage (Ec-ZZ2) that infects Enterococcus faecium was isolated from sewage. The bacteriophage belongs to the family Siphoviridae and has a linear double-stranded DNA genome, with a length of 41,170 bp and a 34.59% G+C content, which is highly similar to Enterococcus phage IME-EF4.

Liu, J. L., Zabetakis, D., Goldman, E. R., and Anderson, G. P. Selection and characterization of single domain antibodies against human CD20. Mol.Immunol. 2016; 78:146-154; PM: 27639717

ABSTRACT: CD20 is a membrane protein with four integral membrane regions and a large extracellular loop between residues 142 and 187, which serves as a target binding region for rituximab (RTX) and most other anti-CD20 monoclonal antibodies. It is highly expressed in B-lymphoma cells and B lymphocytes and often serves as a target for immunotherapy. In this study, we developed single domain antibodies (sdAbs) for the sensitive detection of CD20. To achieve this, an immune sdAb library was prepared from llamas immunized with a fusion between the large loop from CD20 and Hoc, a highly antigenic protein from the T4 bacteriophage, (CD20-Hoc). By subtracting binders to recombinant Hoc during the biopanning, potential anti-CD20 sdAbs were selected, sequenced, and characterized for their binding affinity to CD20-Hoc fusion versus Hoc. Twenty five clones

grouped into three different families based on CDR3 sequence were identified as potential CD20 binders. The binding kinetics of representative sdAbs from each class and RTX were evaluated by surface plasmon resonance (SPR). Most of the sdAbs that were evaluated show binding affinities to CD20-Hoc in the nM range, and class A sdAbs, exhibited >/=40-fold increase in affinity for CD20-Hoc versus Hoc. When the binding of the sdAbs to CD20 on SU-DHL-4 cells was evaluated by flow cytometry, only class A sdAbs displayed strong binding to CD20 and recognized DHL cells in a concentration dependent manner.

Liu, Y., Mi, L., Mi, Z., Huang, Y., Li, P., Zhang, X., Tong, Y., and Bai, C. Complete Genome Sequence of IME207, a Novel Bacteriophage Which Can Lyse Multidrug-Resistant Klebsiella pneumoniae and Salmonella. Genome Announc. 10-27-2016; 4(5); PM: 27789630

ABSTRACT: A novel lytic Salmonella bacteriophage was isolated by using Klebsiella pneumoniae as host cells. The phage's genome was determined to be 47,564 bp and has the highest similarity to Salmonella phage E1 and Salmonella phage 64795_sal3, with coverages of 61% and 56%, respectively. Here, we announce the phage's complete genome.

Liu, Y., Mi, Z., Niu, W., An, X., Yuan, X., Liu, H., Wang, Y., Feng, Y., Huang, Y., Zhang, X., Zhang, Z., Fan, H., Peng, F., Li, P., Tong, Y., and Bai, C. Potential of a lytic bacteriophage to disrupt Acinetobacter baumannii biofilms in vitro. Future.Microbiol. 2016; 11:1383-1393; PM: 27538011

ABSTRACT: **AIM**: The ability of Acinetobacter baumannii to form biofilms and develop antibiotic resistance makes it difficult to control infections caused by this bacterium. In this study, we explored the potential of a lytic bacteriophage to disrupt A. baumannii biofilms. ***MATERIALS & METHODS***: The potential of the lytic bacteriophage to disrupt A. baumannii biofilms was assessed by performing electron microscopy, live/dead bacterial staining, crystal violet staining and by determining adenosine triphosphate release. ***RESULTS***: The bacteriophage inhibited the formation of and disrupted preformed A. baumannii biofilms. Results of disinfection assay showed that the lytic bacteriophage lysed A. baumannii cells suspended in blood or grown on metal surfaces. ***CONCLUSION***: These results suggest the potential of the lytic bacteriophage to disrupt A. baumannii biofilms.

Logger, L., Aschtgen, M. S., Guerin, M., Cascales, E., and Durand, E. Molecular Dissection of the Interface between the Type VI Secretion TssM Cytoplasmic Domain and the TssG Baseplate Component. J.Mol.Biol. 11-6-2016; 428(22):4424-4437; PM: 27600411

ABSTRACT: The type VI secretion system (T6SS) is a multiprotein complex that catalyses toxin secretion through the bacterial cell envelope of various Gram-negative bacteria including important human pathogens. This machine uses a bacteriophage-like contractile tail to puncture the prey cell and inject harmful toxins. The T6SS tail comprises an inner tube capped by the cell-puncturing spike and wrapped by the contractile sheath. This structure is built on an assembly platform, the baseplate, which is anchored

to the bacterial cell envelope by the TssJLM membrane complex (MC). This MC serves as both a tail docking station and a channel for the passage of the inner tube. The TssM transmembrane protein is a key component of the MC as it connects the inner and outer membranes. In this study, we define the TssM topology, highlighting a large but poorly studied 35-kDa cytoplasmic domain, TssMCyto, located between two transmembrane segments. Protein-protein interaction assays further show that TssMCyto oligomerises and makes contacts with several baseplate components. Using computer predictions, we delineate two subdomains in TssMCyto, including a nucleotide triphosphatase (NTPase) domain, followed by a 110-aa extension. Finally, site-directed mutagenesis coupled to functional assays reveals the contribution of these subdomains and conserved motifs to the interaction with T6SS partners and to the function of the secretion apparatus.

Lopez, M. E., Batalha, L. S., Vidigal, P. M., Albino, L. A., Boggione, D. M., Gontijo, M. T., Bazzolli, D. M., and Mendonca, R. C. Genome Sequence of the Enterohemorrhagic Escherichia coli Bacteriophage UFV-AREG1. Genome Announc. 10-13-2016; 4(5); PM: 27738021

ABSTRACT: Here, we present the genome sequence of the Escherichia coli bacteriophage UFV-AREG1. This phage was isolated from cowshed wastewater and showed specificity for enterohemorrhagic E. coli O157:H7 (ATCC 43895), E. coli 0111 (CDC O11ab) and E. coli (ATCC 23229).

Lusiak-Szelachowska, M., Zaczek, M., Weber-Dabrowska, B., Miedzybrodzki, R., Letkiewicz, S., Fortuna, W., Rogoz, P., Szufnarowski, K., Jonczyk-Matysiak, E., Olchawa, E., Walaszek, K. M., and Gorski, A. Antiphage activity of sera during phage therapy in relation to its outcome. Future.Microbiol. 9-19-2016; PM: 27643529

ABSTRACT: ***AIM***: The aim was to study the association between the phage neutralization of patients' sera and the clinical outcome of phage therapy (PT). ***PATIENTS***: About 62 patients with various bacterial infections receiving PT as well as 30 healthy volunteers were studied. ***MATERIALS & METHODS***: Antiphage activity of sera (AAS) was examined using the phage neutralization test of different types of phages before and during PT in relation to the route of phage administration and correlated with the results of PT. ***RESULTS & CONCLUSION***: The analysis of the association between AAS level and clinical results indicated that the level of AAS is not correlated with the outcome of PT.

Ly, M., Jones, M. B., Abeles, S. R., Santiago-Rodriguez, T. M., Gao, J., Chan, I. C., Ghose, C., and Pride, D. T. Transmission of viruses via our microbiomes. Microbiome. 12-2-2016; 4(1):64; PM: 27912785

ABSTRACT: ***BACKGROUND***: Bacteria inhabiting the human body have important roles in a number of physiological processes and are known to be shared amongst genetically-related individuals. Far less is known about viruses inhabiting the human body, but their ecology suggests they may be shared between close contacts. ***RESULTS***: Here, we report the ecology of viruses in the guts and mouths of a cohort and demonstrate that substantial numbers of gut and oral viruses were shared amongst genetically unrelated, cohabitating individuals. Most of these viruses were bacteriophages, and each individual had distinct oral and gut viral ecology from their housemates despite the fact that some

of their bacteriophages were shared. The distribution of bacteriophages over time within households indicated that they were frequently transmitted between the microbiomes of household contacts. ***CONCLUSIONS***: Because bacteriophages may shape human oral and gut bacterial ecology, their transmission to household contacts suggests they could have substantial roles in shaping the microbiota within a household.

Ma, Y. H., Islam, G. S., Wu, Y., Sabour, P. M., Chambers, J. R., Wang, Q., Wu, S. X., and Griffiths, M. W. Temporal distribution of encapsulated bacteriophages during passage through the chick gastrointestinal tract. Poult.Sci. 9-1-2016; PM: 27587723

ABSTRACT: Encapsulation of bacteriophages ("phage") protects phage against environmental deactivation and provides a product that is easy to handle for storage and application with animal feed as an antibiotic alternative. The objective of this study was to evaluate an orally administered, encapsulated phage for efficient phage release in the gastrointestinal tract (GIT) of young chicks receiving feed. An optimized formulation that consisted of 0.8% low molecular weight (MW) alginate, 2% ultra-low molecular weight alginate and 3% whey protein completely released the encapsulated phage within 60 min under simulated intestinal conditions. This product was given to broiler chicks to determine passage time and distribution of the viable phage within the GIT. Following a single oral dose of 109 plaque-forming unit (PFU)/chick, the major portion (peak concentration) of the encapsulated phage passed through the chick's GIT and was detected in the feces within 4 h, with low levels being continuously excreted for up to 24 h. In comparison, the passage of free phage through the GIT occurred faster as indicated by a peak concentration in feces after 1.5 h. In assessing the temporal phage distribution, both encapsulated and free phage treatments showed no apparent difference, both having low levels of 102 to 106 PFU/g of contents along the entire GIT after 1, 2 and 4 h. These low concentrations recovered in vivo led us to examine various exposure conditions (with feed, fecal material, and buffer solutions) that were suspected to have affected phage viability/infectivity during oral delivery, sample recovery, and enumeration by plaque assay. Results showed that the exposure conditions examined did not significantly reduce phage viability and could not account for the observed low phage levels following oral administration in chicks that are on feed. In conclusion, an oral encapsulated phage dose can take more than 4 h to completely move through the GIT of young chicks. Thus, repeated or higher doses may be necessary to attain higher phage concentrations in the GIT.

Maciejewska, B., Roszniowski, B., Espaillat, A., Kesik-Szeloch, A., Majkowska-Skrobek, G., Kropinski, A. M., Briers, Y., Cava, F., Lavigne, R., and Drulis-Kawa, Z. Klebsiella phages representing a novel clade of viruses with an unknown DNA modification and biotechnologically interesting enzymes. Appl.Microbiol.Biotechnol. 10-21-2016; PM: 27766357

ABSTRACT: Lytic bacteriophages and phage-encoded endolysins (peptidoglycan hydrolases) provide a source for the development of novel antimicrobial strategies. In the present study, we focus on the closely related (96 % DNA sequence identity) environmental myoviruses vB_KpnM_KP15 (KP15) and vB_KpnM_KP27 (KP27) infecting multidrug-resistant Klebsiella pneumoniae and Klebsiella oxytoca strains. Their genome organisation and evolutionary relationship are compared to Enterobacter phage phiEap-3 and Klebsiella phages Matisse and Miro. Due to the shared and distinct evolutionary history of these phages, we propose to create a new phage genus

"Kp15virus" within the Tevenvirinae subfamily. In silico genome analysis reveals two unique putative homing endonucleases of KP27 phage, probably involved in unrevealed mechanism of DNA modification and resistance to restriction digestion, resulting in a broader host spectrum. Additionally, we identified in KP15 and KP27 a complete set of lysis genes, containing holin, antiholin, spanin and endolysin. By turbidimetric assays on permeabilized Gram-negative strains, we verified the ability of the KP27 endolysin to destroy the bacterial peptidoglycan. We confirmed high stability, absence of toxicity on a human epithelial cell line and the enzymatic specificity of endolysin, which was found to possess endopeptidase activity, cleaving the peptide stem between L-alanine and D-glutamic acid.

Maervoet, V. E. and Briers, Y. Synthetic biology of modular proteins. Bioengineered. 9-20-2016; 1-7; PM: 27645260

ABSTRACT: The evolution of natural modular proteins and domain swapping by protein engineers have shown the disruptive potential of non-homologous recombination to create proteins with novel functions or traits. Bacteriophage endolysins, cellulosomes and polyketide synthases are 3 examples of natural modular proteins with each module having a dedicated function. These modular architectures have been created by extensive duplication, shuffling of domains and insertion/deletion of new domains. Protein engineers mimic these natural processes in vitro to create chimeras with altered properties or novel functions by swapping modules between different parental genes. Most domain swapping efforts are realized with traditional restriction and ligation techniques, which become particularly restrictive when either a large number of variants, or variants of proteins with multiple domains have to be constructed. Recent advances in homology-independent shuffling techniques increasingly address this need, but to realize the full potential of the synthetic biology of modular proteins a complete homology-independent method for both rational and random shuffling of modules from an unlimited number of parental genes is still needed.

Majkowska-Skrobek, G., Latka, A., Berisio, R., Maciejewska, B., Squeglia, F., Romano, M., Lavigne, R., Struve, C., and Drulis-Kawa, Z. Capsule-Targeting Depolymerase, Derived from Klebsiella KP36 Phage, as a Tool for the Development of Anti-Virulent Strategy. Viruses. 12-1-2016; 8(12); PM: 27916936

ABSTRACT: The rise of antibiotic-resistant Klebsiella pneumoniae, a leading nosocomial pathogen, prompts the need for alternative therapies. We have identified and characterized a novel depolymerase enzyme encoded by Klebsiella phage KP36 (depoKP36), from the Siphoviridae family. To gain insights into the catalytic and structural features of depoKP36, we have recombinantly produced this protein of 93.4 kDa and showed that it is able to hydrolyze a crude exopolysaccharide of a K. pneumoniae host. Using in vitro and in vivo assays, we found that depoKP36 was also effective against a native capsule of clinical K. pneumoniae strains, representing the K63 type, and significantly inhibited Klebsiella-induced mortality of Galleria mellonella larvae in a time-dependent manner. DepoKP36 did not affect the antibiotic susceptibility of Klebsiella strains. The activity of this enzyme was retained in a broad range of pH values (4.0-7.0) and temperatures (up to 45 degrees C). Consistently, the circular dichroism (CD) spectroscopy revealed a highly stability with melting transition temperature (Tm) = 65 degrees C. In contrast to other phage tailspike proteins, this enzyme was susceptible to sodium dodecyl sulfate (SDS) denaturation and proteolytic cleavage. The structural studies in solution showed a trimeric arrangement with a high beta-sheet content. Our

findings identify depoKP36 as a suitable candidate for the development of new treatments for K. pneumoniae infections.

Manrique, P., Bolduc, B., Walk, S. T., van der, Oost J., de Vos, W. M., and Young, M. J. Healthy human gut phageome. Proc.Natl.Acad.Sci.U.S.A. 9-13-2016; 113(37):10400-10405; PM: 27573828

ABSTRACT: The role of bacteriophages in influencing the structure and function of the healthy human gut microbiome is unknown. With few exceptions, previous studies have found a high level of heterogeneity in bacteriophages from healthy individuals. To better estimate and identify the shared phageome of humans, we analyzed a deep DNA sequence dataset of active bacteriophages and available metagenomic datasets of the gut bacteriophage community from healthy individuals. We found 23 shared bacteriophages in more than one-half of 64 healthy individuals from around the world. These shared bacteriophages were found in a significantly smaller percentage of individuals with gastrointestinal/irritable bowel disease. A network analysis identified 44 bacteriophage groups of which 9 (20%) were shared in more than one-half of all 64 individuals. These results provide strong evidence of a healthy gut phageome (HGP) in humans. The bacteriophage community in the human gut is a mixture of three classes: a set of core bacteriophages shared among more than one-half of all people, a common set of bacteriophages found in 20-50% of individuals, and a set of bacteriophages that are either rarely shared or unique to a person. We propose that the core and common bacteriophage communities are globally distributed and comprise the HGP, which plays an important role in maintaining gut microbiome structure/function and thereby contributes significantly to human health.

Manthey, G. M., Clear, A. D., Liddell, L. C., Negritto, M. C., and Bailis, A. M. Homologous recombination in budding yeast expressing the human RAD52 gene reveals a Rad51-independent mechanism of conservative double-strand break repair. Nucleic Acids Res. 12-6-2016; PM: 27923995

ABSTRACT: RAD52 is a homologous recombination (HR) protein that is conserved from bacteriophage to humans. Simultaneously attenuating expression of both the RAD52 gene, and the HR and tumor suppressor gene, BRCA2, in human cells synergistically reduces HR - indicating that RAD52 and BRCA2 control independent mechanisms of HR. We have expressed the human RAD52 gene (HsRAD52) in budding yeast strains lacking the endogenous RAD52 gene and found that HsRAD52 supports repair of DNA double-strand breaks (DSB) by a mechanism of HR that conserves genome structure. Importantly, this mechanism of HR is independent of RAD51, which encodes the central strand exchange protein in yeast required for conservative HR. In contrast, BRCA2 exerts its effect on HR in human cells together with HsRAD51, potentially explaining the synergistic effect of attenuating the expression of both HsRAD52 and BRCA2 This suggests that multiple mechanisms of conservative DSB repair may contribute to tumor suppression in human cells.

Marston, M. F. and Martiny, J. B. Genomic diversification of marine cyanophages into stable ecotypes. Environ.Microbiol. 2016; 18(11):4240-4253; PM: 27696643

ABSTRACT: Understanding the structure and origin of natural bacteriophage genomic diversity is important in elucidating how bacteriophages influence the mortality rates and composition of their host communities. Here, we examine the genetic structure and genomic diversification of naturally occurring bacteriophages by analyzing the full genomic sequences of over 100 isolates of Synechococcus-infecting cyanophages collected over 15 years from coastal waters of Southern New England, USA. Our analysis revealed well-supported cyanophage genomic clusters (genome-wide average nucleotide identity (ANI) >93%) and subclusters (genome-wide ANI >98%) that remained consistent for a decade or longer. Furthermore, by combining the genomic data with genetic analysis of an additional 800 isolates and environmental amplicon sequence data both genomic clusters and subclusters were found to exhibit clear temporal and/or spatial patterns of abundance, suggesting that these units represent distinct viral ecotypes. The processes responsible for diversification of cyanophages into genomic clusters and subclusters were similar across genetic scales and included allelic exchange as well as gene gain and loss. Isolates belonging to different subclusters were found to differ in genes that encoded auxiliary metabolic functions, restriction modification enzymes, and virion structural proteins, although the specific traits and selection pressures responsible for the maintenance of distinct ecotypes remain unknown.

Maslanova, I., Stribna, S., Doskar, J., and Pantucek, R. Efficient plasmid transduction to Staphylococcus aureus strains insensitive to the lytic action of transducing phage. FEMS Microbiol.Lett. 2016; 363(19); PM: 27609232

ABSTRACT: The transduction mediated by bacteriophages is considered to be one of the primary driving forces in horizontal gene transfer in staphylococci, which is crucial to their adaptation and successful evolution. For a transduction to be effective, it is generally accepted that the recipient strain should be susceptible to the transducing phage. In this study, we demonstrate that the plasmid DNAs are effectively transduced into the recipient Staphylococcus aureus strains in spite of their insensitivity to the lytic action of the transducing phage, provided that these phages adsorb effectively to the bacterial cells. The tetracycline and penicillinase plasmids were transduced to insensitive laboratory and clinical strains by bacteriophages varphi29, varphi52A and varphi80alpha as well as by prophage varphi53 and naturally occurring prophages induced from donor lysogenic strains. Comparable frequencies of transduction were achieved in both phage-sensitive and phage-insensitive recipient strains. We have demonstrated that such mechanisms as the restriction of DNA and lysogenic immunity which are responsible for insensitivity of cells to phages may not be a barrier to the transfer, maintenance and effective spread of plasmids to a wider range of potential recipients in the staphylococcal population.

McLellan, N. L., Lee, H., and Habash, M. B. Evaluation of propidium monoazide and long-amplicon qPCR as an infectivity assay for coliphage. J.Virol.Methods. 2016; 238:48-55; PM: 27744093

ABSTRACT: Standardized and rapid assays for viable viral pathogens are needed to inform human health risk assessments. Conventional qPCR is designed to enumerate the

gene copies of an organism in a sample, but does not identify those that originated from a viable pathogen. This study was undertaken to evaluate modified qPCR methods as infectivity assays for the enumeration of infectious MS2 coliphage. Propidium monoazide (PMA) treatment coupled with long-amplicon qPCR assays were assessed for their ability to quantify infectious MS2 in pure cultures and following inactivation by a range of UV light exposures and chlorine doses. The qPCR results were compared to the plaque assay, which was used as the standard to indicate the level of infectious MS2 in each sample. For pure cultures, PMA-qPCR results were not significantly different from the plaque assay ($p>0.05$). At >4 log inactivation, combined PMA and long-amplicon qPCR assays overestimated the level of infectious MS2 remaining ($p<0.05$). The most accurate long-amplicon qPCR infectivity assay targeted a 624-bp region at the 5' end of the genome. Modified qPCR approaches may be useful tools to monitor the loss of infectivity as a result of disinfection processes.

Mehta, A. P., Li, H., Reed, S. A., Supekova, L., Javahishvili, T., and Schultz, P. G. Replacement of 2'-Deoxycytidine by 2'-Deoxycytidine Analogues in the E. coli Genome. J.Am.Chem.Soc. 11-2-2016; 138(43):14230-14233; PM: 27762133

ABSTRACT: Several modified bases have been observed in the genomic DNA of bacteriophages, prokaryotes, and eukaryotes that play a role in restriction systems and/or epigenetic regulation. In our efforts to understand the consequences of replacing a large fraction of a canonical nucleoside with a modified nucleoside, we previously replaced around 75% of thymidine (T) with 5'-hydroxymethyl-2'-deoxyuridine (5hmU) in the Escherichia coli genome. In this study, we engineered the pyrimidine nucleotide biosynthetic pathway using T4 bacteriophage genes to achieve approximately 63% replacement of 2'-deoxycytidine (dC) with 5-hydroxymethyl-2'-deoxycytidine (5hmC) in the E. coli genome and approximately 71% replacement in plasmids. We further engineered the glucose metabolic pathway to transform the 5hmC into glucosyl-5-hydroxymethyl-2'-deoxycytidine (5-gmC) and achieved 20% 5-gmC in the genome and 45% 5-gmC in plasmid DNA.

Merrill, B. D., Ward, A. T., Grose, J. H., and Hope, S. Software-based analysis of bacteriophage genomes, physical ends, and packaging strategies. BMC.Genomics. 8-26-2016; 17:679; PM: 27561606

ABSTRACT: ***BACKGROUND***: Phage genome analysis is a rapidly growing field. Recurrent obstacles include software access and usability, as well as genome sequences that vary in sequence orientation and/or start position. Here we describe modifications to the phage comparative genomics software program, Phamerator, provide public access to the code, and include instructions for creating custom Phamerator databases. We further report genomic analysis techniques to determine phage packaging strategies and identification of the physical ends of phage genomes. ***RESULTS***: The original Phamerator code can be successfully modified and custom databases can be generated using the instructions we provide. Results of genome map comparisons within a custom database reveal obstacles in performing the comparisons if a published genome has an incorrect complementarity or an incorrect location of the first base of the genome, which are common issues in GenBank-downloaded sequence files. To address these issues, we review phage packaging strategies and provide results that demonstrate identification of the genome start location and orientation using raw sequencing data and software programs such as PAUSE and Consed to establish the

location of the physical ends of the genome. These results include determination of exact direct terminal repeats (DTRs) or cohesive ends, or whether phages may use a headful packaging strategy. Phylogenetic analysis using ClustalO and phamily circles in Phamerator demonstrate that the large terminase gene can be used to identify the phage packaging strategy and thereby aide in identifying the physical ends of the genome. ***CONCLUSIONS***: Using available online code, the Phamerator program can be customized and utilized to generate databases with individually selected genomes. These databases can then provide fruitful information in the comparative analysis of phages. Researchers can identify packaging strategies and physical ends of phage genomes using raw data from high-throughput sequencing in conjunction with phylogenetic analyses of large terminase proteins and the use of custom Phamerator databases. We promote publication of phage genomes in an orientation consistent with the physical structure of the phage chromosome and provide guidance for determining this structure.

Meyer, J. R., Dobias, D. T., Medina, S. J., Servilio, L., Gupta, A., and Lenski, R. E. Ecological speciation of bacteriophage lambda in allopatry and sympatry. Science. 11-24-2016; PM: 27884940

ABSTRACT: Understanding the conditions that allow speciation to occur is difficult because most research has focused on either long-lived organisms or asexual microorganisms. We propagated bacteriophage lambda, a virus with rapid generations and frequent recombination, on two Escherichia coli host genotypes that expressed either the LamB or OmpF receptor. When supplied with either single host (allopatry), lambda improved its binding to the available receptor while losing its ability to use the alternative. When evolving on both hosts together (sympatry), the viruses split into two lineages with divergent receptor preferences. Although the level of divergence varied among replicates, some lineages evolved reproductive isolation via genetic incompatibilities. This outcome indicates that, under suitable conditions, allopatric and sympatric speciation can occur with similar ease.

Miller, C. R., Nagel, A. C., Scott, L., Settles, M., Joyce, P., and Wichman, H. A. Love the one you're with: replicate viral adaptations converge on the same phenotypic change. PeerJ. 2016; 4:e2227; PM: 27547540

ABSTRACT: Parallelism is important because it reveals how inherently stochastic adaptation is. Even as we come to better understand evolutionary forces, stochasticity limits how well we can predict evolutionary outcomes. Here we sought to quantify parallelism and some of its underlying causes by adapting a bacteriophage (ID11) with nine different first-step mutations, each with eight-fold replication, for 100 passages. This was followed by whole-genome sequencing five isolates from each endpoint. A large amount of variation arose-281 mutational events occurred representing 112 unique mutations. At least 41% of the mutations and 77% of the events were adaptive. Within wells, populations generally experienced complex interference dynamics. The genome locations and counts of mutations were highly uneven: mutations were concentrated in two regulatory elements and three genes and, while 103 of the 112 (92%) of the mutations were observed in </=4 wells, a few mutations arose many times. 91% of the wells and 81% of the isolates had a mutation in the D-promoter. Parallelism was moderate compared to previous experiments with this system. On average, wells shared 27% of their mutations at the DNA level and 38% when the definition of parallel change is

expanded to include the same regulatory feature or residue. About half of the parallelism came from D-promoter mutations. Background had a small but significant effect on parallelism. Similarly, an analyses of epistasis between mutations and their ancestral background was significant, but the result was mostly driven by four individual mutations. A second analysis of epistasis focused on de novo mutations revealed that no isolate ever had more than one D-promoter mutation and that 56 of the 65 isolates lacking a D-promoter mutation had a mutation in genes D and/or E. We assayed time to lysis in four of these mutually exclusive mutations (the two most frequent D-promoter and two in gene D) across four genetic backgrounds. In all cases lysis was delayed. We postulate that because host cells were generally rare (i.e., high multiplicity of infection conditions developed), selection favored phage that delayed lysis to better exploit their current host (i.e., 'love the one you're with'). Thus, the vast majority of wells (at least 64 of 68, or 94%) arrived at the same phenotypic solution, but through a variety of genetic changes. We conclude that answering questions about the range of possible adaptive trajectories, parallelism, and the predictability of evolution requires attention to the many biological levels where the process of adaptation plays out.

Moon, J. S., Lee, Y., Shin, D. M., Kim, C., Kim, W. G., Park, M., Han, J., Song, H., Kim, K., and Oh, J. W. Identification of Endocrine Disrupting Chemicals using a Virus-Based Colorimetric Sensor. Chem.Asian J. 11-7-2016; 11(21):3097-3101; PM: 27616055

ABSTRACT: A simple and portable colorimetric sensor based on M13 bacteriophage (phage) was devised to identify a class of endocrine disrupting chemicals, including benzene, phthalate, and chlorobenzene derivatives. Arrays of structurally and genetically modified M13 bacteriophage were fabricated so as to produce a biomimetic colorimetric sensor, and color changes in the phage arrays in response to several benzene derivatives were characterized. The sensor was also used to classify phthalate and chlorobenzene derivatives as representatives of endocrine disrupting chemicals. The characteristic color patterns obtained on exposure to various benzene derivatives enabled similar chemical structures in the vapor phase to be classified. Our sensing approach based on the use of a genetically surface modified M13 bacteriophage offers a promising platform for portable, simple environmental monitors that could be extended for use in numerous application areas, including food monitoring, security monitoring, explosive risk assessment, and point of care testing.

Moore, A. W., Zytner, R. G., and Chang, S. Potential Water Reuse for High Strength Fruit and Vegetable Processor Wastewater with an MBR. Water Environ.Res. 2016; 88(9):852-870; PM: 27654083

ABSTRACT: High strength food processing wastewater from two processing plants was studied to determine the effectiveness of an aerobic membrane bioreactor (MBR) to reduce BOD, TSS and nutrients below municipal sewer discharge limits. The MBR comprised a 20 L lab-scale reactor combined with a flat sheet, ultrafiltration membrane module. The parameters studied included the operational flux, solids and hydraulic retention times and recirculation ratio with regards to nitrification/denitrification. The MBR system provided excellent removal efficiency at 97% COD, 99% BOD, 99.9% TSS, 90% TKN, and 60% TP for both processing plants, which eliminated the surcharges, allowing the firms to stay competitive. Effluent reuse tests showed that activated carbon proved effective in removing color from the MBR permeate, while UV treatment was able to

achieve a 5 log reduction in bacteriophage. Overall, these treatment successes show the potential for water reuse in the agrifood sector.

Morley, D., Broniewski, J. M., Westra, E. R., Buckling, A., and van, Houte S. Host diversity limits the evolution of parasite local adaptation. Mol.Ecol. 11-14-2016; PM: 27862566

ABSTRACT: Specificity in the interactions between hosts and their parasites can lead to local adaptation. However, the degree of local adaptation is predicted to depend upon the diversity of resistance alleles within the host population; increasing host diversity should decrease mean parasite infectivity and hence reduce local adaptation. In this study, we empirically test this prediction using the highly specific interactions between bacteria with clustered regularly interspaced short palindromic repeats/CRISPR-associated (CRISPR/Cas) immunity and their bacteriophage. Bacteria acquire immunity to phage by incorporating a phage-derived spacer sequence into CRISPR loci on the host genome, and phage can escape the CRISPR-mediated immunity of a specific clone by mutating the targeted sequence. We found that high levels of CRISPR allele diversity that naturally evolve in host populations exposed to phage (because each bacterial clone captures a unique phage-derived sequence) prevents phage from becoming locally adapted. By manipulating the number of CRISPR alleles in the host population, we show that phage can become locally adapted to their bacterial hosts but only when CRISPR allele diversity is low.

Mostafa, M. M., Nassef, M., and Badr, A. Computational determination of the effects of virulent Escherichia coli and salmonella bacteriophages on human gut. Comput.Methods Programs Biomed. 2016; 135:27-35; PM: 27586477

ABSTRACT: ***BACKGROUND AND OBJECTIVE***: Salmonella and Escherichia coli are different types of bacteria that cause food poisoning in humans. In the elderly, infants and people with chronic conditions, it is very dangerous if Salmonella or E. coli gets into the bloodstream and then they must be treated by phage therapy. Treating Salmonella and E. coli by phage therapy affects the gut flora. This research paper presents a system for detecting the effects of virulent E. coli and Salmonella bacteriophages on human gut. ***METHODS***: A method based on Domain-Domain Interactions (DDIs) model is implemented in the proposed system to determine the interactions between the proteins of human gut bacteria and the proteins of bacteriophages that infect virulent E. coli and Salmonella. The system helps gastroenterologists to realize the effect of injecting bacteriophages that infect virulent E. coli and Salmonella on the human gut. ***RESULTS***: By testing the system over Enterobacteria phage 933W, Enterobacteria phage VT2-Sa and Enterobacteria phage P22, it resulted in four interactions between the proteins of the bacteriophages that infect E. coli O157:H7, E. coli O104:H4 and Salmonella typhimurium and the proteins of human gut bacterium strains. ***CONCLUSION***: Several effects were detected such as: antibacterial activity against a number of bacterial species in human gut, regulation of cellular differentiation and organogenesis during gut, lung, and heart development, ammonia assimilation in bacteria, yeasts, and plants, energizing defense system and its function in the detoxification of lipopolysaccharide, and in the prevention of bacterial translocation in human gut.

Myga-Nowak, M., Godela, A., Glab, T., Lewanska, M., and Boratynski, J. Bacteriophages to combat foodborne infections caused by food contamination by bacteria of the

Campylobacter genus. Postepy Hig.Med.Dosw.(Online.). 9-26-2016; 70(0):989-1000; PM: 27668651

ABSTRACT: It is estimated that each year more than 2 million people suffer from diarrheal diseases, resulting from the consumption of contaminated meat. Foodborne infections are most frequently caused by small Gram-negative rods Campylobacter. The hosts of these bacteria are mainly birds wherein they are part of the normal intestinal flora. During the commercial slaughter, there is a likelihood of contamination of carcasses by the bacteria found in the intestinal content. In Europe, up to 90% of poultry flocks can be a reservoir of the pathogen. According to the European Food Safety Authority report from 2015, the number of reported and confirmed cases of human campylobacteriosis exceeds 200 thousands per year, and such trend remains at constant level for several years. The occurrence of growing antibiotic resistance in bacteria forces the limitation of antibiotic use in the animal production. Therefore, the European Union allows only using stringent preventive and hygienic treatment on farms. Achieving Campylobacter free chickens using these methods is possible, but difficult to implement and expensive. Utilization of bacterial viruses - bacteriophages, can be a path to provide the hygienic conditions of poultry production and food processing. Formulations applied in the food protection should contain strictly lytic bacteriophages, be non-pyrogenic and retain long lasting biological activity. Currently, on the market there are available commercial bacteriophage preparations for agricultural use, but neither includes phages against Campylobacter. However, papers on the application of bacteriophages against Campylobacter in chickens and poultry products were published in the last few years. In accordance with the estimates, 2-logarithm reduction of Campylobacter in poultry carcases will contribute to the 30-fold reduction in the incidence of campylobacteriosis in humans. Research on bacteriophages against Campylobacter have cognitive and economic importance. The paper presents current state of research on bacteriophages targeted against Campylobacter.

Nale, J. Y., Chutia, M., Carr, P., Hickenbotham, P. T., and Clokie, M. R. 'Get in Early'; Biofilm and Wax Moth (Galleria mellonella) Models Reveal New Insights into the Therapeutic Potential of Clostridium difficile Bacteriophages. Front Microbiol. 2016; 7:1383; PM: 27630633

ABSTRACT: Clostridium difficile infection (CDI) is a global health threat associated with high rates of morbidity and mortality. Conventional antibiotic CDI therapy can result in treatment failure and recurrent infection. C. difficile produces biofilms which contribute to its virulence and impair antimicrobial activity. Some bacteriophages (phages) can penetrate biofilms and thus could be developed to either replace or supplement antibiotics. Here, we determined the impact of a previously optimized 4-phage cocktail on C. difficile ribotype 014/020 biofilms, and additionally as adjunct to vancomycin treatment in Galleria mellonella larva CDI model. The phages were applied before or after biofilm establishment in vitro, and the impact was analyzed according to turbidity, viability counts and topography as observed using scanning electron and confocal microscopy. The infectivity profiles and efficacies of orally administered phages and/or vancomycin were ascertained by monitoring colonization levels and larval survival rates. Phages prevented biofilm formation, and penetrated established biofilms. A single phage application reduced colonization causing extended longevity in the remedial treatment and prevented disease in the prophylaxis group. Multiple phage doses significantly improved the larval remedial regimen, and this treatment is comparable to vancomycin and the combined treatments. Taken together, our data suggest that the phages significantly reduce C. difficile biofilms, and prevent colonization in the G.

mellonella model when used alone or in combination with vancomycin. The phages appear to be highly promising therapeutics in the targeted eradication of CDI and the use of these models has revealed that prophylactic use could be a propitious therapeutic option.

Nale, J. Y., Spencer, J., Hargreaves, K. R., Buckley, A. M., Trzepinski, P., Douce, G. R., and Clokie, M. R. Bacteriophage Combinations Significantly Reduce Clostridium difficile Growth In Vitro and Proliferation In Vivo. Antimicrob.Agents Chemother. 2016; 60(2):968-981; PM: 26643348

ABSTRACT: The microbiome dysbiosis caused by antibiotic treatment has been associated with both susceptibility to and relapse of Clostridium difficile infection (CDI). Bacteriophage (phage) therapy offers target specificity and dose amplification in situ, but few studies have focused on its use in CDI treatment. This mainly reflects the lack of strictly virulent phages that target this pathogen. While it is widely accepted that temperate phages are unsuitable for therapeutic purposes due to their transduction potential, analysis of seven C. difficile phages confirmed that this impact could be curtailed by the application of multiple phage types. Here, host range analysis of six myoviruses and one siphovirus was conducted on 80 strains representing 21 major epidemic and clinically severe ribotypes. The phages had complementary coverage, lysing 18 and 62 of the ribotypes and strains tested, respectively. Single-phage treatments of ribotype 076, 014/020, and 027 strains showed an initial reduction in the bacterial load followed by the emergence of phage-resistant colonies. However, these colonies remained susceptible to infection with an unrelated phage. In contrast, specific phage combinations caused the complete lysis of C. difficile in vitro and prevented the appearance of resistant/lysogenic clones. Using a hamster model, the oral delivery of optimized phage combinations resulted in reduced C. difficile colonization at 36 h postinfection. Interestingly, free phages were recovered from the bowel at this time. In a challenge model of the disease, phage treatment delayed the onset of symptoms by 33 h compared to the time of onset of symptoms in untreated animals. These data demonstrate the therapeutic potential of phage combinations to treat CDI.

Negut, A. C., Chifiriuc, M. C., Sandulescu, O., Streinu-Cercel, A., Oprea, M., Dragulescu, E. C., Gheorghe, I., Berciu, I., Coralia, B., Popa, M., Otelea, D., Talapan, D., Dorobat, O., Codita, I., Popa, M. I., and Streinu-Cercel, A. Bacteriophage-driven inhibition of biofilm formation in Staphylococcus strains from patients attending a Romanian reference center for infectious diseases. FEMS Microbiol.Lett. 2016; 363(18); PM: 27521261

ABSTRACT: The increasing burden of invasive biofilm-related staphylococcal infections has led to a dire need for new agents to prevent biofilm formation. Bacteriophages may hypothetically alter a biofilm through several mechanisms, including induction of depolymerizing enzymes and lysis of persistent bacteria. We have assessed the influence of commercially available bacteriophage cocktails on Staphylococcus spp. clinical strains viability and biofilm formation. We analyzed 83 staphylococcal strains from patients consecutively admitted to a Romanian infection reference center from October 2014 through May 2015; the strains were characterized by phenotypic and genetic tools for their resistance and virulence features and for their phyliation. Experiments were performed in triplicate. Methicillin-susceptible strains were significantly more susceptible to all tested phages: 1.7-fold higher susceptibility for PYO, 1.4-fold for INTESTI, 2.9-fold for PHAGYO, 2.7-fold for PHAGESTI and 3.9-fold for STAPHYLOCOCCAL; t030 strains were significantly more susceptible to PYO and INTESTI compared with t127 strains. We

identified a significant decrease in biofilm formation in the presence of both low and high PYO and INTESTI concentrations ($P < 0.001$). In conclusion, Staphylococcus strains from Romania displayed fairly good susceptibility to commercially available bacteriophages. We have also ascertained there is phage-driven in vitro inhibition of biofilm formation, the results potentially impacting prevention of prosthetic infections.

Nzila, A., Razzak, S. A., and Zhu, J. Bioaugmentation: An Emerging Strategy of Industrial Wastewater Treatment for Reuse and Discharge. Int.J.Environ.Res.Public Health. 8-25-2016; 13(9); PM: 27571089

ABSTRACT: A promising long-term and sustainable solution to the growing scarcity of water worldwide is to recycle and reuse wastewater. In wastewater treatment plants, the biodegradation of contaminants or pollutants by harnessing microorganisms present in activated sludge is one of the most important strategies to remove organic contaminants from wastewater. However, this approach has limitations because many pollutants are not efficiently eliminated. To counterbalance the limitations, bioaugmentation has been developed and consists of adding specific and efficient pollutant-biodegrading microorganisms into a microbial community in an effort to enhance the ability of this microbial community to biodegrade contaminants. This approach has been tested for wastewater cleaning with encouraging results, but failure has also been reported, especially during scale-up. In this review, work on the bioaugmentation in the context of removal of important pollutants from industrial wastewater is summarized, with an emphasis on recalcitrant compounds, and strategies that can be used to improve the efficiency of bioaugmentation are also discussed. This review also initiates a discussion regarding new research areas, such as nanotechnology and quorum sensing, that should be investigated to improve the efficiency of wastewater bioaugmentation.

Oduor, J. M., Onkoba, N., Maloba, F., Arodi, W. O., and Nyachieo, A. Efficacy of lytic Staphylococcus aureus bacteriophage against multidrug-resistant Staphylococcus aureus in mice. J.Infect.Dev.Ctries. 11-24-2016; 10(11):1208-1213; PM: 27886033

ABSTRACT: ***INTRODUCTION***: The use of bacteriophages as an alternative treatment method against multidrug-resistant bacteria has not been explored in Kenya. This study sought to determine the efficacy of environmentally obtained lytic bacteriophage against multidrug-resistant Staphylococcus aureus (MDRSA) bacterium in mice. ***METHODOLOGY***: Staphylococcus aureus bacterium and S. aureus-specific lytic phage were isolated from sewage and wastewater collected within Nairobi County, Kenya. Thirty mice were randomly assigned into three groups: MDRSA infection group (n = 20), phage-infection group (n = 5), and non-infection group (n = 5). The MDRSA infection group was further subdivided into three groups: clindamycin treatment (8 mg/kg; n = 5), lytic phage treatment (108 PFU/mL (n = 5), and a combination treatment of clindamycin and lytic phage (n = 5). Treatments were done at either 24 or 72 hours post-infection (p.i), and data on efficacy, bacterial load, and animal physical health were collected. ***RESULTS***: Treatment with phage was more effective (100%) than with clindamycin (62.25% at 24 hours p.i and 87.5% at 72 hours p.i.) or combination treatment (75% at 24 hours p.i. and 90% at 72 hours p.i.) ($p < 0.001$). ***CONCLUSIONS***: The results show that the environmentally obtained S. aureus lytic bacteriophage has therapeutic potential against MDRSA bacterium in mice.

Ozaki, T., Abe, N., Kimura, K., Suzuki, A., and Kaneko, J. Genomic analysis of Bacillus subtilis lytic bacteriophage varphiNIT1 capable of obstructing natto fermentation carrying genes for the capsule-lytic soluble enzymes poly-gamma-glutamate hydrolase and levanase. Biosci.Biotechnol.Biochem. 2017; 81(1):135-146; PM: 27885938

ABSTRACT: Bacillus subtilis strains including the fermented soybean (natto) starter produce capsular polymers consisting of poly-gamma-glutamate and levan. Capsular polymers may protect the cells from phage infection. However, bacteriophage varphiNIT1 carries a gamma-PGA hydrolase gene (pghP) that help it to counteract the host cell's protection strategy. varphiNIT had a linear double stranded DNA genome of 155,631-bp with a terminal redundancy of 5,103-bp, containing a gene encoding an active levan hydrolase. These capsule-lytic enzyme genes were located in the possible foreign gene cluster regions between central core and terminal redundant regions, and were expressed at the late phase of the phage lytic cycle. All tested natto origin Spounavirinae phages carried both genes for capsule degrading enzymes similar to varphiNIT1. A comparative genomic analysis revealed the diversity among varphiNIT1 and Bacillus phages carrying pghP-like and levan-hydrolase genes, and provides novel understanding on the acquisition mechanism of these enzymatic genes.

Ozkan, I., Akturk, E., Yeshenkulov, N., Atmaca, S., Rahmanov, N., and Atabay, H. I. Lytic Activity of Various Phage Cocktails on Multidrug-Resistant Bacteria. Clin.Invest Med. 12-1-2016; 39(6):27504; PM: 27917795

ABSTRACT: ***PURPOSE***: A bacteriophage is a virus that infects and replicates within a bacterium following the injection of phage genome into the bacterial cytoplasm. They are seen as a possible therapy for multi-drug-resistant strains of many bacteria. The aim of this study is to evaluate the lytic activity of the Pyo, Intesti and Fersisi bacteriophage cocktails on P. aeruginosa and S. aureus. ***METHODS***: Ten different S. aureus and P. aeruginosa strains, which were isolated from hospitalized patients in Turkey, were used in the study. The identification and antibiotic susceptibility of the isolates were performed using Vitec 2 system. The identities of the isolates were confirmed by a species-specific Polymerase Chain Reaction (PCR) assay. Lytic activity of the bacteriophage cocktails on bacteria was determined by spot test and plaque assay methods. ***RESULTS***: The lytic activity of the Pyo phage cocktail was evaluated on P. aeruginosa and S. aureus strains. It was found that eight isolates of MDR S. aureus were susceptible to Pyo phage cocktail and two isolates were resistant. Nine isolates of antibiotic-resistant P. aeruginosa were found to be susceptible to this phage cocktail and one isolate was resistant. Thus, the Pyo, Intesti and Fersisi cocktails are very effective in treating clinical strains of multidrug-resistant P. aeruginosa and S. aureus isolated in Turkey. ***CONCLUSION***: The Pyo, Intesti and Fersisi cocktails may prove useful in the treatment of various infections caused by those bacteria.

Pabary, R., Singh, C., Morales, S., Bush, A., Alshafi, K., Bilton, D., Alton, E. W., Smithyman, A., and Davies, J. C. Antipseudomonal Bacteriophage Reduces Infective Burden and Inflammatory Response in Murine Lung. Antimicrob.Agents Chemother. 2016; 60(2):744-751.; PM: 26574007

ABSTRACT: As antibiotic resistance increases, there is a need for new therapies to treat infection, particularly in cystic fibrosis (CF), where Pseudomonas aeruginosa is a ubiquitous pathogen associated with increased morbidity and mortality. Bacteriophages

are an attractive alternative treatment, as they are specific to the target bacteria and have no documented side effects. The efficacy of phage cocktails was established in vitro. Two P. aeruginosa strains were taken forward into an acute murine infection model with bacteriophage administered either prophylactically, simultaneously, or postinfection. The infective burden and inflammation in bronchoalveolar lavage fluid (BALF) were assessed at various times. With low infective doses, both control mice and those undergoing simultaneous phage treatment cleared P. aeruginosa infection at 48 h, but there were fewer neutrophils in BALF of phage-treated mice (median, 73.2 x 10(4)/ml [range, 35.2 to 102.1 x 10(4)/ml] versus 174 x 10(4)/ml [112.1 to 266.8 x 10(4)/ml], P < 0.01 for the clinical strain; median, 122.1 x 10(4)/ml [105.4 to 187.4 x 10(4)/ml] versus 206 x 10(4)/ml [160.1 to 331.6 x 10(4)/ml], P < 0.01 for PAO1). With higher infective doses of PAO1, all phage-treated mice cleared P. aeruginosa infection at 24 h, whereas infection persisted in all control mice (median, 1,305 CFU/ml [range, 190 to 4,700 CFU/ml], P < 0.01). Bacteriophage also reduced CFU/ml in BALF when administered postinfection (24 h) and both CFU/ml and inflammatory cells in BALF when administered prophylactically. A reduction in soluble inflammatory cytokine levels in BALF was also demonstrated under different conditions. Bacteriophages are efficacious in reducing both the bacterial load and inflammation in a murine model of P. aeruginosa lung infection. This study provides proof of concept for future clinical trials in patients with CF.

Paff, M. L., Nuismer, S. L., Ellington, A. D., Molineux, I. J., May, R. H., and Bull, J. J. Design and engineering of a transmissible antiviral defense. J.Biol.Eng. 2016; 10:12PM: 27752283

ABSTRACT: ***BACKGROUND***: We propose, model, and implement a novel system of population-level intervention against a virus. One context is a treatment against a chronic infection such as HIV. The underlying principle is a form of virus 'wars' in which a benign, transmissible agent is engineered to protect against infection by and spread of a lethal virus. In our specific case, the protective agent consists of two entities, a benign virus and a gene therapy vector mobilized by the benign virus. ***RESULTS***: Numerical analysis of a mathematical model identified parameter ranges in which adequate, population-wide protection is achieved. The protective system was implemented and tested using E. coli, bacteriophage M13 and a phagemid vector mobilized by M13 to block infection by the lethal phage T5. Engineering of M13 profoundly improved its dynamical properties for facilitating spread of the gene therapy vector. However, the gene therapy vector converts the host cell to resist T5 too slowly for protection on a time scale appropriate for T5. ***CONCLUSIONS***: Overall, there is a reasonable marriage between the mathematical model and the empirical system, suggesting that such models can be useful guides to the design of such systems even before the models incorporate most of the relevant biological details.

Paradiso, R., Lombardi, S., Iodice, M. G., Riccardi, M. G., Orsini, M., Bolletti, Censi S., Galiero, G., and Borriello, G. Complete Genome Sequence of a Lytic Siphoviridae Bacteriophage Infecting Several Serovars of Salmonella enterica. Genome Announc. 9-29-2016; 4(5); PM: 27688334

ABSTRACT: The bacteriophage 100268_sal2 was isolated from water buffalo feces in southern Italy, exhibiting lytic activity against several subspecies of Salmonella enterica This bacteriophage belongs to the Siphoviridae family and has a 125,114-bp double-stranded DNA (ds-DNA) genome containing 188 coding sequences (CDSs).

Paradiso, R., Orsini, M., Bolletti, Censi S., Borriello, G., and Galiero, G. Complete Genome Sequence of a Myoviridae Bacteriophage Infecting Salmonella enterica Serovar Typhimurium. Genome Announc. 9-29-2016; 4(5); PM: 27688333

ABSTRACT: The bacteriophage 118970_sal3 was isolated from water buffalo feces in southern Italy, exhibiting lytic activity against Salmonella enterica serovar Typhimurium. This bacteriophage belongs to the Myoviridae family and has a 39,464-bp double-stranded DNA (ds-DNA) genome containing 53 coding sequences (CDSs).

Park, H. J., Oh, S., Vinod, N., Ji, S., Noh, H. B., Koo, J. M., Lee, S. H., Kim, S. C., Lee, K. S., and Choi, C. W. Characterization of Chemically-Induced Bacterial Ghosts (BGs) Using Sodium Hydroxide-Induced Vibrio parahaemolyticus Ghosts (VPGs). Int.J.Mol.Sci. 11-15-2016; 17(11); PM: 27854308

ABSTRACT: Acellular bacterial ghosts (BGs) are empty non-living bacterial cell envelopes, commonly generated by controlled expression of the cloned lysis gene E of bacteriophage PhiX174. In this study, Vibrio parahaemolyticus ghosts (VPGs) were generated by chemically-induced lysis and the method is based on minimum inhibitory concentration (MIC) of sodium hydroxide (NaOH), acetic acid, boric acid, citric acid, maleic acid, hydrochloric acid, and sulfuric acid. The MIC values of the respective chemicals were 3.125, 6.25, <50.0, 25.0, 6.25, 1.56, and 0.781 mg/mL. Except for boric acid, the lysis efficiency reached more than 99.99% at 5 min after treatment of all chemicals. Among those chemicals, NaOH-induced VPGs appeared completely DNA-free, which was confirmed by quantitative real-time PCR. Besides, lipopolysaccharides (LPS) extracted from the NaOH-induced VPGs showed no distinctive band on SDS-PAGE gel after silver staining. On the other hand, LPS extracted from wild-type bacterial cells, as well as the organic acids-induced VPGs showed triple major bands and LPS extracted from the inorganic acids-induced VPGs showed double bands. It suggests that some surface structures in LPS of the NaOH-induced VPGs may be lost, weakened, or modified by the MIC of NaOH. Nevertheless, Limulus amoebocyte lysate assay revealed that there is no significant difference in endotoxic activity between the NaOH-induced VPGs and wild-type bacterial cells. Macrophages exposed to the NaOH-induced VPGs at 0.5 x 10(6) CFU/mL showed cell viability of 97.9%, however, the MIC of NaOH did not reduce the cytotoxic effect of wild-type bacterial cells. Like Escherichia coli LPS, the NaOH-induced VPGs are an excellent activator of pro-inflammatory cytokines (IL-1beta and iNOS), anti-inflammatory cytokine (IL-10), and dual activities (IL-6) in the stimulated macrophage cells. On the other hand, the induction of TNF-alpha mRNA was remarkable in the macrophages exposed with wild-type cells. Scanning electron microscopy showed the formation of trans-membrane lysis tunnel structures in the NaOH-induced VPGs. SDS-PAGE and agarose gel electrophoresis also confirmed that cytoplasmic proteins and genomic DNA released from the VPGs to culture medium through the lysis tunnel structures. Taken together, all these data indicate that the NaOH-induced VPGs show the potency of a safe, economical, and effective inactivated bacterial vaccine candidate.

Parker, M. T. An Ecological Framework of the Human Virome Provides Classification of Current Knowledge and Identifies Areas of Forthcoming Discovery. Yale J.Biol.Med. 2016; 89(3):339-351; PM: 27698618

ABSTRACT: Recent advances in sequencing technologies have opened the door for the classification of the human virome. While taxonomic classification can be applied to the viruses identified in such studies, this gives no information as to the type of interaction the virus has with the host. As follow-up studies are performed to address these questions, the description of these virus-host interactions would be greatly enriched by applying a standard set of definitions that typify them. This paper describes a framework with which all members of the human virome can be classified based on principles of ecology. The scaffold not only enables categorization of the human virome, but can also inform research aimed at identifying novel virus-host interactions.

Parmar, K. M., Hathi, Z. J., and Dafale, N. A. Control of Multidrug-Resistant Gene Flow in the Environment Through Bacteriophage Intervention. Appl.Biochem.Biotechnol. 10-8-2016; PM: 27723009

ABSTRACT: The spread of multidrug-resistant (MDR) bacteria is an emerging threat to the environment and public wellness. Inappropriate use and indiscriminate release of antibiotics in the environment through un-metabolized form create a scenario for the emergence of virulent pathogens and MDR bugs in the surroundings. Mechanisms underlying the spread of resistance include horizontal and vertical gene transfers causing the transmittance of MDR genes packed in different host, which pass across different food webs. Several controlling agents have been used for combating pathogens; however, the use of lytic bacteriophages proves to be one of the most eco-friendly due to their specificity, killing only target bacteria without damaging the indigenous beneficial flora of the habitat. Phages are part of the natural microflora present in different environmental niches and are remarkably stable in the environment. Diverse range of phage products, such as phage enzymes, phage peptides having antimicrobial properties, and phage cocktails also have been used to eradicate pathogens along with whole phages. Recently, the ability of phages to control pathogens has extended from the different areas of medicine, agriculture, aquaculture, food industry, and into the environment. To avoid the arrival of pre-antibiotic epoch, phage intervention proves to be a potential option to eradicate harmful pathogens generated by the MDR gene flow which are uneasy to cure by conventional treatments.

Pathak, A., Chauhan, A., Blom, J., Indest, K. J., Jung, C. M., Stothard, P., Bera, G., Green, S. J., and Ogram, A. Comparative Genomics and Metabolic Analysis Reveals Peculiar Characteristics of Rhodococcus opacus Strain M213 Particularly for Naphthalene Degradation. PLoS.One. 2016; 11(8):e0161032; PM: 27532207

ABSTRACT: The genome of Rhodococcus opacus strain M213, isolated from a fuel-oil contaminated soil, was sequenced and annotated which revealed a genome size of 9,194,165 bp encoding 8680 putative genes and a G+C content of 66.72%. Among the protein coding genes, 71.77% were annotated as clusters of orthologous groups of

proteins (COGs); 55% of the COGs were present as paralog clusters. Pulsed field gel electrophoresis (PFGE) analysis of M213 revealed the presence of three different sized replicons- a circular chromosome and two megaplasmids (pNUO1 and pNUO2) estimated to be of 750Kb 350Kb in size, respectively. Conversely, using an alternative approach of optical mapping, the plasmid replicons appeared as a circular ~1.2 Mb megaplasmid and a linear, ~0.7 Mb megaplasmid. Genome-wide comparative analysis of M213 with a cohort of sequenced Rhodococcus species revealed low syntenic affiliation with other R. opacus species including strains B4 and PD630. Conversely, a closer affiliation of M213, at the functional (COG) level, was observed with the catabolically versatile R. jostii strain RHA1 and other Rhodococcii such as R. wratislaviensis strain IFP 2016, R. imtechensis strain RKJ300, Rhodococcus sp. strain JVH1, and Rhodococcus sp. strain DK17, respectively. An in-depth, genome-wide comparison between these functional relatives revealed 971 unique genes in M213 representing 11% of its total genome; many associating with catabolic functions. Of major interest was the identification of as many as 154 genomic islands (GEIs), many with duplicated catabolic genes, in particular for PAHs; a trait that was confirmed by PCR-based identification of naphthalene dioxygenase (NDO) as a representative gene, across PFGE-resolved replicons of strain M213. Interestingly, several plasmid/GEI-encoded genes, that likely participate in degrading naphthalene (NAP) via a peculiar pathway, were also identified in strain M213 using a combination of bioinformatics, metabolic analysis and gene expression measurements of selected catabolic genes by RT-PCR. Taken together, this study provides a comprehensive understanding of the genome plasticity and ecological competitiveness of strain M213 likely facilitated by horizontal gene transfer (HGT), bacteriophage attacks and genomic reshuffling- aspects that continue to be understudied and thus poorly understood, in particular for the soil-borne Rhodococcii.

Peduzzi, P. Virus ecology of fluvial systems: a blank spot on the map? Biol.Rev.Camb.Philos.Soc. 2016; 91(4):937-949; PM: 26105126

ABSTRACT: The ecology of viruses has been studied only in a limited number of rivers and streams. In light of a recent re-appraisal of the global fluvial surface area, issues such as abundance and production, host mortality and the influence of suspended particles and biofilms are addressed. Viral life cycles, potential impacts of viruses on water biochemistry and carbon flow, and viral diversity are considered. Variability in trophic levels along with the heterogeneous nature and hydrological dynamics of fluvial environments suggest a prevailingly physical control of virus-related processes under lotic conditions and more biological control under lentic conditions. Viral lysis likely contributes to a pool of rapidly cycling carbon in environments typically characterized by high proportions of recalcitrant terrestrial carbon. On average, 33.6% (equalling 0.605 Pg C year-1) of the globally respired carbon from fluvial systems may pass through a viral loop. Virus distribution and the proportion of organic material in horizontal transport versus processes in retention zones remain to be determined in detail. The need for up-scaling the contribution of virus-related processes in fluvial systems is of global relevance. Further, the role of climate change and the effect of anthropogenic alterations of fluvial systems on viruses require attention. The identification of these considerable knowledge gaps should foster future research efforts.

Peng, Y. Z. and Huang, G. T. [Phage therapy for bacterial infection of burn]. Zhonghua Shao Shang Za Zhi. 9-20-2016; 32(9):513-516; PM: 27647065

ABSTRACT: With the long-term and widespread use of antibiotics, drug resistance of bacteria has become a major problem in the treatment of burn infection. For treating multidrug resistant bacteria, phage therapy has become the focus of attention. Development of phage therapy to fill the blank of this field in China is extremely urgent.

Pereira, C., Moreirinha, C., Teles, L., Rocha, R. J., Calado, R., Romalde, J. L., Nunes, M. L., and Almeida, A. Application of phage therapy during bivalve depuration improves Escherichia coli decontamination. Food Microbiol. 2017; 61:102-112; PM: 27697159

ABSTRACT: The present study investigated the potential application of the bacteriophage (or phage) phT4A, ECA2 and the phage cocktail phT4A/ECA2 to decrease the concentration of Escherichia coli during the depuration of natural and artificially contaminated cockles. Depuration in static seawater at multiplicity of infection (MOI) of 1 with single phage suspensions of phT4A and ECA2 was the best condition, as it decreased by approximately 2.0 log CFU/g the concentration of E. coli in artificially contaminated cockles after a 4 h of treatment. When naturally contaminated cockles were treated in static seawater with single phage suspensions and the phage cocktail, similar decreases in the concentration of E. coli (approximately 0.7 log CFU/g) were achieved. However, when employing the phage cocktail, a longer treatment time was required to obtain comparable results to those achieved when using single phage suspensions. When naturally contaminated cockles were depurated with phage phT4A in a recirculated seawater system (mimicking industrial depuration conditions), a 0.6 log CFU/g reduction of E. coli was achieved after a 2 h of treatment. When the depuration process was performed without phage addition, a 4 h treatment was necessary to obtain a similar decrease. By combining phage therapy and depuration procedures, a reduction in bivalves depuration period can be achieved for, thus decreasing the cost associated with this procedure and even enhance the quality and safety of depurated bivalves destined for human consumption.

Petrovic, A., Kostanjsek, R., Rakhely, G., and Knezevic, P. The First Siphoviridae Family Bacteriophages Infecting Bordetella bronchiseptica Isolated from Environment. Microb.Ecol. 9-15-2016; PM: 27628741

ABSTRACT: Bordetella bronchiseptica is a well-known etiological agent of kennel cough in dogs and cats and one of the two causative agents of atrophic rhinitis, a serious swine disease. The aim of the study was to isolate B. bronchiseptica bacteriophages from environmental samples for the first time. A total of 29 phages from 65 water samples were isolated using the strain ATCC 10580 as a host. The lytic spectra of the phages were examined at 25 and 37 degrees C, using 12 strains of B. bronchiseptica. All phages were able to plaque on 25.0 % to 41.7 % of the strains. The selected phages showed similar morphology (Siphoviridae, morphotype B2), but variation of RFLP patterns and efficacy of plating on various strains. The partial genome sequence of phage vB_BbrS_CN1 showed its similarity to phages from genus Yuavirus. Using PCR, it was confirmed that the phages do not originate from the host strain, and environmental origin was additionally confirmed by the analysis of host genome sequence in silico and plating heated and unheated samples in parallel. Accordingly, this is the first isolation of B. bronchiseptica phages from environment and the first isolation and characterization of phages of B. bronchiseptica belonging to family Siphoviridae.

Pinto, G., Silva, M. D., Peddey, M., Sillankorva, S., and Azeredo, J. The role of bacteriophages in periodontal health and disease. Future.Microbiol. 2016; 11:1359-1369; PM: 27633580

ABSTRACT: The human periodontium health is commonly compromised by chronic inflammatory conditions and has become a major public health concern. Dental plaque, the precursor of periodontal disease, is a complex biofilm consisting mainly of bacteria, but also archaea, protozoa, fungi and viruses. Viruses that specifically infect bacteria - bacteriophages - are most common in the oral cavity. Despite this, their role in the progression of periodontal disease remains poorly explored. This review aims to summarize how bacteriophages interact with the oral microbiota, their ability to increase bacterial virulence and mediate the transfer of resistance genes and suggests how bacteriophages can be used as an alternative to the current periodontal disease therapies.

Pope, W. H., Guido, M. J., Iyengar, P., Nigra, J. T., Serbin, M. B., Kasturiarachi, N. S., Pressimone, C. A., Schiebel, J. G., Furbee, E. C., Grubb, S. R., Warner, M. H., Montgomery, M. T., Garlena, R. A., Russell, D. A., Jacobs-Sera, D., and Hatfull, G. F. Genome Sequence of Gordonia Phage Emalyn. Genome Announc. 8-11-2016; 4(4); PM: 27516499

ABSTRACT: Emalyn is a newly isolated bacteriophage of Gordonia terrae 3612 and has a double-stranded DNA genome 43,982 bp long with 67 predicted protein-encoding genes, 32 of which we can assign putative functions. Emalyn has a prolate capsid and has extensive nucleotide similarity with several previously sequenced phages.

Pope, W. H., Berryman, E. N., Forrest, K. M., McHale, L., Wertz, A. T., Zhuang, Z., Kasturiarachi, N. S., Pressimone, C. A., Schiebel, J. G., Furbee, E. C., Grubb, S. R., Warner, M. H., Montgomery, M. T., Garlena, R. A., Russell, D. A., Jacobs-Sera, D., and Hatfull, G. F. Genome Sequence of Gordonia Phage BetterKatz. Genome Announc. 8-11-2016; 4(4); PM: 27516497

ABSTRACT: BetterKatz is a bacteriophage isolated from a soil sample collected in Pittsburgh, Pennsylvania using the host Gordonia terrae 3612. BetterKatz's genome is 50,636 bp long and contains 75 predicted protein-coding genes, 35 of which have been assigned putative functions. BetterKatz is not closely related to other sequenced Gordonia phages.

Pretorius, I. S. Synthetic genome engineering forging new frontiers for wine yeast. Crit Rev.Biotechnol. 8-18-2016; 1-25; PM: 27535766

ABSTRACT: Over the past 15 years, the seismic shifts caused by the convergence of biomolecular, chemical, physical, mathematical, and computational sciences alongside cutting-edge developments in information technology and engineering have erupted into a new field of scientific endeavor dubbed Synthetic Biology. Recent rapid advances in high-throughput DNA sequencing and DNA synthesis techniques are enabling the design and construction of new biological parts (genes), devices (gene networks) and modules (biosynthetic pathways), and the redesign of biological systems (cells and organisms) for useful purposes. In 2014, the budding yeast Saccharomyces cerevisiae

became the first eukaryotic cell to be equipped with a fully functional synthetic chromosome. This was achieved following the synthesis of the first viral (poliovirus in 2002 and bacteriophage Phi-X174 in 2003) and bacterial (Mycoplasma genitalium in 2008 and Mycoplasma mycoides in 2010) genomes, and less than two decades after revealing the full genome sequence of a laboratory (S288c in 1996) and wine (AWRI1631 in 2008) yeast strain. A large international project - the Synthetic Yeast Genome (Sc2.0) Project - is now underway to synthesize all 16 chromosomes (approximately 12 Mb carrying approximately 6000 genes) of the sequenced S288c laboratory strain by 2018. If successful, S. cerevisiae will become the first eukaryote to cross the horizon of in silico design of complex cells through de novo synthesis, reshuffling, and editing of genomes. In the meantime, yeasts are being used as cell factories for the semi-synthetic production of high-value compounds, such as the potent antimalarial artemisinin, and food ingredients, such as resveratrol, vanillin, stevia, nootkatone, and saffron. As a continuum of previously genetically engineered industrially important yeast strains, precision genome engineering is bound to also impact the study and development of wine yeast strains supercharged with synthetic DNA. The first taste of what the future holds is the de novo production of the raspberry ketone aroma compound, 4-[4-hydroxyphenyl]butan-2-one, in a wine yeast strain (AWRI1631), which was recently achieved via metabolic pathway engineering and synthetic enzyme fusion. A peek over the horizon is revealing that the future of "Wine Yeast 2.0" is already here. Therefore, this article seeks to help prepare the wine industry - an industry rich in history and tradition on the one hand, and innovation on the other - for the inevitable intersection of the ancient art practiced by winemakers and the inventive science of pioneering "synthetic genomicists". It would be prudent to proactively engage all stakeholders - researchers, industry practitioners, policymakers, regulators, commentators, and consumers - in a meaningful dialog about the potential challenges and opportunities emanating from Synthetic Biology. To capitalize on the new vistas of synthetic yeast genomics, this paper presents wine yeast research in a fresh context, raises important questions and proposes new directions.

Qiao, J., Li, Y., Wei, C., Yang, H., Yu, J., and Wei, H. Rapid detection of viral antibodies based on multifunctional Staphylococcus aureus nanobioprobes. Enzyme Microb.Technol. 2016; 95:94-99; PM: 27866631

ABSTRACT: Biosynthesis of nanoparticles inside S. aureus cells has enhanced the sensitivity of immunoassays based on the S. aureus nanoparticles. However, the current methods are limited to antigen detection by conjugating IgG antibodies on S. aureus nanoparticles. In this study, a simple way to conjugate antigens to the S. aureus nanobioparticles was developed by utilizing a cell wall binding domain (CBD) from a bacteriophage lysin PlyV12. Based on this novel design, simple agglutination tests of the IgG antibodies of Ebola virus (EBOV) nucleoprotein (NP) and Middle East Respiratory Virus (MERS) NP in rabbit sera were successfully developed by conjugating the S. aureus nanobioparticles with two fusion proteins EBOV NP- CBD and MERS NP-CBD, respectively. The conjugation was done easily by just mixing the fusion proteins with the S. aureus nanoparticles. The detection time was within 20 min without any special equipment or expertise. As far as we know, this is the first time to realize the detection of viral antibodies based on S. aureus nanoparticles.

Rahbarnia, L., Farajnia, S., Babaei, H., Majidi, J., Veisi, K., Ahmadzadeh, V., and Akbari, B. Evolution of phage display technology: from discovery to application. J.Drug Target. 11-21-2016; 1-9; PM: 27819143

ABSTRACT: Phage display technology as a selection-based system is an attractive method for evolution of new biological drugs. Unique ability of phage libraries for displaying proteins on bacteriophage surfaces enable them to make a major contribution in diverse fields of researches related to the diagnosis and therapy of diseases. One of the great challenges facing researchers is the modification of phage display technology and the development of new applications. This article reviews the molecular basis of phage display library, and summarizes the novel and specific applications of this technique in the field of biological drugs development including therapeutic antibodies, peptides, vaccines, and catalytic antibodies.

Rahimi-Midani, A., Kim, K. H., Lee, S. W., Jung, S. B., and Choi, T. J. Isolation and Characterization of a Bacteriophage Preying an Antifungal Bacterium. Plant Pathol.J. 2016; 32(6):584-588; PM: 27904467

ABSTRACT: Several Bacillus species were isolated from rice field soils, and 16S rRNA gene sequence analysis showed that Bacillus cereus was the most abundant. A strain named BC1 showed antifungal activity against Rhizoctonia solani. Bacteriophages infecting strain BC1 were isolated from the same soil sample. The isolated phage PK16 had an icosahedral head of 100 +/- 5 nm and tail of 200 +/- 5 nm, indicating that it belonged to the family Myoviridae. Analysis of the complete linear dsDNA genome revealed a 158,127-bp genome with G + C content of 39.9% comprising 235 open reading frames as well as 19 tRNA genes (including 1 pseudogene). Blastp analysis showed that the proteins encoded by the PK16 genome had the closest hits to proteins of seven different bacteriophages. A neighbor-joining phylogenetic tree based on the major capsid protein showed a robust clustering of phage PK16 with phage JBP901 and BCP8-2 isolated from Korean fermented food.

Rashid, S. J., Barylski, J., Hargreaves, K. R., Millard, A. A., Vinner, G. K., and Clokie, M. R. Two Novel Myoviruses from the North of Iraq Reveal Insights into Clostridium difficile Phage Diversity and Biology. Viruses. 11-16-2016; 8(11); PM: 27854339

ABSTRACT: Bacteriophages (phages) are increasingly being explored as therapeutic agents to combat bacterial diseases, including Clostridium difficile infections. Therapeutic phages need to be able to efficiently target and kill a wide range of clinically relevant strains. While many phage groups have yet to be investigated in detail, those with new and useful properties can potentially be identified when phages from newly studied geographies are characterised. Here, we report the isolation of C. difficile phages from soil samples from the north of Iraq. Two myoviruses, CDKM15 and CDKM9, were selected for detailed sequence analysis on the basis of their broad and potentially useful host range. CDKM9 infects 25/80 strains from 12/20 C. difficile ribotypes, and CDKM15 infects 20/80 strains from 9/20 ribotypes. Both phages can infect the clinically relevant ribotypes R027 and R001. Phylogenetic analysis based on whole genome sequencing revealed that the phages are genetically distinct from each other but closely related to other long-tailed myoviruses. A comparative genomic analysis revealed key differences in the genes predicted to encode for proteins involved in bacterial infection. Notably, CDKM15 carries a clustered regularly interspaced short palindromic repeat (CRISPR) array with spacers that are homologous to sequences in the

CDKM9 genome and of phages from diverse localities. The findings presented suggest a possible shared evolutionary past for these phages and provides evidence of their widespread dispersal.

Rees, J. C. and Barr, J. R. Detection of methicillin-resistant Staphylococcus aureus using phage amplification combined with matrix-assisted laser desorption/ionization mass spectrometry. Anal.Bioanal.Chem. 11-19-2016; PM: 27866257

ABSTRACT: Antibiotic resistance continues to contribute significantly to morbidity and mortality across the world. Developing new tests for antibiotic-resistant bacteria is a core action to combat resistant infections. We describe a method that uses phage amplification detection (PAD) combined with matrix-assisted laser desorption/ionization mass spectrometry (MALDI-MS) to rapidly identify Staphylococcus aureus and determine phenotypic susceptibility to cefoxitin. Samples tested for S. aureus are incubated together with bacteriophage in the presence and absence of cefoxitin and subjected to rapid trypsin digestion followed by MALDI-MS analysis. Tryptic peptides derived from amplified phage proteins can be detected by MALDI-MS, as validated by time-of-flight (TOF)/TOF analysis of each peptide combined with database searching. Methicillin-resistant S. aureus show significant phage amplification in the presence of cefoxitin, while methicillin-sensitive S. aureus show no phage amplification relative to a no-antibiotic control. We also show that PAD methodology can be implemented on an FDA-approved commercial MALDI-MS bacterial identification system to identify S. aureus and determine antibiotic susceptibility. The novelty of this assay includes the use of phage-derived tryptic peptides as detected by MALDI-MS to monitor the results of PAD on an instrument common to many modern microbiology laboratories.

Renda, B. A., Chan, C., Parent, K. N., and Barrick, J. E. Emergence of a Competence-Reducing Filamentous Phage from the Genome of Acinetobacter baylyi ADP1. J.Bacteriol. 12-1-2016; 198(23):3209-3219; PM: 27645387

ABSTRACT: Bacterial genomes commonly contain prophage sequences as a result of past infections with lysogenic phages. Many of these integrated viral sequences are believed to be cryptic, but prophage genes are sometimes coopted by the host, and some prophages may be reactivated to form infectious particles when cells are stressed or mutate. We found that a previously uncharacterized filamentous phage emerged from the genome of Acinetobacter baylyi ADP1 during a laboratory evolution experiment. This phage has a genetic organization similar to that of the Vibrio cholerae CTXvarphi phage. The emergence of the ADP1 phage was associated with the evolution of reduced transformability in our experimental populations, so we named it the competence-reducing acinetobacter phage (CRAvarphi). Knocking out ADP1 genes required for competence leads to resistance to CRAvarphi infection. Although filamentous bacteriophages are known to target type IV pili, this is the first report of a phage that apparently uses a competence pilus as a receptor. A. baylyi may be especially susceptible to this route of infection because every cell is competent during normal growth, whereas competence is induced only under certain environmental conditions or in a subpopulation of cells in other bacterial species. It is possible that CRAvarphi-like phages restrict horizontal gene transfer in nature by inhibiting the growth of naturally transformable strains. We also found that prophages with homology to CRAvarphi exist in several strains of Acinetobacter baumannii These CRAvarphi-like A. baumannii prophages encode toxins similar to CTXvarphi that might contribute to the

virulence of this opportunistic multidrug-resistant pathogen. IMPORTANCE: We observed the emergence of a novel filamentous phage (CRAvarphi) from the genome of Acinetobacter baylyi ADP1 during a long-term laboratory evolution experiment. CRAvarphi is the first bacteriophage reported to require the molecular machinery involved in the uptake of environmental DNA for infection. Reactivation and evolution of CRAvarphi reduced the potential for horizontal transfer of genes via natural transformation in our experiment. Risk of infection by similar phages may limit the expression and maintenance of bacterial competence in nature. The closest studied relative of CRAvarphi is the Vibrio cholerae CTXvarphi phage. Variants of CRAvarphi are found in the genomes of Acinetobacter baumannii strains, and it is possible that phage-encoded toxins contribute to the virulence of this opportunistic multidrug-resistant pathogen.

Ricci, D. P., Melfi, M. D., Lasker, K., Dill, D. L., McAdams, H. H., and Shapiro, L. Cell cycle progression in Caulobacter requires a nucleoid-associated protein with high AT sequence recognition. Proc.Natl.Acad.Sci.U.S.A. 10-4-2016; 113(40):E5952-E5961; PM: 27647925

ABSTRACT: Faithful cell cycle progression in the dimorphic bacterium Caulobacter crescentus requires spatiotemporal regulation of gene expression and cell pole differentiation. We discovered an essential DNA-associated protein, GapR, that is required for Caulobacter growth and asymmetric division. GapR interacts with adenine and thymine (AT)-rich chromosomal loci, associates with the promoter regions of cell cycle-regulated genes, and shares hundreds of recognition sites in common with known master regulators of cell cycle-dependent gene expression. GapR target loci are especially enriched in binding sites for the transcription factors GcrA and CtrA and overlap with nearly all of the binding sites for MucR1, a regulator that controls the establishment of swarmer cell fate. Despite constitutive synthesis, GapR accumulates preferentially in the swarmer compartment of the predivisional cell. Homologs of GapR, which are ubiquitous among the alpha-proteobacteria and are encoded on multiple bacteriophage genomes, also accumulate in the predivisional cell swarmer compartment when expressed in Caulobacter The Escherichia coli nucleoid-associated protein H-NS, like GapR, selectively associates with AT-rich DNA, yet it does not localize preferentially to the swarmer compartment when expressed exogenously in Caulobacter, suggesting that recognition of AT-rich DNA is not sufficient for the asymmetric accumulation of GapR. Further, GapR does not silence the expression of H-NS target genes when expressed in E. coli, suggesting that GapR and H-NS have distinct functions. We propose that Caulobacter has co-opted a nucleoid-associated protein with high AT recognition to serve as a mediator of cell cycle progression.

Rios, A. C., Moutinho, C. G., Pinto, F. C., Del Fiol, F. S., Jozala, A., Chaud, M. V., Vila, M. M., Teixeira, J. A., and Balcao, V. M. Alternatives to overcoming bacterial resistances: State-of-the-art. Microbiol.Res. 2016; 191:51-80; PM: 27524653

ABSTRACT: Worldwide, bacterial resistance to chemical antibiotics has reached such a high level that endangers public health. Presently, the adoption of alternative strategies that promote the elimination of resistant microbial strains from the environment is of utmost importance. This review discusses and analyses several (potential) alternative strategies to current chemical antibiotics. Bacteriophage (or phage) therapy, although not new, makes use of strictly lytic phage particles as an alternative, or a complement, in the antimicrobial treatment of bacterial infections. It is being rediscovered as a safe

method, because these biological entities devoid of any metabolic machinery do not possess any affinity whatsoever to eukaryotic cells. Lysin therapy is also recognized as an innovative antimicrobial therapeutic option, since the topical administration of preparations containing purified recombinant lysins with amounts in the order of nanograms, in infections caused by Gram-positive bacteria, demonstrated a high therapeutic potential by causing immediate lysis of the target bacterial cells. Additionally, this therapy exhibits the potential to act synergistically when combined with certain chemical antibiotics already available on the market. Another potential alternative antimicrobial therapy is based on the use of antimicrobial peptides (AMPs), amphiphilic polypeptides that cause disruption of the bacterial membrane and can be used in the treatment of bacterial, fungal and viral infections, in the prevention of biofilm formation, and as antitumoral agents. Interestingly, bacteriocins are a common strategy of bacterial defense against other bacterial agents, eliminating the potential opponents of the former and increasing the number of available nutrients in the environment for their own growth. They can be applied in the food industry as biopreservatives and as probiotics, and also in fighting multi-resistant bacterial strains. The use of antibacterial antibodies promises to be extremely safe and effective. Additionally, vaccination emerges as one of the most promising preventive strategies. All these will be tackled in detail in this review paper.

Ross, A., Ward, S., and Hyman, P. More Is Better: Selecting for Broad Host Range Bacteriophages. Front Microbiol. 2016; 7:1352; PM: 27660623

ABSTRACT: Bacteriophages are viruses that infect bacteria. In this perspective, we discuss several aspects of a characteristic feature of bacteriophages, their host range. Each phage has its own particular host range, the range of bacteria that it can infect. While some phages can only infect one or a few bacterial strains, other phages can infect many species or even bacteria from different genera. Different methods for determining host range may give different results, reflecting the multiple mechanisms bacteria have to resist phage infection and reflecting the different steps of infection each method depends on. This makes defining host range difficult. Another difficulty in describing host range arises from the inconsistent use of the words "narrow" and especially "broad" when describing the breadth of the host range. Nearly all bacteriophages have been isolated using a single host strain of bacteria. While this procedure is fairly standard, it may more likely produce narrow rather than broad host range phage. Our results and those of others suggest that using multiple host strains during isolation can more reliably produce broader host range phages. This challenges the common belief that most bacteriophages have a narrow host range. We highlight the implications of this for several areas that are affected by host range including horizontal gene transfer and phage therapy.

Roszniowski, B., Latka, A., Maciejewska, B., Vandenheuvel, D., Olszak, T., Briers, Y., Holt, G. S., Valvano, M. A., Lavigne, R., Smith, D. L., and Drulis-Kawa, Z. The temperate Burkholderia phage AP3 of the Peduovirinae shows efficient antimicrobial activity against B. cenocepacia of the IIIA lineage. Appl.Microbiol.Biotechnol. 10-21-2016; PM: 27770178

ABSTRACT: Burkholderia phage AP3 (vB_BceM_AP3) is a temperate virus of the

Myoviridae and the Peduovirinae subfamily (P2likevirus genus). This phage specifically infects multidrug-resistant clinical Burkholderia cenocepacia lineage IIIA strains commonly isolated from cystic fibrosis patients. AP3 exhibits high pairwise nucleotide identity (61.7 %) to Burkholderia phage KS5, specific to the same B. cenocepacia host, and has 46.7-49.5 % identity to phages infecting other species of Burkholderia. The lysis cassette of these related phages has a similar organization (putative antiholin, putative holin, endolysin, and spanins) and shows 29-98 % homology between specific lysis genes, in contrast to Enterobacteria phage P2, the hallmark phage of this genus. The AP3 and KS5 lysis genes have conserved locations and high amino acid sequence similarity. The AP3 bacteriophage particles remain infective up to 5 h at pH 4-10 and are stable at 60 degrees C for 30 min, but are sensitive to chloroform, with no remaining infective particles after 24 h of treatment. AP3 lysogeny can occur by stable genomic integration and by pseudo-lysogeny. The lysogenic bacterial mutants did not exhibit any significant changes in virulence compared to wild-type host strain when tested in the Galleria mellonella moth wax model. Moreover, AP3 treatment of larvae infected with B. cenocepacia revealed a significant increase ($P < 0.0001$) in larvae survival in comparison to AP3-untreated infected larvae. AP3 showed robust lytic activity, as evidenced by its broad host range, the absence of increased virulence in lysogenic isolates, the lack of bacterial gene disruption conditioned by bacterial tRNA downstream integration site, and the absence of detected toxin sequences. These data suggest that the AP3 phage is a promising potent agent against bacteria belonging to the most common B. cenocepacia IIIA lineage strains.

Salas, M., Holguera, I., Redrejo-Rodriguez, M., and de, Vega M. DNA-Binding Proteins Essential for Protein-Primed Bacteriophage Phi29 DNA Replication. Front Mol.Biosci. 2016; 3:37; PM: 27547754

ABSTRACT: Bacillus subtilis phage Phi29 has a linear, double-stranded DNA 19 kb long with an inverted terminal repeat of 6 nucleotides and a protein covalently linked to the 5' ends of the DNA. This protein, called terminal protein (TP), is the primer for the initiation of replication, a reaction catalyzed by the viral DNA polymerase at the two DNA ends. The DNA polymerase further elongates the nascent DNA chain in a processive manner, coupling strand displacement with elongation. The viral protein p5 is a single-stranded DNA binding protein (SSB) that binds to the single strands generated by strand displacement during the elongation process. Viral protein p6 is a double-stranded DNA binding protein (DBP) that preferentially binds to the origins of replication at the Phi29 DNA ends and is required for the initiation of replication. Both SSB and DBP are essential for Phi29 DNA amplification. This review focuses on the role of these phage DNA-binding proteins in Phi29 DNA replication both in vitro and in vivo, as well as on the implication of several B. subtilis DNA-binding proteins in different processes of the viral cycle. We will revise the enzymatic activities of the Phi29 DNA polymerase: TP-deoxynucleotidylation, processive DNA polymerization coupled to strand displacement, 3'-5' exonucleolysis and pyrophosphorolysis. The resolution of the Phi29 DNA polymerase structure has shed light on the translocation mechanism and the determinants responsible for processivity and strand displacement. These two properties have made Phi29 DNA polymerase one of the main enzymes used in the current DNA amplification technologies. The determination of the structure of Phi29 TP revealed the existence of three domains: the priming domain, where the primer residue Ser232, as well as Phe230, involved in the determination of the initiating nucleotide, are located, the intermediate domain, involved in DNA polymerase binding, and the N-terminal domain, responsible for DNA binding and localization of the TP at the bacterial nucleoid, where viral DNA replication takes place. The biochemical

properties of the Phi29 DBP and SSB and their function in the initiation and elongation of Phi29 DNA replication, respectively, will be described.

Sarker, S. A., Berger, B., Deng, Y., Kieser, S., Foata, F., Moine, D., Descombes, P., Sultana, S., Huq, S., Bardhan, P. K., Vuillet, V., Praplan, F., and Brussow, H. Oral application of Escherichia coli bacteriophage: safety tests in healthy and diarrheal children from Bangladesh. Environ.Microbiol. 10-17-2016; PM: 27750388

ABSTRACT: A T4-like coliphage cocktail was given with different oral doses to healthy Bangladeshi children in a placebo-controlled randomized phase I safety trial. Fecal phage detection was oral dose dependent suggesting passive gut transit of coliphages through the gut. No adverse effects of phage application were seen clinically and by clinical chemistry. Similar results were obtained for a commercial phage preparation (Coliproteus from Microgen/Russia). By 16S rRNA gene sequencing, only a low degree of fecal microbiota conservation was seen in healthy children from Bangladesh who were sampled over a time interval of 7 days suggesting a substantial temporal fluctuation of the fecal microbiota composition. Microbiota variability was not associated with the age of the children or the presence of phage in the stool. Stool microbiota composition of Bangladeshi children resembled that found in children of other regions of the world. Marked variability in fecal microbiota composition was also seen in 71 pediatric diarrhea patients receiving only oral rehydration therapy and in 38 patients receiving coliphage preparations or placebo when sampled 1.2 or 4 days apart respectively. Temporal stability of the gut microbiota should be assessed in case-control studies involving children before associating fecal microbiota composition with health or disease phenotypes.

Sasikala, D. and Srinivasan, P. Characterization of potential lytic bacteriophage against Vibrio alginolyticus and its therapeutic implications on biofilm dispersal. Microb.Pathog. 2016; 101:24-35; PM: 27793690

ABSTRACT: Vibrio alginolyticus is a leading cause of vibriosis, presenting opportunistic infections to humans associated with raw seafood contamination. At present, phage therapy that acts as an alternative sanitizing agent is explored for targeting V. alginolyticus. The study outcome revealed that the phage VP01 with its extreme lytic effect showed a high potential impact on the growth of V. alginolyticus as well as biofilm formation. Electron microscopy revealed the phage resemblance to Myoviridae, based on its morphology. Further study clarified that the phage VP01 possesses a broad host spectrum and amazing phage sensitivity at different pH, high thermal stability, and high burst size of 415 PFU/cell. In addition, the investigation of phage co-culturing against this pathogen resulted in a significant growth reduction even at less MOIs 0.1 and 1. These results suggest that the phage could be a promising candidate for the control of V. alginolyticus infections.

Saxena, P., He, L., Malyutin, A., Datta, S. A., Rein, A., Bond, K. M., Jarrold, M. F., Spilotros, A., Svergun, D., Douglas, T., and Dragnea, B. Virus Matryoshka: A Bacteriophage Particle-Guided Molecular Assembly Approach to a Monodisperse Model of the Immature Human Immunodeficiency Virus. Small. 9-16-2016; PM: 27634413

ABSTRACT: Immature human immunodeficiency virus type 1 (HIV-1) is approximately spherical, but is constructed from a hexagonal lattice of the Gag protein. As a hexagonal lattice is necessarily flat, the local symmetry cannot be maintained throughout the structure. This geometrical frustration presumably results in bending stress. In natural particles, the stress is relieved by incorporation of packing defects, but the magnitude of this stress and its significance for the particles is not known. In order to control this stress, we have now assembled the Gag protein on a quasi-spherical template derived from bacteriophage P22. This template is monodisperse in size and electron-transparent, enabling the use of cryo-electron microscopy in structural studies. These templated assemblies are far less polydisperse than any previously described virus-like particles (and, while constructed according to the same lattice as natural particles, contain almost no packing defects). This system gives us the ability to study the relationship between packing defects, curvature and elastic energy, and thermodynamic stability. As Gag is bound to the P22 template by single-stranded DNA, treatment of the particles with DNase enabled us to determine the intrinsic radius of curvature of a Gag lattice, unconstrained by DNA or a template. We found that this intrinsic radius is far larger than that of a virion or P22-templated particle. We conclude that Gag is under elastic strain in a particle; this has important implications for the kinetics of shell growth, the stability of the shell, and the type of defects it will assume as it grows.

Sazinas, P., Smith, C., Suhaimi, A., Hobman, J. L., Dodd, C. E., and Millard, A. D. Draft Genome Sequence of the Bacteriophage vB_Eco_slurp01. Genome Announc. 11-17-2016; 4(6); PM: 27856574

ABSTRACT: Bacteriophage vB_Eco_slurp01 was isolated from porcine feces using Escherichia coli MG1655 as a host. With a genome size of 348 kb, vB_Eco_slurp01 is one of the largest bacteriophages isolated to date.

Scanlan, P. D., Hall, A. R., and Buckling, A. Parasite genetic distance and local adaptation in co-evolving bacteria-bacteriophage populations. Mol.Ecol. 10-24-2016; PM: 27775190

ABSTRACT: Antagonistic co-evolution between hosts and parasites can lead to local adaptation (LA) such that parasite fitness is greatest in sympatric hosts (or vice versa). The magnitude of LA typically increases with geographical distance, which is assumed to be because genetic (and hence phenotypic) distance increases with geographical distance. Here, we explicitly test the relationships between parasite genetic and phenotypic distance and LA using isolates of co-evolved viral parasites (lytic bacteriophage varphi2) and the host bacterium Pseudomonas fluorescens SBW25. We find positive relationships between parasite genotype and infectivity phenotype, but the strength of the relationship was greater when infectivity was defined by the identity of hosts that could be infected rather than the actual number of hosts infected (host range), and when measurements were compared within rather than among populations. Crucially, we find a monotonic relationship between LA and genetic distance across phage isolates from different populations, although in contrast to many geographical studies, parasite LA decreased with genetic distance. These results can be

explained by the fact that bacteria can rapidly adapt to phage infectivity mutations, but that evolved resistance has a degree of specificity to the local phage population. Our results show that antagonistic co-evolution alone can result in predictable links between genetic distance and host-parasite local adaptation.

Scanlan, P. D., Bischofberger, A. M., and Hall, A. R. Modification of Escherichia coli-bacteriophage interactions by surfactants and antibiotics in vitro. FEMS Microbiol.Ecol. 2017; 93(1); PM: 27737900

ABSTRACT: Although experiments indicate that the abiotic environment plays an important role in bacterial interactions with their parasitic viruses (bacteriophages or phages), it is not yet clear how exposure to compounds present in nature alters the impact of phages on bacterial growth and evolution. To address this question, we exposed Escherichia coli K12 MG1655, in combination with three lytic phages, to various substances that natural and clinical microbial populations are likely to encounter: bile salts (present in mammalian gastrointestinal tracts), sodium dodecyl sulfate (SDS, a common surfactant in cleaning and hygiene products) and four antibiotics (present at variable concentrations in natural and clinical environments). Our results show that bile salts and SDS can reduce the detrimental effect of phages on bacterial growth. In some cases these compounds completely mitigated any negative effects of phages on bacterial growth and consequently bacteria did not evolve resistance to phages in these conditions. The proportional effects of phages were unaffected by antibiotics in most combinations, excepting three cases of phage-drug synergy. These results suggest that accounting for interactions between phages and environmental factors such as surfactants and antibiotics will improve understanding of both bacterial growth and resistance evolution to phages in vivo and in nature.

Schmidt, A., Rabsch, W., Broeker, N. K., and Barbirz, S. Bacteriophage tailspike protein based assay to monitor phase variable glucosylations in Salmonella O-antigens. BMC.Microbiol. 9-7-2016; 16:207; PM: 27604475

ABSTRACT: ***BACKGROUND***: Non-typhoid Salmonella Typhimurium (S. Typhimurium) accounts for a high number of registered salmonellosis cases, and O-serotyping is one important tool for monitoring epidemiology and spread of the disease. Moreover, variations in glucosylated O-antigens are related to immunogenicity and spread in the host. However, classical autoagglutination tests combined with the analysis of specific genetic markers cannot always reliably register phase variable glucose modifications expressed on Salmonella O-antigens and additional tools to monitor O-antigen glucosylation phenotypes of S. Typhimurium would be desirable. ***RESULTS***: We developed a test for the phase variable O-antigen glucosylation state of S. Typhimurium using the tailspike proteins (TSP) of Salmonella phages 9NA and P22. We used this ELISA like tailspike adsorption (ELITA) assay to analyze a library of 44 Salmonella strains. ELITA was successful in discriminating strains that carried glucose 1-6 linked to the galactose of O-polysaccharide backbone (serotype O1) from non-glucosylated strains. This was shown by O-antigen compositional analyses of the respective strains with mass spectrometry and capillary electrophoresis. The ELITA test worked rapidly in a microtiter plate format and was highly O-antigen specific. Moreover, TSP as probes could also detect glucosylated strains in flow cytometry and distinguish multiphasic cultures differing in their glucosylation state. ***CONCLUSIONS***: Tailspike proteins contain large binding sites with precisely defined specificities and are therefore promising tools to be included in

serotyping procedures as rapid serotyping agents in addition to antibodies. In this study, 9NA and P22TSP as probes could specifically distinguish glucosylation phenotypes of Salmonella on microtiter plate assays and in flow cytometry. This opens the possibility for flow sorting of cell populations for subsequent genetic analyses or for monitoring phase variations during large scale O-antigen preparations necessary for vaccine production.

Secor, P. R., Michaels, L. A., Smigiel, K. S., Rohani, M. G., Jennings, L. K., Hisert, K. B., Arrigoni, A., Braun, K. R., Birkland, T. P., Lai, Y., Hallstrand, T. S., Bollyky, P. L., Singh, P. K., and Parks, W. C. Filamentous bacteriophage produced by Pseudomonas aeruginosa alters the inflammatory response and promotes non-invasive infection in vivo. Infect.Immun. 10-17-2016; PM: 27795361

ABSTRACT: Pseudomonas aeruginosa is an important opportunistic human pathogen that lives in biofilm-like cell aggregates at sites of chronic infection, such as occurs in the lungs of patients with cystic fibrosis and non-healing ulcers. During growth in a biofilm, P. aeruginosa dramatically increases the production of filamentous Pf bacteriophage (Pf phage). Previous work indicated that when in vivo Pf phage production was inhibited, P. aeruginosa was less virulent. However, it is not clear how the production of abundant quantities of Pf phage, similar to those produced by biofilms under in vitro conditions, would affect pathogenesis. Here, using a murine pneumonia model, we show that the production of biofilm-relevant amounts of Pf phage prevents the dissemination of P. aeruginosa from the lung. Furthermore, filamentous phage promoted bacterial adhesion to mucin and inhibited bacterial invasion of airway epithelial cultures, suggesting that Pf phage trap P. aeruginosa within the lung. In vivo production of Pf phage was also associated with reduced lung injury, reduced neutrophil recruitment, and lower cytokine levels. Additionally, when producing Pf phage, P. aeruginosa were less prone to phagocytosis by macrophages compared to bacteria not producing Pf phage. Collectively, these data suggest that filamentous Pf phage alter the progression of the inflammatory response and promote phenotypes typically associated with chronic infection.

Severinov, K., Ispolatov, I., and Semenova, E. The Influence of Copy-Number of Targeted Extrachromosomal Genetic Elements on the Outcome of CRISPR-Cas Defense. Front Mol.Biosci. 2016; 3:45; PM: 27630990

ABSTRACT: Prokaryotic type I CRISPR-Cas systems respond to the presence of mobile genetic elements such as plasmids and phages in two different ways. CRISPR interference efficiently destroys foreign DNA harboring protospacers fully matching CRISPR RNA spacers. In contrast, even a single mismatch between a spacer and a protospacer can render CRISPR interference ineffective but causes primed adaptation-efficient and specific acquisition of additional spacers from foreign DNA into the CRISPR array of the host. It has been proposed that the interference and primed adaptation pathways are mediated by structurally different complexes formed by the effector Cascade complex on matching and mismatched protospacers. Here, we present experimental evidence and present a simple mathematical model that shows that when plasmid copy number maintenance/phage genome replication is taken into account, the two apparently different outcomes of the CRISPR-Cas response can be accounted for by just one kind of effector complex on both targets. The results underscore the importance of consideration of targeted genome biology when considering consequences of CRISPR-Cas systems action.

Shabbir, M. A., Hao, H., Shabbir, M. Z., Wu, Q., Sattar, A., and Yuan, Z. Bacteria vs. Bacteriophages: Parallel Evolution of Immune Arsenals. Front Microbiol. 2016; 7:1292; PM: 27582740

ABSTRACT: Bacteriophages are the most common entities on earth and represent a constant challenge to bacterial populations. To fend off bacteriophage infection, bacteria evolved immune systems to avert phage adsorption and block invader DNA entry. They developed restriction-modification systems and mechanisms to abort infection and interfere with virion assembly, as well as newly recognized clustered regularly interspaced short palindromic repeats (CRISPR). In response to bacterial immune systems, bacteriophages synchronously evolved resistance mechanisms, such as the anti-CRISPR systems to counterattack bacterial CRISPR-cas systems, in a continuing evolutionary arms race between virus and host. In turn, it is fundamental to the survival of the bacterial cell to evolve a system to combat bacteriophage immune strategies.

Shabbir, M. A., Hao, H., Shabbir, M. Z., Hussain, H. I., Iqbal, Z., Ahmed, S., Sattar, A., Iqbal, M., Li, J., and Yuan, Z. Survival and Evolution of CRISPR-Cas System in Prokaryotes and Its Applications. Front Immunol. 2016; 7:375; PM: 27725818

ABSTRACT: Prokaryotes have developed numerous innate immune mechanisms in order to fend off bacteriophage or plasmid attack. One of these immune systems is clustered regularly interspaced short palindromic repeats (CRISPR). CRISPR-associated proteins play a key role in survival of prokaryotes against invaders, as these systems cleave DNA of foreign genetic elements. Beyond providing immunity, these systems have significant impact in altering the bacterial physiology in term of its virulence and pathogenicity, as well as evolution. Also, due to their diverse nature of functionality, cas9 endoribonuclease can be easily reprogrammed with the help of guide RNAs, showing unprecedented potential and significance for gene editing in treating genetic diseases. Here, we also discuss the use of NgAgo-gDNA system in genome editing of human cells.

Shao, Q., Trinh, J. T., McIntosh, C. S., Christenson, B., Balazsi, G., and Zeng, L. Lysis-lysogeny coexistence: prophage integration during lytic development. Microbiologyopen. 8-17-2016; PM: 27530202

ABSTRACT: The infection of Escherichia coli cells by bacteriophage lambda results in bifurcated means of propagation, where the phage decides between the lytic and lysogenic pathways. Although traditionally thought to be mutually exclusive, increasing evidence suggests that this lysis-lysogeny decision is more complex than once believed, but exploring its intricacies requires an improved resolution of study. Here, with a newly developed fluorescent reporter system labeling single phage and E. coli DNAs, these two distinct pathways can be visualized by following the DNA movements in vivo. Surprisingly, we frequently observed an interesting "lyso-lysis" phenomenon in lytic cells, where phage integrates its DNA into the host, a characteristic event of the lysogenic pathway, followed by cell lysis. Furthermore, the frequency of lyso-lysis increases with the number of infecting phages, and specifically, with CII activity. Moreover, in lytic cells, the integration site attB on the E. coli genome migrates toward the polar region over time, leading to more spatial overlap with the phage DNA and frequent colocalization/collision of attB and phage DNA, possibly contributing to a higher chance for DNA integration.

Sharma, M., Kumar, D., and Poluri, K. M. Elucidating the pH-Dependent Structural Transition of T7 Bacteriophage Endolysin. Biochemistry. 8-23-2016; 55(33):4614-4625; PM: 27513288

ABSTRACT: Bacteriophages are the most abundant and diverse biological entities on earth. Bacteriophage endolysins are unique peptidoglycan hydrolases and have huge potential as effective enzybiotics in various infectious models. T7 bacteriophage endolysin (T7L), also known as N-acetylmuramoyl-l-alanine amidase or T7 lysozyme, is a 17 kDa protein that lyses a range of Gram-negative bacteria by hydrolyzing the amide bond between N-acetylmuramoyl residues and the l-alanine of the peptidoglycan layer. Although the activity profiles of several of the T7 family members have been known for many years, the molecular basis for their pH-dependent differential activity is not clear. In this study, we explored the pH-induced structural, stability, and activity characteristics of T7L by applying a variety of biophysical techniques and protein nuclear magnetic resonance (NMR) spectroscopy. Our studies established a reversible structural transition of T7L below pH 6 and the formation of a partially denatured conformation at pH 3. This low-pH conformation is thermally stable and exposed its hydrophobic pockets. Further, NMR relaxation measurements and structural analysis unraveled that T7L is highly dynamic in its native state and a network of His residues are responsible for the observed pH-dependent conformational dynamics and transitions. As bacteriophage chimeric and engineered endolysins are being developed as novel therapeutics against multiple drug resistance pathogens, we believe that our results are of great help in designing these entities as broadband antimicrobial and/or antibacterial agents.

Sharma, S., Chatterjee, S., Datta, S., Prasad, R., Dubey, D., Prasad, R. K., and Vairale, M. G. Bacteriophages and its applications: an overview. Folia Microbiol.(Praha). 10-8-2016; PM: 27718043

ABSTRACT: Bacteriophages (or phages), the most abundant viral entity of the planet, are omni-present in all the ecosystems. On the basis of their unique characteristics and anti-bacterial property, phages are being freshly evaluated taxonomically. Phages replicate inside the host either by lytic or lysogenic mode after infecting and using the cellular machinery of a bacterium. Since their discovery by Twort and d'Herelle in the early 1900s, phage became an important agent for combating pathogenic bacteria in clinical treatments and its related research gained momentum. However, due to recent emergence of bacterial resistance on antibiotics, applications of phage (phage therapy) become an inevitable option of research. Phage particles become popular as a biotechnological tool and treatment of pathogenic bacteria in a range of applied areas. However, there are few concerns over the application of phage-based solutions. This review deals with the updated phage taxonomy (ICTV 2015 Release and subsequent revision) and phage biology and the recent development of its application in the areas of biotechnology, biosensor, therapeutic medicine, food preservation, aquaculture diseases, pollution remediation, and wastewater treatment and issues related with limitations of phage-based remedy.

Shimabuku, Q. L., Arakawa, F. S., Fernandes, Silva M., Ferri, Coldebella P., Ueda-Nakamura, T., Fagundes-Klen, M. R., and Bergamasco, R. Water treatment with exceptional virus inactivation using activated carbon modified with silver (Ag) and copper oxide (CuO) nanoparticles. Environ.Technol. 10-21-2016; 1-12; PM: 27766917

ABSTRACT: Continuous flow experiments (450 mL min-1) were performed in household filter in order to investigate the removal and/or inactivation of T4 bacteriophage, using granular activated carbon (GAC) modified with silver and/or copper oxide nanoparticles at different concentrations. GAC and modified GAC were characterized by X-ray diffractometry, specific surface area, pore size and volume, pore average diameter, scanning electron microscopy, transmission electron microscopy, zeta potential and atomic absorption spectroscopy. The antiviral activity of the produced porous media was evaluated by passing suspensions of T4 bacteriophage (approximately 105 UFP/mL) through filters. The filtered water was analyzed for the presence of the bacteriophage and the release of silver and copper oxide. The porous media containing silver and copper oxide nanoparticles showed high inactivation capacity, even reaching reductions higher than 3 log. GAC6 (GAC/Ag0.5%Cu1.0%) was effective in the bacteriophage inactivation, reaching 5.53 log reduction. The levels of silver and copper released in filtered water were below the recommended limits (100 ppb for silver and 1000 ppb for copper) in drinking water. From this study, it is possible to conclude that activated carbon modified with silver and copper oxide nanoparticles can be used as a filter for virus removal in the treatment of drinking water.

Shimasaki, N., Hara, M., Kikuno, R., and Shinohara, K. A Highly Sensitive Assay Using Synthetic Blood Containing Test Microbes for Evaluation of the Penetration Resistance of Protective Clothing Material under Applied Pressure. Biocontrol.Sci. 2016; 21(3):141-152. PM: 27667519

ABSTRACT: To prevent nosocomial infections caused by even either Ebola virus or methicillin-resistant Staphylococcus aureus (MRSA), healthcare workers must wear the appropriate protective clothing which can inhibit contact transmission of these pathogens. Therefore, it is necessary to evaluate the performance of protective clothing for penetration resistance against infectious agents. In Japan, some standard methods were established to evaluate the penetration resistance of protective clothing fabric materials under applied pressure. However, these methods only roughly classified the penetration resistance of fabrics, and the detection sensitivity of the methods and the penetration amount with respect to the relationship between blood and the pathogen have not been studied in detail. Moreover, no standard method using bacteria for evaluation is known. Here, to evaluate penetration resistance of protective clothing materials under applied pressure, the detection sensitivity and the leak amount were investigated by using synthetic blood containing bacteriophage phi-X174 or S. aureus. And the volume of leaked synthetic blood and the amount of test microbe penetration were simultaneously quantified. Our results showed that the penetration detection sensitivity achieved using a test microbial culture was higher than that achieved using synthetic blood at invisible leak level pressures. This finding suggested that there is a potential risk of pathogen penetration even when visual leak of contaminated blood through the protective clothing was not observed. Moreover, at visible leak level pressures, it was found that the amount of test microbe penetration varied at least ten-fold among protective clothing materials classified into the same class of penetration resistance. Analysis of the penetration amount revealed a significant correlation between the volume of penetrated synthetic blood and the amount of test microbe penetration, indicating that the leaked volume of synthetic blood could be considered as a latent indicator for infection risk, that the amount of exposure to contaminated blood corresponds to the risk of infection. Our study helped us ascertain, with high sensitivity, the differences among fabric materials with respect to their protective

performance, which may facilitate effective selection of protective clothing depending on the risk assessment.

Shishovs, M., Rumnieks, J., Diebolder, C., Jaudzems, K., Andreas, L. B., Stanek, J., Kazaks, A., Kotelovica, S., Akopjana, I., Pintacuda, G., Koning, R. I., and Tars, K. Structure of AP205 Coat Protein Reveals Circular Permutation in ssRNA Bacteriophages. J.Mol.Biol. 10-23-2016; 428(21):4267-4279; PM: 27591890

ABSTRACT: AP205 is a single-stranded RNA bacteriophage that has a coat protein sequence not similar to any other known single-stranded RNA phage. Here, we report an atomic-resolution model of the AP205 virus-like particle based on a crystal structure of an unassembled coat protein dimer and a cryo-electron microscopy reconstruction of the assembled particle, together with secondary structure information from site-specific solid-state NMR data. The AP205 coat protein dimer adopts the conserved Leviviridae coat protein fold except for the N-terminal region, which forms a beta-hairpin in the other known single-stranded RNA phages. AP205 has a similar structure at the same location formed by N- and C-terminal beta-strands, making it a circular permutant compared to the other coat proteins. The permutation moves the coat protein termini to the most surface-exposed part of the assembled particle, which explains its increased tolerance to long N- and C-terminal fusions.

Shlezinger, M., Houri-Haddad, Y., Coppenhagen-Glazer, S., Resch, G., Que, Y. A., Beyth, S., Dorfman, E., Hazan, R., and Beyth, N. Phage therapy: A new horizon in the antibacterial treatment of oral pathogens. Curr.Top.Med.Chem. 9-30-2016; PM: 27770768

ABSTRACT: Dental diseases are perhaps the most prevalent infection-related diseases in humans. Biofilm is involved in almost every infectious disease compromising oral health, notably caries, periodontal disease, gingivitis, endodontic infections and peri-implantitis. Current therapies of biofilm-derived oral infections lack sensitivity; they are not species-specific and kill pathogenic species as well as commensal species, which are protective against the formation of pathogenic biofilms. Moreover, antibiotics have a limited effect on biofilm and are almost unused in oral diseases. A promising alternative approach is bacteriophage (phage) therapy. Phages play a key role in the natural balance in a predator-prey relationship with bacteria and thus have the potential to be efficient anti-bacterial agents. Phages are highly efficient against biofilm, strain specific and easy to isolate and manipulate. Thus, like in many other medicinal fields, phage therapy offers new horizons to dentistry, both therapeutics and research. The present review presents the etiology of common oral diseases, characterization of the infection and the treatment challenges of phage therapy in dentistry. Recent findings and development in the use of phages for prevention, control, and treatment of oral infections as well as possibilities of engineering the oral microbiome are discussed.

Simoes, N. G., Bettencourt, A. F., Monge, N., and Ribeiro, R. A. Novel antibacterial agents: An emergent need to win the battle against infections. Mini.Rev.Med.Chem. 9-7-2016; PM: 27629996

ABSTRACT: Multiple approaches have been advised for control and prevention of antimicrobial resistance. Solutions will need to be found soon if we are not to run the serious risk of losing the ability to treat bacterial infections, especially the ones arising from multi-resistant strains. Thorough understanding of resistance mechanism and innovation in

new drugs, novel therapies and vaccines is needed. A multidisciplinary, collaborative, regulatory approach is demanded by this challenge. This review will be focused on new strategies to control infections. Among strategies to tackle antibiotic resistance that have been under investigation, especially during the last ten years, are the use of antimicrobial peptides, phage therapy and phage enzymes, therapeutic antibodies, quorum sensing inhibitors and, finally, the antibacterial nanomedicines. Although all of the approaches seem to be effective, and at least one of them has been in use for relatively a long time (phage therapy), antibacterial nanomedicines show the most promising results with regards to clinical and industrial applicability. Several advances have been made but a great effort is still mandatory in order to reach feasible, effective and marketable novel antimicrobial products.

Singh, M. I., Ganesh, B., and Jain, V. On the domains of T4 phage sliding clamp gp45: An intermolecular crosstalk governs structural stability and biological activity. Biochim.Biophys.Acta. 8-21-2016; PM: 27554844

ABSTRACT: ***BACKGROUND***: DNA polymerase processivity factors are ubiquitously present in all living organisms. Notwithstanding their high significance, the molecular details of clamps pertaining to the factors contributing to their stability are presently lacking. The bacteriophage T4 sliding clamp gp45 forms a homotrimer that besides being involved in DNA replication, moonlights as a transcription factor. Here we have carried out a detailed characterization of gp45 to understand the role of monomer-monomer interface interactions in stability and functioning of the protein. ***METHODS***: We generated several gp45 mutants harboring either Ala or Pro substitutions at the interface residues and performed a detailed investigation using biochemical and biophysical methods including circular dichroism, fluorescence anisotropy and quenching, differential scanning calorimetry, blue-native PAGE, cross-linking, size exclusion chromatography, and dynamic light scattering. We also carried out both transcription and DNA replication to understand the properties of the wild-type and the mutant proteins. ***RESULTS***: One specific mutation S88P leads not only to monomerization, but also results in an unstable molecule. Most interestingly, mutating either Q125 or K164 in the gp45 C-terminal domain negatively affects the stability of the N-terminal domain. We also report that these residues upon mutation to alanine make gp45 inactive for late promoter transcription, whereas strand-displacement DNA replication ability remains unaltered. ***CONCLUSIONS AND GENERAL SIGNIFICANCE***: The results suggest that the two domains of gp45 demonstrate an "inter-monomer" crosstalk that stabilizes the trimer. We also conclude that the residue-specific interactions at the interface allow the protein to function distinctly as replication and transcription factors.

Skliros, D., Kalatzis, P. G., Katharios, P., and Flemetakis, E. Comparative Functional Genomic Analysis of Two Vibrio Phages Reveals Complex Metabolic Interactions with the Host Cell. Front Microbiol. 2016; 7:1807; PM: 27895630

ABSTRACT: Sequencing and annotation was performed for two large double stranded DNA bacteriophages, phiGrn1 and phiSt2 of the Myoviridae family, considered to be of great interest for phage therapy against Vibrios in aquaculture live feeds. In addition, phage-host metabolic interactions and exploitation was studied by transcript profiling of selected viral and host genes. Comparative genomic analysis with other large Vibrio phages was also performed to establish the presence and location of homing endonucleases highlighting distinct features for both phages. Phylogenetic analysis

revealed that they belong to the "schizoT4like" clade. Although many reports of newly sequenced viruses have provided a large set of information, basic research related to the shift of the bacterial metabolism during infection remains stagnant. The function of many viral protein products in the process of infection is still unknown. Genome annotation identified the presence of several viral open reading frames (ORFs) participating in metabolism, including a Sir2/cobB (sirtuin) protein and a number of genes involved in auxiliary NAD+ and nucleotide biosynthesis, necessary for phage DNA replication. Key genes were subsequently selected for detail study of their expression levels during infection. This work suggests a complex metabolic interaction and exploitation of the host metabolic pathways and biochemical processes, including a possible post-translational protein modification, by the virus during infection.

Smeal, S. W., Schmitt, M. A., Pereira, R. R., Prasad, A., and Fisk, J. D. Simulation of the M13 life cycle II: Investigation of the control mechanisms of M13 infection and establishment of the carrier state. Virology. 2017; 500:275-284; PM: 27569186

ABSTRACT: Bacteriophage M13 is a true parasite of bacteria, able to co-opt the infected cell and control the production of progeny across many cellular generations. Here, our genetically-structured simulation of M13 is applied to quantitatively dissect the interplay between the host cellular environment and the controlling interactions governing the phage life cycle during the initial establishment of infection and across multiple cell generations. Multiple simulations suggest that phage-encoded feedback interactions constrain the utilization of host DNA polymerase, RNA polymerase and ribosomes. The simulation reveals the importance of p5 translational attenuation in controlling the production of phage double-stranded DNA and suggests an underappreciated role for p5 translational self-attenuation in resource allocation. The control elements active in a single generation are sufficient to reproduce the experimentally-observed multigenerational curing of the phage infection. Understanding the subtleties of regulation will be important for maximally exploiting M13 particles as scaffolds for nanoscale devices.

Smeal, S. W., Schmitt, M. A., Pereira, R. R., Prasad, A., and Fisk, J. D. Simulation of the M13 life cycle I: Assembly of a genetically-structured deterministic chemical kinetic simulation. Virology. 2017; 500:259-274; PM: 27644585

ABSTRACT: To expand the quantitative, systems level understanding and foster the expansion of the biotechnological applications of the filamentous bacteriophage M13, we have unified the accumulated quantitative information on M13 biology into a genetically-structured, experimentally-based computational simulation of the entire phage life cycle. The deterministic chemical kinetic simulation explicitly includes the molecular details of DNA replication, mRNA transcription, protein translation and particle assembly, as well as the competing protein-protein and protein-nucleic acid interactions that control the timing and extent of phage production. The simulation reproduces the holistic behavior of M13, closely matching experimentally reported values of the intracellular levels of phage species and the timing of events in the M13 life cycle. The computational model provides a quantitative description of phage biology, highlights gaps in the present understanding of M13, and offers a framework for exploring alternative mechanisms of regulation in the context of the complete M13 life cycle.

Soffer, N., Abuladze, T., Woolston, J., Li, M., Hanna, L. F., Heyse, S., Charbonneau, D., and Sulakvelidze, A. Bacteriophages safely reduce Salmonella contamination in pet food and raw pet food ingredients. Bacteriophage. 2016; 6(3):e1220347; PM: 27738557

ABSTRACT: Contamination of pet food with Salmonella is a serious public health concern, and several disease outbreaks have recently occurred due to human exposure to Salmonella tainted pet food. The problem is especially challenging for raw pet foods (which include raw meats, seafood, fruits, and vegetables). These foods are becoming increasingly popular because of their nutritional qualities, but they are also more difficult to maintain Salmonella-free because they lack heat-treatment. Among various methods examined to improve the safety of pet foods (including raw pet food), one intriguing approach is to use bacteriophages to specifically kill Salmonella serotypes. At least 2 phage preparations (SalmoFresh(R) and Salmonelex) targeting Salmonella are already FDA cleared for commercial applications to improve the safety of human foods. However, similar preparations are not yet available for pet food applications. Here, we report the results of evaluating one such preparation (SalmoLyse(R)) in reducing Salmonella levels in various raw pet food ingredients (chicken, tuna, turkey, cantaloupe, and lettuce). Application of SalmoLyse(R) in low (ca. 2-4x106 PFU/g) and standard (ca. 9x106 PFU/g) concentrations significantly ($P < 0.01$) reduced (by 60-92%) Salmonella contamination in all raw foods examined compared to control treatments. When SalmoLyse(R)-treated (ca. 2x107 PFU/g) dry pet food was fed to cats and dogs, it did not trigger any deleterious side effects in the pets. Our data suggest that the bacteriophage cocktail lytic for Salmonella can significantly and safely reduce Salmonella contamination in various raw pet food ingredients.

Stalin, N. and Srinivasan, P. Characterization of Vibrio parahaemolyticus and its specific phage from shrimp pond in Palk Strait, South East coast of India. Biologicals. 2016; 44(6):526-533; PM: 27697363

ABSTRACT: Phage therapy is an alternative and eco-friendly biocontrol agent to prevent and control multidrug resistant bacteria in the aquatic system. The aim of this study is to isolate and characterize the Vibrio parahaemolyticus and its potential lytic phage from Penaeus monodon growing-out by rearing in shrimp ponds in Palk Strait, South East coast of India. The conventional phenotypic characteristics and molecular identification was confirmed using 16S rRNA sequence and to determine the antibiotic resistant profiles. The V. parahaemolyticus phage was effective against V. parahaemolyticus through one-step growth experiments, phage survival was determined by long-term storage at various temperatures and pH. Further, transmission electron microscope (TEM) revealed that the lytic phage belongs to the Myoviridae family. The isolated lytic phage (VVP1) was more specific against N1A V. parahaemolyticus and was able to infect N7A V. parahaemolyticus, N3B and N13B Vibrio alginolyticus strains. Evaluation of microcosm studies with P. monodon larvae infected with V. parahaemolyticus showed the survival of larvae in the presence of phage treatment at 2.3 x 1010 PFU/mL-1 was enhanced when compared with the control. This study provides the application of phage as a useful strategy to prevent and eliminate or reduce shrimp pathogenic V. parahaemolyticus in the aquaculture system.

Stamereilers, C., LeBlanc, L., Yost, D., Amy, P. S., and Tsourkas, P. K. Comparative genomics of 9 novel Paenibacillus larvae bacteriophages. Bacteriophage. 2016; 6(3):e1220349; PM: 27738559

ABSTRACT: American Foulbrood Disease, caused by the bacterium Paenibacillus larvae, is one of the most destructive diseases of the honeybee, Apis mellifera. Our group recently published the sequences of 9 new phages with the ability to infect and lyse P. larvae. Here, we characterize the genomes of these P. larvae phages, compare them to each other and to other sequenced P. larvae phages, and putatively identify protein function. The phage genomes are 38-45 kb in size and contain 68-86 genes, most of which appear to be unique to P. larvae phages. We classify P. larvae phages into 2 main clusters and one singleton based on nucleotide sequence identity. Three of the new phages show sequence similarity to other sequenced P. larvae phages, while the remaining 6 do not. We identified functions for roughly half of the P. larvae phage proteins, including structural, assembly, host lysis, DNA replication/metabolism, regulatory, and host-related functions. Structural and assembly proteins are highly conserved among our phages and are located at the start of the genome. DNA replication/metabolism, regulatory, and host-related proteins are located in the middle and end of the genome, and are not conserved, with many of these genes found in some of our phages but not others. All nine phages code for a conserved N-acetylmuramoyl-L-alanine amidase. Comparative analysis showed the phages use the "cohesive ends with 3' overhang" DNA packaging strategy. This work is the first in-depth study of P. larvae phage genomics, and serves as a marker for future work in this area.

Sun, Y. and Sun, Y. [Preparation and activity validation of PP7 bacteriophage-like particles displaying PAP114-128 peptide]. Xi.Bao.Yu Fen.Zi.Mian.Yi.Xue.Za Zhi. 2016; 32(10):1356-1361; PM: 27667462

ABSTRACT: Objective To obtain the PP7 bacteriophage-like particles carrying the peptide of prostatic acid phosphatase PAP114-128, and prove that they retain the original biological activity. Methods First, the plasmid pETDuet-2PP7 was constructed as follows: the gene of PP7 coat protein dimer was amplified by gene mutation combined with overlapping PCR technology, and inserted into the vector pETDuet-1. Following that, the plasmid pETDuet-2PP7-PAP114-128 was constructed as follows: the PP7 coat protein gene carrying the coding gene of PAP114-128 peptide was amplified using PCR, and then inserted into the vector pETDuet-2PP7. Both pETDuet-2PP7 and pETDuet-2PP7-PAP114-128 were transformed into E.coli and expressed. The expression product was verified by SDS-PAGE, double immunodiffusion assay and ELISA. Results The gene fragment of PP7 coat protein dimer was obtained by overlapping PCR using Ex Taq DNA polymerase, and the antigenicity of its expression product was the same as that of the coat protein of wild-type PP7 bacteriophage. Moreover, the PAP114-128 peptide epitope that was displayed on the surface of PP7 bacteriophage was identical with the corresponding epitope of natural human PAP, and it was able to induce high levels of antibodies. Conclusion The gene of PP7 coat protein dimer with repeated sequences can be prepared by gene mutation combined with overlapping PCR. Based on this, PP7 bacteriophage-like particles carrying PAP peptide can be prepared, which not only solves the problem of the instability of the peptides, but also lays a foundation for the study on their delivery and function.

Swift, S. M., Rowley, D. T., Young, C., Franks, A., Hyman, P., and Donovan, D. M. The endolysin from the Enterococcus faecalis bacteriophage VD13 and conditions stimulating its lytic activity. FEMS Microbiol.Lett. 9-14-2016; PM: 27634307

ABSTRACT: Bacteriophage produce endolysins (peptidoglycan hydrolases) to lyse the host cell from within and release nascent bacteriophage particles. Recombinant endolysins can lyse Gram-positive bacteria when added exogenously. As a potential alternative antimicrobial, we cloned and expressed the enterococcal VD13 bacteriophage endolysin. VD13 endolysin has a CHAP catalytic domain with 92% identity with the bacteriophage IME-EF1 endolysin. The predicted size of VD13 endolysin is approximately 27 kDa as verified by SDS-PAGE. The VD13 endolysin lyses Enterococcus faecalis strains, but not Enterococcus faecium or other non-enterococci. VD13 endolysin has activity from pH 4 to pH 8, with peak activity at pH 5, and exhibits greater activity in the presence of calcium. Optimum activity at pH 5 occurs in the absence of NaCl. VD13 endolysin, in ammonium acetate (C2H3O2NH4) calcium chloride (CaCl2) buffer pH 5, is stimulated to higher activity upon heating at temperatures up to 65 degrees C for 30 minutes, whereas activity is lost upon heating to 42 degrees C, in pH 7 buffer.

Szot-Karpinska, K., Golec, P., Lesniewski, A., Palys, B., Marken, F., Niedziolka-Jonsson, J., Wegrzyn, G., and Los, M. Modified Filamentous Bacteriophage as a Scaffold for Carbon Nanofiber. Bioconjug.Chem. 10-17-2016; PM: 27748604

ABSTRACT: With the advent of nanotechnology, carbon nanomaterials such as carbon nanofibers (CNF) have aroused substantial interest in various research fields including energy storage and sensing. Further improvement of their properties might be achieved via application of viral particles such as bacteriophages. In this report we present a filamentous M13 bacteriophage, with a point mutation in gene VII (pVII-mutant-M13), that selectively binds to the carbon nanofibers to form 3D structures. The phage-display technique was utilized for the selection of the pVII-mutant-M13 phage from the phage display peptide library. The properties of this phage make it a prospective candidate for a scaffold material for CNFs. The results for binding of CNF by mutant phage were compared with those for maternal bacteriophage (pVII-M13). The efficiency of binding between pVII-mutant-M13 and CNF is about two orders of magnitude higher compared to the pVII-M13. Binding affinity between the pVII-mutant-M13 and CNF was also characterized using atomic force microscopy, scanning electron microscopy and transmission electron microscopy, which confirmed the specificity of the interaction of the phage pVII-mutant-M13 and the.

Tan, C. W., Tee, H. K., Lee, M. H., Sam, I. C., and Chan, Y. F. Enterovirus A71 DNA-Launched Infectious Clone as a Robust Reverse Genetic Tool. PLoS.One. 2016; 11(9):e0162771; PM: 27617744

ABSTRACT: Enterovirus A71 (EV-A71) causes major outbreaks of hand, foot and mouth disease, and is occasionally associated with neurological complications and death in children. Reverse genetics is widely used in the field of virology for functional study of viral genes. For EV-A71, such tools are limited to clones that are transcriptionally controlled by T7/SP6 bacteriophage promoter. This is often time-consuming and expensive. Here, we describe the development of infectious plasmid DNA-based EV-A71 clones, for which EV-A71 genome expression is under transcriptional control by the CMV-intermediate early promoter and SV40 transcriptional-termination signal. Transfection of this EV-A71

infectious DNA produces good virus yield similar to in vitro-transcribed EV-A71 infectious RNA, 6.4 and 5.8 log10PFU/ml, respectively. Infectious plasmid with enhanced green fluorescence protein and Nano luciferase reporter genes also produced good virus titers, with 4.3 and 5.0 log10 PFU/ml, respectively. Another infectious plasmid with both CMV and T7 promoters was also developed for easy manipulation of in vitro transcription or direct plasmid transfection. Transfection with either dual-promoter infectious plasmid DNA or infectious RNA derived from this dual-promoter clone produced infectious viral particles. Incorporation of hepatitis delta virus ribozyme, which yields precise 3' ends of the DNA-launched EV-A71 genomic transcripts, increased infectious viral production. In contrast, the incorporation of hammerhead ribozyme in the DNA-launched EV-A71 resulted in lower virus yield, but improved the virus titers for T7 promoter-derived infectious RNA. This study describes rapid and robust reverse genetic tools for EV-A71.

Thomas, J. A., itez Quintana, A. D., Bosch, M. A., Coll De, Pena A., Aguilera, E., Coulibaly, A., Wu, W., Osier, M. V., Hudson, A. O., Weintraub, S. T., and Black, L. W. Identification of Essential Genes in the Salmonella Phage SPN3US Reveals Novel Insights into Giant Phage Head Structure and Assembly. J.Virol. 11-15-2016; 90(22):10284-10298; PM: 27605673

ABSTRACT: Giant tailed bacterial viruses, or phages, such as Pseudomonas aeruginosa phage varphiKZ, have long genomes packaged into large, atypical virions. Many aspects of varphiKZ and related phage biology are poorly understood, mostly due to the fact that the functions of the majority of their proteins are unknown. We hypothesized that the Salmonella enterica phage SPN3US could be a useful model phage to address this gap in knowledge. The 240-kb SPN3US genome shares a core set of 91 genes with varphiKZ and related phages, approximately 61 of which are virion genes, consistent with the expectation that virion complexity is an ancient, conserved feature. Nucleotide sequencing of 18 mutants enabled assignment of 13 genes as essential, information which could not have been determined by sequence-based searches for 11 genes. Proteome analyses of two SPN3US virion protein mutants with knockouts in 64 and 241 provided new insight into the composition and assembly of giant phage heads. The 64 mutant analyses revealed all the genetic determinants required for assembly of the SPN3US head and a likely head-tail joining role for gp64, and its homologs in related phages, due to the tailless-particle phenotype produced. Analyses of the mutation in 241, which encodes an RNA polymerase beta subunit, revealed that without this subunit, no other subunits are assembled into the head, and enabled identification of a "missing" beta' subunit domain. These findings support SPN3US as an excellent model for giant phage research, laying the groundwork for future analyses of their highly unusual virions, host interactions, and evolution. IMPORTANCE: In recent years, there has been a paradigm shift in virology with the realization that extremely large viruses infecting prokaryotes (giant phages) can be found in many environments. A group of phages related to the prototype giant phage varphiKZ are of great interest due to their virions being among the most complex of prokaryotic viruses and their potential for biocontrol and phage therapy applications. Our understanding of the biology of these phages is limited, as a large proportion of their proteins have not been characterized and/or have been deemed putative without any experimental verification. In this study, we analyzed Salmonella phage SPN3US using a combination of genomics, genetics, and proteomics and in doing so revealed new information regarding giant phage head structure and assembly and virion RNA polymerase composition. Our findings demonstrate the suitability of SPN3US as a model phage for the growing group of phages related to varphiKZ.

Thomason, L. C., Costantino, N., and Court DL. Examining a DNA Replication Requirement for Bacteriophage lambda Red- and Rac Prophage RecET-Promoted Recombination in Escherichia coli. MBio. 9-13-2016; 7(5); PM: 27624131

ABSTRACT: Recombineering, in vivo genetic engineering with bacteriophage homologous recombination systems, is a powerful technique for making genetic modifications in bacteria. Two systems widely used in Escherichia coli are the Red system from phage lambda and RecET from the defective Rac prophage. We investigated the in vivo dependence of recombineering on DNA replication of the recombining substrate using plasmid targets. For lambda Red recombination, when DNA replication of a circular target plasmid is prevented, recombination with single-stranded DNA oligonucleotides is greatly reduced compared to that under replicating conditions. For RecET recombination, when DNA replication of the targeted plasmid is prevented, the recombination frequency is also reduced, to a level identical to that seen for the Red system in the absence of replication. The very low level of oligonucleotide recombination observed in the absence of any phage recombination functions is the same in the presence or absence of DNA replication. In contrast, both the Red and RecET systems recombine a nonreplicating linear dimer plasmid with high efficiency to yield a circular monomer. Therefore, the DNA replication requirement is substrate dependent. Our data are consistent with recombination by both the Red and RecET systems occurring predominately by single-strand annealing rather than by strand invasion. IMPORTANCE: Bacteriophage homologous recombination systems are widely used for in vivo genetic engineering in bacteria. Single- or double-stranded linear DNA substrates containing short flanking homologies to chromosome targets are used to generate precise and accurate genetic modifications when introduced into bacteria expressing phage recombinases. Understanding the molecular mechanism of these recombination systems will facilitate improvements in the technology. Here, two phage-specific systems are shown to require exposure of complementary single-strand homologous targets for efficient recombination; these single-strand regions may be created during DNA replication or by single-strand exonuclease digestion of linear duplex DNA. Previously, in vitro studies reported that these recombinases promote the single-strand annealing of two complementary DNAs and also strand invasion of a single DNA strand into duplex DNA to create a three-stranded region. Here, in vivo experiments show that recombinase-mediated annealing of complementary single-stranded DNA is the predominant recombination pathway in E. coli.

Thummeepak, R., Kitti, T., Kunthalert, D., and Sitthisak, S. Enhanced Antibacterial Activity of Acinetobacter baumannii Bacteriophage OABP-01 Endolysin (LysABP-01) in Combination with Colistin. Front Microbiol. 2016; 7:1402; PM: 27656173

ABSTRACT: Endolysins are lytic enzymes produced by bacteriophages with their ability to degrade the cell wall of bacterial hosts. Endolysin (LysABP-01) from Acinetobacter baumannii bacteriophage OABP-01 was cloned, overexpressed and characterized. Endolysin LysABP-01 has a globular structure consisting of lysozyme-like (N-acetyl-beta-D-muramidase) catalytic domain. It contains 185 amino acids which correspond to a 21.1 kDa protein. The lytic activity of the recombinant endolysin protein was determined by a plate lysis assay for its ability to lyse the autoclaved cell (crude cell wall) of the different bacterial species. LysABP-01 can degrade the crude cell wall of A. baumannii strains, Escherichia coli and Pseudomonas aeruginosa but not of Staphylococcus aureus. The antibacterial activity of LysABP-01 and its synergism with various antibiotics were tested.

The results exhibited elevated antibacterial activity in a combination of the sub-MIC LysABP-01 and colistin. The checkerboard assay for measuring antibiotic synergy of LysABP-01 and colistin was performed. This combination was synergistic against various drug-resistant strains of A. baumannii (FIC index < 0.5). In summary, our study highlights the ability of LysABP-01 endolysin to hydrolyze the A. baumannii cell wall and its synergistic interaction with colistin.

Tian, Y., Yan, X., Saha, M. L., Niu, Z., and Stang, P. J. Hierarchical Self-Assembly of Responsive Organoplatinum(II) Metallacycle-TMV Complexes with Turn-On Fluorescence. J.Am.Chem.Soc. 9-21-2016; 138(37):12033-12036; PM: 27608138

ABSTRACT: Here we report that the rod-like tobacco mosaic virus (TMV), having a negatively charged surface, can be assembled into three-dimensional micrometer-sized bundle-like superstructures via multiple electrostatic interactions with a positively charged molecular "glue", namely, a tetraphenylethylene (TPE)-based discrete organoplatinum(II) metallacycle (TPE-Pt-MC). Due to the nanoconfinement effect in the resultant TMV/TPE-Pt-MC complexes and the aggregation-induced emission (AIE) activity of the TPE units, these hierarchical architectures result in a dramatic fluorescence enhancement that not only provides evidence for the formation of novel metal-organic biohybrid materials but also represents an alternative to turn-on fluorescence. Moreover, the dissociation of these final constructs and subsequent release of individual virus have been achieved by disrupting the TPE-Pt-MC core using tetrabutylammonium bromide (TBAB). This strategy is also compatible with other protein-based nanoparticles such as bacteriophage M13 and ferritin, proving the generality of this approach. Hence, this research will open new routes for the fabrication of functional biohybrid materials involving metal-organic complexes and anisotropically shaped bionanoparticles.

Tripler, T. N., Teschke, C. M., and Alexandrescu, A. T. NMR assignments for the insertion domain of bacteriophage Sf6 coat protein. Biomol.NMR Assign. 10-31-2016; PM: 27798771

ABSTRACT: The P22 bacteriophage group is a subgroup of the lambda phage supercluster, comprised of the three major sequence types Sf6, P22, and CUS-3, based on their capsid proteins. Our goal is to investigate the extent to which structure-function relationships are conserved for the viral coat proteins and I-domains in this subgroup. Sf6 is a phage that infects the human pathogen Shigella flexneri. The coat protein of Sf6 assembles into a procapsid, which further undergoes maturation during DNA packaging into an infectious virion. The Sf6 coat protein contains a genetically inserted domain, termed the I-domain, similar to the ones present in the P22 and CUS-3 coat proteins. Based on the P22 example, I-domains play important functional roles in capsid assembly, stability, viability, and size-determination. Here we report the 1H, 15N, and 13C chemical shift assignments for the I-domain of the Sf6 phage coat protein. Chemical shift-based secondary structure prediction and hydrogen-bond patterns from a long-range HNCO experiment indicate that the Sf6 I-domain adopts a 6-stranded beta-barrel fold like those of P22 and CUS-3 but with important differences, including the absence of the D-loop that is critical for capsid assembly and the addition of a novel disordered loop region.

Tucker, M. B., MacKenzie, S. H., Maciag, J. J., Dirscherl, Ackerman H., Swartz, P., Yoder, J. A., Hamilton, P. T., and Clay, Clark A. Phage display and structural studies reveal plasticity in

substrate specificity of caspase-3a from zebrafish. Protein Sci. 2016; 25(11):2076-2088; PM: 27577093

ABSTRACT: The regulation of caspase-3 enzyme activity is a vital process in cell fate decisions leading to cell differentiation and tissue development or to apoptosis. The zebrafish, Danio rerio, has become an increasingly popular animal model to study several human diseases because of their transparent embryos, short reproductive cycles, and ease of drug administration. While apoptosis is an evolutionarily conserved process in metazoans, little is known about caspases from zebrafish, particularly regarding substrate specificity and allosteric regulation compared to the human caspases. We cloned zebrafish caspase-3a (casp3a) and examined substrate specificity of the recombinant protein, Casp3a, compared to human caspase-3 (CASP3) by utilizing M13 bacteriophage substrate libraries that incorporated either random amino acids at P5-P1' or aspartate fixed at P1. The results show a preference for the tetrapeptide sequence DNLD for both enzymes, but the P4 position of zebrafish Casp3a also accommodates valine equally well. We determined the structure of zebrafish Casp3a to 2.28A resolution by X-ray crystallography, and when combined with molecular dynamics simulations, the results suggest that a limited number of amino acid substitutions near the active site result in plasticity of the S4 sub-site by increasing flexibility of one active site loop and by affecting hydrogen-bonding with substrate. The data show that zebrafish Casp3a exhibits a broader substrate portfolio, suggesting overlap with the functions of caspase-6 in zebrafish development.

Urquhart, T., Daub, E., and Honek, J. F. Bioorthogonal Modification of the Major Sheath Protein of Bacteriophage M13: Extending the Versatility of Bionanomaterial Scaffolds. Bioconjug.Chem. 10-19-2016; 27(10):2276-2280; PM: 27626459

ABSTRACT: With a mass of approximately 1.6 x 107 Daltons and composed of approximately 2700 proteins, bacteriophage M13 has been employed as a molecular scaffold in bionanomaterials fabrication. In order to extend the versatility of M13 in this area, residue-specific unnatural amino acid incorporation was employed to successfully display azide functionalities on specific solvent-exposed positions of the pVIII major sheath protein of this bacteriophage. Employing a combination of engineered mutants of the gene coding for the pVIII protein, the methionine (Met) analog, l-azidohomoalanine (Aha), and a suitable Escherichia coli Met auxotroph for phage production, conditions were developed to produce M13 bacteriophage labeled with over 350 active azides (estimated by fluorescent dye labeling utilizing a strain-promoted azide-alkyne cycloaddition) and capable of azide-selective attachment to 5 nm gold nanoparticles as visualized by transmission electron microscopy. The capability of this system to undergo dual labeling utilizing both chemical acylation and bioorthogonal cycloaddition reactions was also verified. The above stratagem should prove particularly advantageous in the preparation of assemblies of larger and more complex molecular architectures based on the M13 building block.

Van Belleghem, J. D., Merabishvili, M., Vergauwen, B., Lavigne, R., and Vaneechoutte, M. A comparative study of different strategies for removal of endotoxins from bacteriophage preparations. J.Microbiol.Methods. 11-29-2016; PM: 27913133

ABSTRACT: Bacterial endotoxins have high immunogenicity. Phage biology studies as well

as therapeutic phage applications necessitate highly purified phage particles. In this study, we compared combinations of seven different endotoxin removal strategies and validated their endotoxin removal efficacy for five different phages (i.e. four Pseudomonas aeruginosa phages and one Staphylococcus aureus phage). These purification strategies included Endotrap HD column purification and/or CsCl density centrifugation in combination with Endotrap purification, followed by organic solvent (1-octanol), detergent (Triton X-100), enzymatic inactivation of the endotoxin using alkaline phosphatase and CIM monolytic anion exchange chromatography. We show that CsCl density purification of the P. aeruginosa phages, at an initial concentration of 1012-1013pfu/ml, led to the strongest reduction of endotoxins, with an endotoxin removal efficacy of up to 99%, whereas additional purification methods did not result in a complete removal of endotoxins from the phage preparations and only yielded an additional endotoxin removal efficacy of 23 to 99%, sometimes accompanied with strong losses in phage titer.

van Zyl, L. J., Nemavhulani, S., Cass, J., Cowan, D. A., and Trindade, M. Three novel bacteriophages isolated from the East African Rift Valley soda lakes. Virol.J. 12-3-2016; 13(1):204; PM: 27912769

ABSTRACT: ***BACKGROUND***: Soda lakes are unique environments in terms of their physical characteristics and the biology they harbour. Although well studied with respect to their microbial composition, their viral compositions have not, and consequently few bacteriophages that infect bacteria from haloalkaline environments have been described. ***METHODS***: Bacteria were isolated from sediment samples of lakes Magadi and Shala. Three phages were isolated on two different Bacillus species and one Paracoccus species using agar overlays. The growth characteristics of each phage in its host was investigated and the genome sequences determined and analysed by comparison with known phages. ***RESULTS***: Phage Shbh1 belongs to the family Myoviridae while Mgbh1 and Shpa belong to the Siphoviridae family. Tetranucleotide usage frequencies and G + C content suggests that Shbh1 and Mgbh1 do not regularly infect, and have therefore not evolved with, the hosts they were isolated on here. Shbh1 was shown capable of infecting two different Bacillus species from the two different lakes demonstrating its potential broad-host range. Comparative analysis of their genome sequence with known phages revealed that, although novel, Shbh1 does share substantial amino acid similarity with previously described Bacillus infecting phages (Grass, phiNIT1 and phiAGATE) and belongs to the Bastille group, while Mgbh1 and Shpa are highly novel. ***CONCLUSION***: The addition of these phages to current databases should help with metagenome/metavirome annotation efforts. We describe a highly novel Paracoccus infecting virus (Shpa) which together with NgoPhi6 and vB_PmaS_IMEP1 is one of only three phages known to infect Paracoccus species but does not show similarity to these phages.

Vaughan, M. D., Su, Z., Daub, E., and Honek, J. F. Intriguing cellular processing of a fluorinated amino acid during protein biosynthesis in Escherichia coli. Org.Biomol.Chem. 9-26-2016; 14(38):8942-8946; PM: 27722405

ABSTRACT: Bioincorporation of the methionine analogue S-(2-fluoroethyl)-l-homocysteine (l-MFE) into bacteriophage lysozyme overproduced in Escherichia coli results not only in the expected l-MFE incorporation but surprisingly substantial l-vinthionine incorporation into the labeled lysozymes. Synthetic l-vinthionine itself however is not activated by

purified Escherichia coli methionyl-tRNA synthetase. The indirect preparation of vinthionine-containing proteins has the potential to be an alternate strategy to prepare vinyl thioether moieties for click chemistry applications on proteins.

Visweswaran, G. R., Kurek, D., Szeliga, M., Pastrana, F. R., Kuipers, O. P., Kok, J., and Buist, G. Expression of prophage-encoded endolysins contributes to autolysis of Lactococcus lactis. Appl.Microbiol.Biotechnol. 9-22-2016; PM: 27660179

ABSTRACT: Analysis of autolysis of derivatives of Lactococcus lactis subsp. cremoris MG1363 and subsp. lactis IL1403, both lacking the major autolysin AcmA, showed that L. lactis IL1403 still lysed during growth while L. lactis MG1363 did not. Zymographic analysis revealed that a peptidoglycan hydrolase activity of around 30 kDa is present in cell extracts of L. lactis IL1403 that could not be detected in strain MG1363. A comparison of all genes encoding putative peptidoglycan hydrolases of IL1403 and MG1363 led to the assumption that one or more of the 99 % homologous 27.9-kDa endolysins encoded by the prophages bIL285, bIL286 and bIL309 could account for the autolysis phenotype of IL1403. Induced expression of the endolysins from bIL285, bIL286 or bIL309 in L. lactis MG1363 resulted in detectable lysis or lytic activity. Prophage deletion and insertion derivatives of L. lactis IL1403 had a reduced cell lysis phenotype. RT-qPCR and zymogram analysis showed that each of these strains still expressed one or more of the three phage lysins. A homologous gene and an endolysin activity were also identified in the natural starter culture L. lactis subsp. cremoris strains E8, Wg2 and HP, and the lytic activity could be detected under growth conditions that were identical as those used for IL1403. The results presented here show that these endolysins of L. lactis are expressed during normal growth and contribute to autolysis without production of (lytic) phages. Screening for natural strains expressing homologous endolysins could help in the selection of strains with enhanced autolysis and, thus, cheese ripening properties.

Wagner, N., Brinks, E., Samtlebe, M., Hinrichs, J., Atamer, Z., Kot, W., Franz, C. M., Neve, H., and Heller, K. J. Whey powders are a rich source and excellent storage matrix for dairy bacteriophages. Int.J.Food Microbiol. 1-16-2017; 241:308-317; PM: 27835774

ABSTRACT: Thirteen whey powders and 5 whey powder formulations were screened for the presence of dairy bacteriophages using a representative set of 8 acid-producing Lactococcus lactis and 5 Streptococcus thermophilus, and 8 flavour-producing Leuconostoc pseudomesenteroides and Leuconostoc mesenteroides strains. Lytic L. lactis phages were detected in all samples, while S. thermophilus and Leuconostoc phages were present in 50% or 40% of the samples, respectively. Maximal phage titers were 6x107 plaque-forming units (pfu)/g of whey powder for L. lactis phages, 1x107pfu/g for Leuconostoc phages and 1x105pfu/g for S. thermophilus phages. In total, 55 phages were isolated and characterized. Thirty one of the 33 lactococcal phages tested belonged to the wide-spread 936 phage group. In the course of this study, a PCR detection method for Leuconostoc phages (Ali et al., 2013) was adapted to new phage isolates. Furthermore, a remarkably high stability of phages in whey powder samples was documented during a long-term storage period of 4 years.

Wang, J. B., Lin, N. T., Tseng, Y. H., and Weng, S. F. Genomic Characterization of the Novel Aeromonas hydrophila Phage Ahp1 Suggests the Derivation of a New Subgroup from phiKMV-Like Family. PLoS.One. 2016; 11(9):e0162060; PM: 27603936

ABSTRACT: Aeromonas hydrophila is an opportunistic pathogenic bacterium causing diseases in human and fish. The emergence of multidrug-resistant A. hydrophila isolates has been increasing in recent years. In this study, we have isolated a novel virulent podophage of A. hydrophila, designated as Ahp1, from waste water. Ahp1 has a rapid adsorption (96% adsorbed in 2 min), a latent period of 15 min, and a burst size of 112 PFU per infected cell. At least eighteen Ahp1 virion proteins were visualized in SDS-polyacrylamide gel electrophoresis, with a 36-kDa protein being the predicted major capsid protein. Genome analysis of Ahp1 revealed a linear doubled-stranded DNA genome of 42,167 bp with a G + C content of 58.8%. The genome encodes 46 putative open reading frames, 5 putative phage promoters, and 3 transcriptional terminators. Based on high degrees of similarity in overall genome organization and among most of the corresponding ORFs, as well as phylogenetic relatedness among their DNAP, RNAP and major capsid proteins, we propose a new subgroup, designated Ahp1-like subgroup. This subgroup contains Ahp1 and members previously belonging to phiKMV-like subgroup, phiAS7, phi80-18, GAP227, phiR8-01, and ISAO8. Since Ahp1 has a narrow host range, for effective phage therapy, different phages are needed for preparation of cocktails that are capable of killing the heterogeneous A. hydrophila strains.

Wang, S., Liu, Y., Li, D., Zhou, T., Gao, S., Zha, E., and Yue, X. Preparation and evaluation of MS2 bacteriophage-like particles packaging hepatitis E virus RNA. FEMS Microbiol.Lett. 2016; 363(20); PM: 27664054

ABSTRACT: Hepatitis E virus (HEV) is the pathogen causing hepatitis E (HE). It arouses global public health concern since it is a zoonotic disease. The objective of this letter is to report a cost-effective internal control prepared for monitoring procedures of HEV reverse transcriptase (RT)-PCR detection. A selected conserved HEV RNA fragment was integrated into the downstream of the truncated MS2 bacteriophage genome based on Armored RNA technology. The resulting MS2-HEV gene harbored by the pET-28b-MS2-HEV plasmid was transformed into E. coli BL21(DE3) for expression analysis by SDS-PAGE. The expression products were purified and concentrated by ultrasonication and ultrafiltration separation. The morphology and stability properties of the virus-like particles (VLPs) were evaluated by electron microscopy scanning and nuclease challenges, respectively. SDS-PAGE results showed that the constructed MS2-HEV gene expressed efficiently and the purity of the VLPs was highly consistent with the result in electron microscopy. Stability evaluation results demonstrated that the prepared VLPs exhibited strong resistance to DNase I and RNase A attacks and also performed long-lasting protection of coated HEV RNA for at least 4 months at -20 degrees C. These data revealed that the prepared VLPs meet the basic requirements of use as internal control material in the HEV RNA amplification assay.

Wang, X., Liu, J., Yu, Z., Jin, J., Liu, X., and Wang, G. Novel groups of cyanobacterial podovirus DNA polymerase (pol) genes exist in paddy waters in northeast China. FEMS Microbiol.Ecol. 2016; 92(12); PM: 27612493

ABSTRACT: In this study, we surveyed cyanopodovirus DNA polymerase (pol) sequences in paddy waters using the culture-independent PCR and Sanger sequencing methods.

Four paddy waters generated from a pot experiment with different soil types collected from op E: n paddy fields in northeast China were used in this study. A total of 438 DNA pol clones were identified as cyanopodoviruses. The clones from the paddy waters formed nine unique groups of cyanopodoviruses either exclusively or with clones from East Lake in China (subclusters alpha-1 to alpha-8 and cluster beta). None of the clones from open oceans or coastal waters fell into these unique groups. Additionally, the distribution proportions of the clones into different cyanopodovirus groups varied among paddy water samples, which suggested that the cyanopodovirus compositions were spatially distributed in the paddy fields. The comparison of clone libraries in different studies indicated that the diversity of cyanopodoviruses in paddy waters was comparable to the diversity in the open oceans but was less than the diversity in the coastal estuary of Chesapeake Bay. Non-metric multidimensional scaling analysis indicated that the cyanopodovirus communities in paddy waters were similar to those in lake freshwater but distinct from the communities in marine and coastal waters.

Wang, X., Wei, Z., Li, M., Wang, X., Shan, A., Mei, X., Jousset, A., Shen, Q., Xu, Y., and Friman, V. P. Parasites and competitors suppress bacterial pathogen synergistically due to evolutionary trade-offs. Evolution. 12-7-2016; PM: 27925169

ABSTRACT: Parasites and competitors are important for regulating pathogen densities and subsequent disease dynamics. It is, however, unclear to what extent this is driven by ecological and evolutionary processes. Here we used experimental evolution to study the eco-evolutionary feedbacks between Ralstonia solanacearum bacterial pathogen, Ralstonia-specific phage parasite and Bacillus amyloliquefaciens competitor bacterium in the laboratory and plant rhizosphere. We found that while the phage had a small effect on pathogen densities on its own, it considerably increased the R. solanacearum sensitivity to antibiotics produced by B. amyloliquefaciens. Instead of density effects, this synergy was due to phage-driven increase in phage resistance that led to trade-off with the resistance to B. amyloliquefaciens antibiotics. While no evidence was found for pathogen resistance evolution to B. amyloliquefaciens antibiotics, the fitness cost of adaptation (reduced growth) was highest when the pathogen had evolved in the presence of both parasite and competitor. Qualitatively similar patterns were found between laboratory and greenhouse experiments even though the evolution of phage resistance was considerably attenuated in the tomato rhizosphere. These results suggest that evolutionary trade-offs can impose strong constraints on disease dynamics and that combining phages and antibiotic-producing bacteria could be an efficient way to control agricultural pathogens. This article is protected by copyright. All rights reserved.

Weber-Dabrowska, B., Jonczyk-Matysiak, E., Zaczek, M., Lobocka, M., Lusiak-Szelachowska, M., and Gorski, A. Bacteriophage Procurement for Therapeutic Purposes. Front Microbiol. 2016; 7:1177; PM: 27570518

ABSTRACT: Bacteriophages (phages), discovered 100 years ago, are able to infect and destroy only bacterial cells. In the current crisis of antibiotic efficacy, phage therapy is considered as a supplementary or even alternative therapeutic approach. Evolution of multidrug-resistant and pandrug-resistant bacterial strains poses a real threat, so it is extremely important to have the possibility to isolate new phages for therapeutic purposes. Our phage laboratory and therapy center has extensive experience with phage isolation, characterization, and therapeutic application. In this article we present current progress in bacteriophages isolation and use for therapeutic purposes, our

experience in this field and its practical implications for phage therapy. We attempt to summarize the state of the art: properties of phages, the methods for their isolation, criteria of phage selection for therapeutic purposes and limitations of their use. Perspectives for the use of genetically engineered phages to specifically target bacterial virulence-associated genes are also briefly presented.

Weber-Dabrowska, B., Jonczyk-Matysiak, E., Zaczek, M., Lobocka, M. B., Lusiak-Szelachowska, M., and Gorski, A. Corrigendum: Bacteriophage Procurement for Therapeutic Purposes. Front Microbiol. 2016; 7:1813; PM: 27833608

ABSTRACT: [This corrects the article on p. 1177 in vol. 7, PMID: 27570518.].

Weber, B. S., Hennon, S. W., Wright, M. S., Scott, N. E., de, Berardinis, V, Foster, L. J., Ayala, J. A., Adams, M. D., and Feldman, M. F. Genetic Dissection of the Type VI Secretion System in Acinetobacter and Identification of a Novel Peptidoglycan Hydrolase, TagX, Required for Its Biogenesis. MBio. 10-11-2016; 7(5); PM: 27729508

ABSTRACT: The type VI secretion system (T6SS) is a widespread secretory apparatus produced by Gram-negative bacteria that has emerged as a potent mediator of antibacterial activity during interbacterial interactions. Most Acinetobacter species produce a genetically conserved T6SS, although the expression and functionality of this system vary among different strains. Some pathogenic Acinetobacter baumannii strains activate this secretion system via the spontaneous loss of a plasmid carrying T6SS repressors. In this work, we compared the expression of T6SS-related genes via transcriptome sequencing and differential proteomics in cells with and without the plasmid. This approach, together with the mutational analysis of the T6SS clusters, led to the determination of the genetic components required to elaborate a functional T6SS in the nosocomial pathogen A. baumannii and the nonpathogen A. baylyi By constructing a comprehensive combination of mutants with changes in the T6SS-associated vgrG genes, we delineated their relative contributions to T6SS function. We further determined the importance of two effectors, including an effector-immunity pair, for antibacterial activity. Our genetic analysis led to the identification of an essential membrane-associated structural component named TagX, which we have characterized as a peptidoglycan hydrolase possessing l,d-endopeptidase activity. TagX shows homology to known bacteriophage l,d-endopeptidases and is conserved in the T6SS clusters of several bacterial species. We propose that TagX is the first identified enzyme that fulfills the important role of enabling the transit of T6SS machinery across the peptidoglycan layer of the T6SS-producing bacterium. IMPORTANCE: Acinetobacter baumannii is one of the most troublesome and least investigated multidrug-resistant bacterial pathogens. We have previously shown that A. baumannii employs a T6SS to eliminate competing bacteria. Here we provide a comprehensive analysis of the components of the T6SS of Acinetobacter, and our results provide genetic and functional insights into the Acinetobacter T6SS. Through this analysis, we identified a novel peptidoglycan hydrolase, TagX, that is required for biogenesis of the T6SS apparatus. This is the first peptidoglycanase specialized in T6SS function identified in any species. We propose that this enzyme is required for the spatially and temporally regulated digestion of peptidoglycan to allow assembly of the T6SS machinery.

Wright, R. C., Brockhurst, M. A., and Harrison, E. Ecological conditions determine extinction risk in co-evolving bacteria-phage populations. BMC.Evol.Biol. 10-24-2016; 16(1):227; PM: 27776482

ABSTRACT: ***BACKGROUND***: Antagonistic coevolution between bacteria and their viral parasites, phage, drives continual evolution of resistance and infectivity traits through recurrent cycles of adaptation and counter-adaptation. Both partners are vulnerable to extinction through failure of adaptation. Environmental conditions may impose unequal abiotic selection pressures on each partner, destabilising the coevolutionary relationship and increasing the extinction risk of one partner. In this study we explore how the degree of population mixing and resource supply affect coevolution-induced extinction risk by coevolving replicate populations of Pseudomonas fluorescens SBW25 with its associated lytic phage SBW25capital EF, Cyrillic2 under four treatment regimens incorporating low and high resource availability with mixed or static growth conditions. **RESULTS**: We observed an increased risk of phage extinction under population mixing, and in low resource conditions. High levels of evolved bacterial resistance promoted phage extinction at low resources under both mixed and static conditions, whereas phage populations could survive when phage susceptible bacterial genotypes rose to high frequency. ***CONCLUSIONS***: These findings demonstrate that phage extinction risk is influenced by multiple abiotic conditions, which together act to destabilise the bacteria-phage coevolutionary relationship. The risk of coevolution-induced extinction is therefore dependent on the ecological context.

Wu, W., Leavitt, J. C., Cheng, N., Gilcrease, E. B., Motwani, T., Teschke, C. M., Casjens, S. R., and Steven, A. C. Localization of the Houdinisome (Ejection Proteins) inside the Bacteriophage P22 Virion by Bubblegram Imaging. MBio. 8-9-2016; 7(4); PM: 27507825

ABSTRACT: The P22 capsid is a T=7 icosahedrally symmetric protein shell with a portal protein dodecamer at one 5-fold vertex. Extending outwards from that vertex is a short tail, and putatively extending inwards is a 15-nm-long alpha-helical barrel formed by the C-terminal domains of portal protein subunits. In addition to the densely packed genome, the capsid contains three "ejection proteins" (E-proteins [gp7, gp16, and gp20]) destined to exit from the tightly sealed capsid during the process of DNA delivery into target cells. We estimated their copy numbers by quantitative SDS-PAGE as approximately 12 molecules per virion of gp16 and gp7 and 30 copies of gp20. To localize them, we used bubblegram imaging, an adaptation of cryo-electron microscopy in which gaseous bubbles induced in proteins by prolonged irradiation are used to map the proteins' locations. We applied this technique to wild-type P22, a triple mutant lacking all three E-proteins, and three mutants each lacking one E-protein. We conclude that all three E-proteins are loosely clustered around the portal axis, in the region displaced radially inwards from the portal crown. The bubblegram data imply that approximately half of the alpha-helical barrel seen in the portal crystal structure is disordered in the mature virion, and parts of the disordered region present binding sites for E-proteins. Thus positioned, the E-proteins are strategically placed to pass down the shortened barrel and through the portal ring and the tail, as they exit from the capsid during an infection. *IMPORTANCE*: While it has long been appreciated that capsids serve as delivery vehicles for viral genomes, there is now growing awareness that viruses also deliver proteins into their host cells. P22 has three such proteins (ejection proteins [E-proteins]), whose initial locations in the virion have remained unknown despite their copious amounts (total of 2.5 MDa). This study succeeded in localizing them by the novel technique of bubblegram imaging. The P22 E-proteins are seen to be distributed around

the orifice of the portal barrel. Interestingly, this barrel, 15 nm long in a crystal structure, is only about half as long in situ: the remaining, disordered, portion appears to present binding sites for E-proteins. These observations document a spectacular example of a regulatory order-disorder transition in a supramolecular system and demonstrate the potential of bubblegram imaging to map the components of other viruses as well as cellular complexes.

Xu, R., Shieh, Y. C., and Stewart, D. S. Comparison of RNA extraction kits for the purification and detection of an enteric virus surrogate on green onions via RT-PCR. J.Virol.Methods. 2017; 239:61-68; PM: 27836658

ABSTRACT: Real-time reverse transcriptase-polymerase chain reaction (RT-PCR) offers a rapid and sensitive molecular method for detection of enteric viruses. Unfortunately, these assays are often hampered by the low virus titre found in foods and PCR inhibition due to matrix carryover during RNA extraction. Four commercial RNA extraction kits (Qiagen's QIAamp Viral RNA Mini and UltraSens Virus kits, MoBio UltraClean Tissue & Cells RNA Isolation kit, and Ambion MagMAX Viral RNA Isolation kit) were evaluated for their ability to extract and purify MS2 bacteriophage RNA, an enteric virus surrogate, from inoculated green onions, a food which has been associated with viral gastroenteritis outbreaks. Inoculated green onion wash concentrates and green onion pieces with and without Qiagen QIAshredder homogenization were assayed in the kit comparison. MS2 detection and PCR inhibition were evaluated using a duplex real-time RT-PCR for MS2 and an exogenous internal amplification control (IAC) assay. Without homogenization, MS2 inoculated at 40pfu/g was detected in at least 4 lots of green onion wash concentrates using the silica-membrane spin-column kits. Inhibition was a factor for the magnetic silica-based MagMAX kit, which resulted in detection of MS2 in 1 of 5. Addition of QIAshredder homogenization prior to extraction did not adversely affect the silica-membrane kit results but improved the MS2 detection by MagMAX to 5 of 5 lots. Use of a 1:10 dilution of primary RNA extracts also improved detection. The QIAamp Viral RNA Mini and MagMAX kits were further compared for detection of MS2 from green onion pieces inoculated at 20 and 5pfu/g. Using homogenization, the MagMAX kit detected 20pfu/g in only 1 of 2 green onion lots, whereas the QIAamp Viral RNA kit detected 2 of 2 lots at 5 pfu/g without homogenization.

Xu, Y., Buss, E. A., and Boucias, D. G. Impacts of Antibiotic and Bacteriophage Treatments on the Gut-Symbiont-Associated Blissus insularis (Hemiptera: Blissidae). Insects. 11-3-2016; 7(4); PM: 27827869

ABSTRACT: The Southern chinch bug, Blissus insularis, possesses specialized midgut crypts that harbor dense populations of the exocellular symbiont Burkholderia. Oral administration of antibiotics suppressed the gut symbionts in B. insularis and negatively impacted insect host fitness, as reflected by retarded development, smaller body size, and higher susceptibility to an insecticide, bifenthrin. Considering that the antibiotics probably had non-lethal but toxic effects on host fitness, attempts were conducted to reduce gut symbionts using bacteriophage treatment. Soil-lytic phages active against the cultures of specific Burkholderia ribotypes were successfully isolated using a soil enrichment protocol. Characterization of the BiBurk16MC_R phage determined its specificity to the Bi16MC_R_vitro ribotype and placed it within the family Podoviridae. Oral administration of phages to fifth-instar B. insularis, inoculated with Bi16MC_R_vitro as neonates had no deleterious effects on host fitness. However, the ingested phages failed

to impact the crypt-associated Burkholderia. The observed inactivity of the phage was likely due to the blockage of the connection between the anterior and posterior midgut regions. These findings suggest that the initial colonization by Burkholderia programs the ontogeny of the midgut, providing a sheltered residence protected from microbial antagonists.

Yamada, T., Wajima, T., Nakaminami, H., Kobayashi, K., Ikoshi, H., and Noguchi, N. The modified Gingyo-san, a Chinese herbal medicine, has direct antibacterial effects against respiratory pathogens. BMC.Complement Altern.Med. 11-14-2016; 16(1):463; PM: 27842538

ABSTRACT: ***BACKGROUND***: Modified Gingyo-san (MGS) is empirically used to treat various respiratory infections. MGS has been reported to have antiinflammatory and antiviral activities; however, it is not known if it has an antibacterial activity. Therefore, in this study, we aimed to investigate the antimicrobial activity of MGS against respiratory pathogens. ***METHODS***: MGS, which is sold as an over-the-counter drug in Japan, was used for the study. Antimicrobial activity was evaluated using the disk diffusion method. Growth inhibitory activity was evaluated by measuring colony-forming units of the pathogens in the presence of MGS. ***RESULTS***: MGS inhibited the growth of Bacillus subtilis, Streptococcus pneumoniae, and Streptococcus pyogenes, which are gram-positive bacteria. Although the growth of most gram-negative bacteria was not inhibited by MGS, interestingly, the growth of Haemophilus influenzae was inhibited. MGS did not show any activity against Candida albicans or bacteriophage phiX174. ***CONCLUSIONS***: In addition to the antiinflammatory and antiviral activities of MGS, which have already been reported, the data obtained from this study indicates that MGS has an antibacterial activity.

Yang, C., Li, Y., Zhou, Y., Lei, X., Zheng, W., Tian, Y., Van Nostrand, J. D., He, Z., Wu, L., Zhou, J., and Zheng, T. A comprehensive insight into functional profiles of free-living microbial community responses to a toxic Akashiwo sanguinea bloom. Sci.Rep. 10-5-2016; 6:34645; PM: 27703234

ABSTRACT: Phytoplankton blooms are a worldwide problem and can greatly affect ecological processes in aquatic systems, but its impacts on the functional potential of microbial communities are limited. In this study, a high-throughput microarray-based technology (GeoChip) was used to profile the functional potential of free-living microbes from the Xiamen Sea Area in response to a 2011 Akashiwo sanguinea bloom. The bloom altered the overall community functional structure. Genes that were significantly ($p < 0.05$) increased during the bloom included carbon degradation genes and genes involved in nitrogen (N) and/or phosphorus (P) limitation stress. Such significantly changed genes were well explained by chosen environmental factors (COD, nitrite-N, nitrate-N, dissolved inorganic phosphorus, chlorophyll-a and algal density). Overall results suggested that this bloom might enhance the microbial converting of nitrate to N2 and ammonia nitrogen, decrease P removal from seawater, activate the glyoxylate cycle, and reduce infection activity of bacteriophage. This study presents new information on the relationship of algae to other microbes in aquatic systems, and provides new insights into our understanding of ecological impacts of phytoplankton blooms.

Yeh, T. Y. Complete nucleotide sequence of a new filamentous phage, Xf109, which integrates its genome into the chromosomal DNA of Xanthomonas oryzae. Arch.Virol. 10-14-2016; PM: 27743252

ABSTRACT: Unlike Ff-like coliphages, certain filamentous Inoviridae phages integrate their genomes into the host chromosome and enter a prophage state in their infectious cycle. This lysogenic life cycle was first reported for Xanthomonas citri Cf phage. However, except for the X. citri phages Cf and XacF1, complete genome sequence information about lysogenic Xanthomonas phages is not available to date. A proviral sequence of Xf109 phage was identified in the genome of Xanthomonas oryzae, the rice bacterial blight pathogen, and revived as infectious virions to lysogenize its host de novo. The genome of Xf109 phage is 7190 nucleotides in size and contains 12 predicted open reading frames in an organization similar to that of the Cf phage genome. Seven of the Xf109 proteins show significant sequence similarity to Cf and XacF1 phage proteins, while its ORF4 shares 92 % identity with the major coat protein of X. phage oryzae Xf. Integration of Xf109 phage DNA into the host genome is site-specific, and the attP/attB sequence contains the dif core sequence 5'-TATACATTATGCGAA-3', which is identical to that of Cf, XacF1, and Xanthomonas campestris phage varphiLf. To my knowledge, this is the first complete genome sequence of a filamentous bacteriophage that infects X. oryzae.

Yosef, I., Edgar, R., Levy, A., Amitai, G., Sorek, R., Munitz, A., and Qimron, U. Natural selection underlies apparent stress-induced mutagenesis in a bacteriophage infection model. Nat.Microbiol. 4-18-2016; 1(6):16047; PM: 27572836

ABSTRACT: The emergence of mutations following growth-limiting conditions underlies bacterial drug resistance, viral escape from the immune system and fundamental evolution-driven events. Intriguingly, whether mutations are induced by growth limitation conditions or are randomly generated during growth and then selected by growth limitation conditions remains an open question(1). Here, we show that bacteriophage T7 undergoes apparent stress-induced mutagenesis when selected for improved recognition of its host's receptor. In our unique experimental set-up, the growth limitation condition is physically and temporally separated from mutagenesis: growth limitation occurs while phage DNA is outside the host, and spontaneous mutations occur during phage DNA replication inside the host. We show that the selected beneficial mutations are not pre-existing and that the initial slow phage growth is enabled by the phage particle's low-efficiency DNA injection into the host. Thus, the phage particle allows phage populations to initially extend their host range without mutagenesis by virtue of residual recognition of the host receptor. Mutations appear during non-selective intracellular replication, and the frequency of mutant phages increases by natural selection acting on free phages, which are not capable of mutagenesis.

Yuan, W., Zhang, Y., Wang, G., Bai, J., Wang, X., Li, Y., and Jiang, P. Genomic and proteomic characterization of SE-I, a temperate bacteriophage infecting Erysipelothrix rhusiopathiae. Arch.Virol. 2016; 161(11):3137-3150; PM: 27541818

ABSTRACT: A bacteriophage infecting pathogenic Erysipelothrix rhusiopathiae was isolated from a swine farm experiencing an outbreak of acute swine erysipelas; we designated this phage SE-I. SE-I has an icosahedral head, a long tail and a double-stranded DNA genome. The 34,997-bp genome has a GC content of 34 % and contains 43 open reading frames (ORFs) encoding packaging, structural, lysin-holin, and

hypothetical proteins. Components of purified SE-I were separated using SDS-PAGE and analyzed using liquid chromatography-mass spectrometry. Nine proteins were identified, encoded by ORF9, ORF15, ORF23, ORF30, ORF31, ORF33, ORF39, ORF40 and ORF 42. A phylogenetic tree constructed based on the sequence of the large terminase subunit revealed that SE-I is closely related to Staphylococcus phages P954 and phi3396. The CHAP-domain-containing protein encoded by ORF25 was expressed in E. coli and which was able to inactivate host bacteria. SE-I was able to infect 7 of 13 E. rhusiopathiae strains, but was unable to infect Salmonella, Streptococcus suis, and Staphylococcus aureus. This is the first report of the isolation, characterization, and genomic and proteomic analysis of a temperate phage infecting E. rhusiopathiae, and it might lead to the development of new anti- E. rhusiopathiae agents.

Zaczek, M., Lusiak-Szelachowska, M., Jonczyk-Matysiak, E., Weber-Dabrowska, B., Miedzybrodzki, R., Owczarek, B., Kopciuch, A., Fortuna, W., Rogoz, P., and Gorski, A. Antibody Production in Response to Staphylococcal MS-1 Phage Cocktail in Patients Undergoing Phage Therapy. Front Microbiol. 2016; 7:1681; PM: 27822205

ABSTRACT: In this study, we investigated the humoral immune response (through the release of IgG, IgA, and IgM antiphage antibodies) to a staphylococcal phage cocktail in patients undergoing experimental phage therapy at the Phage Therapy Unit, Medical Center of the Ludwik Hirszfeld Institute of Immunology and Experimental Therapy in Wroclaw, Poland. We also evaluated whether occurring antiphage antibodies had neutralizing properties toward applied phages (K rate). Among 20 examined patients receiving the MS-1 phage cocktail orally and/or locally, the majority did not show a noticeably higher level of antiphage antibodies in their sera during phage administration. Even in those individual cases with an increased immune response, mostly by induction of IgG and IgM, the presence of antiphage antibodies did not translate into unsatisfactory clinical results of phage therapy. On the other hand, a negative outcome of the treatment occurred in some patients who showed relatively weak production of antiphage antibodies before and during treatment. This may imply that possible induction of antiphage antibodies is not an obstacle to the implementation of phage therapy and support our assumption that the outcome of the phage treatment does not primarily depend on the appearance of antiphage antibodies in sera of patients during therapy. These conclusions are in line with our previous findings. The confirmation of this thesis is of great interest as regards the efficacy of phage therapy in humans.

Zelasko, S., Gorski, A., and Dabrowska, K. Delivering phage therapy per os: benefits and barriers. Expert.Rev.Anti.Infect.Ther. 11-25-2016; PM: 27885865

ABSTRACT: ***INTRODUCTION***: Multidrug-resistant bacterial infections of the gastrointestinal tract pose a serious public health concern. High levels of antibiotic drug resistance, along with the potential for antibiotics to precipitate disease or alter the gut microbiome has prompted research into alternative treatment methods. Evidence suggests that bacteriophage therapy delivered per os may be well-suited to target such infections. ***AREAS COVERED***: Herein, we discuss the specific advantages and challenges of using orally administered phage therapy. Our literature review encompasses recent works using phages to target various clinically-relevant bacteria in vivo. We also provide insights into methods that aim to overcome the barriers to effective phage transit through the harsh gastrointestinal environment. ***EXPERT COMMENTARY***: Evidence from a number of in vivo animal studies suggests that targeting bacterial infections using phages

delivered orally holds potential. Efficacious oral phage therapy depends on the delivery of sufficient phage titers to the infection site, which may be hindered by the host's gastrointestinal tract and immune response.

Zhang, C., Ma, Y., Wang, T., Sun, H., Lu, G., and Ren, H. Characterization and complete genome sequence of vB_EcoP-Bp4, a novel polyvalent N4-like bacteriophage that infects chicken pathogenic Escherichia coli. Virol.Sin. 2016; 31(4):353-356; PM: 27535068

No Abstract Available.

Zhang, C., Zhang, Z., Li, J., Qin, K., Wei, Y., Zhang, Q., Lin, L., and Ji, X. Complete genome sequence of the lytic cold-active Pseudomonas fluorescens bacteriophage VSW-3 from Napahai plateau wetland. Virus Genes. 10-31-2016; PM: 27796639

ABSTRACT: The lytic cold-active bacteriophage VSW-3, belonging to the Podoviridae family and infecting the host Pseudomonas fluorescens SW-3, was isolated from the Napahai plateau wetland in China. With the development of sequencing technology, the study of Pseudomonas genomic diversity has increased; however, knowledge of cold-active phages infecting Pseudomonas is limited. The newly sequenced phage VSW-3 was classified based on virion morphology by transmission electron microscope. Sequence analysis revealed that the genome size was 40,556 bp with an overall GC content of 57.54 % and 46 open reading frames. The genome was organized into several modules containing genes for packaging, structural proteins, replication/transcription, and phage lysis. The sequence contained 45 potential promoters, 3 transcription terminators, and yet no tRNAs. This is the first report of cold-active Pseudomonas fluorescens bacteriophage genome sequencing.

Zhang, D., Coronel-Aguilera, C. P., Romero, P. L., Perry, L., Minocha, U., Rosenfield, C., Gehring, A. G., Paoli, G. C., Bhunia, A. K., and Applegate, B. The Use of a Novel NanoLuc -Based Reporter Phage for the Detection of Escherichia coli O157:H7. Sci.Rep. 9-14-2016; 6:33235; PM: 27624517

ABSTRACT: Rapid detection of the foodborne pathogen Escherichia coli O157:H7 is of vital importance for public health worldwide. Among detection methods, reporter phages represent unique and sensitive tools for the detection of E. coli O157:H7 from food as they are host-specific and able to differentiate live cells from dead ones. Upon infection, target bacteria become identifiable since reporter genes are expressed from the engineered phage genome. The E. coli O157:H7 bacteriophage PhiV10 was modified to express NanoLuc luciferase (Nluc) derived from the deep-sea shrimp Oplophorus gracilirostris. Once infected by the PhiV10 reporter phage, E. coli O157:H7 produces a strong bioluminescent signal upon addition of commercial luciferin (Nano-Glo((R))). Enrichment assays using E. coli O157:H7 grown in LB broth with a reporter phage concentration of 1.76 x 10(2) pfu ml(-1) are capable of detecting approximately 5 CFU in 7 hours. Comparable detection was achieved within 9 hours using 9.23 x 10(3) pfu ml(-1) of phage in selective culture enrichments of ground beef as a representative food matrix. Therefore we conclude that this NanoLuc reporter phage assay shows promise for detection of E. coli O157:H7 from food in a simple, fast and sensitive manner.

Zhang, L. and Tripathi, A. Archaeal RNA ligase from thermoccocus kodakarensis for template dependent ligation. RNA.Biol. 10-7-2016; 1-9; PM: 27715457

ABSTRACT: Nicking-sealing RNA ligases play a significant biological role in host defense and cellular repair, and have become an important molecular tool in biomedical engineering. Due to the propensity for RNA to form secondary structures, RNA modifying enzymes with elevated optimum temperatures are highly desired. Current characterized double stranded RNA ligases, such as the bacteriophage T4 RNA ligase 2, while possessing good template dependency, are not active at elevated temperatures. The few characterized RNA ligases from thermophiles exhibit high template independency. We synthesize and characterize here, KOD RNA ligase (KOD1RnI), a thermostable and template dependent RNA ligase from the archaeon, Thermoccocus Kodakarensis. We disclose that a 13 time reduction in template independent ligation can be achieved with the addition of a single stranded DNase, such as RecJ. We also elucidate the effects of the presence of blood proteins on the activity of KOD1RnI. Template dependent and thermostable RNA ligases, such as KOD RNA ligase, can be utilized in RNA detection, modification and sequencing.

Zhang, L., Sun, L., Wei, R., Gao, Q., He, T., Xu, C., Liu, X., and Wang, R. Study of intracellular Staphylococcus aureus control by virulent bacteriophage within MAC-T bovine mammary epithelial cells. Antimicrob.Agents Chemother. 12-5-2016; PM: 27919889

ABSTRACT: Bacteriophages (phages) are known to effectively kill extracellular multiplying bacteria. The present study demonstrated that phages penetrated bovine mammary epithelial cells and cleared intracellular Staphylococcus aureus, in a time-dependent manner. In particular, phage vB_SauM_JS25 reached the nucleus within the 3 h post-incubation. The phages had an endocytotic efficiency of 12%. This ability to kill intracellular host bacteria suggests the utility of phage-based therapies, and may protect from recurrent infection and treatment failure.

Zhao, X., Chen, C., Jiang, X., Shen, W., Huang, G., Le, S., Lu, S., Zou, L., Ni, Q., Li, M., Zhao, Y., Wang, J., Rao, X., Hu, F., and Tan, Y. Transcriptomic and Metabolomic Analysis Revealed Multifaceted Effects of Phage Protein Gp70.1 on Pseudomonas aeruginosa. Front Microbiol. 2016; 7:1519; PM: 27725812

ABSTRACT: The impact of phage infection on the host cell is severe. In order to take over the cellular machinery, some phage proteins were produced to shut off the host biosynthesis early in the phage infection. The discovery and identification of these phage-derived inhibitors have a significant prospect of application in antibacterial treatment. This work presented a phage protein, gp70.1, with non-specific inhibitory effects on Pseudomonas aeruginosa and Escherichia coli. Gp70.1 was encoded by early gene - orf 70.1 from P. aeruginosa phage PaP3. The P. aeruginosa with a plasmid encoding gp70.1 showed with delayed growth and had the appearance of a small colony. The combination of multifaceted analysis including microarray-based transcriptomic analysis, RT-qPCR, nuclear magnetic resonance (NMR) spectroscopy-based metabolomics and phenotype experiments were performed to investigate the effects of gp70.1 on P. aeruginosa. A total of 178 genes of P. aeruginosa mainly involved in extracellular function and metabolism were differentially expressed in the presence of gp70.1 at three examined time points. Furthermore, our results indicated that gp70.1 had an extensive impact on the extracellular phenotype of P. aeruginosa, such as motility,

pyocyanin, extracellular protease, polysaccharide, and cellulase. For the metabolism of P. aeruginosa, the main effect of gp70.1 was the reduction of amino acid consumption. Finally, the RNA polymerase sigma factor RpoS was identified as a potential cellular target of gp70.1. Gp70.1 was the first bacterial inhibitor identified from Pseudomonas aeruginosa phage PaP3. It was also the first phage protein that interacted with the global regulator RpoS of bacteria. Our results indicated the potential value of gp70.1 in antibacterial applications. This study preliminarily revealed the biological function of gp70.1 and provided a reference for the study of other phage genes sharing similarities with orf70.1.

Zhao, X. and Skurnik, M. Bacteriophages of Yersinia pestis. Adv.Exp.Med.Biol. 2016; 918:361-37; PM: 27722870

ABSTRACT: Bacteriophage play many varied roles in microbial ecology and evolution. This chapter collates a vast body of knowledge and expertise on Yersinia pestis phages, including the history of their isolation and classical methods for their isolation and identification. The genomic diversity of Y. pestis phage and bacteriophage islands in the Y. pestis genome are also discussed because all phage research represents a branch of genetics. In addition, our knowledge of the receptors that are recognized by Y. pestis phage, advances in phage therapy for Y. pestis infections, the application of phage in the detection of Y. pestis, and clustered regularly interspaced short palindromic repeats (CRISPRs) sequences of Y. pestis from prophage DNA are all reviewed here.

Zhao, Z., De-Donatis, G. M., Schwartz, C., Fang, H., Li, J., and Guo, P. An Arginine Finger Regulates the Sequential Action of Asymmetrical Hexameric ATPase in the Double-Stranded DNA Translocation Motor. Mol.Cell Biol. 10-1-2016; 36(19):2514-2523; PM: 27457616

ABSTRACT: Biological motors are ubiquitous in living systems. Currently, how the motor components coordinate the unidirectional motion is elusive in most cases. Here, we report that the sequential action of the ATPase ring in the DNA packaging motor of bacteriophage varphi29 is regulated by an arginine finger that extends from one ATPase subunit to the adjacent unit to promote noncovalent dimer formation. Mutation of the arginine finger resulted in the interruption of ATPase oligomerization, ATP binding/hydrolysis, and DNA translocation. Dimer formation reappeared when arginine mutants were mixed with other ATPase subunits that can offer the arginine to promote their interaction. Ultracentrifugation and virion assembly assays indicated that the ATPase was presenting as monomers and dimer mixtures. The isolated dimer alone was inactive in DNA translocation, but the addition of monomer could restore the activity, suggesting that the hexameric ATPase ring contained both dimer and monomers. Moreover, ATP binding or hydrolysis resulted in conformation and entropy changes of the ATPase with high or low DNA affinity. Taking these observations together, we concluded that the arginine finger regulates sequential action of the motor ATPase subunit by promoting the formation of the dimer inside the hexamer. The finding of asymmetrical hexameric organization is supported by structural evidence of many other ATPase systems showing the presence of one noncovalent dimer and four monomer subunits. All of these provide clues for why the asymmetrical hexameric ATPase gp16 of varphi29 was previously reported as a pentameric configuration by cryo-electron microscopy (cryo-EM) since the contact by the arginine finger renders two adjacent ATPase subunits closer than other subunits. Thus, the asymmetrical hexamer would appear as a pentamer by cryo-EM, a technology that acquires the average of many images.

Zhou, Z., Ji, Z., Wang, S., Haque, F., and Guo, P. Oriented single directional insertion of nanochannel of bacteriophage SPP1 DNA packaging motor into lipid bilayer via polar hydrophobicity. Biomaterials. 2016; 105:222-227; PM: 27529454

ABSTRACT: Insertion of biological nanopore into artificial membrane is of fundamental importance in nanotechnology. Many applications require control and knowledge of channel orientation. In this work, the insertion orientation of the bacteriophage SPP1 and phi29 DNA packaging motors into lipid membranes was investigated. Single molecule electrophysiological assays and Ni-NTA-nanogold binding assays revealed that both SPP1 and phi29 motor channels exhibited a one-way traffic property for TAT peptide translocation from N- to C-termini of the protein channels. SPP1 motor channels preferentially inserts into liposomes with their C-terminal wider region facing inward. Changing the hydrophobicity of the N- or C-termini of phi29 connector alters the insertion orientation, suggesting that the hydrophobicity and hydrophilicity of the termini of the protein channel governs the orientation of the insertion into lipid membrane. It is proposed that the specificity in motor channel orientation is a result of the hydrophilic/hydrophobic interaction at the air/water interface when the protein channels are incorporating into liposome membranes.



Ian Norman, Phage Camp, September 7 2011, Creative Commons License

I. Probiotics & Bacteriotherapy

Cost-Effectiveness Analysis of the Use of Probiotics for the Prevention of Clostridium difficile-Associated Diarrhea in a Provincial Healthcare System - ERRATUM. Infect.Control Hosp.Epidemiol. 2016; 37(9):1133; PM: 27580173

No Abstract Available.

Fecal Microbiota Therapy for Clostridium difficile Infection: A Health Technology Assessment. Ont.Health Technol.Assess.Ser. 2016; 16(17):1-69; PM: 27516814

ABSTRACT: ***BACKGROUND***: Fecal microbiota therapy is increasingly being used to treat patients with Clostridium difficile infection. This health technology assessment primarily evaluated the effectiveness and cost-effectiveness of fecal microbiota therapy compared with the usual treatment (antibiotic therapy). ***METHODS***: We performed a literature search using Ovid MEDLINE, Embase, Cochrane Database of Systematic Reviews, Database of Abstracts of Reviews of Effects, CRD Health Technology Assessment Database, Cochrane Central Register of Controlled Trials, and NHS Economic Evaluation Database. For the economic review, we applied economic filters to these search results. We also searched the websites of agencies for other health technology assessments. We conducted a meta-analysis to analyze effectiveness. The quality of the body of evidence for each outcome was examined according to the Grading of Recommendations Assessment, Development, and Evaluation (GRADE) Working Group criteria. Using a step-wise, structural methodology, we determined the overall quality to be high, moderate, low, or very low. We used a survey to examine physicians' perception of patients' lived experience, and a modified grounded theory method to analyze information from the survey. ***RESULTS***: For the review of clinical effectiveness, 16 of 1,173 citations met the inclusion criteria. A meta-analysis of two randomized controlled trials found that fecal microbiota therapy significantly improved diarrhea associated with recurrent C. difficile infection versus treatment with vancomycin (relative risk 3.24, 95% confidence interval [CI] 1.85-5.68) (GRADE: moderate). While fecal microbiota therapy is not associated with a significant decrease in mortality compared with antibiotic therapy (relative risk 0.69, 95% CI 0.14-3.39) (GRADE: low), it is associated with a significant increase in adverse events (e.g., short-term diarrhea, relative risk 30.76, 95% CI 4.46-212.44; abdominal cramping, relative risk 14.81, 95% CI 2.07-105.97) (GRADE: low). For the value-for-money component, two of 151 economic evaluations met the inclusion criteria. One reported that fecal microbiota therapy was dominant (more effective and less expensive) compared with vancomycin; the other reported an incremental cost-effectiveness ratio of $17,016 USD per quality-adjusted life-year for fecal microbiota therapy compared with vancomycin. This ratio for the second study indicated that there would be additional cost associated with each recurrent C. difficile infection resolved. In Ontario, if fecal microbiota therapy were adopted to treat recurrent C. difficile infection, considering it from the perspective of the Ministry of Health and Long-Term Care as the payer, an estimated $1.5 million would be saved after the first year of adoption and $2.9 million after 3 years. The contradiction between the second economic evaluation and the savings we estimated may be a result of the lower cost of fecal microbiota therapy and hospitalization in Ontario compared with the cost of therapy used in the US model. Physicians reported that C. difficile infection significantly reduced patients' quality of life. Physicians saw fecal microbiota therapy as improving patients' quality of life because

patients could resume daily activities. Physicians reported that their patients were happy with the procedures required to receive fecal microbiota therapy. ***CONCLUSIONS***: In patients with recurrent C. difficile infection, fecal microbiota therapy improves outcomes that are important to patients and provides good value for money.

Abdurasulova, I. N., Matsulevich, A. V., Tarasova, E. A., Kudryavtsev, I. V., Serebrjakova, M. K., Ermolenko, E. I., Bisaga, G. N., Klimenko, V. M., and Suvorov, A. N. Enterococcus faecium strain L-3 and glatiramer acetate ameliorate experimental allergic encephalomyelitis in rats by affecting different populations of immune cells. Benef.Microbes. 11-30-2016; 7(5):719-729; PM: 27633171

ABSTRACT: The effect of probiotic Enterococcus faecium strain L-3 was studied in rats with experimental allergic encephalomyelitis (EAE). Glatiramer acetate (GA) was used as control drug. E. faecium strain L-3 and GA both were able to reduce the severity of EAE in a similar fashion. Both approaches increased the proportion of EAE resistant rats and rats with mild disease, prolonged the inductive phase of EAE and reduced the disease duration. Study of the phenotypes of immune cells in blood revealed the differences in immunoregulatory pathways that mediate the protective action of probiotic or GA treatment of EAE. The presence of pronounced protective and immunomodulating effects of the probiotic E. faecium strain L-3 opens an opportunity of its application for the treatment of multiple sclerosis.

Aggarwal, A., Sarangi, A. N., Gaur, P., Shukla, A., and Aggarwal, R. Gut microbiome in children with enthesitis-related arthritis in a developing country, and the effect of probiotic administration. Clin.Exp.Immunol. 11-9-2016; PM: 27861762

ABSTRACT: In Asia, enthesitis-related arthritis (ERA) is the most-frequent category of juvenile idiopathic arthritis. ERA has a strong association with HLA-B27 and subclinical gut inflammation. In a HLA-B27 transgenic rat model, presence of Bacteroides bacteria in the gut appears to cause spondyloarthropathy (SpA). Thus, we studied gut microbiota in children with ERA. Stool specimens from 33 patients with ERA and 14 age-matched healthy controls were studied; none had any gastrointestinal symptom, or had received a drug known to affect gut motility or microbiota in the preceding 6 weeks. From each specimen, a cDNA library for V3 region of bacterial 16S rRNA was subjected to high-throughput, massively-parallel sequencing. Relationship of specimens was studied using principal co-ordinate analysis (PCoA), and abundances of various bacterial taxa and alpha diversity were compared between groups. In 8 patients, a repeat fecal specimen was studied after 12 weeks of probiotic therapy. The 55 specimens yielded a median (range) of 397315 (102.

Ahmad, S. and Bromberg, J. S. Current status of the microbiome in renal transplantation. Curr.Opin.Nephrol.Hypertens. 2016; 25(6):570-576; PM: 27517135

ABSTRACT: ***PURPOSE OF REVIEW***: An imbalance between pathogenic and protective microbiota characterizes dysbiosis. Presence of dysbiosis may affect immunity, tolerance, or disease depending on a variety of conditions. In the transplant patient population, the need for immunosuppression and widespread use of prophylactic and therapeutic antimicrobial agents create new posttransplant microbiota communities that remain to be fully defined. ***RECENT FINDINGS***: Studies in mice have demonstrated significant bidirectional interactions between microbiota-derived products and host immune cells.

The stimulation of regulatory T cell and T helper cell type 17 cells by specific products leads to maintenance of immune homeostasis versus activation of inflammation, respectively. Dysbiosis may lead to development of antigen cross-reactivity, which may affect alloreactivity. Certain immunologic sequelae of microbiota are pronounced in chronic kidney disease, because of uremia and renal metabolism of microbiota metabolites. Dietary modifications, probiotics, and fecal microbiota transplant have been investigated for alteration of microbiota in humans. ***SUMMARY***: Researchers have begun to identify dysbioses associated with clinical conditions, including chronic kidney disease, posttransplant infection, and rejection. This information will allow clinicians not only to select at-risk patients for early intervention, but also to develop therapies that restore the microbiota to a state of homeostasis or tolerance.

Ahmed, I., Roy, B. C., Khan, S. A., Septer, S., and Umar, S. Microbiome, Metabolome and Inflammatory Bowel Disease. Microorganisms. 6-15-2016; 4(2); PM: 27681914

ABSTRACT: Inflammatory Bowel Disease (IBD) is a multifactorial disorder that conceptually occurs as a result of altered immune responses to commensal and/or pathogenic gut microbes in individuals most susceptible to the disease. During Crohn's Disease (CD) or Ulcerative Colitis (UC), two components of the human IBD, distinct stages define the disease onset, severity, progression and remission. Epigenetic, environmental (microbiome, metabolome) and nutritional factors are important in IBD pathogenesis. While the dysbiotic microbiota has been proposed to play a role in disease pathogenesis, the data on IBD and diet are still less convincing. Nonetheless, studies are ongoing to examine the effect of pre/probiotics and/or FODMAP reduced diets on both the gut microbiome and its metabolome in an effort to define the healthy diet in patients with IBD. Knowledge of a unique metabolomic fingerprint in IBD could be useful for diagnosis, treatment and detection of disease pathogenesis.

Akbari, E., Asemi, Z., Daneshvar, Kakhaki R., Bahmani, F., Kouchaki, E., Tamtaji, O. R., Hamidi, G. A., and Salami, M. Effect of Probiotic Supplementation on Cognitive Function and Metabolic Status in Alzheimer's Disease: A Randomized, Double-Blind and Controlled Trial. Front Aging Neurosci. 2016; 8:256; PM: 27891089

ABSTRACT: Alzheimer's disease (AD) is associated with severe cognitive impairments as well as some metabolic defects. Scant studies in animal models indicate a link between probiotics and cognitive function. This randomized, double-blind, and controlled clinical trial was conducted among 60 AD patients to assess the effects of probiotic supplementation on cognitive function and metabolic status. The patients were randomly divided into two groups (n = 30 in each group) treating with either milk (control group) or a mixture of probiotics (probiotic group). The probiotic supplemented group took 200 ml/day probiotic milk containing Lactobacillus acidophilus, Lactobacillus casei, Bifidobacterium bifidum, and Lactobacillus fermentum (2 x 109 CFU/g for each) for 12 weeks. Mini-mental state examination (MMSE) score was recorded in all subjects before and after the treatment. Pre- and post-treatment fasting blood samples were obtained to determine the related markers. After 12 weeks intervention, compared with the control group (-5.03% +/- 3.00), the probiotic treated (+27.90% +/- 8.07) patients showed a significant improvement in the MMSE score (P <0.001). In addition, changes in plasma malondialdehyde (-22.01% +/- 4.84 vs. +2.67% +/- 3.86 mumol/L, P <0.001), serum high-sensitivity C-reactive protein (-17.61% +/- 3.70 vs. +45.26% +/- 3.50 mug/mL, P <0.001), homeostasis model of assessment-estimated insulin resistance (+28.84% +/- 13.34 vs.

+76.95% +/- 24.60, P = 0.002), Beta cell function (+3.45% +/- 10.91 vs. +75.62% +/- 23.18, P = 0.001), serum triglycerides (-20.29% +/- 4.49 vs. -0.16% +/- 5.24 mg/dL, P = 0.003), and quantitative insulin sensitivity check index (-1.83 +/- 1.26 vs. -4.66 +/- 1.70, P = 0.006) in the probiotic group were significantly varied compared to the control group. We found that the probiotic treatment had no considerable effect on other biomarkers of oxidative stress and inflammation, fasting plasma glucose, and other lipid profiles. Overall, the current study demonstrated that probiotic consumption for 12 weeks positively affects cognitive function and some metabolic statuses in the AD patients. *CLINICAL TRIAL REGISTRATION*: http://www.irct.ir/, IRCT201511305623N60.

Akbari, V. and Hendijani, F. Effects of probiotic supplementation in patients with type 2 diabetes: systematic review and meta-analysis. Nutr.Rev. 2016; 74(12):774-784; PM: 27864537

ABSTRACT: ***CONTEXT***: The rising prevalence of type 2 diabetes requires increased efforts to find effective therapeutic agents for this complex condition. Following the recent observation that the gut microbiota is altered in diabetic patients, researchers investigated the effect of probiotics in patients with diabetes. ***OBJECTIVE***: The aim of this systematic review was to assess the effects of probiotic consumption on glycemic control in diabetic patients. ***DATA SOURCES***: PubMed, Scopus, Web of Science (formerly ISI Web of Knowledge), Cochrane Central Register of Controlled Trials, ClinicalTrials.gov, and ProQuest Dissertations and Theses databases were searched up to November 2015. ***STUDY SELECTION***: Clinical trials in diabetic patients in whom probiotics were administered as an intervention were included. ***DATA EXTRACTION***: Primary outcomes were fasting blood glucose, insulin concentration, insulin resistance, and hemoglobin A1c. Secondary outcomes were adverse events. ***DATA SYNTHESIS***: Of the 2736 reports that were screened, 13 clinical trials met the inclusion criteria. Pooling data from eligible clinical trials revealed that probiotic supplementation significantly ($P < 0.05$) decreased fasting blood glucose and hemoglobin A1c in diabetic patients, although the participants' characteristics (eg, body mass index) and the number and type of probiotic microorganisms affected the clinical response. **CONCLUSIONS**: Administration of probiotics appears to have a beneficial role in the management of type 2 diabetes; however, more clinical studies with adequate sample sizes and sound methodology are required to inform the development of evidence-based treatment guidelines.

Al-Rubaye, A. A., Ekesi, N. S., Zaki, S., Emami, N. K., Wideman, R. F., Jr., and Rhoads, D. D. Chondronecrosis with osteomyelitis in broilers: Further defining a bacterial challenge model using the wire flooring model. Poult.Sci. 9-1-2016; PM: 27587724

ABSTRACT: Lameness in broiler chickens is a significant animal welfare and financial issue. Bacterial chondronecrosis with osteomyelitis (BCO) leading to lameness can be enhanced by rearing young broilers on wire flooring. Using the wire floor system, we identified Staphylococcus agnetis as the predominant isolate in BCO of the proximal tibiae and femora, and blood of lame broilers. Administration of S. agnetis isolates in water can induce lameness. We now report that the wire floor system increases bacterial translocation into the blood stream. We have also determined that approximately 105 CFU/mL is the minimum effective dose in the drinking water and that challenge at 10, 20, or 30 days of age produces similar incidences of lameness. BCO isolates of S. agnetis are much more effective than other Staphylococcus species and can overwhelm the protective effects of some commercial probiotics. Finally, we also demonstrated that the

BCO lameness induced by administration of S. agnetis in the drinking water is transmissible to unchallenged broilers in the same pen.

Alagawany, M., bd El-Hack, M. E., Arif, M., and Ashour, E. A. Individual and combined effects of crude protein, methionine, and probiotic levels on laying hen productive performance and nitrogen pollution in the manure. Environ.Sci.Pollut.Res.Int. 2016; 23(22):22906-22913; PM: 27572695

ABSTRACT: The present study aimed to investigate the effect of the dietary levels of protein, methionine (Met), as well as probiotic on productive performance, feed utilization, and environmental pollution by N in Lohmann Brown laying hens. A total number of 160 Lohmann Brown laying hens at 20 weeks of age were randomly divided into 8 treatment groups using a 2 x 2 x 2 factorial design experiment. The experiment involved two levels of crude protein (16 and 18 %), two levels of Met (0.45 and 0.50 %), and two concentrations of probiotic (0 or 1 g/kg diet, with a concentration of 1010 CFU/g of Lactobacillus acidophilus) within 20-42 weeks of age. Results revealed that egg production parameters were significantly ($P < 0.01$) improved for hens fed diets of 18 % CP comparing with that of 16 % protein within the period from 26 to 30 weeks of age. Protein utilization and feed efficiency values were enhanced with 0.67 and 0.72 % Met during the period of 26-30 weeks of the age. For the N pollution, results showed that increasing crude protein in the diet from 16 to 18 % caused significant ($P < 0.01$) increase in the excreted N from 0.349 to 0.492 g/d. The methionine level of 0.72 % recorded the highest values of total consumed N being 3.98 g/d and excreted N being 0.527 g/d comparing with the other levels. It could be concluded that the best productive performance could be given by using 0.72 % total sulfur amino acids (TSAA). Furthermore, the dietary level of 18 % CP with 0.72 % Met is preferred in feeding laying hens through the whole experimental period. Ecologically, reducing the level of crude protein in layer diets to be 16 % along with the supplementation of Met can play an important role in minimizing the pollution with N from poultry excretion.

Alexander, D. D., Yan, J., Bylsma, L. C., Northington, R. S., Grathwohl, D., Steenhout, P., Erdmann, P., Spivey-Krobath, E., and Haschke, F. Growth of infants consuming whey-predominant term infant formulas with a protein content of 1.8 g/100 kcal: a multicenter pooled analysis of individual participant data. Am.J.Clin.Nutr. 2016; 104(4):1083-1092; PM: 27604774

ABSTRACT: ***BACKGROUND***: High protein intake during infancy may contribute to obesity later in life in infants who are not exclusively breastfed. Lowering the protein content of infant formula so it is closer to that of mature breast milk may reduce long-term risk of overweight or obesity in formula-fed infants. ***OBJECTIVE***: We assessed the effects of whey-predominant formulas with a protein content of 1.8 g/100 kcal (lower than that in most current formulas and closer to breast milk) on infant growth by comparing against WHO growth standards and breastfed infants. ***DESIGN***: A multicenter pooled analysis was conducted with the use of individual participant data (n = 1882) from 11 randomized controlled trials of healthy term infants. Mixed-effects models that used ANCOVA were generated to estimate weight-for-age z score (WAZ), as well as length-for-age, BMI-for-age, and head circumference-for-age z scores at age 4 mo in infants fed a lower-protein infant formula (LPF) or a lower-protein infant formula with additional active ingredients (probiotics, prebiotics, or both) (LPFA) and breastfed infants. Estimates, including 95% CIs, were compared with a +/-0.5 SD of WHO growth standards, a benchmark for clinically

significant differences. ***RESULTS***: The 95% CIs for pooled estimates of WAZ were within +/- 0.5 SD of WHO growth standards for the LPF [0.07 (-0.16, 0.29)] and LPFA [0.22 (0.01, 0.43)] groups. WAZ was higher in the LPF ($P < 0.001$) and LPFA ($P = 0.003$) groups than in the breastfed infants, likely because breastfed infants had a relatively low WAZ [-0.23 (-0.51, 0.05)] compared with WHO growth standards. The 95% CIs for all other z scores in the LPF and LPFA groups were within +/-0.5 SD of WHO growth standards, except for head circumference, for which the upper limit of the 95% CI slightly exceeded 0.5 SD. No difference was observed in any z scores between the LPF and LPFA groups. ***CONCLUSION***: Whey-predominant infant formula with a lower protein content that more closely resembles that of breast milk supports healthy growth comparable to the WHO growth standards and close to breastfed infants.

Alioua, S., Abdi, A., Fhoula, I., Bringel, F., Boudabous, A., and Ouzari, I. H. Diversity of Vaginal Lactic Acid Bacterial Microbiota in 15 Algerian Pregnant Women with and without Bacterial Vaginosis by using Culture Independent Method. J.Clin.Diagn.Res. 2016; 10(9):DC23-DC27; PM: 27790434

ABSTRACT: ***INTRODUCTION***: Bacterial Vaginosis (BV) is the most common lower genital tract disorder among women of reproductive age (pregnant and non-pregnant) and a better knowledge of Lactobacillus species richness in healthy and infected vaginal microbiota is needed to efficiently design better probiotic products to promote the maintenance of normal flora which will help prevent bacterial vaginosis. AIM: To evaluate and compare the diversity of lactic acid bacterial species in pregnant women with and without BV. ***MATERIALS AND METHODS***: A pilot study was carried out during November-2014 to March-2015 in University Badji Mokhtar, Annaba, Algeria. Vaginal swabs were collected from 15 pregnant women aged between 19 and 35 years (mean 27.6 years; n=15) living in the East of Algeria visiting Gynecology service, hospital Abdallah Nouaouria- El bouni, Annaba. Vaginal samples were gram-stained, and scored by the Nugent method. The cohort included cases of women with healthy "normal" vaginal flora, infected flora with bacterial vaginosis and women with "intermediate" flora. The vaginal LAB community from pregnant women was identified by culture independent method based on Denaturing Gradient Gel Electrophoresis (DGGE), with the 16S rRNA gene sequencing. ***RESULTS***: A majority of LAB affiliated to the genus Lactobacillus was found in "normal" and "intermediate" flora (87.5% and 43.75% respectively), while a majority of LAB affiliated to the genus Enterococcus was identified in women with bacterial vaginosis and intermediate flora (60% and 46.75% respectively). Our results showed that the presence of Lactobacillus iners and Lactobacillus delbruekii promotes stability of the vaginal microbiota. ***CONCLUSION***: This result confirms the findings of previous studies suggesting that the occurrence of predominant Lactobacillus negatively correlates with bacterial vaginosis incidence and their current use as probiotics. Lactobacillus iners and Lactobacillus delbruekii can be defined as critical for defense of the vagina. In addition, Enterococcus feacalis can be considered as an indicator of imbalance of the vaginal ecosystem.

Alkasir, R., Li, J., Li, X., Jin, M., and Zhu, B. Human gut microbiota: the links with dementia development. Protein Cell. 11-19-2016; PM: 27866330

ABSTRACT: Dementia is a comprehensive category of brain diseases that is great enough to affect a person's daily functioning. The most common type of dementia is Alzheimer's disease, which makes most of cases. New researches indicate that gastrointestinal tract microbiota are directly linked to dementia pathogenesis through triggering metabolic diseases and low-grade inflammation progress. A novel strategy is proposed for the management of these disorders and as an adjuvant for psychiatric treatment of dementia and other related diseases through modulation of the microbiota (e.g. with the use of probiotics).

Alkaya, B., Laleman, I., Keceli, S., Ozcelik, O., Cenk, Haytac M., and Teughels, W. Clinical effects of probiotics containing Bacillus species on gingivitis: a pilot randomized controlled trial. J.Periodontal Res. 11-17-2016; PM: 27859252

ABSTRACT: ***BACKGROUND AND OBJECTIVE***: Lactobacillus spp. and bifidobacteria are the most frequently used probiotics in oral health research. However, although probiotic effects have been suggested for other genera, such as bacilli, no trials are available to describe the effect of bacilli probiotics on gingivitis in humans. The aim of the present study was to evaluate the clinical effects of a bacilli-containing toothpaste, a mouthrinse and a toothbrush cleaner versus a placebo in patients with generalized gingivitis. ***MATERIAL AND METHODS***: In this double-blind placebo-controlled randomized clinical trial, nonsmoking, systemically healthy patients with generalized gingivitis were included. They used a placebo or an experimental probiotic Bacillus subtilis-, Bacillus megaterium- and Bacillus pumulus-containing toothpaste, mouthrinse and toothbrush cleaner for 8 wk. Primary outcome measures of interest were plaque and gingivitis index, and the secondary outcome measures were pocket probing depth and bleeding on probing. ***RESULTS***: Twenty male and 20 female patients were randomized over the two groups. All participants could be included in the final analysis. Although plaque and gingivitis indices were significantly reduced after 8 wk, no intergroup differences could be found at any time point. Also, for the secondary outcome measure, intragroup but no intergroup differences could be detected. No harm or unintended effects were reported by the patients after using the study products. ***CONCLUSIONS***: This study did not show any statistically significant differences between a placebo and a bacilli-containing toothpaste, mouthrinse and toothbrush cleaner on gingivitis parameters.

Altieri, C. Dairy propionibacteria as probiotics: recent evidences. World J.Microbiol.Biotechnol. 2016; 32(10):172; PM: 27565782

ABSTRACT: Nowdays there is evidence that dairy propionibacteria display probiotic properties, which as yet have been underestimated. The aim of this paper is to review the recent highlights of data representing the probiotic potential of dairy propionibacteria, studied both by general selection criteria (useful for all probiotic potentials), and by more specific and innovative approach. Dairy propionibacteria show a robust nature, that makes them able to overcome technological hurdles, allowing their future use in various fermented probiotic foods. In addition to the general selection criteria for probiotics in areas such as food safety, technological and digestive stress tolerance, many potential health benefits have been recently described for dairy propionibacteria, including, production of several active molecules and adhesion

capability, that can mean a steady action in modulation of microbiota and of metabolic activity in the gut; their impact on intestinal inflammation, modulation of the immune system, potential modulation of risk factors for cancer development modulation of intestinal absorption.

Amakiri, A. C. and Thantsha, M. S. Survival of Bifidobacterium longum LMG 13197 microencapsulated in Vegetal or Vegetal-inulin matrix in simulated gastrointestinal fluids and yoghurt. Springerplus. 2016; 5(1):1343; PM: 27588236

ABSTRACT: ***BACKGROUND***: Vegetal BM 297 ATO is a food grade lipid based material extracted from vegetables, and certified for human consumption. In this study, Bifidobacterium longum LMG 13197 was encapsulated in Vegetal BM 297 ATO-inulin by freeze drying, followed by evaluation of its survival in simulated gastrointestinal fluids and yoghurt. Furthermore, the effect of incorporation of such microparticles on physico-chemical properties of yoghurt was examined. Unencapsulated and encapsulated B. longum cells were exposed to simulated gastrointestinal fluids for 6 h and yoghurt at 4 degrees C for 6 weeks, and then evaluated for viability using plate counts. ***RESULTS***: By the end of exposure to simulated gastrointestinal fluids, encapsulated cells were >5 log units higher than their unencapsulated counterparts. Furthermore, their levels in yoghurt remained above 10(6) cfu mL(-1) until the end of 6 weeks of storage while unencapsulated levels were at this level up to 5 weeks. There were no significant differences in pH values of yoghurts containing encapsulated cells throughout the storage ($p > 0.05$). However, significant differences in the lightness and yellowness of these yoghurts were recorded, although the total colour change was negligible. ***CONCLUSIONS***: Vegetal-inulin encapsulation protected probiotics in gastrointestinal fluids and yoghurt with negligible effects to its appearance, thus can be used for fortification of yoghurt with probiotics.

Ananthan, A., Balasubramanian, H., Rao, S., and Patole, S. Probiotic supplementation in children with cystic fibrosis-a systematic review. Eur.J.Pediatr. 2016; 175(10):1255-1266; PM: 27576473

ABSTRACT: Probiotics may benefit in cystic fibrosis (CF) as gut dysbiosis is associated with gastrointestinal symptoms and exacerbation of respiratory symptoms in CF. We conducted a systematic review of randomized controlled trials (RCTs) and non-RCTs of probiotic supplementation in children with CF, using the Cochrane methodology, preferred reporting items for systematic reviews (PRISMA) statement, and meta-analysis of observational studies in epidemiology (MOOSE) guidelines. Primary outcomes were pulmonary exacerbations, duration of hospitalization and antibiotics, and all-cause mortality. Secondary outcomes included gastrointestinal symptoms, markers of gut inflammation, and intestinal microbial balance. A total of nine studies (RCTs, 6, non-RCTs, 3; N = 275) with some methodological weaknesses were included in the review. The pooled estimate showed significant reduction in the rate of pulmonary exacerbation (fixed effects model, two parallel group RCTs and one cross-over trial: relative risk (RR) 0.25, (95 % confidence interval (95 % CI) 0.15,0.41); $p < 0.00001$; level of evidence: low) and decrease in fecal calprotectin (FCLP) levels (fixed effect model, three RCTs: mean difference (MD) -16.71, 95 % CI -27.30,-6.13); $p = 0.002$; level of evidence: low) after probiotic supplementation. Probiotic supplementation significantly improved gastrointestinal symptoms (one RCT, one non-RCT) and gut microbial balance (decreased Proteobacteria, increased Firmicutes, and Bacteroides in one RCT, one non-RCT). CONCLUSION: Limited low-quality evidence exists on the effects of probiotics in

children with CF. Well-designed adequately powered RCTs assessing clinically meaningful outcomes are required to study this important issue. WHAT IS KNOWN: * Gut dysbiosis is frequent in children with cystic fibrosis due to frequent exposure to pathogens and antibiotics. * Probiotics decrease gut dysbiosis and improve gut maturity and function. What is New: * This comprehensive systematic review shows that current evidence on the safety and efficacy of probiotics in children with cystic fibrosis is limited and of low quality. * Well-designed and adequately powered trials assessing clinically important outcomes are required considering the health burden of cystic fibrosis and the potential benefits of probiotics.

Anderson, J. L., Miles, C., and Tierney, A. C. Effect of probiotics on respiratory, gastrointestinal and nutritional outcomes in patients with cystic fibrosis: A systematic review. J.Cyst.Fibros. 9-29-2016; PM: 27693010

ABSTRACT: ***BACKGROUND***: An increasing body of research investigating the use of probiotics to improve health outcomes in patients with cystic fibrosis (CF) prompted the need to systematically assess and summarise the relevant literature. ***METHODS***: An electronic search of five databases and three trial databases was conducted. Studies describing the administration of probiotics to patients with CF older than 2years, with a comparator group on respiratory, gastrointestinal and nutritional outcomes were included. ***RESULTS***: Three pre-post studies and six randomised controlled trials met the inclusion criteria. Overall studies showed a positive effect of probiotics on reducing the number of pulmonary exacerbations and decreasing gastrointestinal inflammation. There was limited effect of probiotics on other outcomes and inadequate evidence for the effects of specific probiotic species and strains. ***CONCLUSION***: The findings suggest that probiotics may improve respiratory and gastrointestinal outcomes in a stable CF clinic population with no reported evidence of harm. There is inadequate evidence at this time to recommend a specific species, strain or dose of probiotic as likely to be of significant benefit.

Andrejcakova, Z., Sopkova, D., Vlckova, R., Kulichova, L., Gancarcikova, S., Almasiova, V., Holovska, K., Petrilla, V., and Kresakova, L. Synbiotics suppress the release of lactate dehydrogenase, promote non-specific immunity and integrity of jejunum mucosa in piglets. Anim Sci.J. 2016; 87(9):1157-1166; PM: 27581561

ABSTRACT: The aim of our experiment was to study how synbiotics are able to deal with the problems of post-weaning piglets. Lactobacillus plantarum - Biocenol(TM) LP96 (CCM 7512), Lactobacillus fermentum - Biocenol(TM) LF99 (CCM 7514) and flaxseed (rich in n-3 polyunsaturated fatty acids) were administered to 36 conventional piglets from a problematic breed with confirmed presence of enterotoxigenic Escherichia coli and Coronavirus. The experimental piglets were supplied with probiotic cheeses and crushed flax-seed in the period starting 10 days before weaning and lasting up to 14 days post-weaning. Piglets in the control group were supplied only control cheese. The impact of such additives on the release of lactate dehydrogenase (LDH; spectroscopic and electrophoretic assay), alteration of immunity (index of metabolic activity), jejunum histology (light microscopy), and health of conventional piglets from a problematic breed (monitoring of hematology, consistency and moisture of feces and body temperature) were examined. We found significant decrease in LDH leakage in the blood serum and tissue extracts, indicating better cell membrane integrity in the individual organs of animals. Probiotics and flaxseed applied together seem to be a

good source of nutrients to improve the immune status and the integrity of jejunum mucosa during infection. (c) 2015 Japanese Society of Animal Science.

Angelakis, E. Weight gain by gut microbiota manipulation in productive animals. Microb.Pathog. 11-9-2016; PM: 27836763

ABSTRACT: Antibiotics, prebiotics and probiotics are widely used as growth promoters in agriculture. In the 1940s, use of Streptomyces aureofaciens probiotics resulted in weight gain in animals, which led to the discovery of chlortetracycline. Tetracyclines, macrolides, avoparcin and penicillins have been commonly used in livestock agriculture to promote growth through increased food intake, weight gain, and improved herd health. Prebiotic supplements including oligosaccharides, fructooligosaccharides, and galactosyl-lactose improve the growth performance of animals. Probiotics used in animal feed are mainly bacterial strains of Gram-positive bacteria and have been effectively used for weight gain in chickens, pigs, ruminants and in aquaculture. Antibiotics, prebiotics and probiotics all modify the gut microbiota and the effect of a probiotic species on the digestive flora is probably determined by bacteriocin production. Regulations governing the introduction of novel probiotics and prebiotics vary by geographical region and bias is very common in industry-funded studies. Probiotic and prebiotic foods have been consumed for centuries, either as natural components of food, or as fermented foods and it is possible to cause the same weight gain effects in humans as in animals. This review presents the use of growth promoters in food-producing animals to influence food intake and weight gain.

Anonye, B. O. General Commentary on: Alternatives to Antibiotic Growth Promoters in Animals. Front Vet.Sci. 2016; 3:74; PM: 27656646

No Abstract Available.

Anselmo, A. C., McHugh, K. J., Webster, J., Langer, R., and Jaklenec, A. Layer-by-Layer Encapsulation of Probiotics for Delivery to the Microbiome. Adv.Mater. 2016; 28(43):9486-9490; PM: 27616140

ABSTRACT: The gastrointestinal (GI) microbiome is widely investigated for its role in many diseases. However, technologies designed for microbiome delivery are lacking. Here, a layer-by-layer (LbL) approach is reported for probiotic encapsulation to protect probiotics against GI tract insults and improve their adhesion and growth on the intestines. These advantages translate to significantly enhanced survival of LbL-probiotics in vivo.

Aoki, R., Tsuchida, S., Arai, Y., Ohno, K., Nishijima, T., Mawatari, T., Mikami, Y., and Ushida, K. Effect of Bifidobacterium animalis subsp. lactis GCL2505 on the physiological function of intestine in a rat model. Food Sci.Nutr. 2016; 4(6):782-790; PM: 27826427

ABSTRACT: Bifidobacterium animalis ssp. lactis GCL2505 has been shown to proliferate in

the human intestine. The intestinal dynamics and physiological effects of GCL2505 as well as the mechanism underlying proliferation in the gut were investigated. GCL2505 showed markedly higher resistance to free bile acids (cholic and deoxycholic acids) than other bifidobacterial species. The intestinal dynamics of GCL2505 and B. longum ssp. longum JCM1217T was compared. The level of B. animalis ssp. lactis in the GCL2505-administered group was remarkably higher than that of B. longum in the JCM1217T-administered group. The distribution of B. animalis ssp. lactis through the intestine of the GCL2505-administered group revealed that GCL2505 proliferated in the cecum. The physiological effects of GCL2505 and JCM 1217T were investigated. The cecal IgA level in the GCL2505-administered group was significantly higher than that in the nontreated control group. In contrast, the JCM 1217T-administered group did not manifest any change in the cecal IgA level. Mucin excretion in the GCL2505-administered group was significantly higher than that in the JCM 1217T-administered group. The thickness of the sulfomucin layer of the colon in the GCL2505-administered group tended to be higher than that in the JCM 1217T-administered group. In a loperamide-induced constipation model, fecal excretion in the GCL2505-administered group was significantly increased compared with that in the loperamide-treated control group. Short-chain fatty acid concentration in the GCL2505-administered group was significantly higher than that in the loperamide-treated control group. These results indicate that the level of proliferation of probiotics in the intestine correlates with the magnitude of host physiological responses, such as IgA production and mucin secretion, which possibly affect gastrointestinal functions such as bowel movement to counteract constipation. GCL2505 exhibits high tolerance to secondary bile acids, which partially explains its higher rate of proliferation in the large intestine.

Aoyagi, Y., Park, S., Matsubara, S., Honda, Y., Amamoto, R., Kushiro, A., Miyazaki, K., and Shephard, R. J. Habitual intake of fermented milk products containing Lactobacillus casei strain Shirota and a reduced risk of hypertension in older people. Benef.Microbes. 12-1-2016; 1-8; PM: 27903092

ABSTRACT: This study investigated relationships between the frequent intake of fermented milk products containing Lactobacillus casei strain Shirota (LcS) and the onset of hypertension (resting systemic pressure >/=140 mmHg [systolic]/>/=90 mmHg [diastolic], a doctor's diagnosis and/or antihypertensive medicine use) during a 5-year period in 352 communityliving Japanese aged 65 to 93 years (125 men and 227 women). Initially normotensive subjects were divided into two groups (n=254 and n=98) on the basis of their intake of fermented milk products (<3 or >/=3 times/week, respectively), as estimated during an interview by a certified nutritionist. The incidence of hypertension over the 5-year interval was significantly lower in those who took fermented milk products >/=3 rather than <3 times/week (6.1 vs 14.2%, P=0.037). A multivariate-adjusted proportional hazards model predicted that blood pressures were significantly more likely to remain normal over 5 years in subjects who took >/=3 fermented milk products rather than <3 times/ week (relative risk 0.398 [95% confidence interval 0.167-0.948], P=0.037). These results suggest that after adjustment for potential confounders, the risk of developing hypertension is substantially lower in elderly people who take fermented milk products containing LcS at least 3 times a week.

Arboleya, S., Watkins, C., Stanton, C., and Ross, R. P. Gut Bifidobacteria Populations in Human Health and Aging. Front Microbiol. 2016; 7:1204; PM: 27594848

ABSTRACT: The intestinal microbiota has increasingly been shown to have a vital role in various aspects of human health. Indeed, several studies have linked alterations in the gut microbiota with the development of different diseases. Among the vast gut bacterial community, Bifidobacterium is a genus which dominates the intestine of healthy breast-fed infants whereas in adulthood the levels are lower but relatively stable. The presence of different species of bifidobacteria changes with age, from childhood to old age. Bifidobacterium longum, B. breve, and B. bifidum are generally dominant in infants, whereas B. catenulatum, B. adolescentis and, as well as B. longum are more prevalent in adults. Increasingly, evidence is accumulating which shows beneficial effects of supplementation with bifidobacteria for the improvement of human health conditions ranging from protection against infection to different extra- and intra-intestinal positive effects. Moreover, bifidobacteria have been associated with the production of a number of potentially health promoting metabolites including short chain fatty acids, conjugated linoleic acid and bacteriocins. The aim of this mini-review is to describe the bifidobacteria compositional changes associated with different stages in life, highlighting their beneficial role, as well as their presence or absence in many disease states.

Arbulu, S., Jimenez, J. J., Gutiez, L., Campanero, C., Del, Campo R., Cintas, L. M., Herranz, C., and Hernandez, P. E. Evaluation of bacteriocinogenic activity, safety traits and biotechnological potential of fecal lactic acid bacteria (LAB), isolated from Griffon Vultures (Gyps fulvus subsp. fulvus). BMC.Microbiol. 9-29-2016; 16(1):228; PM: 27688001

ABSTRACT: ***BACKGROUND***: Lactic acid bacteria (LAB) are part of the gut microbiota and produce ribosomally synthesized antimicrobial peptides or bacteriocins with interest as natural food preservatives and therapeutic agents. Bacteriocin-producing LAB are also attractive as probiotics. Griffon vultures (Gyps fulvus subspecies fulvus) are scavenger birds that feed almost exclusively on carrion without suffering apparent ill effects. Therefore, griffon vultures might be considered a reservoir of bacteriocin-producing lactic acid bacteria (LAB) with potential biotechnological applications. ***RESULTS***: Griffon vulture feces were screened for LAB with antimicrobial activity, genes encoding bacteriocins, potential virulence determinants, susceptibility to antibiotics, genotyping and characterization of bacteriocins. In this study, from 924 LAB evaluated 332 isolates (36 %) showed direct antimicrobial activity against Gram-positive bacteria only. The molecular identification of the most antagonistic 95 isolates showed that enterococci was the largest LAB group with antimicrobial activity (91 %) and E. faecium (40 %) the most identified antagonistic species. The evaluation of the presence of bacteriocin structural genes in 28 LAB isolates with the highest bacteriocinogenic activity in their supernatants determined that most enterococcal isolates (75 %) encoded multiple bacteriocins, being enterocin A (EntA) the largest identified (46 %) bacteriocin. Most enterococci (88 %) were resistant to multiple antibiotics. ERIC-PCR and MLST techniques permitted genotyping and recognition of the potential safety of the bacteriocinogenic enterococci. A multiple-step chromatographic procedure, determination of the N-terminal amino acid sequence of purified bacteriocins by Edman degradation and a MALDI TOF/TOF tandem MS procedure permitted characterization of bacteriocins present in supernatants of producer cells. ***CONCLUSIONS***: Enterococci was the largest LAB group with bacteriocinogenic activity isolated from griffon vulture feces. Among the isolates, E. faecium M3K31 has been identified as producer of enterocin HF (EntHF), a bacteriocin with remarkable antimicrobial activity against most evaluated Listeria spp. and of elevated interest as a natural food preservative. E. faecium M3K31 would be also considered a safe probiotic strain for use in animal nutrition.

Argenta, A., Satish, L., Gallo, P., Liu, F., and Kathju, S. Local Application of Probiotic Bacteria Prophylaxes against Sepsis and Death Resulting from Burn Wound Infection. PLoS.One. 2016; 11(10):e0165294; PM: 27780258

ABSTRACT: ***OBJECTIVE***: To determine if local prophylactic application of probiotic bacteria to burn wounds will prevent death in a mouse model of burn wound sepsis. ***BACKGROUND***: Infection remains the most common complication after burn injury and can result in sepsis and death, despite the use of topical and systemic antibiotics. Pseudomonas aeruginosa is a frequently implicated pathogen. Local application of probiotics directly to burn wounds is an attractive novel intervention that avoids the pitfalls of standard antibiotic therapies. ***METHODS***: A burn-sepsis model was established using a sub-eschar injection of bioluminescent P. aeruginosa; infection was tracked using a charge-coupled camera. Full-thickness burn injuries were placed on the dorsums of adult mice; the injured sites were then treated with vehicle (burn wound control), probiotics (Lactobacillus plantarum only), pathogenic bacteria (Pseudomonas aeruginosa only), or probiotics plus pathogen (Lactobacillus plus Pseudomonas). Animals were monitored until death/moribundity or for one week, then sacrificed. Harvested tissues were subjected to imaging and molecular assays. ***RESULTS***: Control and probiotic-only animals showed no mortality (100% survival) at one week. Pseudomonas-only animals showed > 90% mortality within 40 hours of infection. In contrast, animals treated with probiotics plus Pseudomonas showed less than 10% mortality. Use of bioluminescent Pseudomonas bacteria demonstrated that probiotic therapy inhibited septicemic accumulation of the pathogen in remote organs. In addition, probiotic therapy successfully suppressed the infection-dependent induction of TNF-alpha and interleukins 6 and 10 in the liver. ***CONCLUSIONS***: Local probiotic therapy shows great potential as a valuable adjunct in the management of complicated burn injury.

Arief, I. I., Afiyah, D. N., Wulandari, Z., and Budiman, C. Physicochemical Properties, Fatty Acid Profiles, and Sensory Characteristics of Fermented Beef Sausage by Probiotics Lactobacillus plantarum IIA-2C12 or Lactobacillus acidophilus IIA-2B4. J.Food Sci. 10-6-2016; PM: 27712046

ABSTRACT: Probiotics may be used to enhance the functionality and nutritional values of fermented sausages. This study aims to evaluate the physicochemical and sensory properties of beef sausages fermented by lactic acid bacteria of Lactobacillus plantarum IIA-2C12 and L. acidophilus IIA-2B4. These strains were isolated from beef cattle and have shown to display probiotic features. While the nutrient contents were not affected by the probiotics, the pH, texture, and color varied among the sausages. Further analysis on fatty acids showed different profiles of saturated (C14:0, C17:0, and C20:0) and unsaturated (C14:1, C18:1n9c, C18:2n6c, and C22:6n3) fatty acids in sausages with probiotics. Gas chromatography-mass spectrometry further revealed some flavor development compounds including acid, alcohols, aldehydes, aromatic, ketones, sulfur, hydrocarbons and terpenes, varied among the sausages. Hedonic test showed no difference in the preference toward aroma, texture, and color for untrained panelists.

Aroniadis, O. C., Drossman, D. A., and Simren, M. A Perspective on Brain-Gut Communication: The American Gastroenterology Association and American Psychosomatic Society Joint

Symposium on Brain-gut Interactions and the Intestinal Microenvironment. Psychosom.Med. 12-2-2016; PM: 27922565

ABSTRACT: ***BACKGROUND***: Alterations in brain-gut communication and the intestinal microenvironment have been implicated in a variety of medical and neuropsychiatric diseases. Three central areas require basic and clinical research: (1) how the intestinal microenvironment interacts with the host immune system, central nervous system and enteric nervous system; (2) the role of the intestinal microenvironment in the pathogenesis of medical and neuropsychiatric disease; (3) the effects of diet, prebiotics, probiotics and fecal microbiota transplantation on the intestinal microenvironment and the treatment of disease. ***METHODS***: This review article is based on a symposium convened by the American Gastroenterology Association and American Psychosomatic Society to foster interest in the role of the intestinal microenvironment in brain-gut communication and pathogenesis of neuropsychiatric and biopsychosocial disorders. The aim was to define the stat-of-the-art of the current scientific knowledge base and to identify guidelines and future directions for new research in this area. ***RESULTS***: This review provides a characterization of the intestinal microbial composition and function. We also provide evidence for the interactions between the intestinal microbiome, the host and the environment. The role of the intestinal microbiome in medical and neuropsychiatric diseases is reviewed as well as the treatment effects of manipulation of the intestinal microbiome. ***CONCLUSIONS***: Based on this review, opportunities and challenges for conducting research in the field are described, leading to potential avenues for future research.

Asha, M. K., Debraj, D., Dethe, S., Bhaskar, A., Muruganantham, N., and Deepak, M. Effect of Flavonoid-Rich Extract of Glycyrrhiza glabra on Gut-Friendly Microorganisms, Commercial Probiotic Preparations, and Digestive Enzymes. J.Diet.Suppl. 9-2-2016; 1-11; PM: 27588327

ABSTRACT: Flavonoid-rich extract prepared from Glycyrrhiza glabra has been found to be beneficial in patients with functional dyspepsia and was reported to possess some gut health-promoting properties such as antioxidant, anti-inflammatory and anti-Helicobacter pylori activities. In the present study, the flavonoid-rich extract of Glycyrrhiza glabra was evaluated for its compatibility with probiotic strains (Lactobacillus casei, Lactobacillus fermentum, Lactobacillus plantarum, and Streptococcus thermophilus), commercial probiotic drinks, and digestive enzymes (pancreatic alpha-amylase, alpha-glucosidase, phytase, xylanase, and pancreatic lipase). Results of this study indicated that the flavonoid-rich extract of Glycyrrhiza glabra is compatible with the tested probiotic strains, probiotic drinks and digestive enzymes.

Ayoub, D., Lopetuso, L. R., Chamseddine, F., Dajani, A., Lahiri, K., Mahmoud, H., Miqdady, M. S., Zirizzotti, G., Sultan, M. A., Franceschi, F., and Gasbarrini, A. Epidemiological evaluation of acute gastroenteritis and therapeutic approaches in Middle East Countries. Eur.Rev.Med.Pharmacol.Sci. 2016; 20(18):3891-3901; PM: 27735025

ABSTRACT: ***OBJECTIVE***: Gastroenteritis represents with respiratory tract infection the most common infectious disease syndrome of humans in developing countries. Gut microbiota regional variation and dysbiosis play a crucial role in triggering and worsening this devastating GI disease. ***MATERIALS AND METHODS***: With this manuscript, we want to explore and emphasize the critical aspect of acute gastroenteritis in Middle-East

Countries and its correlation with the clinical aspect of gut microbiota modification and intestinal homeostasis. ***RESULTS***: Approximately 1 of 50 children born each year in developed nations is hospitalized for acute gastroenteritis sometime during childhood. The highest rate of illness occurs in children between 3 and 24 months of age. The common causes of diarrhea are infections with viruses and bacteria, diarrhea due to a systemic infection other than gastrointestinal, diarrhea associated with antibiotic administration, and feeding related diarrhea. The single most common diarrheal disorder observed in the Emergency Department and general practice is viral gastroenteritis. In particular, Rotavirus is the cause of more than 2 million hospitalizations and over half a million deaths from acute GE in infants and young children. This burden produces also direct and indirect economic costs. The use of probiotics to counterbalance commensal dysbiosis is emerging as a standard medical practice in these countries. ***CONCLUSIONS***: In this scenario, one of the most interesting aspects is that probiotics and gut microbiota modulation could deeply improve the prevention and treatment of this devastating GI pathology. At the same time, vaccination might represent a cost-effective strategy to reduce the health and economic burden of some pathogens, such as rotavirus.

Azimirad, M., Alebouyeh, M., and Naji, T. Inhibition of Lipopolysaccharide-Induced Interleukin 8 in Human Adenocarcinoma Cell Line HT-29 by Spore Probiotics: B. coagulans and B. subtilis (natto). Probiotics.Antimicrob.Proteins. 10-26-2016; PM: 27785697

ABSTRACT: Probiotics are used as a treatment for different intestinal disorders. They confer health benefits by different ways. This study was aimed to investigate immunomodulatory effect of Bacillus probiotic spores on the production of lipopolysaccharide (LPS)-induced interleukin 8 (IL-8) in HT-29 intestinal epithelial cells. Differentiated intestinal epithelial cell line was used as a model for the study of colonization of purified spores (Bacillus subtilis (natto) and B. coagulans) and their anti-inflammatory effects. MTT assay and trypan blue staining were used for the detection of optimal concentration of the purified spores and LPS. Pre-treatment assay was done by treatment of the cells with the purified spores for 2 h, followed by challenges with LPS for 3 and 18 h. Post-treatment assay was done by initial treatment of the cells with LPS for 18 h, followed by the spores for 3 and 6 h. Levels of IL-8 secretion and its mRNA expression were measured by ELISA and relative Q real-time PCR. Our results showed similar rates of adherence to intestinal epithelial cells by the spore probiotics, while displaying no cytotoxic effect. In the pre-treatment assay, a significant decrease in IL-8, at both protein and mRNA levels, was measured for B. coagulans spores after the addition of LPS, which was higher than those observed for Bacillus subtilis (natto) spores. In the post-treatment assay, while Bacillus subtilis (but not B. coagulans) diminished the LPS-stimulated IL-8 levels after 3 h of incubation, the inhibitory effect was not constant. In conclusion, ability of Bacillus spore probiotics for adherence to intestinal epithelial cell and their anti-inflammatory effects, through interference with LPS/IL-8 signaling, was shown in this study. Further studies are needed to characterize responsible bacterial compounds associated with these effects.

Babashahi, M., Mirlohi, M., Ghiasvand, R., and Torki-Baghbadorani, S. Evaluation of response surface methodology to predict optimum growth conditions for Lactobacillus plantarum

A7 (KC 355240) in probiotic soy milk containing essential oil of Cuminum cyminum. Recent Pat Food Nutr.Agric. 10-26-2016; PM: 27784227

ABSTRACT: ***BACKGROUND***: A new product from soy milk, which is a combination of probiotics and essential oils, can be regarded as a functional food with new added health benefit. ***OBJECTIVES***: This study aimed at evaluation of the results of response surface methodology (RSM) in prediction of the optimum growth condition of Lactobacillus plantarum A7 (KC 355240) (LA7) cultured in soy milk enriched with different concentrations of essential oil of Cuminum cyminum in comparison with the classical growth curve monitoring method. ***MATERIALS AND METHODS***: Bacterial growth was investigated in the samples of probiotic soy milk containing three concentration levels (0.01%, 0.02% and 0.03% (v/v)) of the essential oil of C. cyminum. The experiments were repeated three times. Concentration of C. cyminum essential oil and the fermentation time were considered as factors and experiments were conducted base on a small composite design of response surface methodology. ***RESULTS***: According to the tests carried out, the optimum conditions were determined as the concentration of 0.02% (v/v) essential oil and 9 hours fermentation. ***CONCLUSIONS***: These results were in accordance with the outcomes of the classical growth curve monitoring method. There are several patents that have recently shown a diverse mix of soy milk in functional foods. Results of this study can be used in the preparation of functional products, enriched with medicinal plants.

Bahi, A., Guardiola, F. A., Messina, C., Mahdhi, A., Cerezuela, R., Santulli, A., Bakhrouf, A., and Esteban, M. A. Effects of dietary administration of fenugreek seeds, alone or in combination with probiotics, on growth performance parameters, humoral immune response and gene expression of gilthead seabream (Sparus aurata L.). Fish.Shellfish.Immunol. 11-14-2016; 60:50-58; PM: 27856325

ABSTRACT: The use of immunostimulants is considered a promising preventive practice that may help to maintain animal welfare and a healthy environment, while increasing production and providing higher profits. The purpose of this study was to evaluate the effects on gilthead seabream (Sparus aurata L.) of the dietary administration of fenugreek (Trigonella foenum graecum) seeds, alone or combined with one of the following probiotic strains: Bacillus licheniformis (TSB27), Lactobacillus plantarum or Bacillus subtilis (B46). Gilthead seabream were fed a control or one of the supplemented diets for 3 weeks. The effects of these supplemented diets on growth performance parameters and the humoral immune response (natural haemolytic complement, peroxidase, total IgM levels, proteases and antiproteases activities) were evaluated after 2 and 3 weeks of feeding. Simultaneously, the expression levels of some immune-relevant genes (igm, tcr-beta, csfr1 and bd) were measured in the head-kidney. Interestingly, all probiotic supplemented diets increased seabream growth rates, especially the B. licheniformis supplemented diet. Generally, humoral immune parameters were enhanced by the dietary supplementation at the different time points measured. The results showed a significant increases in the immune parameters, principally in fish fed only fenugreek or fenugreek combined with B. subtilis. Furthermore, real time qPCR revealed that dietary supplementation significantly enhances the expression of immune-associated genes in the head-kidney, particularly igm gene expression. These results suggest that fenugreek alone or combined with one of the probiotic strains mentioned enhances the immune response of gilthead seabream, a species with one of the highest rates of production in marine aquaculture.

Bai, Z., Zhang, H., Li, N., Bai, Z., Zhang, L., Xue, Z., Jiang, H., Song, Y., and Zhou, D. Impact of Environmental Microbes on the Composition of the Gut Microbiota of Adult BALB/c Mice. PLoS.One. 2016; 11(8):e0160568; PM: 27518814

ABSTRACT: To investigate the impact of microbes within the living environment on the gut microbiota of adults, we raised three groups of BALB/c mice from 3-4 weeks age in the same specific-pathogen-free animal room for 8 weeks. The control group lived in cages with sterilized bedding (pelletized cardboard), the probiotics group had three probiotics added to the sterilized bedding, and the intestinal microbes (IM) group had the intestinal microbes of a healthy goat added to the bedding. All other variables such as diet, age, genetic background, physiological status, original gut microbiota, and living room were controlled. Using high-throughput sequencing of the 16S rRNA gene, we observed that the control and probiotics groups had similar diversity and richness of gut microbiota. The two groups had significantly lower diversity than the IM group. We also observed that the IM group had a specific structure of gut microbial community compared with the control and probiotics groups. However, the dominate bacteria changed slightly upon exposure to intestinal microbes, and the abundance of the non-dominate species changed significantly. In addition, exposure to intestinal microbes inhibited DNFB-induced elevation of serum IgE levels. Our results provide new evidence in support of the microflora and hygiene hypotheses.

Balakumar, M., Prabhu, D., Sathishkumar, C., Prabu, P., Rokana, N., Kumar, R., Raghavan, S., Soundarajan, A., Grover, S., Batish, V. K., Mohan, V., and Balasubramanyam, M. Improvement in glucose tolerance and insulin sensitivity by probiotic strains of Indian gut origin in high-fat diet-fed C57BL/6J mice. Eur.J.Nutr. 10-18-2016; PM: 27757592

ABSTRACT: ***PURPOSE***: Diabetes and obesity are characterized by glucose intolerance, fat deposition, inflammation, and dyslipidemia. Recent reports postulated that distinct gut microbiota alterations were observed in obese/diabetic subjects and modulating gut microbiota beneficially through specific probiotics could be a potential therapeutic option for type 2 diabetes/obesity. Therefore, we attempted to study the efficacy of probiotics of Indian gut origin (Lactobacillus plantarum MTCC5690 and Lactobacillus fermentum MTCC5689) along with a positive control, Lactobacillus rhamnosus (LGG) on glucose/lipid homeostasis in high-fat-diet-induced diabetic animal model. ***METHODS***: C57BL/6J male mice were divided into seven groups (n = 6 per group) comprising feeding on: (1) Normal Pellet Diet (NPD), (2) High-Fat Diet (HFD), (3) HFD with LGG, (4) HFD with MTCC5690, (5) HFD with MTCC5689, (6) HFD with metformin, and 7) HFD with vildagliptin for a period of 6 months. Biochemical markers, glucose tolerance, insulin resistance, and GLP-1 and LPS levels were assessed by standard protocols. Gut integrity was measured by intestinal permeability test. Transcriptional levels of tight junction proteins (TJPs) were probed in small intestinal tissues while inflammatory signals and other pathway specific genes were profiled in liver, visceral adipose tissue, and skeletal muscle. ***RESULTS***: Mice fed with HFD became insulin resistant, glucose intolerant, hyperglycemic, and dyslipidemic. Diabetic mice were characterized to exhibit decreased levels of GLP-1, increased gut permeability, increased circulatory levels of LPS, decrease in the gene expression patterns of intestinal tight junction markers (occludin and ZO-1), and increased proinflammatory gene markers (TNFalpha and IL6) in visceral fat along with decreased mRNA expression of FIAF and adiponectin. Diabetic mice also exhibited increased mRNA expression of ER stress markers in skeletal muscle. In addition, liver from HFD-fed diabetic mice showed increased gene expressions of proinflammation, lipogenesis, and gluconeogenesis. Probiotic interventions (most

prominently the MTCC5689) resisted insulin resistance and development of diabetes in mice under HFD feeding and beneficially modulated all the biochemical and molecular alterations in a mechanistic way in several tissues. The metabolic benefits offered by the probiotics were also more or less similar to that of standard drugs such as metformin and vildagliptin. ***CONCLUSION***: Native probiotic strains MTCC 5690 and MTCC 5689 appear to have potential against insulin resistance and type 2 diabetes with mechanistic, multiple tissue-specific mode of actions.

Balamuralikrishnan, B., Lee, S. I., and Kim, I. H. Dietary inclusion of different multi-strain complex probiotics on performance in broilers. Br.Poult.Sci. 12-5-2016; PM: 27918205

ABSTRACT: 1. This study was designed to evaluate the effects of two different complex probiotic supplementations on the growth performance, meat quality, excreta microflora, nutrient retention, blood metabolic profile and noxious gas emissions in broilers. 2. A total of 612 conventional healthy 1-d-old Ross 308 broilers with body weight of 41 +/- 0.3 g were randomly divided into 3 treatments with 12 replicate cages, with 17 broilers in each cage and fed with the following diets: CON-Basal diet, T1-CON + Probiotic A, T2-CON + Probiotic B. 3. Significant results were observed on body weight gain, but not on feed conversation ratio and feed intake, in the whole experimental period. Increased faecal lactobacillus counts were found with probiotics supplementation. However, no significant effects were found for meat quality, nutrient retention, blood metabolic profile or noxious gas emissions. 4. In conclusion, both multi-strain probiotics had beneficial effects on growth performance, drip loss percentage and faecal Lactobacillus counts in broilers.

Baldassarre, M. E., Di, Mauro A., Mastromarino, P., Fanelli, M., Martinelli, D., Urbano, F., Capobianco, D., and Laforgia, N. Administration of a Multi-Strain Probiotic Product to Women in the Perinatal Period Differentially Affects the Breast Milk Cytokine Profile and May Have Beneficial Effects on Neonatal Gastrointestinal Functional Symptoms. A Randomized Clinical Trial. Nutrients. 10-27-2016; 8(11); PM: 27801789

ABSTRACT: ***BACKGROUND***: Probiotic supplementation to women during pregnancy and lactation can modulate breast milk composition, with immune benefits being transferred to their infants. AIM: The aim of the study was to evaluate the effect of high-dose probiotic supplementation to women during late pregnancy and lactation on cytokine profile and secretory IgA (sIgA) in breast milk and thus to study if differences in breast milk composition can affect lactoferrin and sIgA levels in stool samples of newborns. The safety of maternal probiotic administration on neonatal growth pattern and gastrointestinal symptoms were also evaluated. ***METHODS***: In a double-blind, placebo-controlled, randomized trial, 66 women took either the probiotic (n = 33) or a placebo (n = 33) daily. Levels of interleukins (IL-6, IL-10 and IL-1beta), transforming growth factor-beta1 (TGF-beta1), and sIgA in breast milk; and the level of sIgA and lactoferrin in newborn stool samples were analyzed at birth and then again at one month of life. Antropometrical evaluation and analysis of gastrointestinal events in newborns was also performed. ***RESULTS***: Probiotic maternal consumption had a significant impact on IL6 mean values in colostrum and on IL10 and TGF-beta1 mean values in mature breast milk. Fecal sIgA mean values were higher in newborns whose mothers took the probiotic product than in the control group. Probiotic maternal supplementation seems to decrease incidence of infantile colic and regurgitation in infants. ***CONCLUSION***: High-dose multi-strain probiotic administration to women during pregnancy influences breast

milk cytokines pattern and sIgA production in newborns, and seems to improve gastrointestinal functional symptoms in infants.

Balzaretti, S., Taverniti, V., Guglielmetti, S., Fiore, W., Minuzzo, M., Ngo, H. N., Ngere, J. B., Sadiq, S., Humphreys, P. N., and Laws, A. P. A novel rhamnose-rich hetero-exopolysaccharide isolated from Lactobacillus paracasei DG activates THP-1 human monocytic cells. Appl.Environ.Microbiol. 12-2-2016; PM: 27913418

ABSTRACT: Lactobacillus paracasei DG is a bacterial strain with recognized probiotic properties and is used in commercial probiotic products. However, the mechanisms underlying its probiotic properties are mainly unknown. In this study, we tested the hypothesis that the capability of strain DG to interact with the host is, at least partly, associated with its ability to synthesize a surface-associated exopolysaccharide (EPS). Comparative genomics revealed the presence of putative EPS gene clusters in DG genome; accordingly, EPS was isolated from the surface of the bacterium. A sample of the pure EPS from strain DG (DG-EPS), upon NMR and chemical analyses, was shown to be a novel branched hetero-EPS with a repeat unit composed of L-rhamnose, D-galactose, and N-acetyl-D-galactosamine in a ratio of 4:1:1. Subsequently, we demonstrated that the DG-EPS displays immunostimulating properties by enhancing the gene expression of the pro-inflammatory cytokines TNF-alpha and IL-6, and, particularly, the chemokines IL-8 and CCL20 in the THP-1 human monocytic cell line. In contrast, the expression of the cyclooxygenase enzyme COX-2 was not affected. In conclusion, the DG-EPS is a bacterial macromolecule with the potential ability to boost the immune system as either a secreted molecule released from the bacterium or as a capsular envelope on the bacterial cell wall. This study provides additional information about the mechanisms supporting the cross-talk between L. paracasei DG and the host. IMPORTANCE: The consumption of food products and supplements named probiotics, i.e. containing live microbial cells, to potentially prevent or treat specific diseases, is constantly gaining popularity. The lack of knowledge on the precise mechanisms supporting their potential health-promoting properties, however, greatly limits a more appropriate use of each single probiotic strain. In this context, we studied a well-known probiotic, Lactobacillus paracasei DG, in order to identify the constitutive molecules that can explain the documented health-promoting properties of this bacterium. We found a novel polysaccharide molecule, named DG-EPS, that is secreted by and covers the bacterium. We demonstrated that this molecule, which has a chemical structure never identified before, has immunostimulatory properties and, therefore, may contribute to the ability of the probiotic L. paracasei DG to interact with the immune system.

Barbosa, I. C., Oliveira, M. E., Madruga, M. S., Gullon, B., Pacheco, M. T., Gomes, A. M., Batista, A. S., Pintado, M. M., Souza, E. L., and Queiroga, R. C. Influence of the addition of Lactobacillus acidophilus La-05, Bifidobacterium animalis subsp. lactis Bb-12 and inulin on the technological, physicochemical, microbiological and sensory features of creamy goat cheese. Food Funct. 10-12-2016; 7(10):4356-4371. PM: 27711907

ABSTRACT: The effects of the addition of Lactobacillus acidophilus LA-05, Bifidobacterium animalis subsp. lactis BB-12 and inulin on the quality characteristics of creamy goat cheese during refrigerated storage were evaluated. The manufactured cheeses included the addition of starter culture (Lactococcus lactis subsp. lactis and

Lactococcus lactis subsp. cremoris - R-704) (CC); starter culture, L. acidophilus LA-05 and inulin (CLA); starter culture, B. lactis BB-12 and inulin (CBB); or starter culture, L. acidophilus LA-05, B. lactis BB-12 and inulin (CLB). In the synbiotic cheeses (CLA, CBB and CLB), the counts of L. acidophilus LA-05 and B. lactis BB-12 were greater than 6log CFU g-1, the amount of inulin was greater than 6 g per 100 g, and the firmness was reduced. The cheeses evaluated had high brightness values (L*), with a predominance of yellow (b*). CC had higher contents of proteins, lipids and minerals compared to the other cheeses. There was a decrease in the amount of short-chain fatty acids (SCFAs) and an increase of medium-chain (MCFAs) and long-chain fatty acids (LCFAs) in the synbiotic cheeses compared to CC. The amount of conjugated linoleic acid increased in CLA, CBB and CLB. The highest depth of proteolysis and the greatest changes in the release of free amino acids were found in CLB. The addition of inulin and probiotics, alone or in co-culture, did not affect the cheese acceptance. Inulin and probiotics can be used together for the production of creamy goat cheese without negatively affecting the general quality characteristics of the product, and to add value because of its synbiotic potential.

Barichella, M., Pacchetti, C., Bolliri, C., Cassani, E., Iorio, L., Pusani, C., Pinelli, G., Privitera, G., Cesari, I., Faierman, S. A., Caccialanza, R., Pezzoli, G., and Cereda, E. Probiotics and prebiotic fiber for constipation associated with Parkinson disease: An RCT. Neurology. 9-20-2016; 87(12):1274-1280; PM: 27543643

ABSTRACT: ***OBJECTIVES***: Our objective was to evaluate the efficacy of probiotics and prebiotics in patients with Parkinson disease (PD) and constipation. ***METHODS***: We conducted a tertiary setting, randomized, double-blind, placebo-controlled trial in patients with PD with Rome III-confirmed constipation based on 2-week stool diary data at baseline. Patients (n = 120) were randomly assigned (2:1) to either a fermented milk, containing multiple probiotic strains and prebiotic fiber, or placebo, once daily for 4 weeks. The primary efficacy endpoint was the increase in the number of complete bowel movements (CBMs) per week. The key secondary endpoints were 3 or more CBMs and an increase by one or more CBMs per week during weeks 3 and 4. ***RESULTS***: For the primary endpoint, the consumption of a fermented milk containing probiotics and prebiotics resulted in a higher increase in the number of CBMs (mean 1.2, 95% confidence interval [CI] 0.8-1.6) than placebo (0.1, 95% CI -0.4% to 0.6%) (mean difference 1.1, 95% CI 0.4-1.8; p = 0.002). For the key secondary endpoints, a higher number of patients in the probiotics-prebiotics group vs the placebo group reported 3 or more CBMs (p = 0.030; 58.8% vs 37.5%; odds ratio = 2.4, 95% CI 1.1-5.2) and an increase by one or more CBMs (p = 0.004; 53.8% vs 25.0%; odds ratio = 3.5, 95% CI 1.8-8.1) during weeks 3 and 4. ***CONCLUSIONS***: The consumption of a fermented milk containing multiple probiotic strains and prebiotic fiber was superior to placebo in improving constipation in patients with PD. ***CLINICALTRIALSGOV IDENTIFIER***: NCT02459717. ***CLASSIFICATION OF EVIDENCE***: This study provides Class I evidence that for patients with PD who have constipation, fermented milk containing probiotics and prebiotics increases the frequency of CBMs.

Basturk, A., Artan, R., and Yilmaz, A. Efficacy of synbiotic, probiotic, and prebiotic treatments for irritable bowel syndrome in children: A randomized controlled trial. Turk.J.Gastroenterol. 2016; 27(5):439-443; PM: 27782892

ABSTRACT: ***BACKGROUND/AIMS***: Irritable bowel syndrome (IBS) is an important health

problem that presents serious social burdens and high costs. Our study investigated the efficacy of synbiotic (Bifidobacterium lactis B94 with inulin), probiotic (B. lactis B94), and prebiotic (inulin) treatment for IBS in a pediatric age group. ***MATERIAL AND METHODS***: This study was randomized, double-blind, controlled, and prospective in design and included 71 children between the ages of 4 and 16 years who were diagnosed with IBS according to the Rome III criteria. The first group received synbiotic treatment [5x109 colony forming units (CFU) of B. lactis B94 and 900 mg inulin]; the second group received probiotic treatment (5x109 CFU B. lactis B94), and the third group received prebiotic treatment (900 mg inulin) twice daily for 4 weeks. ***RESULTS***: Probiotic treatment improved belching-abdominal fullness (p<0.001), bloating after meals (p=0.016), and constipation (p=0.031), and synbiotic treatment improved belching-abdominal fullness (p=<0.001), bloating after meals (p=0.004), constipation (p=0.021), and mucus in the feces (p=0.021). The synbiotic group had a significantly higher percentage of patients with full recovery than the prebiotic group (39.1% vs. 12.5%, p=0.036). ***CONCLUSION***: Administration of synbiotics and probiotics resulted in significant improvements in initial complaints when compared to prebiotics. Additionally, there was a significantly higher number of patients with full recovery from IBS symptoms in the synbiotic group than in the prebiotic group. Therefore, the twice daily administration of synbiotics is suggested for the treatment of children with IBS.

Baucells, B. J., Mercadal, Hally M., varez Sanchez, A. T., and Figueras, Aloy J. [Probiotic associations in the prevention of necrotising enterocolitis and the reduction of late-onset sepsis and neonatal mortality in preterm infants under 1,500g: A systematic review]. An.Pediatr.(Barc.). 2016; 85(5):247-255; PM: 26611880

ABSTRACT: ***INTRODUCTION AND OBJECTIVE***: Necrotising enterocolitis (NEC) is one of the most common and serious acquired bowel diseases a premature newborn can face. This meta-analysis was performed comparing different probiotic mixtures to ascertain their benefits as a routine tool for preventing necrotising enterocolitis and reducing late-onset sepsis and mortality in premature neonates of less than 1500g. ***MATERIAL AND METHODS***: A systematic review of randomised controlled trials, between January 1980 and March 2014, on MEDLINE, the Cochrane Central Register of Controlled Trials, together with EMBASE, was carried out. Studies with infants <1500g or <34 weeks were selected, discarding those with Jadad scores lower than 4. ***RESULTS***: 9 studies were selected for further investigation, pooling a total of 3521 newborns. Probiotics were found to reduce the NEC incidence (RR 0.39; 95%CI: 0.26-0.57) and mortality (RR 0.70; 95%CI: 0.52-0.93), with no difference to placebo regarding late-onset sepsis (RR 0.91; 95%CI: 0.78-1.06). Finally, when analysing the different strands, the use of a 2-probiotic combination (Lactobacillus acidophilus with Bifidobacterium bifidum) proved to be statistically significant in reducing all-cause mortality when compared to other probiotic combinations (RR 0.32; 95%CI: 0.15-0.66, NNT 20; 95%CI: 12-50). ***CONCLUSIONS***: Probiotics are a beneficial tool in the prevention of NEC and mortality in preterm neonates. Moreover, the combination of 2 probiotics (Lactobacillus acidophilus with Bifidobacterium bifidum) seems to produce the greatest benefits. However, due to the differences in probiotic components and administration, it would be wise to perform a randomised controlled trial comparing different probiotic mixtures.

Berry, E. D. and Wells, J. E. Reducing Foodborne Pathogen Persistence and Transmission in Animal Production Environments: Challenges and Opportunities. Microbiol.Spectr. 2016; 4(4); PM: 27726803

ABSTRACT: Preharvest strategies to reduce zoonotic pathogens in food animals are important components of the farm-to-table food safety continuum. The problem is complex; there are multiple pathogens of concern, multiple animal species under different production and management systems, and a variety of sources of pathogens, including other livestock and domestic animals, wild animals and birds, insects, water, and feed. Preharvest food safety research has identified a number of intervention strategies, including probiotics, direct-fed microbials, competitive exclusion cultures, vaccines, and bacteriophages, in addition to factors that can impact pathogens on-farm, such as seasonality, production systems, diet, and dietary additives. Moreover, this work has revealed both challenges and opportunities for reducing pathogens in food animals. Animals that shed high levels of pathogens and predominant pathogen strains that exhibit long-term persistence appear to play significant roles in maintaining the prevalence of pathogens in animals and their production environment. Continued investigation and advancements in sequencing and other technologies are expected to reveal the mechanisms that result in super-shedding and persistence, in addition to increasing the prospects for selection of pathogen-resistant food animals and understanding of the microbial ecology of the gastrointestinal tract with regard to zoonotic pathogen colonization. It is likely that this continued research will reveal other challenges, which may further indicate potential targets or critical control points for pathogen reduction in livestock. Additional benefits of the preharvest reduction of pathogens in food animals are the reduction of produce, water, and environmental contamination, and thereby lower risk for human illnesses linked to these sources.

Bhat, M., Arendt, B. M., Bhat, V., Renner, E. L., Humar, A., and Allard, J. P. Implication of the intestinal microbiome in complications of cirrhosis. World J.Hepatol. 9-28-2016; 8(27):1128-1136; PM: 27721918

ABSTRACT: The intestinal microbiome (IM) is altered in patients with cirrhosis, and emerging literature suggests that this impacts on the development of complications. The PubMed database was searched from January 2000 to May 2015 for studies and review articles on the composition, pathophysiologic effects and therapeutic modulation of the IM in cirrhosis. The following combination of relevant text words and MeSH terms were used, namely intestinal microbiome, microbiota, or dysbiosis, and cirrhosis, encephalopathy, spontaneous bacterial peritonitis, hepatorenal syndrome, variceal bleeding, hepatopulmonary syndrome, portopulmonary hypertension and hepatocellular carcinoma. The search results were evaluated for pertinence to the subject of IM and cirrhosis, as well as for quality of study design. The IM in cirrhosis is characterized by a decreased proportion of Bacteroides and Lactobacilli, and an increased proportion of Enterobacteriaceae compared to healthy controls. Except for alcoholic cirrhosis, the composition of the IM in cirrhosis is not affected by the etiology of the liver disease. The percentage of Enterobacteriaceae increases with worsening liver disease severity and decompensation and is associated with bacteremia, spontaneous bacterial peritonitis and hepatic encephalopathy. Lactulose, rifaximin and Lactobacillus-containing probiotics have been shown to partially reverse the cirrhosis associated enteric dysbiosis, in conjunction with improvement in encephalopathy. The IM is altered in cirrhosis, and this may contribute to the development of complications associated with end-stage liver disease. Therapies such as lactulose, rifaximin and probiotics may, at least partially, reverse the cirrhosis-associated changes in the IM. This, in turn, may prevent or alleviate the severity of complications.

Biggs, M. B., Medlock, G. L., Moutinho, T. J., Lees, H. J., Swann, J. R., Kolling, G. L., and Papin, J. A. Systems-level metabolism of the altered Schaedler flora, a complete gut microbiota. ISME.J. 11-8-2016; PM: 27824342

ABSTRACT: The altered Schaedler flora (ASF) is a model microbial community with both in vivo and in vitro relevance. Here we provide the first characterization of the ASF community in vitro, independent of a murine host. We compared the functional genetic content of the ASF to wild murine metagenomes and found that the ASF functionally represents wild microbiomes better than random consortia of similar taxonomic composition. We developed a chemically defined medium that supported growth of seven of the eight ASF members. To elucidate the metabolic capabilities of these ASF species-including potential for interactions such as cross-feeding-we performed a spent media screen and analyzed the results through dynamic growth measurements and non-targeted metabolic profiling. We found that cross-feeding is relatively rare (32 of 3570 possible cases), but is enriched between Clostridium ASF356 and Parabacteroides ASF519. We identified many cases of emergent metabolism (856 of 3570 possible cases). These data will inform efforts to understand ASF dynamics and spatial distribution in vivo, to design pre- and probiotics that modulate relative abundances of ASF members, and will be essential for validating computational models of ASF metabolism. Well-characterized, experimentally tractable microbial communities enable research that can translate into more effective microbiome-targeted therapies to improve human health.The ISME Journal advance online publication, 8 November 2016; doi:10.1038/ismej.2016.130.

Billimoria, Z. C., Pandya, S., Bhatt, P., and Pandya, B. Probiotics-To Use, or Not to Use? An Updated Meta-analysis. Clin.Pediatr.(Phila). 2016; 55(13):1242-1244; PM: 27609512

No Abstract Available.

Blajman, J. E., Astesana, D. M., Zimmermann, J. A., Rossler, E., Scharpen, A. R., Berisvil, A. P., Zbrun, M. V., Soto, L. P., Rosmini, M. R., and Frizzo, L. S. Quantification of FITC-labelled probiotic Lactobacillus salivarius DSPV 001P during gastrointestinal transit in broilers. Benef.Microbes. 12-1-2016; 1-10; PM: 27903091

ABSTRACT: The knowledge related to the fate of probiotics in the complex environment of the intestinal microbiota in broilers is just beginning to be elucidated; however, it is not yet well understood. A good method to investigate the mechanisms by which probiotics mediate their effects is to mark probiotic bacteria and trace them. The aim of this research was to develop a new method to estimate in vivo fluorescein isothiocyanate (FITC)-labelled Lactobacillus salivarius DSPV 001P counts during passage through the gastrointestinal tract (GIT) of broilers. Forty-five, 1 d old Cobb broilers were used in this trial. Programmed necropsies were performed 30 min, 6 h, and 12 h after the administration of the probiotic bacterium, and samples of liver, crop, duodenum, caecum, and bursa of fabricius were collected. To determine the spatial and temporal transit of L. salivarius DSPV 001P in broilers, the number of bacteria as well as its respective fluorescent signal produced by FITC were measured. In order to observe the relationship between the variables, a logistic regression analysis was applied. The amount of fluorescence could be used as an indicator of fluorescent probiotic bacteria in the crop and duodenum 30 min after probiotic bacterium supplementation. In addition, the

fluorescent signal could be used to estimate bacterial counts in caecum 6 and 12 h after L. salivarius DSPV 001P administration. To the best of our knowledge, this research is the first in vivo trial to employ the bacterial FITC-labelling technique in order to enumerate probiotic bacteria during gastrointestinal transit in broilers.

Bluemel, S., Williams, B., Knight, R., and Schnabl, B. Precision medicine in alcoholic and nonalcoholic fatty liver disease via modulating the gut microbiota. Am.J.Physiol Gastrointest.Liver Physiol. 12-1-2016; 311(6):G1018-G1036; PM: 27686615

ABSTRACT: Alcoholic liver disease (ALD) and nonalcoholic fatty liver disease (NAFLD) represent a major health burden in industrialized countries. Although alcohol abuse and nutrition play a central role in disease pathogenesis, preclinical models support a contribution of the gut microbiota to ALD and NAFLD. This review describes changes in the intestinal microbiota compositions related to ALD and NAFLD. Findings from in vitro, animal, and human studies are used to explain how intestinal pathology contributes to disease progression. This review summarizes the effects of untargeted microbiome modifications using antibiotics and probiotics on liver disease in animals and humans. While both affect humoral inflammation, regression of advanced liver disease or mortality has not been demonstrated. This review further describes products secreted by Lactobacillus- and microbiota-derived metabolites, such as fatty acids and antioxidants, that could be used for precision medicine in the treatment of liver disease. A better understanding of host-microbial interactions is allowing discovery of novel therapeutic targets in the gut microbiota, enabling new treatment options that restore the intestinal ecosystem precisely and influence liver disease. The modulation options of the gut microbiota and precision medicine employing the gut microbiota presented in this review have excellent prospects to improve treatment of liver disease.

Bonacina, J., Suarez, N., Hormigo, R., Fadda, S., Lechner, M., and Saavedra, L. A genomic view of food-related and probiotic Enterococcus strains. DNA Res. 10-23-2016; PM: 27773878

ABSTRACT: The study of enterococcal genomes has grown considerably in recent years. While special attention is paid to comparative genomic analysis among clinical relevant isolates, in this study we performed an exhaustive comparative analysis of enterococcal genomes of food origin and/or with potential to be used as probiotics. Beyond common genetic features, we especially aimed to identify those that are specific to enterococcal strains isolated from a certain food-related source as well as features present in a species-specific manner. Thus, the genome sequences of 25 Enterococcus strains, from 7 different species, were examined and compared. Their phylogenetic relationship was reconstructed based on orthologous proteins and whole genomes. Likewise, markers associated with a successful colonization (bacteriocin genes and genomic islands) and genome plasticity (phages and clustered regularly interspaced short palindromic repeats) were investigated for lifestyle specific genetic features. At the same time, a search for antibiotic resistance genes was carried out, since they are of big concern in the food industry. Finally, it was possible to locate 1617 FIGfam families as a core proteome universally present among the genera and to determine that most of the accessory genes code for hypothetical proteins, providing reasonable hints to support their functional characterization.

Bozzetto, S., Pirillo, P., Carraro, S., Berardi, M., Cesca, L., Stocchero, M., Giordano, G., Zanconato, S., and Baraldi, E. Metabolomic profile of children with recurrent respiratory infections. Pharmacol.Res. 11-22-2016; 115:162-167; PM: 27888158

ABSTRACT: Recurrent respiratory infections (RRI) represent a widespread condition which has a severe social and economic impact. Immunostimulants are used for their prevention. It is crucial to better characterize children with RRI to refine their diagnosis and identify effective personalized prevention strategies. Metabolomics is a high-dimensional biological method that can be used for hypothesis-free biomarker profiling, examining a large number of metabolites in a given sample using spectroscopic techniques. Multivariate statistical data analysis then enables us to infer which metabolic information is relevant to the biological characterization of a given physiological or pathological condition. This can lead to the emergence of new, sometimes unexpected metabolites, and hitherto unknown metabolic pathways, enabling the formulation of new pathogenetic hypotheses, and the identification of new therapeutic targets. The aim of our pilot study was to apply mass-spectrometry-based metabolomics to the analysis of urine samples from children with RRI, comparing these children's biochemical metabolic profiles with those of healthy peers. We also compared the RRI children's and healthy controls' metabolomic urinary profiles after the former had received pidotimod treatment for 3 months to see whether this immunostimulant was associated with biochemical changes in the RRI children's metabolic profile. 13 children (age range 3-6 yeas) with RRI and 15 matched per age healthy peers with no history of respiratory diseases or allergies were enrolled. Their metabolomic urine samples were compared before and after the RRI children had been treated with pidotimod for a period of 3 months. Metabolomic analyses on the urine samples were done using mass spectrometry combined with ultra-performance liquid chromatography (UPLC-MS). The resulting spectroscopic data then underwent multivariate statistical analysis and the most relevant variables characterizing the two groups were identified. Data modeling with post-transformation of PLS2-Discriminant Analysis (ptPLS2-DA) generated a robust model capable of discriminating the urine samples from children with RRI from those of healthy controls (R2=0.92,Q2CV7-fold=0.75, p-value<0.001). The dataset included 1502 time per mass variables, and 138 of them characterized the difference between the two groups. Thirty-five of these distinctive 138 variables persisted in the profiles of the children with RRI after pidotimod treatment. Metabolomics can discriminate children with RRI from healthy controls, suggesting that the former have a dysregulated metabolic profile. Among the variables characterizing children with RRI there are metabolites that may reflect the presence of a different microbiome. After pidotimod treatment, the metabolic profile of the children with RRI was no longer very different from that of the healthy controls, except for the persistence of some microbiome-related variables. We surmise that pidotimod partially "restores" the altered metabolic profile of children with RRI, without modifying the metabolites related to the composition of the gut microbiota. In the light of these results, we hypothesize a potential synergic effect of the combined use of immunostimulants and probiotics for the purpose of prevention in children with RRI.

Brabant, G. [Bacterial vaginosis and spontaneous preterm birth]. J.Gynecol.Obstet.Biol.Reprod.(Paris). 10-26-2016; PM: 27793493

ABSTRACT: ***OBJECTIVE***: To determine if bacterial vaginosis is a marker for risk of spontaneous preterm delivery and if its detection and treatment can reduce this risk. ***METHODS***: Consultation of the database Pubmed/Medline, Science Direct, and international guidelines of medical societies. ***RESULTS***: Bacterial vaginosis (BV) is a

dysbiosis resulting in an imbalance in the vaginal flora through the multiplication of anaerobic bacteria and jointly of a disappearance of well-known protective Lactobacilli. His diagnosis is based on clinical Amsel criteria and/or a Gram stain with establishment of the Nugent score. The prevalence of the BV extraordinarily varies according to ethnic and/or geographical origin (4-58 %), in France, it is close to 7 % in the first trimester of pregnancy (EL2). The link between BV and spontaneous premature delivery is low with an odds ratio between 1.5 and 2 in the most recent studies (EL3). Metronidazole or clindamycin is effective to treat BV (EL3). It is recommended to prescribe one of these antibiotics in the case of symptomatic BV (Professional Consensus). The testing associated with the treatment of BV in the global population showed no benefit in the prevention of the risk of spontaneous preterm delivery (EL2). Concerning low-risk asymptomatic population (defined by the absence of antecedent of premature delivery), it has been failed profit to track and treat the BV in the prevention of the risk of spontaneous preterm delivery (EL1). Concerning the high-risk population (defined by a history of preterm delivery), it has been failed profit to track and treat the VB in the prevention of the risk of spontaneous preterm delivery (EL3). However, in the sub population of patients with a history of preterm delivery occurred in a context of materno-fetal bacterial infection, there may be a benefit to detect and treat early and systematically genital infection, and in particular the BV (Professional Consensus). ***CONCLUSION***: The screening and treatment of BV during pregnancy in asymptomatic low-risk population is not recommended in the prevention of the risk of spontaneous preterm delivery (grade A). In the population at high risk with the only notion of antecedent of premature delivery, screening and treatment of the BV is not recommended (grade C).

Bryan, P. F., Karla, C., Edgar Alejandro, M. T., Sara Elva, E. P., Gemma, F., and Luz, C. Sphingolipids as Mediators in the Crosstalk between Microbiota and Intestinal Cells: Implications for Inflammatory Bowel Disease. Mediators.Inflamm. 2016; 2016:9890141; PM: 27656050

ABSTRACT: Inflammatory bowel disease (IBD) describes different illnesses characterized by chronic inflammation of the gastrointestinal tract. Although the pathogenic mechanisms leading to IBD are poorly understood, immune system disturbances likely underlie its development. Sphingolipids (SLs) have been identified as important players and promising therapeutic targets to control inflammation in IBD. Interestingly, it seems that microorganisms of the normal gut microbiota and probiotics are involved in sphingolipid function. However, there is a great need to investigate the role of SLs as intermediates in the crosstalk between intestinal immunity and microorganisms. This review focuses on recent investigations that describe some mechanisms involved in the regulation of cytokine profiles by SLs. We also describe the importance of gut microbiota in providing signaling molecules that favor the communication between resident bacteria and intestinal cells. This, in turn, modulates the immune response in the bowel and likely in other peripheral organs. The potential of SLs and gut microbiota as targets or therapeutic agents for IBD is also discussed.

Bustamante, M., Oomah, B. D., Rubilar, M., and Shene, C. Effective Lactobacillus plantarum and Bifidobacterium infantis encapsulation with chia seed (Salvia hispanica L.) and flaxseed (Linum usitatissimum L.) mucilage and soluble protein by spray drying. Food Chem. 2-1-2017; 216:97-105; PM: 27596397

ABSTRACT: Mucilage (M) and soluble protein (SP) extracted from chia seed and flaxseed were used as encapsulating material for two probiotic bacteria: Bifidobacterium infantis and Lactobacillus plantarum by spray drying. Probiotic survival and viability after spray drying and during storage were evaluated. B. infantis and L. plantarum displayed high survival (98%) after encapsulation with mixtures of maltodextrin (MD) combined with M and SP from flaxseed (MD:FM.

Byun, S. J., Lim, T. J., Lim, Y. J., Seo, J. G., and Chung, M. J. In vivo effects of s-pantoprazole, polaprenzinc, and probiotic blend on chronic small intestinal injury induced by indomethacin. Benef.Microbes. 11-30-2016; 7(5):731-737; PM: 27824272

ABSTRACT: Treatment and prevention methods for non-steroidal anti-inflammatory drug-induced enteropathy have not yet been established. We tested the preventive effects of s-pantoprazole sodium trihydrate (PAN), polaprezinc (PZ), and probiotics on an indomethacin (Indo)-induced small intestinal injury in a rat model. Rats were randomised into 6 groups to receive: normal saline (control), Indo (6 mg/kg), PZ plus Indo, PAN plus Indo, or probiotics plus Indo (at 108 and 109 cfu/head) for 2 weeks. We measured body weight, food intake, severity of small intestinal damage, haemoglobin (Hb) levels in the small intestinal fluid, intestinal inflammatory cytokines, and a few groups of faecal bacteria. The experimental groups were found to have the following survival rates: 0% for the Indo, PZ, and PAN groups; 50% for both probiotic groups; and 100% for control. Treatment with probiotics of different concentrations reduced small intestinal lesion scores and intestinal fluid Hb as compared with the Indo group, while these parameters did not reduce in the PZ and PAN groups. The anti-inflammatory marker interleukin 10 increased in both probiotic groups. Analysis of a few groups of faecal bacteria revealed that Indo-induced a significant increase in Gram-negative bacteria and decreases in Bifidobacterium and Lactobacillus. Similar changes were also observed in the PZ and PAN groups. However, opposite effects were found in both probiotic groups. The use of probiotics appeared to be beneficial in preventing Indo-induced chronic small intestinal injury.

Caffarelli, C., Santamaria, F., Di, Mauro D., Mastrorilli, C., Mirra, V., and Bernasconi, S. Progress in pediatrics in 2015: choices in allergy, endocrinology, gastroenterology, genetics, haematology, infectious diseases, neonatology, nephrology, neurology, nutrition, oncology and pulmonology. Ital.J.Pediatr. 8-27-2016; 42(1):75; PM: 27566421

ABSTRACT: This review focuses key advances in different pediatric fields that were published in Italian Journal of Pediatrics and in international journals in 2015. Weaning studies continue to show promise for preventing food allergy. New diagnostic tools are available for identifying the allergic origin of allergic-like symptoms. Advances have been reported in obesity, short stature and autoimmune endocrine disorders. New molecules are offered to reduce weight gain and insulin-resistance in obese children. Regional investigations may provide suggestions for preventing short stature. Epidemiological studies have evidenced the high incidence of Graves' disease and Hashimoto's thyroiditis in patients with Down syndrome. Documentation of novel risk factors for celiac disease are of use to develop strategies for prevention in the

population at-risk. Diagnostic criteria for non-celiac gluten sensitivity have been reported. Negative effect on nervous system development of the supernumerary X chromosome in Klinefelter syndrome has emerged. Improvements have been made in understanding rare diseases such as Rubinstein-Taybi syndrome. Eltrombopag is an effective therapy for immune trombocytopenia. Children with sickle-cell anemia are at risk for nocturnal enuresis. Invasive diseases caused by Streptococcus pyogenes are still common despite of vaccination. No difference in frequency of antibiotic prescriptions for acute otitis media between before the publication of the national guideline and after has been found. The importance of timing of iron administration in low birth weight infants, the effect of probiotics for preventing necrotising enterocolitis and perspectives for managing jaundice and cholestasis in neonates have been highlighted. New strategies have been developed to reduce the risk for relapse in nephrotic syndrome including prednisolone during upper respiratory infection. Insights into the pathophysiology of cerebral palsy, arterial ischemic stroke and acute encephalitis may drive advances in treatment. Recommendations on breastfeeding and complementary feeding have been updated. Novel treatments for rhabdomyosarcoma should be considered for paediatric patients. Control of risk factors for bronchiolitis and administration of pavilizumab for preventing respiratory syncytial virus infection may reduce hospitalization. Identification of risk factors for hospitalization in children with wheezing can improve the management of this disease. Deletions or mutations in genes encoding proteins for surfactant function may cause diffuse lung disease.

Callewaert, C., Lambert, J., and Van de, Wiele T. Towards a bacterial treatment for armpit malodour. Exp.Dermatol. 11-28-2016; PM: 27892611

ABSTRACT: Axillary malodour is a frustrating condition for many people. It can lead to significant discomforts and various psychological effects. The underarm microbiome generally plays a major role in axillary malodour formation. Not only the bacteria on the armpit epidermis, but especially those living in the sweat glands, sweat pores and hair follicles play a pivotal role in malodour development. In order to treat underarm malodour, this viewpoint paper envisions a bacterial treatment. Replacing the autochthonous malodour causing microbiome with a non-odour causing microbiome, through an armpit bacterial transplantation or direct application of probiotics/non-odour causing bacteria could resolve the condition. Selective steering of the microbiome with prebiotics, biochemicals or plant extracts could likewise greatly help in improving the underarm odour. Elimination/inhibition of the 'bad bugs' and application/stimulation of the 'good bugs' should be part of the future treatment for axillary body odour. This article is protected by copyright. All rights reserved.

Camilleri, M., Park, S. Y., Scarpato, E., and Staiano, A. Exploring hypotheses and rationale for causes of infantile colic. Neurogastroenterol.Motil. 9-20-2016; PM: 27647578

*ABSTRACT: **BACKGROUND***: Infantile colic is a frequent problem in neonates and infants. This review addresses current management including the results for nutrient modifications; soy-based formulas; and prebiotics, probiotics, and synbiotics. ***PURPOSE***: Given the evidence that there is still an unmet clinical need, as current treatments are incompletely efficacious, we have examined the evidence around three hypothetical mechanisms that could potentially be involved in etiopathogenesis of infantile colic: immaturity of bile acid mechanisms that alter intraluminal and absorptive mechanisms, immaturity in motility and alterations in the microbiome. Understanding these potential

mechanisms may lead to the introduction of diagnostic procedures that should enhance the selection or individualization of therapy for infantile colic.

Campanini, A., De, Conto U., Cavasin, F., Bastiani, F., Camarotto, A., Gardini, L., Geremia, A., Marastoni, C., Missorini, C., Quarantelli, E., Sassi, U., Scarabello, F., Dal, Bo N., Ricco, M., Grillo, S., Landi, S., and Di, Mario F. A Primary-Care Interventional Model on the Diverticular Disease: Searching for the Optimal Therapeutic Schedule. J.Clin.Gastroenterol. 2016; 50 Suppl 1:S93-S96; PM: 27622379

ABSTRACT: ***INTRODUCTION***: In routine colonoscopy, diverticulosis is the most commonly found feature, but only a minority of these cases show symptoms of diverticular disease.From June 2014 to December 2014, we enrolled prospectively 178 patients affected by symptomatic uncomplicated diverticular disease (Male/Female=0.47, mean age 71.7+/-11.5 y, range 41 to 95 y) from 15 General Pratictioners patient files. All patients were symptomatic; in all cases, diagnosis was been confirmed by a colonoscopy performed at least 1 year before. Patients with acute diverticulitis were excluded.On the basis of the predominant symptoms (abdominal complaints or constipation), patients were addressed to 4 different therapeutic approaches using mesalamine, rifaximine, probiotics (in a consortium of different species of Lactobacillus and Bifidobacterium), and fibers (Plantago Ovata Husk). All treatments lasted 3 months. ***RESULTS***: Sixty-three patients were enrolled in group A (rifaximine), 43 in group A1 (rifaximine+fibers+probiotics), 23 in group B (mesalamine), and 31 in group B1 (mesalamine+fibers).Analysis of variance suggested a statistically significant difference (P<0.003) among groups at the end of the observation period, with Groups A1 and B1 showing a higher number of bowel movement per week. Global linear measurement confirmed the role of treatment as a significant factor (F=2.858; P=0.039) associated with body mass index (F=6.972; P<0.009). ***CONCLUSIONS***: In accordance with the baseline clinical presentation, the supplementation of fiber and/or probiotics is associated with a statistically significant improvement in the clinical pattern of symptoms in patients with diverticular disease in a primary-care/family physician setting.

Campbell, A. W. Inflammation and a Solution: Probiotics. Altern.Ther.Health Med. 2016; 22(5):8-10 PM: 27622954

No Abstract Available.

Cardile, S., Alterio, T., Arrigo, T., and Salpietro, C. Role of prebiotics and probiotics in pediatric diseases. Minerva Pediatr. 2016; 68(6):487-497; PM: 25926157

ABSTRACT: The increasing knowledge about the composition and activities of the microflora has shown the close link between the bacteria and the health of the human organism. For this reason it has focused attention on the possibility of modulating the gut flora. The use of probiotics and prebiotics has increased enormously in recent years, more for real beneficial effects demonstrated in patients than for their safety profiles. However, it is recorded an indiscriminate use also in conditions in which there are no scientific evidence to support. Their use in case of immunocompromised patients or with severe and debilitating chronic diseases should be very prudent, because of the risk of complications including sepsis. The use of a probiotic cannot ignore the knowledge of the genus and species of the strain and in pediatric patients has been demonstrated

their role for treating acute viral gastroenteritis and preventing antibiotic-associated diarrhea in healthy children. Moreover probiotics are considered as an option for recurrent and relapsing antibiotic sensitive pouchitis and in select patients with mild ulcerative colitis. Further studies are needed to evaluate clinical conditions that may require their use and to define the optimal doses and intake durations.

Carter, G. M., Esmaeili, A., Shah, H., Indyk, D., Johnson, M., Andreae, M., and Sacks, H. S. Probiotics in Human Immunodeficiency Virus Infection: A Systematic Review and Evidence Synthesis of Benefits and Risks. Open.Forum Infect.Dis. 2016; 3(4):ofw164; PM: 27747250

ABSTRACT: People living with human immunodeficiency virus frequently use dietary supplements, including probiotics, but concern exists about ingesting live organisms. We performed a systematic review of the benefits of probiotics and a meta-analysis of sepsis risk. We undertook a protocol-driven, comprehensive review to identify all relevant studies, assess their quality, and summarize the evidence. Of 2068 references, 27 were analyzed. The data suggest possible benefits for CD4 count, recurrence or management of bacterial vaginosis, and diarrhea management. We examined randomized, controlled studies explicitly assessing sepsis in any patient population, and we found zero cases of supplement-associated bacteremia or fungemia in 39 randomized controlled trials comprising 9402 subjects. The estimated number needed to harm is 7369 in Bayesian approach (95% credible interval: 1689, infinity), which should reassure clinicians. No or mild adverse effects were reported. Longer duration studies investigating different individual and mixed strains for plausible indications are needed to establish best practices.

Casado Munoz, Mdel C., Benomar, N., Lavilla, Lerma L., Knapp, C. W., Galvez, A., and Abriouel, H. Biocide tolerance, phenotypic and molecular response of lactic acid bacteria isolated from naturally-fermented Alorena table to different physico-chemical stresses. Food Microbiol. 2016; 60:1-12; PM: 27554140

ABSTRACT: Lactic acid bacteria (LAB) isolated throughout the fermentation process of Alorena table olives were found to be resistant at least to three antibiotics (Casado Munoz et al., 2014); however, most were sensitive to the biocides tested in this study (with minimum inhibitory concentrations [MIC] below the epidemiological cut-off values). 2-15% of the isolates were found to be biocide resistant: Leuconostoc Pseudomesenteroides, which were resistant to hexachlorophene, and Lactobacillus pentosus to cetrimide and hexadecylpiridinium. We analyzed the effect of different physico-chemical stresses, including antimicrobials, on the phenotypic and genotypic responses of LAB, providing new insights on how they become resistant in a changing environment. Results indicated that similar phenotypic responses were obtained under three stress conditions: antimicrobials, chemicals and UV light. Susceptibility patterns to antibiotics changed: increasing MICs for ampicillin, chloramphenicol, ciprofloxacin, teicoplanin and tetracycline, and decreasing the MICs for clindamycin, erythromycin, streptomycin and trimethoprim in most strains. Statistically, cross resistance between different antibiotics was detected in all stress conditions. However, expression profiles of selected genes involved in stress/resistance response (rpsL, recA, uvrB and srtA) differed depending on the stress parameter, LAB species and strain, and the target gene. We conclude that, despite the uniform phenotypic response to stresses, the repertoire of induced and repressed genes differs. So, a search for a target to improve stress

tolerance of LAB, especially those of importance as starter/protective cultures or probiotics, may depend on the individual screening of each strain, even though we could predict the antibiotic phenotypic response to all stresses.

Cassard, L., Lalanne, A. I., Garault, P., Cotillard, A., Chervaux, C., Wels, M., Smokvina, T., Daeron, M., and Bourdet-Sicard, R. Individual strains of Lactobacillus paracasei differentially inhibit human basophil and mouse mast cell activation. Immun.Inflamm.Dis. 2016; 4(3):289-299; PM: 27621812

ABSTRACT: ***INTRODUCTION***: The microbiota controls a variety of biological functions, including immunity, and alterations of the microbiota in early life are associated with a higher risk of developing allergies later in life. Several probiotic bacteria, and particularly lactic acid bacteria, were described to reduce both the induction of allergic responses and allergic manifestations. Although specific probiotic strains were used in these studies, their protective effects on allergic responses also might be common for all lactobacilli. ***METHODS***: To determine whether allergic effector cells inhibition is a common feature of lactobacilli or whether it varies among lactobacilli strains, we compared the ability of 40 strains of the same Lactobacillus paracasei species to inhibit IgE-dependent mouse mast cell and human basophil activation. ***RESULTS***: We uncovered a marked heterogeneity in the inhibitory properties of the 40 Lactobacillus strains tested. These segregated into three to four clusters depending on the intensity of inhibition. Some strains inhibited both mouse mast cell and human basophil activation, others strains inhibited only one cell type and another group induced no inhibition of activation for either cell type. ***CONCLUSIONS***: Individual Lactobacillus strains of the same species differentially inhibit IgE-dependent activation of mouse mast cells and human basophils, two cell types that are critical in the onset of allergic manifestations. Although we failed to identify specific bacterial genes associated with inhibition by gene-trait matching analysis, our findings demonstrate the complexity of the interactions between the microbiota and the host. These results suggest that some L. paracasei strains might be more beneficial in allergies than others strains and provide the bases for a rational screening of lactic acid bacteria strains as next-generation probiotics in the field of allergy.

Cepeda, M. S., Katz, E. G., and Blacketer, C. Microbiome-Gut-Brain Axis: Probiotics and Their Association With Depression. J.Neuropsychiatry Clin.Neurosci. 8-19-2016; appineuropsych15120410; PM: 27539375

ABSTRACT: To assess the association of probiotics with depression, a large population-based cross-sectional study was conducted. National Health and Nutrition Examination Survey adult participants from 2005 through 2012 were included. Exposure was defined as having consumed any probiotic food or supplement on any of the interview days. Subjects were classified as depressed if Patient Health Questionnaire scores were >/=10. Of the 18,019 subjects included, 14.11% consumed probiotics. Unadjusted analysis suggested that subjects who consumed probiotics had lower odds of depression (OR=0.58, 95% CI=0.45-0.75). After adjustment for characteristics associated with depression and probiotic exposure, the effect was attenuated (OR=0.82, 95% CI=0.61-1.1) and no longer significant. Use of probiotics is not associated with lower rates of depression in this national sample.

Chan, Y. K., Brar, M. S., Kirjavainen, P. V., Chen, Y., Peng, J., Li, D., Leung, F. C., and El-Nezami, H. High fat diet induced atherosclerosis is accompanied with low colonic bacterial diversity and altered abundances that correlates with plaque size, plasma A-FABP and cholesterol: a pilot study of high fat diet and its intervention with Lactobacillus rhamnosus GG (LGG) or telmisartan in ApoE-/- mice. BMC.Microbiol. 11-8-2016; 16(1):264; PM: 27821063

ABSTRACT: ***BACKGROUND***: Atherosclerosis appears to have multifactorial causes - microbial component like lipopolysaccharides (LPS) and other pathogen associated molecular patterns may be plausible factors. The gut microbiota is an ample source of such stimulants, and its dependent metabolites and altered gut metagenome has been an established link to atherosclerosis. In this exploratory pilot study, we aimed to elucidate whether microbial intervention with probiotics L. rhamnosus GG (LGG) or pharmaceuticals telmisartan (TLM) could improve atherosclerosis in a gut microbiota associated manner. ***METHODS***: Atherosclerotic phenotype was established by 12 weeks feeding of high fat (HF) diet as opposed to normal chow diet (ND) in apolipoprotein E knockout (ApoE-/-) mice. LGG or TLM supplementation to HF diet was studied. ***RESULTS***: Both LGG and TLM significantly reduced atherosclerotic plaque size and improved various biomarkers including endotoxin to different extents. Colonial microbiota analysis revealed that TLM restored HF diet induced increase in Firmicutes/Bacteroidetes ratio and decrease in alpha diversity; and led to a more distinct microbial clustering closer to ND in PCoA plot. Eubacteria, Anaeroplasma, Roseburia, Oscillospira and Dehalobacteria appeared to be protective against atherosclerosis and showed significant negative correlation with atherosclerotic plaque size and plasma adipocyte - fatty acid binding protein (A-FABP) and cholesterol. ***CONCLUSION***: LGG and TLM improved atherosclerosis with TLM having a more distinct alteration in the colonic gut microbiota. Altered bacteria genera and reduced alpha diversity had significant correlations to atherosclerotic plaque size, plasma A-FABP and cholesterol. Future studies on such bacterial functional influence in lipid metabolism will be warranted.

Chan, Y. K., El-Nezami, H., Chen, Y., Kinnunen, K., and Kirjavainen, P. V. Probiotic mixture VSL#3 reduce high fat diet induced vascular inflammation and atherosclerosis in ApoE(-/-) mice. AMB.Express. 2016; 6(1):61; PM: 27576894

ABSTRACT: Atherosclerosis results from chronic inflammation potentially caused by translocation of bacterial components from the oro-gastrointestinal tract to circulation. Specific probiotics have anti-inflammatory effects and may reduce bacterial translocation. We thereby tested whether a probiotic mixture with documented anti-inflammatory potential could reduce atherosclerosis. ApoE(-/-) mice were fed high fat diet alone or with VSL#3 or a positive control treatment, telmisartan or both for 12 weeks. All treatments reduced atherosclerotic plaques significantly compared to high fat diet alone. VSL#3 significantly reduced proinflammatory adhesion molecules and risk factors of plaque rupture, reduced vascular inflammation and atherosclerosis to a comparable extent to telmisartan; and VSL#3 treated mice had the most distinctly different intestinal microbiota composition from the control groups. Combining the VSL#3 and telmisartan brought no further benefits. Our findings showed the therapeutic potential of VSL#3 in reducing atherosclerosis and vascular inflammation.

Cheifetz, A. S., Gianotti, R., Luber, R., and Gibson, P. R. Complementary and Alternative Medicines Used by Patients With Inflammatory Bowel Diseases. Gastroenterology. 10-12-2016; PM: 27743873

ABSTRACT: Patients and physicians often have many questions regarding the role of complementary and alternative medicines (CAMs), or non-allopathic therapies, for inflammatory bowel diseases (IBD). CAMs of various forms are used by more than half of patients with IBD during some point in their disease course. We summarize the available evidence for the most commonly used and discussed CAMs. We discuss evidence for the effects of herbs (such as cannabis and curcumin), probiotics, acupuncture, exercise, and mind-body therapy. There have been few controlled studies of these therapies, which have been limited by their small sample sizes; most studies have been uncontrolled. In addition, there has been a lack of quality control for herbal preparations. It has been a challenge to design rigorous randomized placebo controlled trials, due in part to problems of adequate blinding for psychological interventions, acupuncture, and exercise. These barriers have limited the acceptance of CAM by physicians. However, such therapies might be used to supplement conventional therapies and help ease patient symptoms. We conclude that physicians should understand the nature of and evidence for CAMs for IBD, so that rational advice can be offered to patients who inquire about their use. CAMs have the potential to aid in treatment of IBD, but further research is needed to validate these approaches.

Chelliah, R., Ramakrishnan, S. R., Prabhu, P. R., and Antony, U. Evaluation of antimicrobial activity and probiotic properties of wild-strain Pichia kudriavzevii isolated from frozen idli batter. Yeast. 2016; 33(8):385-401; PM: 27370793

ABSTRACT: The present research was undertaken to study the probiotic characteristics of Pichia kudriavzevii isolated from frozen idli batter. Polymerase chain reaction amplification with 18S rRNA primers confirmed Pichia kudriavzevii, a xylose-utilizing probiotic strain. It was resistant to physiological concentrations of bile salts, pepsin and pancreatic enzyme. It also showed efficient auto-aggregation as well as co-aggregation ability with four commercial probiotic yeasts and exhibited good hydrophobicity in xylene and toluene. The strain inhibited the growth of 13 enteropathogens and showed a commensal relationship with four commercial probiotic yeast and bacteria. Moreover, it was resistant to 30 antibiotics with different modes of action. The yeast exhibited thermotolerance up to 95 degrees C for 2 h. The cell-free supernatants were also found to be heat stable, indicating the presence of thermostable secondary metabolites. Hence it could be exploited as starter culture, co-culture or probiotic in the preparation of fermented products or incorporated in heatable foods as well. Copyright (c) 2016 John Wiley & Sons, Ltd.

Chen, J., Bechman, A., Klu, Y. A., and Phillips, R. D. Survival of Probiotics in Hypromellose Capsules with Rice or Potato Maltodextrin Excipient. J.Food Sci. 9-28-2016; PM: 27681165

ABSTRACT: There is currently no authorized or established therapeutic level/dose of probiotics for proposed health benefits; however, a daily probiotic consumption of 108 to 1010 CFU has been recommended. This study determined the survival of 5 individual probiotic strains, Lactobacillus rhamnosus, Lactobacillus paracasei, Lactobacillus

plantarum, Lactobacillus acidophilus, and Bifidobacterium lactis, along with a mixture of the 5 strains in hypromellose capsules with rice or potato maltodextrin at 4, 25, and 37 degrees C for 12 mo. Samples were collected monthly and plated on deMan-Rogosa Sharpe agar with 0.05% l-cysteine hydrochloride. Results showed that samples stored at 4 degrees C had an average count of 108 to 1011 CFU/g of probiotic cells during the 12 mo period, whereas at 25 degrees C, L. rhamnosus and L. paracasei had an average counts below 108 CFU/g during the storage period. L. rhamnosus was the most vulnerable strain used in this study, having the least viable counts at all 3 storage temperatures. Probiotics stored in rice maltodextrin, on average, had higher probiotic counts compared to those stored in potato maltodextrin. Study suggests that to provide consumers with 108 to 1010 CFU/d of probiotic cells, robust bacterial strains, suitable carriers, and a storage temperature of 4 degrees C are required.

Chen, L., Xu, K., Gui, Q., Chen, Y., Chen, D., and Yang, Y. Probiotic pre-administration reduces mortality in a mouse model of cecal ligation and puncture-induced sepsis. Exp.Ther.Med. 2016; 12(3):1836-1842; PM: 27588102

ABSTRACT: A number of clinical trials have demonstrated that the use of probiotics has the potential to prevent nosocomial infections. However, the mechanism underlying probiotic-induced anti-infection and sepsis remains to be investigated. In the present study, 200 microl/day of Lactobacillus rhamnosus GG (LGG) or normal saline (control) was orally administrated to 4-week-old C57BL6 mice 4 weeks prior to cecal ligation and puncture (CLP). A number of mice were sacrificed 24 h after CLP, and the remaining mice were used for survival studies. Ileum tissues were collected to evaluate the injury on the intestine. Blood samples were also obtained to investigate the changed metabolic pattern in mice that underwent different treatments using ultra-performance liquid chromatography coupled with quadrupole time-of-flight mass spectrometry (UPLC-QTOF-MS). In the survival studies, the mortality of CLP-induced septic mice pretreated with LGG was significantly lower compared with untreated mice (P=0.029). Ileum mucosal damage was evident in the control septic mice. Based on the data of UPLC-QTOF-MS, phosphatidylcholines were increased and lysophosphatidylcholines (LPCs) that contained polyunsaturated fatty acids were decreased in septic mice, whereas saturated fatty acid LPCs reveal no significant difference between septic and sham mice. In addition, the metabolic profile in the septic mice pretreated with LGG was much closer to that of sham mice compared with control septic mice. The results of the present study suggest that probiotic pre-administration reduces the mortality in septic mice by decreasing ileum mucosal damage, increasing the gut barrier integrity and altering global serum metabolic profiles.

Chen, S., Sanderson, M. W., Lee, C., Cernicchiaro, N., Renter, D. G., and Lanzas, C. Basic reproduction number and transmission dynamics of common serogroups of enterohemorrhagic Escherichia coli. Appl.Environ.Microbiol. 7-8-2016; PM: 27401976

ABSTRACT: Understanding pathogens' transmission dynamics are essential to determine the epidemiology, ecology and ways of controlling enterohemorrhagic Escherichia coli (EHEC) in the animals and their environment. Our objective was to estimate the epidemiological fitness of common EHEC strains in cattle populations. For that purpose, we developed a Markov chain model to characterize the dynamics of 7 serogroups of enterohemorrhagic Escherichia coli (O26, O45, O103, O111, O121, O145, and O157) in cattle production environments based on a set of cross-sectional data on infection

prevalence in two years at two states. The basic reproduction number (R0) was estimated using a Bayesian framework for each serogroup based on two criteria (using serogroup alone: the O-group data; and using O serogroup, Shiga toxin gene(s), and intimin (eae) gene together: the EHEC data). In addition, correlations between external covariates (e.g., location, ambient temperature, dietary, and probiotics usage) and prevalence/R0 were quantified. R0 estimates varied substantially among different EHEC serogroups, with EHEC O157 having R0 >1 (approximately 1.5) and all other six EHEC serogroups less than 1. Using the O-group data substantially increased R0 estimates for O26, O45, and O103 serogroups (R0 >1), but not for others. Different covariates had distinct influences on different serogroups: the coefficients for each covariate were different among serogroups. Our modeling and analysis of this system can be readily expanded to other pathogen systems in order to estimate the pathogen and external factors that influence spread of infectious agents. IMPORTANCE AND HIGHLIGHTS: In this paper we describe a Bayesian modeling framework to estimate basic reproduction numbers of multiple serotypes of Shiga toxin-producing Escherichia coli respectively according to a cross-sectional study.We then couple compartmental model to reconstruct infection dynamics of these serotypes and quantify their risk in the population.We incorporate different sensitivity levels of detecting different serotypes and evaluate their potential influence on the estimation of basic reproduction numbers.

Chiodelli, G., Pellizzoni, M., Ruzickova, G., and Lucini, L. Effect of Different Aloe Fractions on the Growth of Lactic Acid Bacteria. J.Food Sci. 11-25-2016; PM: 27886374

ABSTRACT: Several foods on the market, such as yogurt and fermented milk, include mixtures of prebiotics and probiotic microorganisms effective in promoting the proliferation and equilibrium of intestinal bacteria, thus improving gut health. Particularly, researchers and the public have shown increasing interest in the combination of probiotics with natural substances that promote health or that can act as substrates to promote bacterial growth. The aim of this study is to evaluate the effects of different extracts of Aloe barbadensis and Aloe arborescens in fermented milk, taking into account both the prebiotic effect of aloe polysaccharides and the antimicrobial activity of several secondary metabolites. The results demonstrate a beneficial effect of 5% aloe inner gel on Lactobacillus growth and confirm the antimicrobial activity of the phenolic compounds peculiar of green rind extracts.

Chowdhury, T., Ali, M. M., Hossain, M. M., Singh, J., Yousuf, A. N., Yasmin, F., and Chowdhury, F. R. Efficacy of Probiotics Versus Placebo in the Prevention of Necrotizing Enterocolitis in Preterm Very Low Birth Weight Infants: A Double-Blind Randomized Controlled Trial. J.Coll.Physicians Surg.Pak. 2016; 26(9):770-774; PM: 27671183

ABSTRACT: ***OBJECTIVE***: To evaluate the efficacy of orally administered probiotics in preventing necrotizing enterocolitis (NEC) in preterm very low birth weight (VLBW) infants. ***STUDY DESIGN***: Arandomized double blind controlled trial. ***PLACE AND DURATION OF STUDY***: The Paediatrics Department of Sylhet M.A.G. Osmani Medical College Hospital, Sylhet Bangladesh, from July 2012 to June 2015. ***METHODOLOGY***: Preterm (28 - 33 weeks gestation) VLBW (birth weight 1000 - 1499 g) neonates were enrolled. The study group was fed with probiotics once daily with breast milk from first feeding, and the control group with only breast milk without the addition of probiotics. Both the groups received other standard care. The primary outcome was the development of NEC (stage II and III), categorized by modified Bell's classification. **RESULTS**: In 108 neonates, development

of NEC was significantly lower in the study group than that of control group [1 (1.9%) vs. 6 (11.5%); p=0.044]. Age of achievement of full oral feeding was significantly earlier in the study group than that in the control group (14.88 ±3.15 and 18.80 ±4.32 days; p < 0.001). Duration of hospital stay was significantly short in the study group compared to the control group (15.82 ±2.94 days vs. 19.57 ±4.26 days; p < 0.001). **CONCLUSION**: Probiotic supplementation reduces the frequency of necrotising enterocolitis in preterm neonates with very low birth weight. It is also associated with faster achievement of full oral feeding and short duration of hospital stay.

Cicenia, A., Santangelo, F., Gambardella, L., Pallotta, L., Iebba, V., Scirocco, A., Marignani, M., Tellan, G., Carabotti, M., Corazziari, E. S., Schippa, S., and Severi, C. Protective Role of Postbiotic Mediators Secreted by Lactobacillus rhamnosus GG Versus Lipopolysaccharide-induced Damage in Human Colonic Smooth Muscle Cells. J.Clin.Gastroenterol. 2016; 50 Suppl 2, Proceedings from the 8th Probiotics, Prebiotics & New Foods for Microbiota and Human Health meeting held in Rome, Italy on September 13-15, 2015:S140-S144; PM: 27741159

ABSTRACT: ***BACKGROUND***: Some beneficial effects of probiotics may be due to secreted probiotic-derived factors, identified as "postbiotic" mediators. The aim of this study was to evaluate whether supernatants harvested from Lactobacillus rhamnosus GG (LGG) cultures (ATCC53103 strain) protect colonic human smooth muscle cells (HSMCs) from lipopolysaccharide (LPS)-induced myogenic damage. ***MATERIALS AND METHODS***: LGG was grown in de Man, Rogosa, Share medium at 37 degrees C and samples were collected in middle and late exponential, stationary, and overnight phases. Supernatants were recovered by centrifugation, filtered, and stored at -20 degrees C. The primary HSMCs culture was exposed for 24 hours to purified LPS of a pathogen strain of Escherichia coli (O111:B4) (1 mug/mL) with and without supernatants. Postbiotic effects were evaluated on the basis of HSMCs morphofunctional alterations and interleukin-6 (IL-6) production. Data are expressed as mean+/-SE (P<0.05 significant). ***RESULTS***: LPS induced persistent, significant, 20.5%+/-0.7% cell shortening and 34.5%+/-2.2% decrease in acetylcholine-induced contraction of human HSMCs. These morphofunctional alterations were paralleled to a 365.65%+/-203.13% increase in IL-6 production. All these effects were dose-dependently reduced by LGG supernatants. Supernatants of the middle exponential phase already partially restored LPS-induced cell shortening by 57.34%+/-12.7% and IL-6 increase by 145.8%+/-4.3% but had no effect on LPS-induced inhibition of contraction. Maximal protective effects were obtained with supernatants of the late stationary phase with LPS-induced cell shortening restored by 84.1%+/-4.7%, inhibition of contraction by 85.5%+/-6.4%, and IL-6 basal production by 92.7%+/-1.2%. ***CONCLUSIONS***: LGG-derived products are able to protect human SMCs from LPS-induced myogenic damage. Novel insights have been provided for the possibility that LGG-derived products could reduce the risk of progression to postinfective motor disorders.

Cicero, A. F. and Colletti, A. Combinations of phytomedicines with different lipid lowering activity for dyslipidemia management: The available clinical data. Phytomedicine. 10-15-2016; 23(11):1113-1118; PM: 26621556

ABSTRACT: ***BACKGROUND***: Cardiovascular diseases are the primary cause of death and the leading cause of disability in industrialized countries. Dyslipidemia is a major independent and reversible risk factor for these diseases: it is estimated that a reduction

of 1 mmol/l (38 mg/dl) of LDL cholesterol is associated with a risk of developing a cardiovascular complication reduced by 25%, a reduction potentially achieved by life-style improvement associated to adequate dietary supplementation with bioactive substances. ***AIM***: The aim of this review is to focus on the major phytochemical nutraceuticals combinations supported by clinical trials that have demonstrated positive effects in the treatment of dyslipidemia. ***MAIN TEXT***: There are many nutraceuticals with significant lipid-lowering properties: most of them are used in association with a low dosage, because that permits to reduce the risk of side effects and theoretically to improve efficacy. In fact, natural products with different synergetic lipid-lowering could be combined: they can reduce the absorption of lipids from the bowel and/or increase their excretion (soluble fibers, plant sterols, probiotics), enhance the hepatic uptake of cholesterol (berberine, soybean proteins), inhibit Hydroxy-Methil-Gglutaryl Coenzyme A reductase enzyme and consequently the hepatic synthesis of cholesterol (monacolins, policosanols, allicin, soybean proteins, bergamot); furthermore some products are able to reduce the oxidation of the LDL and increase the thermogenesis and lipid metabolism (chlorogenic acid). ***CONCLUSION***: Rational combinations of nutraceuticals with different lipid-lowering activities, whether associated with an appropriate lifestyle, should provide an alternative to drug treatment in patients in primary cardiovascular disease prevention with mildly added cardiovascular risk and in some statin-intolerant patients.

Cigarran, Guldris S., Gonzalez, Parra E., and Cases, Amenos A. Gut microbiota in chronic kidney disease. Nefrologia. 8-20-2016; PM: 27553986

ABSTRACT: The intestinal microflora maintains a symbiotic relationship with the host under normal conditions, but its imbalance has recently been associated with several diseases. In chronic kidney disease (CKD), dysbiotic intestinal microflora has been reported with an increase in pathogenic flora compared to symbiotic flora. An enhanced permeability of the intestinal barrier, allowing the passage of endotoxins and other bacterial products to the blood, has also been shown in CKD. By fermenting undigested products that reach the colon, the intestinal microflora produce indoles, phenols and amines, among others, that are absorbed by the host, accumulate in CKD and have harmful effects on the body. These gut-derived uraemic toxins and the increased permeability of the intestinal barrier in CKD have been associated with increased inflammation and oxidative stress and have been involved in various CKD-related complications, including cardiovascular disease, anaemia, mineral metabolism disorders or the progression of CKD. The use of prebiotics, probiotics or synbiotics, among other approaches, could improve the dysbiosis and/or the increased permeability of the intestinal barrier in CKD. This article describes the situation of the intestinal microflora in CKD, the alteration of the intestinal barrier and its clinical consequences, the harmful effects of intestinal flora-derived uraemic toxins, and possible therapeutic options to improve this dysbiosis and reduce CKD-related complications.

Cinque, B., La, Torre C., Lombardi, F., Palumbo, P., Van der, Rest M., and Cifone, M. G. Production Conditions Affect the In Vitro Anti-Tumoral Effects of a High Concentration Multi-Strain Probiotic Preparation. PLoS.One. 2016; 11(9):e0163216; PM: 27657913

ABSTRACT: A careful selection of the probiotic agent, standardization of the dose and detailed characterization of the beneficial effects are essential when considering use of a probiotic for the dietary management of serious diseases. However, changes in the manufacturing processes, equipment or facilities can result in differences in the product

itself due to the live nature of probiotics. The need to reconfirm safety and/or efficacy for any probiotic product made at a different factory is therefore mandatory. Recently, under the brand VSL#3(R), a formulation produced by a manufacturer different from the previous one, has been commercialized in some European countries (the UK and Holland). VSL#3 is a high concentration multi-strain preparation which has been recognized by the main Gastroenterology Associations for the dietary management of pouchitis as well as ulcerative colitis. We have compared the "original" VSL#3 produced in USA with the "newfound" VSL#3 produced in Italy. According to our results, the "newfound" VSL#3 has 130-150% more "dead bacteria" compared to the "original" product, raising concerns for the well-known association between dead microbes with adverse effects. The abilities of bacterial lysates from the two formulations to influence in vitro viability and proliferation of different tumor cell lines also resulted different. The repair of previously scratched monolayers of various adherent tumor cell lines (i.e. HT1080, and Caco-2 cells) was inhibited more significantly by the "original" VSL#3 when compared to the "newfound" VSL#3. Tumor cell cycle profile, in particular cell cycle arrest and apoptotic death of the cancer cells, further confirms that the "original" VSL#3 has a better functional profile than the "newfound" VSL#3, at least in in vitro. Our data stress the importance of the production conditions for the "newfound" VSL#3 considering that this product is intended to be used for the dietary management of patients with very serious diseases, such as chronic inflammatory bowel diseases.

Clark, A. and Mach, N. Exercise-induced stress behavior, gut-microbiota-brain axis and diet: a systematic review for athletes. J.Int.Soc.Sports Nutr. 2016; 13:43; PM: 27924137

ABSTRACT: Fatigue, mood disturbances, under performance and gastrointestinal distress are common among athletes during training and competition. The psychosocial and physical demands during intense exercise can initiate a stress response activating the sympathetic-adrenomedullary and hypothalamus-pituitary-adrenal (HPA) axes, resulting in the release of stress and catabolic hormones, inflammatory cytokines and microbial molecules. The gut is home to trillions of microorganisms that have fundamental roles in many aspects of human biology, including metabolism, endocrine, neuronal and immune function. The gut microbiome and its influence on host behavior, intestinal barrier and immune function are believed to be a critical aspect of the brain-gut axis. Recent evidence in murine models shows that there is a high correlation between physical and emotional stress during exercise and changes in gastrointestinal microbiota composition. For instance, induced exercise-stress decreased cecal levels of Turicibacter spp and increased Ruminococcus gnavus, which have well defined roles in intestinal mucus degradation and immune function. Diet is known to dramatically modulate the composition of the gut microbiota. Due to the considerable complexity of stress responses in elite athletes (from leaky gut to increased catabolism and depression), defining standard diet regimes is difficult. However, some preliminary experimental data obtained from studies using probiotics and prebiotics studies show some interesting results, indicating that the microbiota acts like an endocrine organ (e.g. secreting serotonin, dopamine or other neurotransmitters) and may control the HPA axis in athletes. What is troubling is that dietary recommendations for elite athletes are primarily based on a low consumption of plant polysaccharides, which is associated with reduced microbiota diversity and functionality (e.g. less synthesis of byproducts such as short chain fatty acids and neurotransmitters). As more elite athletes suffer from psychological and gastrointestinal conditions that can be linked to the gut, targeting the microbiota therapeutically may need to be incorporated in athletes' diets that take into consideration dietary fiber as well as microbial taxa not currently present in athlete's gut.

Coghetto, C. C., Vasconcelos, C. B., Brinques, G. B., and Ayub, M. A. Lactobacillus plantarum BL011 cultivation in industrial isolated soybean protein acid residue. Braz.J.Microbiol. 2016; 47(4):941-948; PM: 27522926

ABSTRACT: In this study, physiological aspects of Lactobacillus plantarum BL011 growing in a new, all-animal free medium in bioreactors were evaluated aiming at the production of this important lactic acid bacterium. Cultivations were performed in submerged batch bioreactors using the Plackett-Burman methodology to evaluate the influence of temperature, aeration rate and stirring speed as well as the concentrations of liquid acid protein residue of soybean, soy peptone, corn steep liquor, and raw yeast extract. The results showed that all variables, except for corn steep liquor, significantly influenced biomass production. The best condition was applied to bioreactor cultures, which produced a maximal biomass of 17.87gL-1, whereas lactic acid, the most important lactic acid bacteria metabolite, peaked at 37.59gL-1, corresponding to a productivity of 1.46gL-1h-1. This is the first report on the use of liquid acid protein residue of soybean medium for L. plantarum growth. These results support the industrial use of this system as an alternative to produce probiotics without animal-derived ingredients to obtain high biomass concentrations in batch bioreactors.

Consoli, M. L., da Silva, R. S., Nicoli, J. R., Bruna-Romero, O., da Silva, R. G., de Vasconcelos, Generoso S., and Correia, M. I. Randomized Clinical Trial: Impact of Oral Administration of Saccharomyces boulardii on Gene Expression of Intestinal Cytokines in Patients Undergoing Colon Resection. JPEN J.Parenter.Enteral Nutr. 2016; 40(8):1114-1121; PM: 25917895

ABSTRACT: ***BACKGROUND***: When intestinal microbiota is imbalanced, a patient becomes more vulnerable to infectious complications; intervention with beneficial probiotics may help lower risk for infection. The aim of this study was to measure levels of inflammatory cytokine messenger RNA (mRNA) in surgical samples of intestinal mucosal tissues from patients who were given the probiotic Saccharomyces boulardii before undergoing colon surgery. **METHODS**: Thirty-three patients undergoing colon resection were randomly assigned to receive at least 7-day preoperative probiotic treatment (n = 15) or conventional (n = 18) treatment. Probiotic treatment consisted of oral lyophilized S boulardii Cytokine mRNA levels (interleukin [IL]-10, IL-1beta, IL-23A, tumor necrosis factor [TNF]-alpha, IL-12B, interferon-gamma [INF-gamma], and IL-17A) were measured in samples obtained during the operation. Postoperative infections were also assessed. **RESULTS**: Patients who received probiotics had significantly lower mucosal IL-1beta, IL-10, and IL-23A mRNA levels than the control group (P = .001, P = .04, and P = .03, respectively). However, mRNA expression of other cytokines did not differ between the 2 groups (P > .05). The incidence of postoperative infectious complications was 13.3% and 38.8% in probiotic and control groups, respectively (P > .05). There was no perioperative mortality in either group. The mean total length of hospital stay was similar between the groups (P > .05). **CONCLUSIONS**: Probiotic treatment with S boulardii downregulates both pro- and anti-inflammatory cytokines in the intestinal colonic mucosa with no statistical impact on postoperative infection rates.

Cordonnier, C., Thevenot, J., Etienne-Mesmin, L., Alric, M., Livrelli, V., and Blanquet-Diot, S. Probiotic and enterohemorrhagic Escherichia coli: An effective strategy against a deadly enemy? Crit Rev.Microbiol. 11-1-2016; 1-17. PM: 27798976

ABSTRACT: Enterohemorrhagic Escherichia coli (EHEC) are major food-borne pathogens that constitute a serious public health threat. Currently, there is no specific treatment available for EHEC infections in human creating an urgent need for the development of alternative therapeutic strategies. Among them, one of the most promising approaches is the use of probiotic microorganisms. Even if many studies have shown the antagonistic effects of probiotic bacteria or yeast on EHEC survival, virulence, adhesion on intestinal epithelium or pathogen-induced inflammatory responses, mechanisms mediating their beneficial effects remain unclear. This review describes EHEC pathogenesis and novel therapeutic strategies, with a particular emphasis on probiotics. The interests and limits of a probiotic-based approach and the way it might be incorporated into global health strategies against EHEC infections will be discussed.

Costantini, E., Giannitsas, K., and Illiano, E. The role of nonantibiotic treatment of community-acquired urinary tract infections. Curr.Opin.Urol. 11-24-2016; PM: 27898456

ABSTRACT: ***PURPOSE OF REVIEW***: To summarize evidence on the efficacy of nonantibiotic treatments in the prevention of recurrent urinary tract infections (UTIs). The need for antibiotic-free regimens seems to be imperative given the worldwide rates of resistance of uropathogens to available antibiotics. ***RECENT FINDINGS***: In the recently published literature, cranberry products and probiotics are the focus of research. They both seem to be effective in preventing recurrent UTIs compared with placebo, but their benefit becomes less clear when they are compared with antibiotics. ***SUMMARY***: A number of nonantibiotic-containing alternatives are available for the prevention of recurrent UTIs. For the majority of these alternatives, contradictory results have been published. These can be, at least partially, explained by variation in doses administered and duration of treatment. Selection of patients more likely to benefit from these interventions seems to be a realistic approach from a clinical-practice perspective, as well a worthwhile focus for future research.

Costeloe, K., Bowler, U., Brocklehurst, P., Hardy, P., Heal, P., Juszczak, E., King, A., Panton, N., Stacey, F., Whiley, A., Wilks, M., and Millar, M. R. A randomised controlled trial of the probiotic Bifidobacterium breve BBG-001 in preterm babies to prevent sepsis, necrotising enterocolitis and death: the Probiotics in Preterm infantS (PiPS) trial. Health Technol.Assess. 2016; 20(66):1-194; PM: 27594381

ABSTRACT: ***BACKGROUND***: Necrotising enterocolitis (NEC) and late-onset sepsis remain important causes of death and morbidity in preterm babies. Probiotic administration might strengthen intestinal barrier function and provide protection; this is supported by published meta-analyses, but there is a lack of large well-designed trials. ***OBJECTIVE***: To test the use of the probiotic Bifidobacterium breve strain BBG-001 to prevent NEC, late-onset sepsis and death in preterm babies while monitoring probiotic colonisation of participants. ***DESIGN***: Double-blind, randomised, placebo-controlled trial. ***SETTING***: Recruitment was carried out in 24 hospitals, and the randomisation programme used a minimisation algorithm. Parents, clinicians and outcome assessors were blinded to the allocation. ***PARTICIPANTS***: Babies born between 23 and 30 weeks' gestation and randomised within 48 hours of birth. Exclusions included life-threatening or any gastrointestinal malformation detected within 48 hours of birth and no realistic chance of

survival. ***INTERVENTIONS***: Active intervention: 1 ml of B. breve BBG-001 in one-eighth-strength infant formula Neocate((R)) (Nutricia Ltd, Trowbridge, UK), (6.7 x 10(7) to 6.7 x 10(9) colony-forming units) per dose administered enterally. Placebo: 1 ml of one-eighth-strength infant formula Neocate. Started as soon as practicable and continued daily until 36 weeks' postmenstrual age. ***MAIN OUTCOME MEASURES***: Primary outcomes were an episode of bloodstream infection, with any organism other than a skin commensal, in any baby between 72 hours and 46 weeks' postmenstrual age; an episode of NEC Bell stage >/= 2 in any baby; and death before discharge from hospital. Secondary outcomes included stool colonisation with B. breve. *RESULTS*: In total, 654 babies were allocated to receive probiotic and 661 to receive placebo over 37 months from July 2010. Five babies were withdrawn; 650 babies from the probiotic group and 660 from the placebo group were included in the primary analysis. Baseline characteristics were well balanced. There was no evidence of benefit for the primary outcomes {sepsis: 11.2% vs. 11.7% [adjusted relative risk (RR) 0.97, 95% confidence interval (CI) 0.73 to 1.29]; NEC Bell stage >/= 2: 9.4% vs. 10.0% [adjusted RR 0.93, 95% CI 0.68 to 1.27]; and death: 8.3% vs. 8.5% [adjusted RR 0.93, 95% CI 0.67 to 1.30]}. B. breve colonisation status was available for 1186 (94%) survivors at 2 weeks' postnatal age, of whom 724 (61%) were positive: 85% of the probiotic group and 37% of the placebo group. There were no differences for subgroup analyses by minimisation criteria and by stool colonisation with B. breve at 2 weeks. No harms associated with the interventions were reported. ***LIMITATIONS***: Cross-colonisation of the placebo arm could have reduced statistical power and confounded results; analyses suggest that this did not happen. ***CONCLUSIONS***: This is the largest trial to date of a probiotic intervention. It shows no evidence of benefit and does not support routine use of probiotics for preterm infants. ***FUTURE WORK RECOMMENDATIONS***: The increasing understanding of the pathogenesis of NEC and sepsis will inform the choice of probiotics for testing and better define the target population. Future Phase III trials should incorporate monitoring of the quality and viability of the intervention and colonisation rates of participants; cluster design should be considered. ***TRIAL REGISTRATION***: Current Controlled Trials ISRCTN05511098 and EudraCT 2006-003445-17. ***FUNDING***: This project was funded by the National Institute for Health Research (NIHR) Health Technology Assessment programme and will be published in full in Health Technology Assessment; Vol. 20, No. 66. See the NIHR Journals Library website for further project information.

Crepin, V. F., Collins, J. W., Habibzay, M., and Frankel, G. Citrobacter rodentium mouse model of bacterial infection. Nat.Protoc. 2016; 11(10):1851-1876; PM: 27606775

ABSTRACT: Infection of mice with Citrobacter rodentium is a robust model to study bacterial pathogenesis, mucosal immunology, the health benefits of probiotics and the role of the microbiota during infection. C. rodentium was first isolated by Barthold from an outbreak of mouse diarrhea in Yale University in 1972 and was 'rediscovered' by Falkow and Schauer in 1993. Since then the use of the model has proliferated, and it is now the gold standard for studying virulence of the closely related human pathogens enteropathogenic and enterohemorrhagic Escherichia coli (EPEC and EHEC, respectively). Here we provide a detailed protocol for various applications of the model, including bacterial growth, site-directed mutagenesis, mouse inoculation (from cultured cells and after cohabitation), monitoring of bacterial colonization, tissue extraction and analysis, immune responses, probiotic treatment and microbiota analysis. The main protocol, from mouse infection to clearance and analysis of tissues and host responses, takes approximately 5 weeks to complete.

Culligan, E. P. and Sleator, R. D. Advances in the Microbiome: Applications to Clostridium difficile Infection. J.Clin.Med. 9-21-2016; 5(9); PM: 27657145

ABSTRACT: Clostridium difficile is a major cause of morbidity and mortality worldwide, causing over 400,000 infections and approximately 29,000 deaths in the United States alone each year. C. difficile is the most common cause of nosocomial diarrhoea in the developed world, and, in recent years, the emergence of hyper-virulent (mainly ribotypes 027 and 078, sometimes characterised by increased toxin production), epidemic strains and an increase in the number of community-acquired infections has caused further concern. Antibiotic therapy with metronidazole, vancomycin or fidaxomicin is the primary treatment for C. difficile infection (CDI). However, CDI is unique, in that, antibiotic use is also a major risk factor for acquiring CDI or recurrent CDI due to disruption of the normal gut microbiota. Therefore, there is an urgent need for alternative, non-antibiotic therapeutics to treat or prevent CDI. Here, we review a number of such potential treatments which have emerged from advances in the field of microbiome research.

Curro, D., Ianiro, G., Pecere, S., Bibbo, S., and Cammarota, G. Probiotics, fibre and herbal medicinal products for functional and inflammatory bowel disorders. Br.J.Pharmacol. 10-3-2016; PM: 27696378

ABSTRACT: Functional bowel disorders (FBD), mainly irritable bowel syndrome (IBS) and functional constipation (FC, also called chronic idiopathic constipation), are very common worldwide. Inflammatory bowel disease (IBD), including ulcerative colitis and Crohn's disease, although less common, has a strong impact on patients' quality of life, as well as being highly expensive for our healthcare. A definite cure for those disorders is still yet to come. Over the years, several therapeutic approaches complementary or alternative to traditional pharmacological treatments, including probiotics, prebiotics, synbiotics, fibre and herbal medicinal products, have been investigated for the management of both groups of diseases. However, most available studies are biased by several drawbacks, including small samples and poor methodological quality. Probiotics, in particular Saccharomyces boulardii and Lactobacilli (among which Lactobacillus rhamnosus), synbiotics, psyllium, and some herbal medicinal products, primarily peppermint oil, seem to be effective in ameliorating IBS symptoms. Synbiotics and fibre seem to be beneficial in FC patients. The probiotic combination VSL#3 may be effective in inducing remission in patients with mild-to-moderate ulcerative colitis, in whom Escherichia coli Nissle 1917 seems to be as effective as mesalamine in maintaining remission. No definite conclusions can be drawn as to the efficacy of fibre and herbal medicinal products in IBD patients due to the low number of studies and the lack of randomized controlled trials that replicate the results obtained in the individual studies conducted so far. Thus, further, well-designed studies are needed to address the real role of these therapeutic options in the management of both FBD and IBD.

Czaja, A. J. Factoring the intestinal microbiome into the pathogenesis of autoimmune hepatitis. World J.Gastroenterol. 11-14-2016; 22(42):9257-9278; PM: 27895415

ABSTRACT: The intestinal microbiome is a reservoir of microbial antigens and activated immune cells. The aims of this review were to describe the role of the intestinal microbiome in generating innate and adaptive immune responses, indicate how these

responses contribute to the development of systemic immune-mediated diseases, and encourage investigations that improve the understanding and management of autoimmune hepatitis. Alterations in the composition of the intestinal microflora (dysbiosis) can disrupt intestinal and systemic immune tolerances for commensal bacteria. Toll-like receptors within the intestine can recognize microbe-associated molecular patterns and shape subsets of T helper lymphocytes that may cross-react with host antigens (molecular mimicry). Activated gut-derived lymphocytes can migrate to lymph nodes, and gut-derived microbial antigens can translocate to extra-intestinal sites. Inflammasomes can form within hepatocytes and hepatic stellate cells, and they can drive the pro-inflammatory, immune-mediated, and fibrotic responses. Diet, designer probiotics, vitamin supplements, re-colonization methods, antibiotics, drugs that decrease intestinal permeability, and molecular interventions that block signaling pathways may emerge as adjunctive regimens that complement conventional immunosuppressive management. In conclusion, investigations of the intestinal microbiome are warranted in autoimmune hepatitis and promise to clarify pathogenic mechanisms and suggest alternative management strategies.

Daliri, E. B., Lee, B. H., and Oh, D. H. Current Perspectives on Antihypertensive Probiotics. Probiotics.Antimicrob.Proteins. 11-29-2016; PM: 27900619

ABSTRACT: Hypertension is a major risk factor for cardiovascular diseases. Optimizing blood pressure results in an overall health outcome. Over the years, the gut microbiota has been found to play a significant role in host metabolic processes, immunity, and physiology. Dietary strategies have therefore become a target for restoring disturbed gut microbiota to treat metabolic diseases. Probiotics and their fermented products have been shown in many studies to lower blood pressure by suppressing nitrogen oxide production in microphages, reducing reactive oxygen species, and enhancing dietary calcium absorption. Other studies have shown that hypertension could be caused by many factors including hypercholesterolemia, chronic inflammation, and inconsistent modulation of the renin-angiotensin system. This review discusses the antihypertensive roles of probiotics and their fermented products via the reduction of serum cholesterol levels, anti-inflammation, and inhibition of angiotensin-converting enzyme. The ability of recombinant probiotics to reduce high blood pressure has also been discussed.

Darbaky, Y., Evrard, B., Patrier, S., Falenta, J., Garcin, S., Tridon, A., Dapoigny, M., Silberberg, C., Nivoliez, A., and Diop, L. Oral probiotic treatment of Lactobacillus rhamnosus Lcr35(R) prevents visceral hypersensitivity to a colonic inflammation and an acute psychological stress. J.Appl.Microbiol. 10-8-2016; PM: 27718511

*ABSTRACT: **AIMS***: This study evaluated the efficacy of a repeated oral treatment with two active pharmaceutical ingredients (Lcr Lenio(R) and Lcr Restituo(R)) derivated from the probiotic bacterial strain Lactobacillus rhamnosus Lcr35(R) in two animal models mimicking different features of irritable bowel syndrome (IBS). IBS is characterized by visceral pain associated with alteration of bowel transit. IBS patients present visceral hypersensitivity with peripheral and central origins. ***METHODS AND RESULTS***: The injection of 2,4,6-trinitrobenzenesulfonic acid (TNBS) into the proximal colon as well as an acute partial restraint stress (PRS) produces colonic hypersensitivity measured in conscious rats by a decrease in pain threshold in response to distal colonic distension. Visceral hypersensitivity was produced by injection of TNBS 7 days before colonic distension or by acute PRS on testing day. Treatments were performed once a day during eight

consecutive days. ***CONCLUSIONS***: This study indicates that an 8-day probiotic treatment (Lcr Lenio and Lcr Restituo) produces an antihypersensitivity activity in both TNBS and PRS visceral pain models. As this probiotic strain attenuates peripherally and centrally induced visceral hypersensitivity in rats, it may be active in treatment of IBS symptoms. An immunomodulatory effect of the probiotics was highlighted in the TNBS model on the IL-23 secretion, suggesting a mechanism of action involving a regulation of the local IL-23/Th17 immune activation. ***SIGNIFICANCE AND IMPACT OF THE STUDY***: Two formulas of Lcr35(R) probiotic strain show very encouraging results for the treatment of IBS patients. Further studies are needed to better understand the role and mechanisms of probiotics on the pathogenesis of IBS.

Dardmeh, F., Nielsen, H. I., Alipour, H., Kjaergaard, B., Brandsborg, E., and Gazerani, P. Potential Nociceptive Regulatory Effect of Probiotic Lactobacillus rhamnosus PB01 (DSM 14870) on Mechanical Sensitivity in Diet-Induced Obesity Model. Pain Res.Manag. 2016; 2016:5080438; PM: 27647980

ABSTRACT: Treatments for obesity have been shown to reduce pain secondary to weight loss. Intestinal microbiota, as an endogenous factor, influences obesity and pain sensitivity but the effect of oral probiotic supplementation on musculoskeletal pain perception has not been studied systematically. The present study examined the effect of a single daily oral dose (1 x 10(9) CFU) of probiotics (Lactobacillus rhamnosus PB01, DSM14870) supplement on mechanical pain thresholds in behaving diet-induced obese (DIO) mice and their normal weight (NW) controls. The mice (N = 24, 6-week-old male) were randomly divided into four groups on either standard or high fat diet with and without probiotic supplementation. Both DIO and NW groups with probiotic supplementation maintained an insignificant weight gain while the control groups gained significant weight ($P < 0.05$). Similarly, both DIO and NW probiotics supplemented groups demonstrated a significantly ($P < 0.05$) lower sensitivity to mechanical stimulation compared to their corresponding control. The results of this study suggest a protective effect of probiotics on nociception circuits, which propose a direct result of the weight reduction or an indirect result of anti-inflammatory properties of the probiotics. Deciphering the exact underlying mechanism of the weight loss and lowering nociception effect of the probiotic applied in this study require further investigation.

Dasari, S., Kathera, C., Janardhan, A., Praveen, Kumar A., and Viswanath, B. Surfacing role of probiotics in cancer prophylaxis and therapy: A systematic review. Clin.Nutr. 11-24-2016; PM: 27923508

ABSTRACT: Cancers figure among the most important causes of morbidity and mortality worldwide. Cancer and its associated infections are always complicated even when specific cancer regimens are available. It is well proved that Lactobacillus and other probiotic bacteria can modulate-ameliorate specific mechanisms against various infections including cancers. The present systematic review is intended to focus on the 'cellular and molecular mechanisms' of probiotic bacteria in the prevention and treatment of various cancers. The clinical and experimental findings of various studies explain the mechanisms such as apoptosis, antioxidant activity, immune response and epigenetics and illustrate the role of probiotics in cancer management and prophylaxis. In addition, the present review also discusses the safety aspects of probiotics when they are used in therapeutic and nutritional diet management. However, further investigations

are required to reveal the effectiveness of probiotics in cancer treatment in clinical settings.

Daskalakis, G. J. and Karambelas, A. K. Vaginal Probiotic Administration in the Management of Preterm Premature Rupture of Membranes. Fetal Diagn.Ther. 10-15-2016; PM: 27744438

ABSTRACT: ***OBJECTIVE***: To examine the influence of vaginal probiotic administration as an adjunct to standard antibiotic treatment on perinatal outcome in women with preterm premature rupture of membranes (PPROM). ***MATERIALS AND METHODS***: This was a prospective randomized trial of cases with PPROM (24-34 weeks) that were admitted to our department between 2011 and 2015. Forty-nine cases received vaginal probiotics for 10 days in combination with antibiotic prophylaxis and were compared to 57 others that received only antibiotics for the same time period. ***RESULTS***: The mean gestational age at birth (35.49 vs. 32.53 weeks), the mean duration of the latency period (5.60 vs. 2.48 weeks), and the mean birth weight (2,439.08 vs. 2,004.81 g) were significantly higher in the study group in comparison to the controls. Moreover, the neonates of the study group had a lower chance to enter the neonatal intensive care unit or the neonatal special care unit, shorter total hospitalization time, and lower need for oxygen administration and mechanical ventilation, as well as lower length of oxygen administration. ***CONCLUSIONS***: Vaginal probiotics as an adjunct to antibiotic prophylaxis in women with PPROM prolonged the latency period and improved the perinatal outcome.

Davis, C. D. The Gut Microbiome and Its Role in Obesity. Nutr.Today. 2016; 51(4):167-174; PM: 27795585

ABSTRACT: The human body is host to a vast number of microbes, including bacterial, fungal and protozoal microoganisms, which together constitute our microbiota. Evidence is emerging that the intestinal microbiome is intrinsically linked with overall health, including obesity risk. Obesity and obesity-related metabolic disorders are characterized by specific alterations in the composition and function of the human gut microbiome. Mechanistic studies have indicated that the gastrointestinal microbiota can influence both sides of the energy balance equation; namely, as a factor influencing energy utilization from the diet and as a factor that influences host genes that regulate energy expenditure and storage. Moreover, its composition is not fixed and can be influenced by several dietary components. This fact raises the attractive possibility that manipulating the gut microbiota could facilitate weight loss or prevent obesity in humans. Emerging as possible strategies for obesity prevention and/or treatment are targeting the microbiota, in order to restore or modulate its composition through the consumption of live bacteria (probiotics), nondigestible or limited digestible food constituents such as oligosaccharides (prebiotics), or both (synbiotics), or even fecal transplants.

Davis, D. J., Doerr, H. M., Grzelak, A. K., Busi, S. B., Jasarevic, E., Ericsson, A. C., and Bryda, E. C. Lactobacillus plantarum attenuates anxiety-related behavior and protects against stress-induced dysbiosis in adult zebrafish. Sci.Rep. 9-19-2016; 6:33726; PM: 27641717

ABSTRACT: The consumption of probiotics has become increasingly popular as a means to try to improve health and well-being. Not only are probiotics considered beneficial to

digestive health, but increasing evidence suggests direct and indirect interactions between gut microbiota (GM) and the central nervous system (CNS). Here, adult zebrafish were supplemented with Lactobacillus plantarum to determine the effects of probiotic treatment on structural and functional changes of the GM, as well as host neurological and behavioral changes. L. plantarum administration altered the beta-diversity of the GM while leaving the major core architecture intact. These minor structural changes were accompanied by significant enrichment of several predicted metabolic pathways. In addition to GM modifications, L. plantarum treatment also significantly reduced anxiety-related behavior and altered GABAergic and serotonergic signaling in the brain. Lastly, L. plantarum supplementation provided protection against stress-induced dysbiosis of the GM. These results underscore the influence commensal microbes have on physiological function in the host, and demonstrate bidirectional communication between the GM and the host.

Dawood, M. A., Koshio, S., Ishikawa, M., El-Sabagh, M., Yokoyama, S., Wang, W. L., Yukun, Z., and Olivier, A. Physiological response, blood chemistry profile and mucus secretion of red sea bream (Pagrus major) fed diets supplemented with Lactobacillus rhamnosus under low salinity stress. Fish.Physiol Biochem. 8-19-2016; PM: 27542150

ABSTRACT: Environmental stressors caused by inadequate aquaculture management strategies suppress the immune response of fish and make them more susceptible to diseases. Therefore, efforts have been made to relieve stress in fish by using various functional feed additives in the diet, including probiotics. The present work evaluates the effects of Lactobacillus rhamnosus (LR) on physiological stress response, blood chemistry and mucus secretion of red sea bream (Pagrus major) under low salinity stress. Fish were fed four diets supplemented with LR at [0 (LR0), 1 x 102 (LR1), 1 x 104 (LR2) and 1 x 106 (LR3) cells g-1] for 56 days. Before stress, blood cortisol, urea nitrogen (BUN) and total bilirubin (T-BIL) showed no significant difference ($P > 0.05$), whereas plasma glucose and triglyceride (TG) of fish-fed LR2 and LR3 diets were significantly lower ($P < 0.05$) than those of the other groups. Plasma total cholesterol (T-CHO) of fish-fed LR3 diet was significantly ($P < 0.05$) lower than that of the other groups. Furthermore, total plasma protein, mucus myeloperoxidase activity and the amount of mucus secretion were significantly enhanced in LR-supplemented groups when compared with the control group ($P < 0.05$). After the application of the low salinity stress test, plasma cortisol, glucose, T-CHO and TG contents in all groups showed an increased trend significantly ($P < 0.01$) compared to the fish before the stress challenge. However, plasma total protein and the amount of secreted mucus showed a decreased trend in all groups. On the other hand, BUN, T-BIL and mucus myeloperoxidase activity showed no significant difference after exposure to the low salinity stress ($P > 0.05$). In addition, the fish that received LR-supplemented diets showed significantly higher tolerance against low salinity stress than the fish-fed LR-free diet ($P < 0.05$). The physiological status and the detected immune responses, including total plasma protein and mucus myeloperoxidase activity in red sea bream, will provide a more comprehensive outlook of the effects of probiotics to relieve stress in fish.

Dawood, M. A., Koshio, S., Ishikawa, M., El-Sabagh, M., Esteban, M. A., and Zaineldin, A. I. Probiotics as an environment-friendly approach to enhance red sea bream, Pagrus major growth, immune response and oxidative status. Fish.Shellfish.Immunol. 2016; 57:170-178; PM: 27542618

ABSTRACT: A usual strategy in modern aquaculture to combat production bottlenecks

associated with intensification is preventive health care through the use of consumer and environment-friendly alternatives including probiotics. The current study evaluates the influence of Lactobacillus rhamnosus (LR), a lyophilized probiotic bacterium, on health status and performance of red sea bream (Pagrus major). Probiotics were incorporated in the diets at four different concentrations: 0 (control diet, LR0), 10(2) (LR1), 10(4) (LR2) and 10(6) (LR3) cells g(-1) and diets were administered to the fish for a period of 8 weeks. After the feeding trial, final body weight, body weight gain, specific growth rate, protease activity, protein digestibility, Lactobacillus sp. intestinal count, and superoxide dismutase were significantly higher in all probiotic-fed groups ($P < 0.05$). In addition, lipid and dry matter digestibility, reactive oxygen metabolites, biological antioxidant potential, and humoral and mucosal immune parameters including (total serum protein, alternative complement pathway, bactericidal and peroxidase activities) were also significantly elevated in fish fed probiotic supplementations being the effects dose-dependent. All growth, feed utilization, immune and oxidative parameters were significantly improved following probiotic administration. Present results revealed that L. rhamnosus is a promising probiotic candidate employed to help red sea bream protect themselves, thus promoting safe farming that would be less dependent on chemotherapy against infectious diseases.

de Brito Alves, J. L., de, Sousa, V, Cavalcanti Neto, M. P., Magnani, M., Braga, V. A., da Costa-Silva, J. H., Leandro, C. G., Vidal, H., and Pirola, L. New Insights on the Use of Dietary Polyphenols or Probiotics for the Management of Arterial Hypertension. Front Physiol. 2016; 7:448; PM: 27766081

ABSTRACT: Arterial hypertension (AH) is one of the most prevalent risk factors for cardiovascular diseases (CD) and is the main cause of deaths worldwide. Current research establish that dietary polyphenols may help to lower blood pressure (BP), thus contributing to the reduction of cardiovascular complications. In addition, the health benefits of probiotics on BP have also attracted increased attention, as probiotics administration modulates the microbiota, which, by interacting with ingested polyphenols, controls their bioavalability. The aim of the present mini-review is to summarize and clarify the effects of dietary polyphenols and probiotics administration on BP using combined evidence from clinical and experimental studies, as well as to discuss the current debate in the literature about the usefulness of this nutritional approach to manage BP. Clinical trials and experimental studies have demonstrated that consuming dietary polyphenols or probiotics in adequate amounts may improve BP, ranging from modest to greater effects. However, the mechanisms linking probiotic intake and reduced BP levels need to be further elucidated as a definitive consensus on the link between intake of polyphenols or probiotics and improvement of AH has not been reached yet.

De, Bastiani R., Sanna, G., Fracasso, P., D'Urso, M., Benedetto, E., and Tursi, A. The Management of Patients With Diverticulosis and Diverticular Disease in Primary Care: An Online Survey Among Italian General Pratictioners. J.Clin.Gastroenterol. 2016; 50 Suppl 1:S89-S92; PM: 27622377

ABSTRACT: ***GOALS***: To investigate the current opinion of Italian general practitioners (GPs) on the management of patients with diverticular disease (DD) of the colon. *BACKGROUND*: The management of DD remains a point of debate, and guidelines are not uniform in their advice. ***STUDY***: A web-based survey was conducted among Italian

GPs. Twelve questions were aimed at the diagnosis, treatment, and management options for diverticulosis and symptomatic DD. ***RESULTS***: In total, 245 surveys were filled out. A high-fiber diet was prescribed widely in diverticulosis (44%), together with advice to allow seeds (30%). Rifaximin (26%) and probiotics (25%) were the most frequently prescribed drugs in this population. Colonoscopy was the most prescribed instrumental tool in the diagnosis (77%) and follow-up (21%) of symptomatic uncomplicated diverticular disease patients. Rifaximin, probiotics, and mesalazine were the most frequently prescribed drugs in symptomatic uncomplicated diverticular disease patients (82.8, 59.5%, and 36.3%, respectively). Finally, 77% of the Italian GPs prescribed laboratory exams in the follow-up of these patients. The vast majority of the Italian GPs (83%) managed suspected acute diverticulitis at home, and did not consider two episodes of acute diverticulitis as a strict surgical indication (86%). Rifaximin, probiotics, and mesalazine were the most frequently prescribed drugs to prevent recurrence of the disease (42.5%, 28.2%, and 12.4%, respectively). Finally, 87% of the Italian GPs prescribed laboratory examinations in the follow-up of these patients. ***CONCLUSIONS***: This survey shows that the current management of DD in primary care by Italian GPs is not fully in line with current guidelines and more recent literature data.

Del, Piano M., Coggiola, F., Pane, M., Amoruso, A., Nicola, S., and Mogna, L. Can Probiotics Reduce Diarrhea and Infant Mortality in Africa?: The Project of a Pilot Study. J.Clin.Gastroenterol. 2016; 50 Suppl 2, Proceedings from the 8th Probiotics, Prebiotics & New Foods for Microbiota and Human Health meeting held in Rome, Italy on September 13-15, 2015:S120-S123; PM: 27741153

ABSTRACT: ***BACKGROUND***: Diarrhea accounts for 9% of the mortality among children under 5 years of age worldwide, and it is significantly associated with malnutrition. Each year, diarrhea kills around 760,000 children under 5 years of age and most of these are in sub-Saharan Africa.In Uganda, the infant mortality rate of 58 per 1000 is unacceptably high, and the major contributors include malnutrition, diarrhea, pneumonia, malaria, prematurity, sepsis, and newborn illnesses.There is an urgent need for intervention to prevent and control diarrheal diseases. ***STUDY DESIGN***: Our open-label, randomized controlled study has the primary endpoint of reducing diarrhea and infectious diseases (number of episodes/severity) and the secondary endpoint of decreasing infant mortality. The trial is currently conducted in Luzira, a suburb of Kampala, the capital of Uganda, and in Gulu and Lira, in the north of Uganda.The study is projected to enroll 4000 babies (control=2000 and treatment=2000) who will be followed till 1 year of life. As controls, 2000 babies of the same community are planned to be considered.The probiotic product selected for the trial is composed of 3 designated microorganisms, namely Bifidobacterium breve BR03 (DSM 16604), B. breve B632 (DSM 24706), and Lactobacillus delbrueckii subsp. delbrueckii LDD01 (DSM 22106). The concentration of the 3 bacteria is 10 viable cells/strain/daily dose (5 drops). ***PERSPECTIVES***: For a total sample of 4000 babies, the study has an 80% power at a 5% significance level.

Derrien, M. and Veiga, P. Rethinking Diet to Aid Human-Microbe Symbiosis. Trends Microbiol. 10-26-2016; PM: 27916707

ABSTRACT: The spread of the Western lifestyle has been accompanied by microbial changes thought to underlie the emergence of chronic, nontransmissible, immune-related diseases. The past decade has seen the unprecedented development of therapies for 'replenishing' the microbiota of sick individuals. However, functional and

ecological solutions helping the host and the gut microbiota to cope with the ecological stressors of modern life are still lacking. In this review, we discuss how recent advances in gut microbiome science are leading to the identification of microbe-derived and health-relevant metabolites. These molecules will guide the selection of the next-generation of probiotics and dietary recommendations, which should also take the resident gut microbiota into account, to optimise efficacy. These solutions for maintaining a well-functioning gut ecosystem and promoting good health should be customised, palatable, and as widely accessible as possible.

Deshpande, G., Rao, S., thalye-Jape, G., Conway, P., and Patole, S. Probiotics in very preterm infants: the PiPS trial. Lancet. 8-13-2016; 388(10045):655; PM: 27533430

No Abstract Available.

Dhawan, S., Singh, R., Kaur, R., and Kaur, J. A beta-mannanase from Paenibacillus sp.: Optimization of production and its possible prebiotic potential. Biotechnol.Appl.Biochem. 2016; 63(5):669-678; PM: 26224294

ABSTRACT: A thermotolerant bacterium Paenibacillus thiaminolyticus with an ability to produce extracellular beta-mannanase was isolated from a soil sample. Bacterium produced 45 U/mL beta-mannanase at 50 degrees C. The culture conditions for high-level production of beta-mannanase were optimized. Optimized MS medium [wheat bran 2% (w/v), ammonium sulfate 0.3% (w/v), yeast extract, and peptone (0.025% each) pH 6.5] was inoculated with 2% of 16 H old culture. The culture was incubated at 50 degrees C for 48 H resulting in 24-folds higher beta-mannanase production (1,100 +/- 50 U/mL). Optimum pH and temperature for enzyme activity of the crude enzyme was 6.0 and 60 degrees C, respectively. The enzyme demonstrated 65% relative enzyme activity at 37 degrees C. The hydrolytic activity of the crude enzymatic preparation was assessed on various agro residues. Thin-layer chromatographic analysis showed that the enzyme activity to saccharify heteromannans resulted in production of a mixture of manno-oligosaccharides (MOS) and enzyme exhibited classic endo-activity. To evaluate the possible prebiotic potential of the MOS thus obtained, initial screening for their ability to support the growth of probiotics was carried out by the pure culture method. Bifidobacterium and Lactobacillus sp. responded positively to the addition of enzymatically derived oligosaccharides and their numbers increased significantly.

di, Mauro G., Bernardini, R., Barberi, S., Capuano, A., Correra, A., De' Angelis, G. L., Iacono, I. D., de, Martino M., Ghiglioni, D., Di, Mauro D., Giovannini, M., Landi, M., Marseglia, G. L., Martelli, A., Miniello, V. L., Peroni, D., Sullo, L. R., Terracciano, L., Vascone, C., Verduci, E., Verga, M. C., and Chiappini, E. Prevention of food and airway allergy: consensus of the Italian Society of Preventive and Social Paediatrics, the Italian Society of Paediatric Allergy and Immunology, and Italian Society of Pediatrics. World Allergy Organ J. 2016; 9:28; PM: 27583103

ABSTRACT: ***BACKGROUND***: Allergic sensitization in children and allergic diseases arising therefrom are increasing for decades. Several interventions, functional foods, pro- and prebiotics, vitamins are proposed for the prevention of allergies and they can't be uncritically adopted. ***OBJECTIVE***: This Consensus document was developed by the Italian

Society of Preventive and Social Paediatrics and the Italian Society of Paediatric Allergy and Immunology. The aim is to provide updated recommendations regarding allergy prevention in children. ***METHODS***: The document has been issued by a multidisciplinary expert panel and it is intended to be mainly directed to primary care paediatricians. It includes 19 questions which have been preliminarily considered relevant by the panel. Relatively to each question, a literature search has been performed, according to the Italian National Guideline Program. Methodology, and a brief summary of the available literature data, has been provided. Many topics have been analyzed including the role of mother's diet restriction, use of breast/formula/hydrolyzed milk; timing of introduction of complementary foods, role (if any) of probiotics, prebiotics, vitamins, exposure to dust mites, animals and to tobacco smoke. ***RESULTS***: Some preventive interventions have a strong level of recommendation. (e.g., the dehumidifier to reduce exposure to mite allergens). With regard to other types of intervention, such as the use of partially and extensively hydrolyzed formulas, the document underlines the lack of evidence of effectiveness. No preventive effect of dietary supplementation with polyunsaturated fatty acids, vitamins or minerals has been demonstrated. There is no preventive effect of probiotics on asthma, rhinitis and allergic diseases. It has demonstrated a modest effect, but steady, in the prevention of atopic dermatitis. ***CONCLUSIONS***: The recommendations of the Consensus are based on a careful analysis of the evidence available. The lack of evidence of efficacy does not necessarily imply that some interventions may not be effective, but currently they can't be recommended.

djei-Fremah, S., Ekwemalor, K., Asiamah, E., Ismail, H., and Worku, M. Transcriptional profiling of the effect of lipopolysaccharide (LPS) pretreatment in blood from probiotics-treated dairy cows. Genom.Data. 2016; 10:15-18; PM: 27656413

ABSTRACT: Probiotic supplements are beneficial for animal health and rumen function; and lipopolysaccharides (LPS) from gram negative bacteria have been associated with inflammatory diseases. In this study the transcriptional profile in whole blood collected from probiotics-treated cows was investigated in response to stimulation with lipopolysaccharides (LPS) in vitro. Microarray experiment was performed between LPS-treated and control samples using the Agilent one-color bovine v2 bovine (v2) 4x44K array slides. Global gene expression analysis identified 13,658 differentially expressed genes (fold change cutoff >/= 2, P < 0.05), 3816 upregulated genes and 9842 downregulated genes in blood in response to LPS. Treatment with LPS resulted in increased expression of TLR4 (Fold change (FC) = 3.16) and transcription factor NFkB (FC = 5.4) and decreased the expression of genes including TLR1 (FC = - 2.54), TLR3 (FC = - 2.43), TLR10 (FC = - 3.88), NOD2 (FC = - 2.4), NOD1 (FC = - 2.45) and pro-inflammatory cytokine IL1B (- 3.27). The regulation of the genes involved in inflammation signaling pathway suggests that probiotics may stimulate the innate immune response of animal against parasitic and bacterial infections. We have provided a detailed description of the experimental design, microarray experiment and normalization and analysis of data which have been deposited into NCBI Gene Expression Omnibus (GEO): GSE75240.

Dos Santos Pereira Andrade AC, Lima, M. T., Oliveira, G. P., Calixto, R. S., de Sales E Souza EL, da Gloria de, Souza D., de Almeida Leite, C. M., Ferreira, J. M., Kroon, E. G., de Oliveira, D. B., Dos Santos, Martins F., and Abrahao, J. S. Daily ingestion of the probiotic Lactobacillus paracasei ST11 decreases Vaccinia virus dissemination and lethality in a mouse model. Benef.Microbes. 11-22-2016; 1-8; PM: 27873547

ABSTRACT: Vaccinia virus (VACV) is an important pathogen. Although studies have shown relationships between probiotics and viruses, the effect of probiotics on VACV infection is unknown. Therefore, this work aims to investigate the probiotics effects on VACV infection. Mice were divided into four groups, two non-infected groups, one receiving the probiotic, the other one not receiving it, and two groups infected intranasally with VACV Western Reserve (VACV-WR) receiving or not receiving the probiotic. Viral titres in organs and cytokine production in the lungs were analysed. Lung samples were also subjected to histological analysis. The intake of probiotic results in reduction in viral spread with a significant decrease of VACV titer on lung, liver and brain of treated group. In addition,treatment with the probiotic results in attenuated mice lung inflammation showing fewer lesions on histological findings and decreased lethality in mice infected with VACV. The ingestion of Lactobacillus paracasei ST11 (LPST11) after VACV infection resulted in 2/9 animal lethality compared with 4/9 in the VACV group. This is the first study on probiotics and VACV interactions, providing not only information about this interaction, but also proposing a model for future studies involving probiotics and other poxvirus.

Dubreuil, J. D., Isaacson, R. E., and Schifferli, D. M. Animal Enterotoxigenic Escherichia coli. EcoSal.Plus. 2016; 7(1); PM: 27735786

ABSTRACT: Enterotoxigenic Escherichia coli (ETEC) is the most common cause of E. coli diarrhea in farm animals. ETEC are characterized by the ability to produce two types of virulence factors: adhesins that promote binding to specific enterocyte receptors for intestinal colonization and enterotoxins responsible for fluid secretion. The best-characterized adhesins are expressed in the context of fimbriae, such as the F4 (also designated K88), F5 (K99), F6 (987P), F17, and F18 fimbriae. Once established in the animal small intestine, ETEC produce enterotoxin(s) that lead to diarrhea. The enterotoxins belong to two major classes: heat-labile toxins that consist of one active and five binding subunits (LT), and heat-stable toxins that are small polypeptides (STa, STb, and EAST1). This review describes the disease and pathogenesis of animal ETEC, the corresponding virulence genes and protein products of these bacteria, their regulation and targets in animal hosts, as well as mechanisms of action. Furthermore, vaccines, inhibitors, probiotics, and the identification of potential new targets by genomics are presented in the context of animal ETEC.

Eeckhaut, V., Wang, J., Van, Parys A., Haesebrouck, F., Joossens, M., Falony, G., Raes, J., Ducatelle, R., and Van, Immerseel F. The Probiotic Butyricicoccus pullicaecorum Reduces Feed Conversion and Protects from Potentially Harmful Intestinal Microorganisms and Necrotic Enteritis in Broilers. Front Microbiol. 2016; 7:1416; PM: 27708624

ABSTRACT: Probiotics which do not result in the development and spread of microbial resistance are among the candidate replacements for antibiotics previously used as growth promotors. In this study the effect of in-feed supplementation of the butyrate producing Butyricicoccus pullicaecorum strain 25-3T on performance, intestinal microbiota and prevention of necrotic enteritis (NE), a disease caused by Clostridium perfringens was evaluated in broilers. For the performance study, day old Ross 308 chicks were randomly allocated into two treatment groups and fed either a non-supplemented diet or a diet supplemented with 109 cfu lyophilized B. pullicaecorum per kg feed for 40 days. On day 40 broilers administered B. pullicaecorum had a significant lower bodyweight (2675 g vs. 2762 g; p = 0.0025) but supplementation of B. pullicaecorum

decreased the feed conversion ratio significantly (1.518 vs. 1.632; $p < 0.0001$). Additionally, ingestion of the Butyricicoccus strain significantly lowered the abundance of Campylobacter spp. in the caecum and Enterococcus and Escherichia/Shigella spp. in the ileum at day 40. In feed supplementation of B. pullicaecorum in the NE trials resulted in a significant decrease in the number of birds with necrotic lesions compared with the untreated control group. These studies show that supplementation of B. pullicaecorum is able to improve feed conversion, to reduce the abundance of some potentially important pathogens in the caeca and ileum and to contribute to the prevention of NE in broilers, making the strain a potential valuable probiotic.

Eggers, S., Barker, A., Valentine, S., Hess, T., Duster, M., and Safdar, N. Impact of Probiotics for Reducing Infections in Veterans (IMPROVE): Study protocol for a double-blind, randomized controlled trial to reduce carriage of Staphylococcus aureus. Contemp.Clin.Trials. 11-9-2016; 52:39-45; PM: 27836508

ABSTRACT: ***BACKGROUND***: Staphylococcus aureus (S. aureus) is an organism of great public health importance, causing 20,000 deaths annually. Decolonization of patients with S. aureus may prevent infections, yet current options are limited to antimicrobials that promote antibiotic resistance and can cause adverse side effects. Probiotics have potential to reduce colonization of pathogenic bacteria, representing a promising alternative for S. aureus decolonization, but thus far lack rigorous evaluation. ***METHODS***: Potential subjects were recruited from inpatient and outpatient settings within a VA medical center and screened for S. aureus gastrointestinal (GI) or extra-GI colonization using swabs at multiple body sites. Positive, eligible, consenting participants were stratified by colonization site and randomized in a 1:1 ratio to 4-weeks of daily placebo or Lactobacillus rhamnosus (L. rhamnosus) HN001 probiotic treatment. Blood and stool samples, and treatment adherence reports were collected from each subject throughout the study, along with a final set of swabs at study completion to detect S. aureus carriage. The outcomes of this study are GI or extra-GI carriage by S. aureus at the end of 4weeks of therapy, change in phagocytic activity of polymorphonuclear cells from pre-intervention to post-intervention, and symptomatic S. aureus infection at any site during the study period. ***CONCLUSION***: 114 participants have been recruited for this study. Analysis of outcomes is underway. This is the first clinical trial to examine the efficacy of L. rhamnosus HN001 for decolonization of S. aureus, and investigates the mechanism by which L. rhamnosus HN001 mediates its effect on S. aureus colonization. ClinicalTrials.govIdentifier NCT01321606.

Elavarasu, S., Suthanthiran, T., Thangavelu, A., Kanagaraj, S. S., Mohandas, L., and Sekar, S. Evaluation of efficacy of probiotic (BIFILAC) on Porphyromonas gingivalis: In vitro study. J.Pharm.Bioallied.Sci. 2016; 8(Suppl 1):S45-S47; PM: 27829746

ABSTRACT: ***BACKGROUND***: Periodontitis is inflammation of the periodontium and causes destruction of the connective tissue attachment of the teeth and alveolar bone. Porphyromonas gingivalis is the primary pathogen for the destructive periodontal diseases. The aim of the study is to evaluate the efficacy of probiotic on P. gingivalis. ***MATERIALS AND METHODS***: An in vitro study was done to analyze the effectiveness of probiotic BIFILAC on P. gingivalis was determined using disc diffusion method. The minimum inhibitory concentration for BIFILAC lozenges was also determined using microdilution method. ***RESULTS***: In disc diffusion method, the antibacterial activity of BIFILAC was analyzed using various concentrations such as 2.5, 5, 10, and 20 mug/ml, of

which 20 mug/ml was proved to have a maximum inhibitory zone of 22 mm. In microdilution method, concentration ranging from 7.25 to 100 mug/ml was used and 25 mug/ml was found to have the minimum inhibitory effect on P. gingivalis. ***CONCLUSION***: The present in vitro study confirms that probiotic BIFILAC has an antimicrobial effect against P. gingivalis. Thus, proving that BIFILAC probiotic can be used as an adjunctive therapeutic modality in periodontitis.

Elzouki, A. N. Probiotics and Liver Disease: Where Are We Now and Where Are We Going? J.Clin.Gastroenterol. 2016; 50 Suppl 2, Proceedings from the 8th Probiotics, Prebiotics & New Foods for Microbiota and Human Health meeting held in Rome, Italy on September 13-15, 2015:S188-S190; PM: 27741172

ABSTRACT: Probiotics are live, nonpathogenic bacteria capable of colonizing the colonic mucosa. The most common probiotics include strains of Lactobacillus or Bifidobacteria, which are part of the normal gastrointestinal microbiota. Initial studies of selected probiotic species have suggested potential efficacy in several gastrointestinal diseases including inflammatory bowel diseases (particularly pouchitis), antibiotic-related diarrhea, Clostridium difficile toxin-induced colitis, infectious diarrhea, irritable bowel syndrome, and allergy. The so-called "gut-liver axis" involves complex interaction between the liver parenchyma and gut microbiota. There is growing evidence to suggest that alteration in gut microbial components may affect the liver and can be a precipitating cofactor in development and modulating of chronic liver damage through ethanol, production of ammonia and endotoxin. This may allow for a better understanding of its role in the pathogenesis of verities of liver diseases and help to identify a microbial target for prevention and treatment of such diseases. This paper discusses the growing evidence that highlights the relationship between gut microbiota and development, prevention and treatment of numbers of liver diseases.

Erdman, S. E. and Poutahidis, T. Microbes and Oxytocin: Benefits for Host Physiology and Behavior. Int.Rev.Neurobiol. 2016; 131:91-126; PM: 27793228

ABSTRACT: It is now understood that gut bacteria exert effects beyond the local boundaries of the gastrointestinal tract to include distant tissues and overall health. Prototype probiotic bacterium Lactobacillus reuteri has been found to upregulate hormone oxytocin and systemic immune responses to achieve a wide array of health benefits involving wound healing, mental health, metabolism, and myoskeletal maintenance. Together these display that the gut microbiome and host animal interact via immune-endocrine-brain signaling networks. Such findings provide novel therapeutic strategies to stimulate powerful homeostatic pathways and genetic programs, stemming from the coevolution of mammals and their microbiome.

Esaiassen, E., Cavanagh, P., Hjerde, E., Simonsen, G. S., Stoen, R., and Klingenberg, C. Bifidobacterium longum Subspecies infantis Bacteremia in 3 Extremely Preterm Infants Receiving Probiotics. Emerg.Infect.Dis. 2016; 22(9):1664-1666; PM: 27532215

No Abstract Available.

Espinola-Docio, B., Costa-Romero, M., az-Gomez, N. M., and Paricio-Talayero, J. M. [Mastitis update.]. Arch.Argent Pediatr. 12-1-2016; 114(6):576-584; PM: 27869418

No Abstract Available.

Eun, S. H., Lim, S. M., Jang, S. E., Han, M. J., and Kim, D. H. Lactobacillus sakei K17, an inducer of IL-10 expression in antigen-presenting cells, attenuates TNBS-induced colitis in mice. Immunopharmacol.Immunotoxicol. 9-27-2016; 1-8; PM: 27600362

ABSTRACT: To understand the anti-colitic effects of probiotics that up-regulate interleukin (IL)-10 expression in dendritic cells (DCs) and macrophages, we isolated Lactobacillus sakei K17, which potently induced IL-10 expression in DCs and peritoneal macrophages in vitro, among the lactic acid bacteria strains collected from kimchi and investigated its anti-inflammatory effect in mice with 2,4,6-trinitrobenzene sulfonic acid (TNBS)-induced colitis. Oral administration of K17 (2 x 109 CFU.mouse-1.day-1) in mice with TNBS-induced colitis suppressed colon shortening and myeloperoxidase activity, as well as infiltration of CD86+ cells into the colon. Treatment with K17 also increased TNBS-suppressed expression of tight junction proteins and IL-10, but inhibited activation of nuclear factor-kappaB (NF-kappaB) and mitogen-activated protein kinases and expression of tumor necrosis factor alpha and IL-17. Its effect was comparable with that of sulfasalazine (50 mg/kg), a positive commercial ant-colitic drug. Furthermore, treatment with K17 (1 x 105 CFU/mL) potently inhibited lipopolysaccharide (LPS)-stimulated NF-kappaB activation in DCs and peritoneal macrophages and restored tight junction protein expression in LPS-stimulated Caco-2 cells. These findings suggest that Lactobacillus sakei K17 may ameliorate colitis by up-regulating the expression of IL-10 and tight junction proteins and inhibiting NF-kappaB activation.

Evenepoel, P., Poesen, R., and Meijers, B. The gut-kidney axis. Pediatr.Nephrol. 11-15-2016; PM: 27848096

ABSTRACT: The host-gut microbiota interaction has been the focus of increasing interest in recent years. It has been determined that this complex interaction is not only essential to many aspects of normal "mammalian" physiology but that it may also contribute to a multitude of ailments, from the obvious case of inflammatory bowel disease to (complex) diseases residing in organs outside the gut. An increasing body of evidence indicates that crosstalk between host and microbiota is pathophysiologically relevant in patients with chronic kidney disease (CKD). Interactions are bidirectional; on the one hand, uremia affects both the composition and metabolism of the gut microbiota and, on the other hand, important uremic toxins originate from microbial metabolism. In addition, gut dysbiosis may induce a disruption of the epithelial barrier, ultimately resulting in increased exposure of the host to endotoxins. Due to dietary restrictions and gastrointestinal dysfunctions, microbial metabolism shifts to a predominantly proteolytic fermentation pattern in CKD. Indoxyl sulfate and p-cresyl sulfate, both end-products of protein fermentation, and trimethylamine-N-oxide, an end-product of microbial choline and carnitine metabolism, are prototypes of uremic toxins originating from microbial metabolism. The vascular and renal toxicity of these co-metabolites has been demonstrated extensively in experimental and clinical studies. These co-metabolites are an appealing target for adjuvant therapy in CKD. Treatment options include dietary therapy, prebiotics, probiotics and host and bacterial enzyme inhibitors. Final proof of the concept should come from randomized controlled and adequately powered intervention studies.

Fabbrocini, G., Bertona, M., Picazo, O., Pareja-Galeano, H., Monfrecola, G., and Emanuele, E. Supplementation with Lactobacillus rhamnosus SP1 normalises skin expression of genes implicated in insulin signalling and improves adult acne. Benef.Microbes. 11-30-2016; 7(5):625-630; PM: 27596801

ABSTRACT: Systemic supplementation with probiotics is increasingly being explored as a potential treatment strategy for skin disorders. Because both the gut-skin axis and dysregulation of insulin signalling have been implicated in the pathogenesis of adult acne, we designed the current study to evaluate the effect of supplementation with the probiotic strain Lactobacillus rhamnosus SP1 (LSP1) on skin expression of genes involved in insulin signalling and acne improvement in adult subjects. A pilot, randomised, double-blinded, placebo-controlled study was conducted with 20 adult subjects (14 females and 6 males; mean age: 33.7+/-3.3 years) with acne. Over a 12-week period, the probiotic group (n=10) consumed a liquid supplement containing LSP1 at a dose of 3x109 cfu/day (75 mg/day), whereas the placebo group (n=10) received a liquid lacking probiotics. Paired skin biopsies - one obtained before treatment initiation and one obtained at the end of the 12-week treatment period - were analysed for insulin-like growth factor 1 (IGF1) and forkhead box protein O1 (FOXO1) gene expression. The clinical criterion for efficacy was the investigator's global improvement rating on a five-point scale. Compared with baseline, the probiotic group showed a 32% (P<0.001) reduction, as well as a 65% increase (P<0.001) in IGF1 and FOXO1 gene expression in the skin, respectively. No such differences were observed in the placebo group. Patients in the probiotic group had an adjusted odds ratio of 28.4 (95% confidence interval = 2.2-411.1, P<0.05) to be rated by physicians as improved/markedly improved (versus worsened or unchanged) compared with the placebo group. We conclude that supplementation with the probiotic strain LSP1 normalises skin expression of genes involved in insulin signalling and improves the appearance of adult acne.

Famouri, F., Shariat, Z., Hashemipour, M., Keikha, M., and Kelishadi, R. Effects of Probiotics on Non-Alcoholic Fatty Liver Disease in Obese Children and Adolescents: A Randomized Clinical Trial. J.Pediatr.Gastroenterol.Nutr. 10-13-2016; PM: 27753706

ABSTRACT: ***OBJECTIVES***: This trial aims to evaluate the effects of some probiotics on sonographic and biochemical non-alcoholic fatty liver disease (NAFLD). **METHODS**: This randomized triple-blind trial was conducted among 64 obese children with sonographic NAFLD. They were randomly allocated to receive probiotic capsule (containing Lactobacillus acidophilus ATCC B3208, 3 x 10 colony forming units, CFU; Bifidobacterium lactis DSMZ 32269, 6 x 10 CFU; B bifidum ATCC SD6576, 2 x 10 CFU; L rhamnosus DSMZ 21690, 2 x 10 CFU) or placebo for 12 weeks. ***RESULTS***: After intervention, in the probiotic group the mean levels of alanine aminotransferase (ALT) decreased from 32.8 (19.6) to 23.1(9.9) U/L (P = 0.02); and mean aspartate aminotransferase (AST) decreased from 32.2 (15.7) to 24.3 (7.7) U/L (P = 0.02). Likewise the mean cholesterol, LDL-C, and triglycerides, as well as waist circumference decreased in the intervention group, without significant change in weight, BMI, and BMI Z-score. After the trial, normal liver sonography was reported in 17 (53.1%) and 5 (16.5%) of patients in the intervention and placebo groups, respectively. ***CONCLUSIONS***: The current findings suggest that a course of the abovementioned probiotic compound can be effective in improving pediatric NAFLD.

Feizollahzadeh, S., Ghiasvand, R., Rezaei, A., Khanahmad, H., Sadeghi, A., and Hariri, M. Effect of Probiotic Soy Milk on Serum Levels of Adiponectin, Inflammatory Mediators, Lipid Profile, and Fasting Blood Glucose Among Patients with Type II Diabetes Mellitus. Probiotics.Antimicrob.Proteins. 10-18-2016; PM: 27757829

ABSTRACT: Probiotic therapies are going to be an effective alternative therapeutic strategy in the treatment and management of diabetes. The mechanism behind the essential effects of probiotic therapies in diabetic patients was not fully understood. The objective of this study was to evaluate the effects of probiotic soy milk containing Lactobacillus planetarum A7 on inflammation, lipid profile, fasting blood glucose, and serum adiponectin among patients with type 2 diabetes mellitus. Forty patients with type 2 diabetes, at the age of 35-68 years old, were assigned to two groups in this randomized, double-blind, controlled clinical trial. The patients in the intervention group consumed 200 ml/day of probiotic soy milk containing L. planetarum A7 and those in control group consumed 200 ml/day of pure soy milk for 8 weeks. Serum TNF-alpha, C reactive protein, adiponectin, lipid profile, and fasting blood glucose were determined before and after intervention. In intervention group, serum adiponectin in pre- and post-treatment did not show any significant changes (2.52 +/- 0.74 vs 2.84 +/- 0.61, P = 0.658), as well as changes in serum TNF-alpha and C reactive protein (172.44 +/- 5.7 vs 172.83 +/- 7.6, P = 0.278, 4.2 +/- 1.4 vs 4.5 +/- 1.9, P = 0.765, respectively). Low-density cholesterol and high-density cholesterol changed significantly (P = 0.023, P = 0.017, respectively), but fasting blood glucose did not show any significant changes. The results of this study showed that consumption of probiotic soy milk and soy milk has no effect on serum adiponectin and inflammation, but it can change lipid profile among type 2 diabetic patients.

Ferrari, Ida S., de Souza, J. V., Ramos, C. L., da Costa, M. M., Schwan, R. F., and Dias, F. S. Selection of autochthonous lactic acid bacteria from goat dairies and their addition to evaluate the inhibition of Salmonella typhi in artisanal cheese. Food Microbiol. 2016; 60:29-38; PM: 27554143

ABSTRACT: This study aimed to select autochthonous lactic acid bacteria (LAB) with probiotic and functional properties from goat dairies and test their addition to artisanal cheese for the inhibition of Salmonella typhi. In vitro tests, including survival in the gastrointestinal tract (GIT), auto- and co-aggregation, the hemolytic test, DNase activity, antimicrobial susceptibility, antibacterial activity, tolerance to NaCl and exopolysaccharide (EPS), gas and diacetyl production were conducted for sixty isolates. Based on these tests, four LAB isolates (UNIVASF CAP 16, 45, 84 and 279) were selected and identified. Additional tests, such as production of lactic and citric acids by UNIVASF CAP isolates were performed in addition to assays of bile salt hydrolase (BSH), beta-galactosidase and decarboxylase activity. The four selected LAB produced high lactic acid (>17 g/L) and low citric acid (0.2 g/L) concentrations. All selected strains showed BSH and beta-galactosidase activity and none showed decarboxylase activity. Three goat cheeses (1, 2 and control) were produced and evaluated for the inhibitory action of selected LAB against Salmonella typhi. The cheese inoculated with LAB (cheese 2) decreased 0.38 log10 CFU/g of S. Typhy population while in the cheese without LAB

inoculation (cheese 1) the pathogen population increased by 0.29 log units. Further, the pH value increased linearly over time, by 0.004 units per day in cheese 1. In the cheese 2, the pH value decreased linearly over time, by 0.066 units per day. The cocktail containing selected Lactobacillus strains with potential probiotic and technological properties showed antibacterial activity against S. typhi in vitro and in artisanal goat cheese. Thus, goat milk is important source of potential probiotic LAB which may be used to inhibit the growth of Salmonella population in cheese goat, contributing to safety and functional value of the product.

Fleming, D., Kesey, J., Rumbaugh, K., and Dissanaike, S. Comparing the Survivability of Lactobacillus Species in Various Probiotic Delivery Vehicles. JPEN J.Parenter.Enteral Nutr. 9-29-2016; PM: 27687914

ABSTRACT: ***BACKGROUND***: Probiotics are widely used in healthy and nonhealthy individuals to maintain a favorable gut microbiome and inhibit pathogen takeover. Currently, there are many varieties of probiotic delivery vehicles on the market, with no real research indicating which is the most effective at allowing for colon colonization. In this study, we sought to determine if probiotic preparation influences the ability of Lactobacillus species, one of the most common genera of probiotic bacteria, to survive gastric acidity. ***OBJECTIVE***: To determine the survivability of lactobacilli in stomach-level acidity for a variety of probiotic delivery vehicles. ***MATERIALS AND METHODS***: We tested the ability of 6 different probiotic preparations-enterically encapsulated pills, nonencapsulated pills, lyophilized powder, yogurt, yogurt drink, and kefir-to enhance survivability of lactobacilli in pH 2.0-adjusted phosphate-buffered saline over time. Colony-forming units were enumerated by spot plating on both nonselective (brain-heart infusion) and selective (Rogosa SL) agar. ***RESULTS***: Regardless of delivery vehicle, lactobacilli are very capable of surviving gastric acidity, allowing for passage into the intestines.

Fong, F. L., Shah, N. P., Kirjavainen, P., and El-Nezami, H. Mechanism of Action of Probiotic Bacteria on Intestinal and Systemic Immunities and Antigen-Presenting Cells. Int.Rev.Immunol. 5-3-2016; 35(3):179-188; PM: 26606641

ABSTRACT: Immunomodulation has been shown to be one of the major functions of probiotic bacteria. This review is presented to provide detailed information on the immunomodulatory properties of probiotics in various animal models and clinical practices. Probiotics can regulate helper T (Th) responses and release of cytokines in a strain-specific manner. For example, Lactobacillus rhamnosus GG can induce beneficial Th1 immunomodulatory effect in infants with cow's milk allergy and relieve intestinal inflammation in atopic children by promoting IL-10 generation. Mechanism of action of probiotics on antigen-presenting cells at gastrointestinal tract is also postulated in this review. Probiotic bacterial cells and their soluble factors may activate dendritic cells, macrophages, and to certain extent monocytes via toll-like-receptor recognition and may further provoke specific Th responses. They are speculated to elicit immunomodulatory effects on intestinal and systemic immunities.

Forrest, C. M., McNair, K., Vincenten, M. C., Darlington, L. G., and Stone, T. W. Selective depletion of tumour suppressors Deleted in Colorectal Cancer (DCC) and neogenin by environmental and endogenous serine proteases: linking diet and cancer. BMC.Cancer. 10-6-2016; 16(1):772; PM: 27716118

ABSTRACT: ***BACKGROUND***: The related tumour suppressor proteins Deleted in Colorectal Cancer (DCC) and neogenin are absent or weakly expressed in many cancers, whereas their insertion into cells suppresses oncogenic behaviour. Serine proteases influence the initiation and progression of cancers although the mechanisms are unknown. ***METHODS***: The effects of environmental (bacterial subtilisin) and endogenous mammalian (chymotrypsin) serine proteases were examined on protein expression in fresh, normal tissue and human neuroblastoma and mammary adenocarcinoma lines. Cell proliferation and migration assays (chemoattraction and wound closure) were used to examine cell function. Cells lacking DCC were transfected with an ectopic dcc plasmid. ***RESULTS***: Subtilisin and chymotrypsin selectively depleted DCC and neogenin from cells at nanomolar concentrations without affecting related proteins. Cells showed reduced adherence and increased migration, but after washing they re-attached within 24 h, with recovery of protein expression. These effects are induced by chymotryptic activity as they are prevented by chymostatin and the soybean Bowman-Birk inhibitor typical of many plant protease inhibitors. ***CONCLUSIONS***: Bacillus subtilis, which secretes subtilisin is widely present in soil, the environment and the intestinal contents, while subtilisin itself is used in meat processing, animal feed probiotics and many household cleaning agents. With chymotrypsin present in chyme, blood and tissues, these proteases may contribute to cancer development by depleting DCC and neogenin. Blocking their activity by Bowman-Birk inhibitors may explain the protective effects of a plant diet. Our findings identify a potential non-genetic contribution to cancer cell behaviour which may explain both the association of processed meats and other factors with cancer incidence and the protection afforded by plant-rich diets, with significant implications for cancer prevention.

Forsberg, A., West, C. E., Prescott, S. L., and Jenmalm, M. C. Pre- and probiotics for allergy prevention: time to revisit recommendations? Clin.Exp.Allergy. 2016; 46(12):1506-1521; PM: 27770467

ABSTRACT: Reduced intensity and diversity of microbial exposure is considered a major factor driving abnormal postnatal immune maturation and increasing allergy prevalence, particularly in more affluent regions. Quantitatively, the largest important source of early immune-microbial interaction, the gut microbiota, is of particular interest in this context, with variations in composition and diversity in the first months of life associated with subsequent allergy development. Attempting to restore the health consequences of the 'dysbiotic drift' in modern society, interventions modulating gut microbiota for allergy prevention have been evaluated in several randomized placebo-controlled trials. In this review, we provide an overview of these trials and discuss recommendations from international expert bodies regarding prebiotic, probiotic and synbiotic interventions. Recent guidelines from the World Allergy Organization recommend the use of probiotics for the primary prevention of eczema in pregnant and breastfeeding mothers of infants at high risk for developing allergy and in high-risk infants. It is however stressed that these recommendations are conditional, based on very low-quality evidence and great heterogeneity between studies, which also impedes specific and practical advice to consumers on the most effective regimens. We discuss how the choice of probiotic strains, timing and duration of administration can critically influence

the outcome due to different effects on immune modulation and gut microbiota composition. Furthermore, we propose strategies to potentially improve allergy-preventive effects and enable future evidence-based implementation.

Forte, C., Moscati, L., Acuti, G., Mugnai, C., Franciosini, M. P., Costarelli, S., Cobellis, G., and Trabalza-Marinucci, M. Effects of dietary Lactobacillus acidophilus and Bacillus subtilis on laying performance, egg quality, blood biochemistry and immune response of organic laying hens. J.Anim Physiol Anim Nutr.(Berl). 2016; 100(5):977-987; PM: 26614687

ABSTRACT: The objective of this study was to evaluate the effects of two different probiotic micro-organisms on the performance, egg quality and blood parameters of organically reared hens. A total of 900 16-week-old Hy-Line layer hybrids were randomly assigned to three groups of 300 birds each. The control (CTR) group was fed a corn-soya bean cake-based diet; the L group was fed the same diet supplemented with 0.1% Lactobacillus acidophilus, while the B group was fed the same diet supplemented with 0.05% Bacillus subtilis. Data were recorded at the beginning (weeks 5 and 6: T1) and at the end (weeks 19 and 20: T2) of the experiment, and no differences in hen performance were recorded between dietary groups or sampling times. All of the investigated clinical chemistry parameters, except GGT, were affected by diet ($p < 0.05$), with the best results recorded for the probiotic-treated groups. The immune-response values showed higher blood bactericidal activity in the B and L groups at T2 ($p < 0.05$) and a lower lysozime concentration in the B group at T1. Higher antibody production against Newcastle disease virus was observed in the L group compared to the CTR ($p = 0.013$). No differences in oxidative status were recorded, and no effects of diet on egg quality were observed. Among the physical egg characteristics, only the Roche scale colour was affected by diet ($p < 0.05$): the egg yolk was paler in the L group. The age of the hen was the most relevant factor affecting physical egg characteristics. The chemical parameters of the egg were almost unaffected by supplementation with probiotics except for the lipid content, which decreased with the L diet ($p < 0.05$). Both probiotic inclusions had beneficial effects on hen metabolism and welfare, and L. acidophilus induced the best immune response.

Fredua-Agyeman, M., Stapleton, P., Basit, A. W., Beezer, A. E., and Gaisford, S. In vitro inhibition of Clostridium difficile by commercial probiotics: A microcalorimetric study. Int.J.Pharm. 12-3-2016; PM: 27923699

ABSTRACT: The aim of the study was to investigate the influence of some commercial probiotics on the growth of Clostridium difficile using the isothermal microcalorimeter, an instrument which can monitor the real time growth of bacteria. Commercial probiotic strains and products, Lactobacillus acidophilus LA-5(R), Bifidobacterium lactis BB-12(R), Probio 7(R) and Symprove were co-cultured with C. difficile in Brain Heart Infusion (BHI) broth supplemented with 0.1% (w/v) l-cysteine hydrochloride and 0.1% (w/v) sodium taurocholate and monitored in the microcalorimeter. Pseudomonas aeruginosa NCIMB 8628 was also co-cultured with C. difficile and studied. The results indicated inhibition of C. difficile by the probiotics. The inhibition of C. difficile was shown to be pH-dependent using neutralized and unmodified cell free supernatant (CFS) produced by the probiotic strains. However, concentrated CFS of the probiotics also inhibited the intestinal pathogen in a non pH-dependent manner, likely due to specific antimicrobial substances produced. The results also indicated that C. difficile growth was greatly influenced by the presence of sodium taurocholate and by the pH of the medium. A

medium pH of between 6.45 and 6.9 demonstrated maximum growth of the organism in the microcalorimeter.

Frost, B. L., Modi, B. P., Jaksic, T., and Caplan, M. S. New Medical and Surgical Insights Into Neonatal Necrotizing Enterocolitis: A Review. JAMA Pediatr. 11-21-2016; PM: 27893069

ABSTRACT: Importance: Necrotizing enterocolitis (NEC) has long remained a significant cause of morbidity and mortality in neonatal intensive care units. While the mainstay of treatment for this devastating condition remains largely supportive, research efforts continue to be directed toward understanding pathophysiology as well as how best to approach surgical management when indicated. Observations: In this review, we first examine recent medical observations, including overviews on the microbiome and a brief review of the use of probiotics. Next, we discuss the use of biomarkers and how clinicians may be able to use them in the future to predict the course of disease and, perhaps, the need for surgical intervention. We then provide an overview on the use of exclusive human milk feeding and the utility of this approach in preventing NEC. Finally, we discuss recent developments in the surgical management of NEC, beginning with indications for surgery and following with a section on technical surgical considerations, including peritoneal drain vs laparotomy. The review concludes with outcomes from infants with surgically treated NEC. Conclusions and Relevance: Although medical treatment options for NEC are largely unchanged, understanding of the disease continues to evolve. As new research methods are developed, NEC pathophysiology can be more completely understood. In time, it is hoped that data from ongoing and planned clinical trials will allow us to routinely add targeted preventive measures in addition to human milk, such as prebiotics and probiotics, to the management of high-risk infants. In addition, the discovery of novel biomarkers may not only prove useful in predicting severity of illness but also will hopefully allow for identification of the disease prior to onset of clinical signs. Finally, continued investigation into optimizing surgical outcomes is essential in this population of infants, many of whom require long-term parenteral therapy and intestinal rehabilitation.

Gagnon, M., Vimont, A., Darveau, A., Fliss, I., and Jean, J. Study of the Ability of Bifidobacteria of Human Origin to Prevent and Treat Rotavirus Infection Using Colonic Cell and Mouse Models. PLoS.One. 2016; 11(10):e0164512; PM: 27727323

ABSTRACT: Rotavirus is the leading cause of severe acute gastroenteritis among children worldwide. Despite effective vaccines, inexpensive alternatives such as probiotics are needed. The aim of this study was to assess the ability of probiotic candidate Bifidobacterium thermophilum RBL67 to inhibit rotavirus infection. Bacterial adhesion to intestinal cells and interference with viral attachment were evaluated in vitro. B. thermophilum RBL67 displayed adhesion indexes of 625 +/- 84 and 1958 +/- 318 on Caco-2 and HT-29 cells respectively and was comparable or superior to four other bifidobacteria, including B. longum ATCC 15707 and B. pseudolongum ATCC 25526 strains. Incubation of B. thermophilum RBL67 for 30 min before (exclusion) and simultaneously (competition) with human rotavirus strain Wa decreased virus attachment by 2.0 +/- 0.1 and 1.5 +/- 0.1 log10 (by 99.0% and 96.8% respectively). Displacement of virus already present was negligible. In CD-1 suckling mice fed B. thermophilum RBL67 challenged with simian rotavirus SA-11, pre-infection feeding with RBL 67 was more effective than post-infection feeding, reducing the duration of diarrhea, limiting epithelial lesions, reducing viral replication in the intestine, accelerating recovery, and stimulating the humoral specific IgG and IgM response, without inducing any adverse effect. B.

thermophilum RBL67 had little effect on intestinal IgA titer. These results suggest that humoral immunoglobulin might provide protection against the virus and that B. thermophilum RBL67 has potential as a probiotic able to inhibit rotavirus infection and ultimately reduce its spread.

Gallo, A., Passaro, G., Gasbarrini, A., Landolfi, R., and Montalto, M. Modulation of microbiota as treatment for intestinal inflammatory disorders: An uptodate. World J.Gastroenterol. 8-28-2016; 22(32):7186-7202; PM: 27621567

ABSTRACT: Alterations of intestinal microflora may significantly contribute to the pathogenesis of different inflammatory and autoimmune disorders. There is emerging interest on the role of selective modulation of microflora in inducing benefits in inflammatory intestinal disorders, by as probiotics, prebiotics, synbiotics, antibiotics, and fecal microbiota transplantation (FMT). To summarize recent evidences on microflora modulation in main intestinal inflammatory disorders, PubMed was searched using terms microbiota, intestinal flora, probiotics, prebiotics, fecal transplantation. More than three hundred articles published up to 2015 were selected and reviewed. Randomized placebo-controlled trials and meta-analysis were firstly included, mainly for probiotics. A meta-analysis was not performed because of the heterogeneity of these studies. Most of relevant data derived from studies on probiotics, reporting some efficacy in ulcerative colitis and in pouchitis, while disappointing results are available for Crohn's disease. Probiotic supplementation may significantly reduce rates of rotavirus diarrhea. Efficacy of probiotics in NSAID enteropathy and irritable bowel syndrome is still controversial. Finally, FMT has been recently recognized as an efficacious treatment for recurrent Clostridium difficile infection. Modulation of intestinal flora represents a very interesting therapeutic target, although it still deserves some doubts and limitations. Future studies should be encouraged to provide new understanding about its therapeutical role.

Gan, X. and Li, J. [Research progress of the application of probiotics in preterm infants]. Zhongguo Dang.Dai Er.Ke.Za Zhi. 2016; 18(9):909-914; PM: 27655553

ABSTRACT: The gastrointestinal structure, function and immunity of preterm infants are immature. Furthermore the gastrointestinal microbe colonization is abnormal. Therefore the preterm infants are prone to a variety of gastrointestinal diseases. Probiotics can regulate gastrointestinal microbe constitute, improve gastrointestinal barrier function, reduce gastrointestinal inflammation response and regulate the immunity. At present, it is used for the prevention of necrotizing enterocolitis, late-onset sepsis, and feeding intolerance. The safety and efficacy of probiotics for preterm infants are still controversial.

Gao, X., Jia, R., Xie, L., Kuang, L., Feng, L., and Wan, C. Comparing three preparation methods of standard substances for intestinal bifidobacteria of childhood obesity. Acta Paediatr. 5-5-2016; PM: 27149162

ABSTRACT: Bifidobacteria are normal intestinal flora that play a physiological role in maintaining intestinal microecology balance, biological antagonism, immune regulation and nutrition (1,2). In recent years, bifidobacteria and probiotics have been widely used in medical treatment, healthcare and food (1-3) and have increasingly become the academic focus of microecology. We published a study that showed that childhood

obesity was connected with the change in the proportion of intestinal bifidobacteria (4). This article is protected by copyright. All rights reserved.

Gao, X., Huang, L., Zhu, L., Mou, C., Hou, Q., and Yu, Q. Inhibition of H9N2 Virus Invasion into Dendritic Cells by the S-Layer Protein from L. acidophilus ATCC 4356. Front Cell Infect.Microbiol. 2016; 6:137; PM: 27826541

ABSTRACT: Probiotics are essential for the prevention of virus invasion and the maintenance of the immune balance. However, the mechanism of competition between probiotics and virus are unknown. The objectives of this study were to isolate the surface layer (S-layer) protein from L. acidophilus ATCC 4356 as a new antiviral material, to evaluate the stimulatory effects of the S-layer protein on mouse dendritic cells (DCs) and to verify its ability to inhibit the invasion of H9N2 avian influenza virus (AIV) in DCs. We found that the S-layer protein induced DCs activation and up-regulated the IL-10 secretion. The invasion and replication of the H9N2 virus in mouse DCs was successfully demonstrated. However, the invasion of H9N2 virus into DCs could be inhibited by treatment with the S-layer protein prior to infection, which was verified by the reduced hemagglutinin (HA) and neuraminidase (NA) mRNA expression, and nucleoprotein (NP) protein expression in the DCs. Furthermore, treatment with the S-layer protein increases the Mx1, Isg15, and Ddx58 mRNA expressions, and remits the inflammatory process to inhibit H9N2 AIV infection. In conclusion, the S-layer protein stimulates the activation of mouse DCs, inhibits H9N2 virus invasion of DCs, and stimulates the IFN-I signaling pathway. Thus, the S-layer protein from Lactobacillus is a promising biological antiviral material for AIV prevention.

Garcia-Hernandez, Y., Perez-Sanchez, T., Boucourt, R., Balcazar, J. L., Nicoli, J. R., Moreira-Silva, J., Rodriguez, Z., Fuertes, H., Nunez, O., Albelo, N., and Halaihel, N. Isolation, characterization and evaluation of probiotic lactic acid bacteria for potential use in animal production. Res.Vet.Sci. 2016; 108:125-132; PM: 27663381

ABSTRACT: In livestock production, lactic acid bacteria (LAB) are the most common microorganisms used as probiotics. For such use, these bacteria must be correctly identified and characterized to ensure their safety and efficiency. In the present study, LAB were isolated from broiler excreta, where a fermentation process was used. Nine among sixteen isolates were identified by biochemical and molecular (sequencing of the 16S rRNA gene) methods as Lactobacillus crispatus (n=1), Lactobacillus pentosus (n=1), Weissella cibaria (n=1), Pediococcus pentosaceus (n=2) and Enterococcus hirae (n=4). Subsequently, these bacteria were characterized for their growth capabilities, lactic acid production, acidic pH and bile salts tolerance, cell surface hydrophobicity, antimicrobial susceptibility and antagonistic activity. Lactobacillus pentosus strain LB-31, which showed the best characteristics, was selected for further analysis. This strain was administered to broilers and showed the ability of modulating the immune response and producing beneficial effects on morpho-physiological, productive and health indicators of the animals.

Gargari, G., Taverniti, V., Balzaretti, S., Ferrario, C., Gardana, C., Simonetti, P., and Guglielmetti, S. Consumption of a Bifidobacterium bifidum Strain for 4 Weeks Modulates Dominant Intestinal Bacterial Taxa and Fecal Butyrate in Healthy Adults. Appl.Environ.Microbiol. 10-1-2016; 82(19):5850-5859; PM: 27451450

ABSTRACT: Modulation of the intestinal microbial ecosystem (IME) is a useful target to establish probiotic efficacy in a healthy population. We conducted a randomized, double-blind, crossover, and placebo-controlled intervention study to determine the impact of Bifidobacterium bifidum strain Bb on the IME of adult healthy volunteers of both sexes. High-throughput 16S rRNA gene sequencing was used to characterize the fecal microbiota before and after 4 weeks of daily probiotic cell consumption. The intake of approximately one billion live B. bifidum cells affected the relative abundance of dominant taxa in the fecal microbiota and modulated fecal butyrate levels. Specifically, Prevotellaceae (P = 0.041) and Prevotella (P = 0.034) were significantly decreased, whereas Ruminococcaceae (P = 0.039) and Rikenellaceae (P = 0.010) were significantly increased. We also observed that the probiotic intervention modulated the fecal concentrations of butyrate in a manner dependent on the initial levels of short-chain fatty acids (SCFAs). In conclusion, our study demonstrates that a single daily administration of Bifidobacterium bifidum strain Bb can significantly modify the IME in healthy (not diseased) adults. These findings demonstrate the need to reassess the notion that probiotics do not influence the complex and stable IME of a healthy individual. IMPORTANCE: Foods and supplements claimed to contain health-promoting probiotic microorganisms are everywhere these days and mainly intended for consumption by healthy people. However, it is still debated what actual effects probiotic products may have on the healthy population. In this study, we report the results of an intervention trial aimed at assessing the modifications induced in the intestinal microbial ecosystem of healthy adults from the consumption of a probiotic product. Our results demonstrate that the introduction of a probiotic product in the dietary habits of healthy people may significantly modify dominant taxa of the intestinal microbiota, resulting in the modulation of short-chain fatty acid concentrations in the gut. The overall changes witnessed in the probiotic intervention indicate a mechanism of microbiota modulation that could have potential effects on human health.

Gasbarrini, G., Bonvicini, F., and Gramenzi, A. Probiotics History. J.Clin.Gastroenterol. 2016; 50 Suppl 2, Proceedings from the 8th Probiotics, Prebiotics & New Foods for Microbiota and Human Health meeting held in Rome, Italy on September 13-15, 2015:S116-S119; PM: 27741152

ABSTRACT: Gut microbiota promotes healthy effects on the host and prevents diseases. Probiotic (probios, for life) are defined as "live microorganisms which when administered in adequate amounts confer a health benefit on the host." At the beginning of 1900s Louis Pasteur identified the microorganisms responsible for the process of fermentation, whereas E. Metchnikoff associated the enhanced longevity of Bulgarian rural people to the regular consumption of fermented dairy products such as yogurt. He suggested that lactobacilli might counteract the putrefactive effects of gastrointestinal metabolism that contributed to illness and aging. Hippocrates declared, 2000 years earlier, that "death sits in the bowels." Metchnikoff considered the lactobacilli as probiotics ("probios," conducive to life of the host as opposed to antibiotics); probiotics could have a positive influence on health and prevent aging. During the neolitic period of the age of the stone, the domestication of animals occurred and man began to get fermented food.

Probably serendipitous contaminations in favorable environments played a major role. Fecal microbiota transplantation dates to a fourth-century Chinese handbook for food poisoning or severe diarrhea. To date fecal transplant cures Clostridium difficile infections with more efficacy than vancomycin, and prevents recurrence.

Geldart, K., Forkus, B., McChesney, E., McCue, M., and Kaznessis, Y. N. pMPES: A Modular Peptide Expression System for the Delivery of Antimicrobial Peptides to the Site of Gastrointestinal Infections Using Probiotics. Pharmaceuticals.(Basel). 10-5-2016; 9(4); PM: 27782051

ABSTRACT: Antimicrobial peptides are a promising alternative to traditional antibiotics, but their utility is limited by high production costs and poor bioavailability pro fi les. Bacterial production and delivery of antimicrobial peptides (AMPs) directly at the site of infection may offer a path for effective therapeutic application. In this study, we have developed a vector that can be used for the production and secretion of seven antimicrobial peptides from both Escherichia coli MC1061 F' and probiotic E.coli Nissle 1917. The vector pMPES (Modular Peptide Expression System) employs the Microcin V (MccV) secretion system and a powerful synthetic promoter to drive AMP production. Herein, we demonstrate the capacity of pMPES to produce inhibitory levels of MccV, Microcin L (MccL), Microcin N (McnN), Enterocin A (EntA), Enterocin P (EntP), Hiracin JM79 (HirJM79) and Enterocin B (EntB). To our knowledge, this is the fi rst demonstration of such a broadly-applicable secretion system for AMP production. This type of modular expression system could expedite the development of sorely needed antimicrobial technologies.

Genis, S., Sanchez-Chardi, A., Bach, A., Fabregas, F., and Aris, A. A combination of lactic acid bacteria regulates Escherichia coli infection and inflammation of the bovine endometrium. J.Dairy Sci. 11-9-2016; PM: 27837977

ABSTRACT: Uterine function in cattle is compromised by bacterial contamination and inflammation after calving. The objective of this study was to select a combination of lactic acid bacteria (LAB) to decrease endometrium inflammation and Escherichia coli infection. Primary endometrial epithelial cells were cultured in vitro to select the most favorable LAB combination modulating basal tissue inflammation and E. coli infection. Supernatants were obtained to determine expression of pro-inflammatory cytokines, and E. coli infection was evaluated after harvesting the tissue and plate counting. The selected LAB combination was tested in uterus explants to assess its capacity to modulate basal and acute inflammation (associated with E. coli infection). The combination of Lactobacillus rhamnosus, Pediococcus acidilactici, and Lactobacillus reuteri at a ratio of 25:25:2, respectively, reduced E. coli infection in vitro with (89.77%) or without basal tissue inflammation (95.10%) compared with single LAB strains. Lactic acid bacteria treatment reduced CXCL8 and IL1B expression 4.7- and 2.2-fold, respectively, under acute inflammation. Ex vivo, the tested LAB combination reduced acute inflammation under E. coli infection, decreasing IL-8, IL-1beta, and IL-6 up to 2.2-, 2.5-, and 2.2-fold, respectively. In the total inflammation model, the LAB combination decreased IL-8 1.6-fold and IL-6 1.2-fold. Ultrastructural evaluation of the tissue suggested no direct interaction between the LAB and E. coli, although pathological effects of E. coli in endometrial cells were greatly diminished or even reversed by the LAB combination. This study shows the promising potential of LAB probiotics for therapeutic use against endometrial inflammation and infection.

Giangioppo, S., Kalaci, O., Radhakrishnan, A., Fleischer, E., Itterman, J., Lyttle, B., Price, A., and Radhakrishnan, D. Complementary and alternative medicine use in children with cystic fibrosis. Complement Ther.Clin.Pract. 2016; 25:68-74; PM: 27863612

ABSTRACT: ***PURPOSE***: To estimate the overall prevalence of complementary and alternative medicine use among children with cystic fibrosis, determine specific modalities used, predictors of use and subjective helpfulness or harm from individual modalities. ***RESULTS***: Of 53 children attending the cystic fibrosis clinic in London, Ontario (100% recruitment), 79% had used complementary and alternative medicine. The most commonly used modalities were air purifiers, humidifiers, probiotics, and omega-3 fatty acids. Family complementary and alternative medicine use was the only independent predictor of overall use. The majority of patients perceived benefit from specific modalities for cystic fibrosis symptoms. ***CONCLUSIONS***: Given the high frequency and number of modalities used and lack of patient and disease characteristics predicting use, we recommend that health care providers should routinely ask about complementary and alternative medicine among all pediatric cystic fibrosis patients and assist patients in understanding the potential benefits and risks to make informed decisions about its use.

Giglione, E., Prodam, F., Bellone, S., Monticone, S., Beux, S., Marolda, A., Pagani, A., Di, Gioia D., Del, Piano M., Mogna, G., and Bona, G. The Association of Bifidobacterium breve BR03 and B632 is Effective to Prevent Colics in Bottle-fed Infants: A Pilot, Controlled, Randomized, and Double-Blind Study. J.Clin.Gastroenterol. 2016; 50 Suppl 2, Proceedings from the 8th Probiotics, Prebiotics & New Foods for Microbiota and Human Health meeting held in Rome, Italy on September 13-15, 2015:S164-S167; PM: 27741166

ABSTRACT: ***GOALS***: To assess the effectiveness of Bifidobacterium breve B632 and BR03 association in the reduction of infants crying over time. The second endpoint was to observe the effect of the same strains on daily evacuations and on the number of regurgitations and vomits. ***BACKGROUND***: Infant colics represent a clinical condition in childhood, characterized by an uncontrollable crying that occurs without any apparent organic cause. An altered intestinal microbiota composition in the very first months may induce intestinal colics in infants. Thus far, no treatment is really effective for this problem, but recent literature shows an increasing attention toward probiotics. ***STUDY***: A total of 83 subjects were enrolled, 60 breastfed infants and 23 bottle-fed infants. Sixty of them carried out the study: 29 infants were given probiotics, whereas 31 placebo. During the 90 days of the study, parents were asked to give 5 drops of active product (10 viable cells/strain) or placebo and to daily take note of: minutes of crying, number, color, and consistency of evacuations, and number of regurgitations or vomits. ***RESULTS***: No significant differences were detected in the infants treated with probiotics, compared with placebo group (P=0.75). The analysis of the 3 months of treatment demonstrated that during the third month, the probiotic group cried 12.14 minutes on average and the placebo cried 46.65 minutes. This difference is statistically significant (P=0.016). ***CONCLUSIONS***: The evidence of the usefulness of some probiotic strains in the treatment and prevention of infant colics is growing, and therefore their use in clinical practice is spreading.

Gisbert, J. P. Helicobacter pylori-related diseases. Gastroenterol.Hepatol. 2016; 39 Suppl 1:36-46; PM: 27888863

ABSTRACT: This article describes the main conclusions drawn from the presentations on Helicobacter pylori infection in Digestive Diseases Week, 2016. Despite the undeniable widespread reduction in the prevalence of this infection, infection rates continue to be high in developing countries. The prevalence of clarithromycin, metronidazole and quinolone resistance is markedly high in most countries and continues to rise. The management of H. pylori infection in patients with peptic ulcers still leaves much to be desired. Although H. pylori eradication reduces the incidence of gastric adenocarcinoma, it does not completely avoid its appearance. The new rapid stool antigen tests show promising results. The efficacy of standard triple therapy is clearly inadequate and continues to decline, and cannot therefore be recommended. Vonoprazan, when associated with 2 antibiotics, is more effective than traditional proton pump inhibitors, especially in clarithromycin-resistant patients. Non-bismuth quadruple (concomitant) therapy achieves eradication rates of around 90% and has a good safety profile. Concomitant therapy is more effective and simpler than sequential therapy. Although some probiotics can increase the efficacy and tolerability of triple therapy, the utility of its association with quadruple concomitant therapy has not been established. If a first treatment with clarithromycin fails, both bismuth-containing quadruple therapy and levofloxacin-containing triple therapy achieve good-but still suboptimal-results. The combination of bismuth and levofloxacin in the same regimen increases the efficacy of rescue therapy. The management of H. pylori infection by European gastroenterologists is widely heterogeneous and the eradication rates achieved by them are generally unacceptable. In Spain, the highest first-line eradication rate is obtained with quadruple concomitant therapy in 14-day regimens and with double doses of proton pump inhibitors; in second-line therapy, the use of levofloxacin- and bismuth-containing quadruple therapy improves the rates traditionally obtained with quinolones. On the other hand, there is a clear disconnect between the recommendations made in consensus documents and clinical practice in primary care.

Gomez-Arango, L. F., Barrett, H. L., McIntyre, H. D., Callaway, L. K., Morrison, M., and Dekker, Nitert M. Increased Systolic and Diastolic Blood Pressure Is Associated With Altered Gut Microbiota Composition and Butyrate Production in Early Pregnancy. Hypertension. 2016; 68(4):974-981; PM: 27528065

ABSTRACT: The risk of developing pregnancy-induced hypertension and preeclampsia is higher in obese pregnant women. In obesity, the composition of the gut microbiota is altered. Obesity is also associated with low-grade inflammation. Metabolites from the gut microbiota may contribute to both hypertension and inflammation. The aim of this study is to investigate whether the composition of the gut microbiota in overweight and obese pregnant women is associated with blood pressure and levels of plasminogen activator inhibitor-1. The composition of the gut microbiota was determined with 16S ribosomal RNA sequencing in 205 women at 16 weeks gestation from the SPRING study (the Study of Probiotics in Gestational Diabetes). Expression of butyrate-producing genes in the gut microbiota was assessed by real-time polymerase chain reaction. Plasminogen activator inhibitor-1 levels were measured in fasting serum of a subset of 70 women. Blood pressure was slightly but significantly higher in obese compared with overweight women. The abundance of the butyrate-producing genus Odoribacter was inversely correlated with systolic blood pressure. Butyrate production capacity was decreased, but plasminogen activator inhibitor-1 concentrations increased in obese pregnant women. Plasminogen activator inhibitor-1 levels were inversely correlated with expression of butyrate kinase and Odoribacter abundance. This study shows that in overweight and obese pregnant

women at 16 weeks gestation, the abundance of butyrate-producing bacteria and butyrate production in the gut microbiota is significantly negatively associated with blood pressure and with plasminogen activator inhibitor-1 levels. Increasing butyrate-producing capacity may contribute to maintenance of normal blood pressure in obese pregnant women.

Gorska, S., Sandstrom, C., Wojas-Turek, J., Rossowska, J., Pajtasz-Piasecka, E., Brzozowska, E., and Gamian, A. Structural and immunomodulatory differences among lactobacilli exopolysaccharides isolated from intestines of mice with experimentally induced inflammatory bowel disease. Sci.Rep. 11-21-2016; 6:37613; PM: 27869188

ABSTRACT: Characteristic changes in the microbiota biostructure and a decreased tolerance to intestinal bacteria have been associated with inflammatory bowel disease (IBD). However, few studies have examined the constituents of the intestinal microbiota, including the surface molecules of the bacteria, in healthy and IBD subsets. Here, we compare the chemical structures and immunomodulatory properties of the exopolysaccharides (EPS) of lactobacilli isolated from mice with induced IBD (IBD "+") versus those of healthy mice (IBD "-"). Classical structural analyses were performed using nuclear magnetic resonance spectroscopy and mass spectrometry. Immunomodulatory properties were assessed by stimulation of dendritic cells derived from mouse bone marrow or human peripheral mononuclear blood cells. Our results revealed that EPS produced by IBD "+" species are structurally different from those isolated from IBD "-". Moreover, the structurally different EPS generate different immune responses by dendritic cells. We speculate that resident strains could, upon gut inflammation, switch to producing EPS with specific motifs that are absent from lactobacilli IBD "-", and/or that bacteria with a particular EPS structure might inhabit the inflamed intestinal mucosa. This study may shed light on the role of EPS in IBD and help the development of a specific probiotic therapy for this disease.

Gorska, S., Dylus, E., Rudawska, A., Brzozowska, E., Srutkova, D., Schwarzer, M., Razim, A., Kozakova, H., and Gamian, A. Immunoreactive Proteins of Bifidobacterium longum ssp. longum CCM 7952 and Bifidobacterium longum ssp. longum CCDM 372 Identified by Gnotobiotic Mono-Colonized Mice Sera, Immune Rabbit Sera and Non-immune Human Sera. Front Microbiol. 2016; 7:1537; PM: 27746766

ABSTRACT: The Bifidobacteria show great diversity in the cell surface architecture which may influence the physicochemical properties of the bacterial cell and strain specific properties. The immunomodulatory role of bifidobacteria has been extensively studied, however studies on the immunoreactivity of their protein molecules are very limited. Here, we compared six different methods of protein isolation and purification and we report identification of immunogenic and immunoreactive protein of two human Bifidobacterium longum ssp. longum strains. We evaluated potential immunoreactive properties of proteins employing polyclonal sera obtained from germ free mouse, rabbit and human. The protein yield was isolation method-dependent and the reactivity of proteins detected by SDS-PAGE and Western blotting was heterogeneous and varied between different serum samples. The proteins with the highest immunoreactivity were isolated, purified and have them sequenced. Among the immunoreactive proteins we identified enolase, aspartokinase, pyruvate kinase, DnaK (B. longum ssp. longum CCM 7952) and sugar ABC transporter ATP-binding protein, phosphoglycerate kinase,

peptidoglycan synthethase penicillin-binding protein 3, transaldolase, ribosomal proteins and glyceraldehyde 3-phosphate dehydrogenase (B. longum ssp. longum CCDM 372).

Goudarzvand, M., Rasouli, Koohi S., Khodaii, Z., and Soleymanzadeh, Moghadam S. Probiotics Lactobacillus plantarum and bifidobacterium B94: cognitive function in demyelinated model. Med.J.Islam Repub.Iran. 2016; 30:391; PM: 27579282

ABSTRACT: ***BACKGROUND***: Multiple Sclerosis (MS) is a disease of the immune system that creates damage of Learning and memory in that. Using probiotic supplements is recommended for preventing MS disease and improving memory. This study aimed to investigate the effect of Lactobacillus plantarum (LP) and bifidobacterium B94 (BB94), on acquisition phase of spatial memory in the local demyelination of rats` hippocampus. ***METHODS***: In this study, 32 male Wistar rats were divided into control, damage group and treatment groups. Treatment groups were including (LP) and (BB94). After the induction of demyelination by 3 mul of EB into the right dentate gyrus of the hippocampus in treatment groups, 1.5x10(8) probiotic bacteria were administered by gavage for 28 days. Data was analyzed using one-way ANOVA and Tukey post-hoc tests (p</=0.05). ***RESULTS***: Findings demonstrated that injection of EB caused a significant increase in traveled distance (p<0.01) and also escape latency (p<0.05) compared with control group. Also, effect administrations of (LP) and (BB94) on traveled distance and escape latency were reviewed, and it was determined that administration of them do not cause significant reduction in the traveled distance compared with the lesion group. Also mentioned probiotics has no significant effect on swimming speed compared with lesion and saline groups. ***CONCLUSION***: According to some studies, probiotics have a positive impact on improving the performance of spatial memory and learning, although the results of the current study could not indicate finality of this assumption. It seems that more researches is needed on this subject.

Govender, M., Choonara, Y. E., van, Vuuren S., Kumar, P., du Toit, L. C., and Pillay, V. Design and evaluation of an oral multiparticulate system for dual delivery of amoxicillin and Lactobacillus acidophilus. Future.Microbiol. 2016; 11:1133-1145; PM: 27545143

ABSTRACT: **AIM**: A delayed-release dual delivery system for amoxicillin and the probiotic Lactobacillus acidophilus was developed and evaluated. ***MATERIALS & METHODS***: Statistical optimization of a cross-linked denatured ovalbumin protective matrix was first synthesized using a Box-Behnken experimental design prior to encapsulation with glyceryl monostereate. The encapsulated ovalbumin matrix was thereafter incorporated with amoxicillin in a gastro-resistant capsule. In vitro characterization and stability analysis of the ovalbumin and encapsulated components were also performed. ***RESULTS***: Protection of L. acidophilus probiotic against the bactericidal effects of amoxicillin within the dual formulation was determined. ***CONCLUSION***: The dual formulation in this study proved effective and provides insight into current microbiome research to identify, classify and use functional healthy bacteria to develop novel probiotic delivery technologies.

Govender, M., Choonara, Y. E., van, Vuuren S., Kumar, P., du Toit, L. C., Erlwanger, K., and Pillay, V. A Dual-Biotic System for the Concurrent Delivery of Antibiotics and Probiotics: In Vitro, Ex Vivo, In Vivo and In Silico Evaluation and Correlation. Pharm.Res. 2016; 33(12):3057-3071; PM: 27599992

ABSTRACT: ***PURPOSE***: A delayed release bio-polymeric Dual-Biotic system has been extensively evaluated in this study to overcome the therapeutic issue of probiotic killing due to incorrect administration with the antibiotic. ***METHODS***: In vitro and ex vivo release and characterization studies have been undertaken on the Dual-Biotic system. In vivo analyses utilizing a Large White pig model were also performed with commercial products used as a comparison. Intestinal fluid for probiotic quantification was aspirated using a surgically implanted intestinal cannula with Lactobacillus acidophilus cell counts determined through luminescence and inoculation onto Lactobacilli-specific agar. Plasma amoxicillin concentrations were determined through Ultra-Performance Liquid Chromatography. The reactional profile and crosslinking mechanism of ovalbumin and genipin was elucidated using molecular mechanic energy relationships in a vacuum system by exploring the spatial disposition of different concentrations of genipin with respect to ovalbumin with ovalbumin/genipin ratios of 1:1, 1:5 and 1:10. ***RESULTS***: In vivo evaluation of the Dual-Biotic system detailed maximum Lactobacillus viability (~455% baseline viability) 6 h after oral administration. Concurrent administration of the commercial products revealed a 75% decrease in bacterial viability when compared to the controls analyzed. A level A in vitro-in vivo correlation was also established with 96.9% predictability of amoxicillin release ascertained. The computational results achieved corroborated well with the experimental findings and physicochemical data. ***CONCLUSIONS***: Evaluation and correlation of the Dual-Biotic system has detailed the success of the formulation for the concurrent delivery of an antibiotic and probiotic.

Graham, K., Rea, R., Simpson, P., and Stack, H. Development of a rapid, one-step screening method for the isolation of presumptive proteolytic enterococci. J.Microbiol.Methods. 11-25-2016; 132:99-105; PM: 27894832

ABSTRACT: Enterococci show higher proteolytic activities than other lactic acid bacteria and thus have received considerable attention in scientific literature in recent years. Proteolytic enzymes of enterococci have warranted the use of some species as starter, adjuncts or protective cultures and as probiotics, while in some strains they have also been linked with virulence. Consequently, the isolation and identification of proteolytic enterococci is becoming of increasing interest and importance. However, current screening methods for proteolytic enterococci can be time consuming, requiring a two-step procedure which may take up to 96h. This study describes a method, utilising Kanamycin Skim Milk Aesculin Azide (KSMEA) agar, for the isolation of proteolytic enterococci in one-step, thereby significantly reducing screening time. KSMEA combines the selective properties of Kanamycin Aesculin Azide Agar (KAA) with skim milk powder for the detection of proteolytic enterococci. Enterococci produced colonies with a black halo on KSMEA which were accompanied by a zone of clearing in the media when enterococci were proteolytic. KSMEA medium retained the selectivity of KAA, while proteolytic enterococci were easily distinguished from non-proteolytic enterococci when two known strains were propagated on KSMEA. KSMEA also proved effective at isolating and detecting enterococci in raw milk, faeces and soil. Isolates recovered from the screen were confirmed as enterococci using genus-specific primers. Proteolytic enterococci were present in the raw milk sample only and were easily distinguishable from non-proteolytic enterococci and other microorganisms. Therefore, KSMEA provides a rapid, one-step screening method for the isolation of presumptive proteolytic enterococci.

Green, Corkins K. and Shurley, T. What's in the Bottle? A Review of Infant Formulas. Nutr.Clin.Pract. 9-19-2016; PM: 27646861

ABSTRACT: Infant formulas are designed to be a substitute for breast milk. Since they are sole source of nutrition for growing and developing infants, they are highly regulated by the government. All ingredients in infant formulas must be considered "generally recognized as safe." Manufacturers are continually modifying their products to make them more like breast milk. Functional ingredients added to infant formula include long-chain polyunsaturated fatty acids, nucleotides, prebiotics, and probiotics. The most common breast milk substitutes are standard cow's milk-based term infant formulas, which include subcategories of organic and breast milk supplementation, and come in standard dilutions of 19 or 20 calories per ounce. In addition to standard cow's milk-based term infant formulas, there is a line of term infant formulas marketed for signs and symptoms of intolerance. These products include modifications in lactose content, partially hydrolyzed protein, added probiotics, or added rice starch. There are also specialized formulas for medical conditions such as prematurity, gastrointestinal disorders, allergy, disorders of fat metabolism, and renal insufficiency. Infants on specialty formulas should be monitored closely by medical professionals. Formulas come in ready-to-feed, liquid concentrate, and powder forms. Each offers advantages and disadvantages. Each step in the formula mixing process or each manipulation required for the feeding is another opportunity to introduce bacteria to the formula. There are guidelines for preparing formula in institutions. Standard dilution and mixing instructions are different for each formula, so individual recipes are needed. Caregivers should also be educated on proper hygiene when preparing formula at home.

Greppi, A., Saubade, F., Botta, C., Humblot, C., Guyot, J. P., and Cocolin, L. Potential probiotic Pichia kudriavzevii strains and their ability to enhance folate content of traditional cereal-based African fermented food. Food Microbiol. 2017; 62:169-177; PM: 27889145

ABSTRACT: With the aim of selecting starter cultures with interesting probiotic potential and with the ability to produce folate in a food matrix, yeast strains isolated from fermented cereal-based African foods were investigated. A total of 93 yeast strains were screened for their tolerance to pH 2 and 0.3% of bile salts. Pichia kudriavzevii isolates gave the best results. Selected P. kudriavzevii strains were tested for survival to the simulated human digestion and for adhesion to Caco-2 cells. Moreover, presence of folate biosynthesis genes was verified and production of extra and intra-cellular folate determined during growth in culture medium. 31% of yeast strains could tolerate pH 2, while 99% bile salts. Survival rate after simulated digestion ranged between 11 and 45%, while adhesion rate between 12 and 40%. Folate production was mainly intracellular, maximum after 24 h of growth. To be closer to traditional cereal-based fermentations, a P. kudriavzevii strain with good probiotic potential was co-inoculated with Lactobacillus fermentum strains in a pearl millet gruel. This resulted in in situ folate production that peaked after 4 h. The use of strains with both probiotic and nutritional enrichment properties may have a greater impact for the consumers.

Grimshaw, K., Logan, K., O'Donovan, S., Kiely, M., Patient, K., van, Bilsen J., Beyer, K., Campbell, D. E., Garcia-Larsen, V., Grabenhenrich, L., Lack, G., Mills, C., Wal, J. M., and Roberts, G.

Modifying the infant's diet to prevent food allergy. Arch.Dis.Child. 8-16-2016; PM: 27530541

ABSTRACT: Recommendations and guidelines on the prevention of food allergy have changed in recent decades. The aim of this review of the current evidence and ongoing studies is to provide a comprehensive and up to date picture of prevention of food allergy for healthcare professionals. The review was undertaken as part of the European Union funded Integrated Approaches to Food Allergy and Allergen Management (iFAAM) study. This is a wide ranging project bringing together expertise across the breadth of food allergy research. Specifically, the review discusses dietary manipulation in food allergy prevention, and covers the possible preventive strategies of allergen avoidance, early allergen introduction, general nutrition and supplements, as well as other strategies, such as prebiotics and probiotics. The review concludes that despite agreement that allergen avoidance strategies should not be undertaken for allergy prevention, there is currently no consensus regarding what actions should be recommended beyond exclusive breastfeeding for the first 4-6 months of life. Recent and upcoming trial results, which are detailed in this review, should help inform the debate and add clarity to the topic.

Grossi, E., Melli, S., Dunca, D., and Terruzzi, V. Unexpected improvement in core autism spectrum disorder symptoms after long-term treatment with probiotics. SAGE Open.Med.Case.Rep. 2016; 4:2050313X16666231; PM: 27621806

ABSTRACT: ***OBJECTIVES***: Autism spectrum disorder is a neurodevelopmental condition that typically displays socio-communicative impairment as well as restricted stereotyped interests and activities, in which gastrointestinal disturbances are commonly reported. We report the case of a boy with Autism Spectrum Disorder (ASD) diagnosis, severe cognitive disability and celiac disease in which an unexpected improvement of autistic core symptoms was observed after four months of probiotic treatment. **METHODS**: The case study refers to a 12 years old boy with ASD and severe cognitive disability attending the Villa Santa Maria Institute in resident care since 2009. Diagnosis of ASDs according to DSM-V criteria was confirmed by ADOS-2 assessment (Autism Diagnostic Observation Schedule). The medication used was VSL#3, a multi-strain mixture of ten probiotics. The treatment lasted 4 weeks followed by a four month follow-up. The rehabilitation program and the diet was maintained stable in the treatment period and in the follow up. ADOS-2 was assessed six times: two times before starting treatment; two times during the treatment and two times after interruption of the treatment. ***RESULTS***: The probiotic treatment reduced the severity of abdominal symptoms as expected but an improvement in Autistic core symptoms was unexpectedly clinically evident already after few weeks from probiotic treatment start. The score of Social Affect domain of ADOS improved changing from 20 to 18 after two months treatment with a further reduction of 1 point in the following two months. The level 17 of severity remained stable in the follow up period. It is well known that ADOS score does not fluctuate spontaneously along time in ASD and is absolutely stable. ***CONCLUSIONS***: The appropriate use of probiotics deserves further research, which hopefully will open new avenues in the fight against ASD.

Haghighat, L. and Crum-Cianflone, N. F. The potential risks of probiotics among HIV-infected persons: Bacteraemia due to Lactobacillus acidophilus and review of the literature. Int.J.STD AIDS. 2016; 27(13):1223-1230; PM: 26130690

ABSTRACT: Lactobacillus sp. are commensal organisms that are increasingly reported to cause invasive infections among immunosuppressed persons. However, few data exist regarding the occurrence and risk factors of these infections among HIV-infected persons. Further, the safety of products that contain lactobacilli (e.g. probiotics) in certain populations, including those with HIV/AIDS, is unclear. We report a case of Lactobacillus acidophilus bacteraemia in a patient with AIDS temporally related to excessive consumption of probiotic-enriched yogurt, and provide a comprehensive review of the literature of Lactobacillus sp. infections among HIV-infected persons.

Haldar, L. and Gandhi, D. N. Effect of oral administration of Bacillus coagulans B37 and Bacillus pumilus B9 strains on fecal coliforms, Lactobacillus and Bacillus spp. in rat animal model. Vet.World. 2016; 9(7):766-772; PM: 27536040

ABSTRACT: ***AIM***: To investigate the effect of oral administration of two Bacillus strains on fecal coliforms, Lactobacillus and Bacillus spp. in rat animal model. MATERIALS AND ***METHODS***: An in vivo experiment was conducted for 49-day period on 36 adult male albino Wister rats divided equally into to four groups. After 7-day adaptation period, one group (T1) was fed on sterile skim milk along with basal diet for the next 28 days. Second (T2) and (T3) groups received spore biomass of Bacillus coagulans B37 and Bacillus pumilus B9, respectively, suspended in sterilized skim milk at 8-9 log colony-forming units/ml plus basal diet for 28 days, while control group (T4) was supplied with clean water along with basal diet. There was a 14-day post-treatment period. A total of 288 fecal samples (8 fecal collections per rat) were collected at every 7-day interval starting from 0 to 49 days and subjected to the enumeration of the counts of coliforms and lactobacilli and Bacillus spores using respective agar media. In vitro acid and bile tolerance tests on both the strains were performed. ***RESULTS***: The rats those (T2 and T3) received either B. coagulans B37 or B. pumilus B9 spore along with non-fermented skim milk showed decrease ($p<0.01$) in fecal coliform counts and increase ($p<0.05$) in both fecal lactobacilli and Bacillus spore counts as compared to the control group (T4) and the group fed only skim milk (T1). In vitro study indicated that both the strains were found to survive at pH 2.0 and 3.0 even up to 3 h and tolerate bile up to 2.0% concentration even after 12 h of exposure. ***CONCLUSIONS***: This study revealed that oral administration of either B. coagulans B37 or B. pumilus B9 strains might be useful in reducing coliform counts accompanied by concurrent increase in lactobacilli counts in the intestinal flora in rats.

Halkjaer, S. I., Nilas, L., Carlsen, E. M., Cortes, D., Halldorsson, T. I., Olsen, S. F., Pedersen, A. E., Krogfelt, K. A., and Petersen, A. M. Effects of probiotics (Vivomixx(R)) in obese pregnant women and their newborn: study protocol for a randomized controlled trial. Trials. 10-11-2016; 17(1):491; PM: 27724923

ABSTRACT: ***BACKGROUND***: Maternal obesity is associated with increased risks of adverse pregnancy-related complications and outcomes for both mothers and infants. Overweight and obese women have an increased risk of pregnancy-induced hypertension, preeclampsia and gestational diabetes mellitus (GDM). Infant Body Mass index (BMI) and the risk of obesity in adulthood are related to maternal gestational weight gain (GWG). Preventive lifestyle and dietary interventions are time-consuming and do not always reduce GWG or the risk of maternal pregnancy complications. Recent research has indicated that the gut microbiota may play a significant role in the development of obesity. Some studies have indicated that the daily consumption of

probiotics may reduce the risk of preeclampsia, maintain serum insulin levels and reduce the frequency of GDM in pregnant women. The aims of this study are to investigate whether daily probiotic supplements in obese women during pregnancy can limit gestational weight gain, improve glucose homeostasis and thereby improve maternal, fetal and infant health outcomes. ***METHODS***: A pilot study including 50 obese pregnant nulliparous women with a prepregnancy BMI of between 30 and 35 kg/m2 will be randomized to receive daily probiotics (four capsules of Vivomixx(R); total of 450 billion CFU/day, including eight probiotic bacterial strains) or placebo from gestational age 14-20 weeks until delivery. The infants will be followed until 9 months of age. The women will be monitored by weight, blood, fecal, vaginal and urine samples, diet questionnaires and hospital record review. Primary outcomes are: maternal weight gain, glycated hemoglobin (HbA1c) level and changes in glucose concentration measured during an oral glucose tolerance test. Secondary outcomes are: microbiota and inflammatory markers in mother and child, pregnancy complications, pregnancy outcomes, physical activity and the body composition of the neonate. ***DISCUSSION***: We expect to find alterations in the metabolic profiles, microbiota and possibly pregnancy outcomes. From a clinical point of view the effects of Vivomixx(R) could control weight gain and reduce complications during pregnancy by inducing changes in the gut microbiota. Furthermore, this intervention during pregnancy could influence the infant's microbiota, which could have important implications for infant development and health. ***TRIAL REGISTRATION***: ClincalTrials.gov Identifier: NCT02508844 , registered on 11 May 2015.

Harnett, J., Myers, S. P., and Rolfe, M. Probiotics and the Microbiome in Celiac Disease: A Randomised Controlled Trial. Evid.Based.Complement Alternat.Med. 2016; 2016:9048574; PM: 27525027

ABSTRACT: ***Background***: There is limited research investigating the composition of the gastrointestinal microbiota in individuals with celiac disease (CoeD) reporting only partial symptom improvement despite adherence to a strict gluten-free diet (GFD). The aim of this research was to determine if the gastrointestinal microbiota could be altered by probiotic bacteria and provide a potential new therapy for this subgroup. ***Methods***: A multicentre RCT was conducted between January and August 2011 in Australia. Participants included 45 people with CoeD reporting only partial symptom improvement despite adherence to a strict GFD for a minimum of 12 months. Participants took 5 g of VSL# probiotic formulation (n = 23) or 5 g placebo (n = 22) orally twice daily for 12 weeks. The main outcome measured was the efficacy of the probiotic formula in altering faecal microbiota counts between baseline and week 12. Safety was determined by safety blood and monitoring adverse events. ***Results***: SPSS multivariate repeated measures analysis (95th confidence level) revealed no statistically significant changes between the groups in the faecal microbiota counts or blood safety measures over the course of the study. ***Conclusion***: The probiotic formula when taken orally over the 12-week period did not significantly alter the microbiota measured in this population. The trial was registered with Australian and New Zealand Clinical Trials Register ACTRN12610000630011.

Harrell, C. S., Gillespie, C. F., and Neigh, G. N. Energetic stress: The reciprocal relationship between energy availability and the stress response. Physiol Behav. 11-1-2016; 166:43-55; PM: 26454211

ABSTRACT: The worldwide epidemic of metabolic syndromes and the recognized burden of mental health disorders have driven increased research into the relationship between

the two. A maladaptive stress response is implicated in both mental health disorders and metabolic disorders, implicating the hypothalamic-pituitary-adrenal (HPA) axis as a key mediator of this relationship. This review explores how an altered energetic state, such as hyper- or hypoglycemia, as may be manifested in obesity or diabetes, affects the stress response and the HPA axis in particular. We propose that changes in energetic state or energetic demands can result in "energetic stress" that can, if prolonged, lead to a dysfunctional stress response. In this review, we summarize the role of the hypothalamus in modulating energy homeostasis and then briefly discuss the relationship between metabolism and stress-induced activation of the HPA axis. Next, we examine seven mechanisms whereby energetic stress interacts with neuroendocrine stress response systems, including by glucocorticoid signaling both within and beyond the HPA axis; by nutrient-induced changes in glucocorticoid signaling; by impacting the sympathetic nervous system; through changes in other neuroendocrine factors; by inducing inflammatory changes; and by altering the gut-brain axis. Recognizing these effects of energetic stress can drive novel therapies and prevention strategies for mental health disorders, including dietary intervention, probiotics, and even fecal transplant.

Hashizume, N., Yagi, M., Ushijima, K., Seki, Y., Fukahori, S., Muto, M., Matsufuji, H., and Taguchi, T. Pharmacotherapy in patients with pediatric CIPO -a nationwide survey in Japan. Pediatr.Int. 11-12-2016; PM: 27862707

ABSTRACT: ***PURPOSE***: Chronic intestinal pseudo-obstruction (CIPO) is a rare disabling and life-threatening disorder characterized by a severe impairment of gastrointestinal peristalsis. With respect to pharmacotherapy, while a number of agents have been developed, only a few trials have attempted to improve the pathological condition of patients with CIPO. This report presents the actual findings of pharmacotherapy in the patients with pediatric CIPO from the view point of a nationwide survey on the actual situation in Japan. ***METHODS***: In 2012, a nationwide survey was conducted to identify the clinical presentation of CIPO in Japan. Information was gathered on patient pharmacotherapy treatments. Four categories were created for medicines were administrered in pharmacotherapy: "probiotics", "Japanese herbal medicines (Kampo medicines)", "laxatives", and "prokinetics". ***RESULTS***: Ninety-two responses were collected from 47 facilities. Of the 62 cases that met the diagnostic criteria. Of these, 52 were treated with medications, while the remaining 10 were not. Thirty-four cases were administered a total of 49 probiotics. 39 cases were administered a total of 50 Kampo medicines, 20 cases were administered a total of 28 laxatives, and 26 cases were administered a total of 30 prokinetics, 70% of which were administered mosapride specifically. ***CONCLUSION***: Traditional Japanese medicines such as Kampo medicines and mosapride are often administered to treat CIPO in Japan.Two combined therapy such as probiotics and Kampo medicines, and Kampo medicines and prokinetics were often administered in the patients with pediatric CIPO in Japan. This article is protected by copyright. All rights reserved.

Hassan, S. T. and Sudomova, M. Probiotics as Dietary Supplements for Eradication of Helicobacter pylori Infection in Children: A Role Beyond Infection. Children.(Basel). 11-10-2016; 3(4); PM: 27834907

ABSTRACT: For decades, treatment of infectious diseases has been a strong focus of interest, for both researchers and healthcare providers. Chronic infection with Helicobacter pylori (H. pylori) has been reported to be associated with several diseases, such as ulcer disease, gastric adenocarcinoma and mucosa-associated lymphoid tissue

(MALT) lymphoma. Infection with H. pylori is generally acquired during childhood and can persist indefinitely, if not treated systematically. Unfortunately, although several strategies have shown high efficacy results, treatment of the H. pylori infection fails in about 25%-30% of infected children. One main reason for this is due to the extensive use of antibiotics, which has created antibiotic resistance, associated with other adverse effects as well. Therefore, it is crucial to find alternative strategies to combat this resistance, and increase treatment efficacy results. Probiotics, which are live microorganisms that are orally administrated, have been found to be a useful regimen in the treatment of the H. pylori infection in children. Their use as a dietary supplement alone, or in combination with antibiotics, resulted in reduced side effects and higher efficacy rates of the H. pylori infection in children. Some probiotics can be considered an adjunctive treatment, especially when eradication of the H. pylori infection fails during initial treatment, and to help reduce adverse effects. However, the evidence of the beneficial role of probiotics is limited due to the small number of clinical trials that have been conducted and heterogeneity across studies in strains and dosage. Additionally, no investigations have been carried out in asymptomatic children. Therefore, large well-conducted studies are needed to evaluate the efficacy and safety of probiotics as an adjuvant therapy of the H. pylori infection.

Hayes, S. R. and Vargas, A. J. Probiotics for the Prevention of Pediatric Antibiotic-Associated Diarrhea. Explore.(NY). 2016; 12(6):463-466; PM: 27688016

ABSTRACT: ***BACKGROUND***: Antibiotics are frequently prescribed in children. They alter the microbial balance within the gastrointestinal tract, commonly resulting in antibiotic-associated diarrhea (AAD). Probiotics may prevent AAD via restoration of the gut microflora. ***OBJECTIVES***: The primary objectives were to assess the efficacy and safety of probiotics (any specified strain or dose) used for the prevention of AAD in children. ***SEARCH METHODS***: MEDLINE, EMBASE, CENTRAL, CINAHL, AMED, and the Web of Science (inception to November 2014) were searched along with specialized registers including the Cochrane IBD/FBD review group, CISCOM (Centralized Information Service for Complementary Medicine), NHS Evidence, the International Bibliographic Information on Dietary Supplements, as well as trial registries. Letters were sent to authors of included trials, nutraceutical and pharmaceutical companies, and experts in the field requesting additional information on ongoing or unpublished trials. Conference proceedings, dissertation abstracts, and reference lists from included and relevant articles were also searched. ***SELECTION CRITERIA***: Randomized, parallel, controlled trials in children (0-18 years) receiving antibiotics, that compare probiotics to placebo, active alternative prophylaxis, or no treatment and measure the incidence of diarrhea secondary to antibiotic use were considered for inclusion. ***DATA COLLECTION AND ANALYSIS***: Study selection, data extraction, and methodological quality assessment using the risk of bias instrument were conducted independently and in duplicate by two authors. Dichotomous data (incidence of diarrhea and adverse events) were combined using a pooled risk ratio (RR) or risk difference (RD), and continuous data (mean duration of diarrhea and mean daily stool frequency) as mean difference (MD), along with their corresponding 95% confidence interval (95% CI). For overall pooled results on the incidence of diarrhea, sensitivity analyses included available case versus extreme-plausible analyses and random- versus fixed-effect models. To explore possible explanations for heterogeneity, a priori subgroup analysis was conducted on probiotic strain, dose, definition of antibiotic-associated diarrhea, and risk of bias. We also conducted post hoc subgroup analyses by patient diagnosis, single versus multi-strain, industry sponsorship, and inpatient status. The overall quality of the evidence supporting

the outcomes was evaluated using the GRADE criteria. ***MAIN RESULTS***: Overall, 23 studies (3938 participants) met the inclusion criteria. Trials included treatment with either Bacillus spp., Bifidobacterium spp., Clostridium butyricum, Lactobacilli spp., Lactococcus spp., Leuconostoc cremoris, Saccharomyces spp., or Streptococcus spp., alone or in combination. Eleven studies used a single-strain probiotic, four combined two probiotic strains, three combined three probiotic strains, one combined four probiotic strains, two combined seven probiotic strains, one included ten probiotic strains, and one study included two probiotic arms that used three and two strains, respectively. The risk of bias was determined to be high or unclear in 13 studies and low in 10 studies. Available case (patients who did not complete the studies were not included in the analysis) results from 22/23 trials reporting on the incidence of diarrhea show a precise benefit from probiotics compared to active, placebo, or no treatment control. The incidence of AAD in the probiotic group was 8% (163/1992) compared to 19% (364/1906) in the control group (RR = 0.46; 95% CI: 0.35-0.61; I2 = 55%, 3898 participants). A GRADE analysis indicated that the overall quality of the evidence for this outcome was moderate. This benefit remained statistically significant in an extreme-plausible (60% of children lost to follow-up in probiotic group and 20% lost to follow-up in the control group had diarrhea) sensitivity analysis, where the incidence of AAD in the probiotic group was 14% (330/2294) compared to 19% (426/2235) in the control group (RR = 0.69; 95% CI: 0.54-0.89; I2 = 63%, 4529 participants). None of the 16 trials (n = 2455) that reported on adverse events documented any serious adverse events attributable to probiotics. Meta-analysis excluded all but an extremely small non-significant difference in adverse events between treatment and control (RD = 0.00, 95% CI: -0.01 to 0.01). The majority of adverse events were in placebo, standard care, or no treatment group. Adverse events reported in the studies include rash, nausea, gas, flatulence, abdominal bloating, abdominal pain, vomiting, increased phlegm, chest pain, constipation, taste disturbance, and low appetite. ***AUTHORS CONCLUSIONS***: Moderate quality evidence suggests a protective effect of probiotics in preventing AAD. Our pooled estimate suggests a precise (RR 0.46; 95% CI: 0.35-0.61) probiotic effect with an NNT of 10. Among the various probiotics evaluated, Lactobacillus rhamnosus or Saccharomyces boulardii at 5-40 billion colony-forming units/day may be appropriate given the modest NNT and the likelihood that adverse events are very rare. It is premature to draw conclusions about the efficacy and safety of other probiotic agents for pediatric AAD. Although no serious adverse events were observed among otherwise healthy children, serious adverse events have been observed in severely debilitated or immunocompromised children with underlying risk factors including central venous catheter use and disorders associated with bacterial/fungal translocation. Until further research has been conducted, probiotic use should be avoided in pediatric populations at risk for adverse events. Future trials would benefit from a standard and valid outcomes to measure AAD.

He, X., Slupsky, C. M., Dekker, J. W., Haggarty, N. W., and Lonnerdal, B. Integrated Role of Bifidobacterium animalis subsp. lactis Supplementation in Gut Microbiota, Immunity, and Metabolism of Infant Rhesus Monkeys. mSystems. 2016; 1(6); PM: 27921083

ABSTRACT: To investigate the impact of probiotic supplementation of infant formula on immune parameters, intestinal microbiota, and metabolism, five individually housed infant rhesus monkeys exclusively fed standard infant formula supplemented with probiotics (Bifidobacterium animalis subsp. lactis HN019) from birth until 3 months of age were compared with five standard formula-fed and five breast-fed monkeys. Anthropometric measurements, serum insulin, immune parameters, fecal microbiota, and metabolic profiles of serum, urine, and feces were evaluated. Consumption of B. lactis-

supplemented formula reduced microbial diversity, restructured the fecal microbial community, and altered the fecal metabolome at the last two time points, in addition to increasing short-chain fatty acids in serum and urine. Circulating CCL22 was lower and threonine, branched-chain amino acids, urea, and allantoin, as well as dimethylglycine in serum and urine, were increased in the group supplemented with B. lactis compared with the standard formula-fed group. These results support a role of probiotics as effectors of gut microbial activity regulating amino acid utilization and nitrogen cycling. Future risk-benefit analyses are still needed to consolidate the existing knowledge on the long-term consequences of probiotic administration during infancy. *IMPORTANCE* Probiotics are becoming increasingly popular due to their perceived effects on health, despite a lack of mechanistic information on how they impart these benefits. Infant formula and complementary foods are common targets for supplementation with probiotics. However, different probiotic strains have different properties, and there is a lack of data on long-term health effects on the consumer. Given the increasing interest in supplementation with probiotics and the fact that the gastrointestinal tracts of infants are still immature, we sought to determine whether consumption of infant formula containing the probiotic Bifidobacterium animalis subsp. lactis HN019 for 3 months starting at birth would impact gut microbial colonization, as well as infant immunity and metabolism, when compared with consumption of formula alone.

Hegarty, J. W., Guinane, C. M., Ross, R. P., Hill, C., and Cotter, P. D. Bacteriocin production: a relatively unharnessed probiotic trait? F1000Res. 2016; 5:2587; PM: 27853525

ABSTRACT: Probiotics are "live microorganisms which, when consumed in adequate amounts, confer a health benefit to the host". A number of attributes are highly sought after among these microorganisms, including immunomodulation, epithelial barrier maintenance, competitive exclusion, production of short-chain fatty acids, and bile salt metabolism. Bacteriocin production is also generally regarded as a probiotic trait, but it can be argued that, in contrast to other traits, it is often considered a feature that is desirable, rather than a key probiotic trait. As such, the true potential of these antimicrobials has yet to be realised.

Heo, J., Seo, M., Park, H., Lee, W. K., Guan, L. L., Yoon, J., Caetano-Anolles, K., Ahn, H., Kim, S. Y., Kang, Y. M., Cho, S., and Kim, H. Gut microbiota Modulated by Probiotics and Garcinia cambogia Extract Correlate with Weight Gain and Adipocyte Sizes in High Fat-Fed Mice. Sci.Rep. 9-23-2016; 6:33566; PM: 27658722

ABSTRACT: Results of recent studies on gut microbiota have suggested that obesogenic bacteria exacerbate obesity and metabolic dysfunction in the host when fed a high fat diet (HFD). In order to explore obesity-associated bacterial candidates and their response to diet, the composition of faecal bacterial communities was investigated by analyzing 16S rRNA gene sequences in mice. Dietary intervention with probiotics and Garcinia cambogia extract attenuated weight gain and adipocyte size in HFD-fed mice. To identify obesity-causative microbiota, two statistical analyses were performed. Forty-eight bacterial species were found to overlap between the two analyses, indicating the commonly identified species as diet-driven and obesity-associated, which would make them strong candidates for host-microbiome interaction on obesity. Finally, correlation based network analysis between diet, microbe, and host revealed that Clostridium aminophilum, a hyper-ammonia-producing bacterium, was highly correlated with obesity phenotypes and other associated bacteria, and shown to be suppressed by the

combination of probiotics and Garcinia cambogia extract. Results of the present study suggest that probiotics and Garcinia cambogia extract alleviate weight gain and adiposity, in part via differentially modulating the composition of gut microbiota in HFD fed mice.

Hessenberger, S., Schatzmayr, G., and Teichmann, K. In vitro inhibition of Eimeria tenella sporozoite invasion into host cells by probiotics. Vet.Parasitol. 10-15-2016; 229:93-98; PM: 27809987

ABSTRACT: The aim was to study the effects of probiotics isolated from the intestinal tract of livestock animals on Eimeria tenella invasion into Madin-Darby bovine kidney (MDBK) cells in vitro. E. tenella sporozoites were purified and labeled with 5(6)-carboxyfluorescein diacetate N-succinimidyl ester before seeding on cell cultures, and invasion was evaluated by fluorescence microscopy. Two protocols (A and B) were used. In protocol A, Enterococcus faecium # 589 or Lactobacillus salivarius subsp. salivarius # 505 were added together with sporozoites to MDBK cell cultures and invasion was evaluated after incubation for approximately 20h. Viable, dead, or spent culture supernatants of probiotics were tested. In protocol B, viable probiotics were incubated with MDBK cells for one hour before sporozoites were added and invasion was evaluated after two more hours of incubation. Parasite invasion of viable, dead, or spent culture supernatant of E. faecium # 589 was assessed. Using protocol A, it was shown that parasite invasion was inhibited by viable (80%) or dead (75%) E. faecium # 589. While inhibition by viable L. salivarius subsp. salivarius # 505 was not valid at the highest concentration and not significant at the other test concentrations, dead cells inhibited parasite invasion up to 45%. Spent culture supernatants of both probiotics had no influence on parasite invasion. Using protocol B, it was shown that viable Bifidobacterium animalis subsp. animalis # 503, E. faecium # 497, E. faecium # 589, L. reuteri # 514, L. salivarius subsp. salivarius # 505, and Bacillus subtilis # 588 inhibited parasite invasion into MDBK cells up to 80%. Anticoccidial activity was strain-specific for E. faecium strains, and the strongest effect was shown by E. faecium # 589. Anticoccidial effects of some of the tested probiotics have already been shown in vivo, which makes them candidates to prevent coccidiosis. These findings have now been confirmed in vitro. The used parasite invasion assay is a fast and inexpensive tool to screen probiotics for prevention of coccidiosis.

Hiergeist, A. and Gessner, A. Clinical implications of the microbiome in urinary tract diseases. Curr.Opin.Urol. 11-24-2016; PM: 27898455

ABSTRACT: ***PURPOSE OF REVIEW***: The purpose of this review is to outline and evaluate the most recent literature on the role of the microbiome in urinary tract diseases. ***RECENT FINDINGS***: High throughput molecular DNA sequencing of bacterial 16S rRNA genes enabled the analysis of complex microbial communities inhabiting the human urinary tract. Several recent studies have identified bacterial taxa of the urinary microbiome to impact urinary tract diseases including interstitial cystitis, urgency urinary incontinence or calcium oxalate stone formation. Furthermore, treatment of urinary tract infections by antibiotics globally impacts community profiles of the intestinal microbiota and might indirectly influence human health. Alternative treatment options like application of probiotics for the treatment of urinary tract infections are currently under investigation. ***SUMMARY***: The urinary microbiome and its relationship to urinary tract diseases is currently under comprehensive investigation. Further studies are needed to shed light on the role of commensal microbiota for urinary tract infections.

Hijova, E., Kuzma, J., Strojny, L., Bomba, A., Bertkova, I., Chmelarova, A., Hertelyova, Z., Benetinova, V., Stofilova, J., and Ambro, L. Ability of Lactobacillus plantarum LS/07 to modify intestinal enzymes activity in chronic diseases prevention. Acta Biochim.Pol. 11-7-2016; PM: 27824363

ABSTRACT: The ability of probiotic strain Lactobacillus plantarum LS/07 to modify the activity of intestinal bacterial enzymes - beta-glucuronidase (beta-GLUCUR), beta-galactosidase (beta-GAL), and beta-glucosidase (beta-GLU) in prevention of chronic diseases - cancer, atherosclerosis and dysbiosis was investigated. The male Sprague-Dawley rats were randomly divided into 12 experimental groups: controls groups - C (control), AT (atherosclerotic), CC (carcinogenic), dysbiotic groups - each group in combination with antibiotics (ATB), probiotics groups - in combinatioan with probiotic (PRO) alone, and each group with combination of antibiotic and probiotic (ATB+PRO). In the control group the beta-glucuronidase activity did not change throughout the experiment. High fat diet in atherosclerotic group significantly increased the activity of beta-glucuronidase ($P < 0.001$) and beta-glucosidase ($P < 0.01$). Azoxymethane application in carcinogenic group significantly increased beta-glucuronidase ($P < 0.01$), but reduced beta-glucosidase ($P < 0.01$) activity. Daily application of probiotics alone and in combination with antibiotic increased beta-galactosidase, of beta-glucosidase, and decreased beta-glucuronidase activity. In control antibiotic group we observed significant increase in beta-glucuronidase ($P < 0.05$) and decreased beta-glucosidase ($P < 0.01$) activity which can be caused by the change of microflora in favor of coliform bacteria. These findings indicate the positive effects of probiotic Lactobacillus plantarum LS/07 and suggest its use in disease prevention in human medicine and some animal species.

Ho, M., Chang, Y. Y., Chang, W. C., Lin, H. C., Wang, M. H., Lin, W. C., and Chiu, T. H. Oral Lactobacillus rhamnosus GR-1 and Lactobacillus reuteri RC-14 to reduce Group B Streptococcus colonization in pregnant women: A randomized controlled trial. Taiwan.J.Obstet.Gynecol. 2016; 55(4):515-518; PM: 27590374

ABSTRACT: ***OBJECTIVE***: This study is to examine the effect of Lactobacillus rhamnosus GR-1 and Lactobacillus reuteri RC-14 taken orally before bedtime on Group B Streptococcus (GBS)-positive pregnant women with respect to becoming GBS negative. ***MATERIALS AND METHODS***: In total, 110 pregnant women at 35-37 weeks of gestation who were diagnosed by GBS culture as being GBS positive for both vaginal and rectal GBS colonization were randomly assigned to be orally treated with two placebo capsules or two probiotic capsules (containing L. rhamnosus GR-1 and L. reuteri RC-14) before bedtime until delivery. All women were tested for vaginal and rectal GBS colonization again by GBS culture on admission for delivery. ***RESULTS***: Of the 110 participants, 99 completed the study (49 in the probiotic group and 50 in the placebo group). The GBS colonization results changed from positive to negative in 21 women in the probiotic group (42.9%) and in nine women in the placebo group (18.0%) during this period (Chi-square p=0.007). ***CONCLUSIONS***: Oral probiotic containing L. rhamnosus GR-1 and L. reuteri RC-14 could reduce the vaginal and rectal GBS colonization rate in pregnant women.

Horvath, A., Leber, B., Schmerboeck, B., Tawdrous, M., Zettel, G., Hartl, A., Madl, T., Stryeck, S., Fuchs, D., Lemesch, S., Douschan, P., Krones, E., Spindelboeck, W., Durchschein, F., Rainer, F., Zollner, G., Stauber, R. E., Fickert, P., Stiegler, P., and Stadlbauer, V. Randomised

clinical trial: the effects of a multispecies probiotic vs. placebo on innate immune function, bacterial translocation and gut permeability in patients with cirrhosis. Aliment.Pharmacol.Ther. 2016; 44(9):926-935; PM: 27593544

ABSTRACT: ***BACKGROUND***: Probiotics may correct intestinal dysbiosis and proinflammatory conditions in patients with liver cirrhosis. AIM: To test the effects of a multispecies probiotic on innate immune function, bacterial translocation and gut permeability. ***METHODS***: In a randomised, double blind, placebo-controlled study, stable cirrhotic out-patients either received a daily dose of a probiotic powder containing eight different bacterial strains (Ecologic Barrier, Winclove, Amsterdam, The Netherlands) (n = 44) or a placebo (n = 36) for 6 months and were followed up for another 6 months. ***RESULTS***: We found a significant but subclinical increase in neutrophil resting burst (2.6-3.2%, P = 0.0134) and neopterin levels (7.7-8.4 nmol/L, P = 0.001) with probiotics but not with placebo. Probiotic supplementation did not have a significant influence on neutrophil phagocytosis, endotoxin load, gut permeability or inflammatory markers. Ten severe infections occurred in total; one during intervention in the placebo group, and five and four after the intervention has ended in the probiotic and placebo group, respectively. Liver function showed some improvement with probiotics but not with placebo. ***CONCLUSIONS***: Probiotic supplementation significantly increased serum neopterin levels and the production of reactive oxygen species by neutrophils. These findings might explain the beneficial effects of probiotics on immune function. Furthermore, probiotic supplementation may be a well-tolerated method to maintain or even improve liver function in stable cirrhosis. However, its influence on gut barrier function and bacterial translocation in cirrhotic patients is minimal.

Hrdy, J., Kocourkova, I., Lodinova-Zadnikova, R., Kolarova, L., and Prokesova, L. The effect of a probiotic Escherichia coli strain on regulatory T-cells in six year-old children. Benef.Microbes. 11-30-2016; 7(5):639-648; PM: 27633175

ABSTRACT: Probiotics are believed to prevent or reduce allergy development but the mechanism of their beneficial effect is still poorly understood. Immune characteristics of regulatory T cells (Tregs) in peripheral blood of perinatally probiotic-supplemented children of allergic mothers (51 children), non-supplemented children of allergic mothers (42 children), and non-supplemented children of healthy mothers (28 children) were compared at the age of 6-7 years. A first dose of a probiotic Escherichia coli strain (E. coli O83:K24:H31) was administered within 2 days after the birth and then 12 times during the first months of life and children were followed longitudinally. Proportion and functional properties of Tregs were estimated by flow cytometry in relation to the children's allergy status. Proportion of Tregs in the peripheral blood of children suffering from allergy tends to be higher whereas median of fluorescence intensity (MFI) of FoxP3 was significantly decreased in allergic group. Intracellular presence of regulatory cytokine interleukin (IL)-10 was also lower in allergic children. Immune functions of Tregs reflected by both MFI of FoxP3 and IL-10 in the group of probiotic-supplemented children of allergic mothers were nearly comparable with children of healthy mothers while probiotic non-supplemented children of allergic mothers have decreased immune function of Tregs. Supplementation by probiotic E. coli strain decreases allergy incidence in high-risk children. In contrast to our expectation, proportion of Tregs has not been increased in probiotic supplemented children. Beneficial effect of probiotics on newborn immature immune system could be, at least partially, explained by the modulating immune function of Tregs. In summary, we detected increased proportion of Tregs in peripheral blood of allergic children, their functional properties were decreased in comparison with the Tregs of healthy children. A

unifying hypothesis for these findings is that Treg numbers in allergic children are increased in order to compensate for decreased function.

Hu, H. J., Zhang, G. Q., Zhang, Q., Shakya, S., and Li, Z. Y. Probiotics Prevent Candida Colonization and Invasive Fungal Sepsis in Preterm Neonates: A Systematic Review and Meta-Analysis of Randomized Controlled Trials. Pediatr.Neonatol. 9-14-2016; PM: 27793494

ABSTRACT: To investigate whether probiotic supplementation could reduce the risk of fungal infection in preterm neonates in neonatal intensive care units (NICUs), we systematically searched PubMed, EMBASE, and the Cochrane Central Register of Controlled Trials databases for randomized controlled trials (RCTs) focusing on the effect of probiotics on fungal infection in preterm neonates. The outcomes of interest were Candida colonization and invasive fungal sepsis. Seven trials involving 1371 preterm neonates were included. Meta-analysis (fixed-effects model) showed that probiotic supplementation was significantly associated with a lower risk of Candida colonization (2 RCTs, n = 329; relative risk (RR), 0.43; 95% confidence interval (CI), 0.27-0.67; p = 0.0002; I2 = 0%), and invasive fungal sepsis (7 RCTs, n = 1371; RR, 0.64; 95% CI, 0.46-0.88; p = 0.006; I2 = 13%). After excluding one study with a high baseline incidence (75%) of fungal sepsis, the effect of probiotics on invasive fungal sepsis became statistically insignificant (RR, 0.88; 95% CI, 0.44-1.78; p = 0.72; I2 = 15%). When using the random-effects model, the effect of probiotics remained favorable for Candida colonization (RR, 0.43; 95% CI 0.27-0.68; p = 0.0002; I2 = 0%) but not for fungal sepsis (RR, 0.64; 95% CI 0.38-1.08; p = 0.10; I2 = 13%). Current evidence indicates that probiotics can reduce the risk of Candida colonization in preterm neonates in NICUs. Limited data support that probiotic supplementation prevents invasive fungal sepsis in preterm neonates. High-quality and adequately powered RCTs are warranted.

Hu, X., Wang, T., and Jin, F. Alzheimer's disease and gut microbiota. Sci.China Life Sci. 2016; 59(10):1006-1023; PM: 27566465

ABSTRACT: Alzheimer's disease (AD) is a most common neurodegenerative disorder, which associates with impaired cognition. Gut microbiota can modulate host brain function and behavior via microbiota-gut-brain axis, including cognitive behavior. Germ-free animals, antibiotics, probiotics intervention and diet can induce alterations of gut microbiota and gut physiology and also host cognitive behavior, increasing or decreasing risks of AD. The increased permeability of intestine and blood-brain barrier induced by gut microbiota disturbance will increase the incidence of neurodegeneration disorders. Gut microbial metabolites and their effects on host neurochemical changes may increase or decrease the risk of AD. Pathogenic microbes infection will also increase the risk of AD, and meanwhile, the onset of AD support the "hygiene hypothesis". All the results suggest that AD may begin in the gut, and is closely related to the imbalance of gut microbiota. Modulation of gut microbiota through personalized diet or beneficial microbiota intervention will probably become a new treatment for AD.

Huang, F. C. and Huang, S. C. The different effects of probiotics treatment on Salmonella-induced interleukin-8 response in intestinal epithelia cells via PI3K/Akt and NOD2 expression. Benef.Microbes. 11-30-2016; 7(5):739-748; PM: 27680208

ABSTRACT: Salmonella spp. remains a major public health problem for the whole world. Intestinal epithelial cells serve as an essential component of the innate mucosal immune system to defend against Salmonella infection. A substantial amount of evidence has accumulated that probiotics can regulate interleukin 8 (IL-8) involved in innate immunity. However, the exact effect of probiotics on epithelial IL-8 response to Salmonella infection is not well understood. Therefore, we investigated the action of probiotics on Salmonella-infected Caco-2 cells and its novel mechanisms. Two probiotic strains were examined for Salmonella-induced IL-8 responses and regulating proteins using Caco-2 cell cultures. We demonstrated probiotic, either Lactobacillus rhamnosus GG or Bifidobacterium animalis subsp. lactis DSM10140, administered before Salmonella infection conferred significantly suppressive effect on Salmonella-induced IL-8 responses in Caco-2 cells, either in secreted protein or mRNA, via the PI3K/Akt signal pathway while probiotic administered after infection enhanced Salmonella-induced IL-8 responses via nucleotide-binding oligomerisation domain-containing protein 2 expression in membrane. These findings suggest that the different regulation of probiotics on Salmonella-induced IL-8 responses in Caco-2 cells according to the administered timing supports a rationale for the therapeutic use of probiotics in the treatment of Salmonella colitis and inflammatory bowel disease. This can explain the reported controversial effect of probiotics on these diseases.

Huang, R., Wang, K., and Hu, J. Effect of Probiotics on Depression: A Systematic Review and Meta-Analysis of Randomized Controlled Trials. Nutrients. 8-6-2016; 8(8); PM: 27509521

ABSTRACT: It has been reported that gut probiotics play a major role in the bidirectional communication between the gut and the brain. Probiotics may be essential to people with depression, which remains a global health challenge, as depression is a metabolic brain disorder. However, the efficacy of probiotics for depression is controversial. This study aimed to systematically review the existing evidence on the effect of probiotics-based interventions on depression. Randomized, controlled trials, identified through screening multiple databases and grey literature, were included in the meta-analysis. The meta-analysis was performed using Review Manager 5.3 software using a fixed-effects model. The meta-analysis showed that probiotics significantly decreased the depression scale score (MD (depressive disorder) = -0.30, 95% CI (-0.51--0.09), p = 0.005) in the subjects. Probiotics had an effect on both the healthy population (MD = -0.25, 95% CI (-0.47--0.03), p = 0.03) and patients with major depressive disorder (MDD) (MD = -0.73, 95% CI (-1.37--0.09), p = 0.03). Probiotics had an effect on the population aged under 60 (MD = -0.43, 95% CI (-0.72--0.13), p = 0.005), while it had no effect on people aged over 65 (MD = -0.18, 95% CI (-0.47-0.11), p = 0.22). This is the first systematic review and meta-analysis with the goal of determining the effect of probiotics on depression. We found that probiotics were associated with a significant reduction in depression, underscoring the need for additional research on this potential preventive strategy for depression.

Huang, T., Zhang, X., Pan, J., Su, X., Jin, X., and Guan, X. Purification and Characterization of a Novel Cold Shock Protein-Like Bacteriocin Synthesized by Bacillus thuringiensis. Sci.Rep. 10-20-2016; 6:35560; PM: 27762322

ABSTRACT: Bacillus thuringiensis (Bt), one of the most successful biopesticides, may expand its potential by producing bacteriocins (thuricins). The aim of this study was to investigate the antimicrobial potential of a novel Bt bacteriocin, thuricin BtCspB,

produced by Bt BRC-ZYR2. The results showed that this bacteriocin has a high similarity with cold-shock protein B (CspB). BtCspB lost its activity after proteinase K treatment; however it was active at 60 degrees C for 30 min and was stable in the pH range 5-7. The partial loss of activity after the treatments of lipase II and catalase were likely due to the change in BtCspB structure and the partial degradation of BtCspB, respectively. The loss of activity at high temperatures and the activity variation at different pHs were not due to degradation or large conformational change. BtCspB did not inhibit four probiotics. It was only active against B. cereus strains 0938 and ATCC 10987 with MIC values of 3.125 mug/mL and 0.781 mug/mL, and MBC values of 12.5 mug/mL and 6.25 mug/mL, respectively. Taken together, these results provide new insights into a novel cold shock protein-like bacteriocin, BtCspB, which displayed promise for its use in food preservation and treatment of B. cereus-associated diseases.

Hussain, S. A., Patil, G. R., Reddi, S., Yadav, V., Pothuraju, R., Singh, R. R., and Kapila, S. Aloe vera (Aloe barbadensis Miller) supplemented probiotic lassi prevents Shigella infiltration from epithelial barrier into systemic blood flow in mice model. Microb.Pathog. 11-30-2016; PM: 27914960

ABSTRACT: The aim of present work was to investigate preventive role of orally administered Aloe vera supplemented probiotic lassi (APL) on Shigella dysenteriae infection in mice. At the end of experimental period (2, 5 and 7 days of challenging), different organs such as spleen, liver, small intestine, large intestine, and peritoneal fluid were collected and assessed for Shigella colonization. Secretary IgA was estimated in intestinal fluid. Blood was collected in heparinized tubes for various haematological studies. Oral administration of APL showed a significant ($p < 0.05$) reduction in the Shigella counts (log cfu/mL) in all organs as compared to other treatment groups at different intervals after post feeding. Similarly, secretary IgA antibody levels (mug/mL) in intestinal fluid were significantly ($p < 0.05$) increased in case of APL fed mice. Further, feeding of APL also demonstrated a positive effect on different haematological parameters viz. Hb (gm %), RBC and WBC count. The results indicated the immunoprotective effects of APL against Shigella dysenteriae induced infection in mice.

Islam, M. M. and Yang, C. J. Efficacy of mealworm and super mealworm larvae probiotics as an alternative to antibiotics challenged orally with Salmonella and E. coli infection in broiler chicks. Poult.Sci. 8-12-2016; PM: 27520069

ABSTRACT: This study was conducted to evaluate dry mealworm (Tenebrio molitor) (DMLP) and super mealworm (Zophobas morio) (DSMLP) larvae probiotics as alternatives to antibiotics in broiler chicks. A total of 240 one-day old Ross 308 male broiler chicks were randomly assigned to three dietary treatments consisting of ten replications with eight birds each in a completely randomized design. The dietary treatments were, (i) control (basal diet), (ii) 0.4% DMLP (basal diet + 0.4% DMLP, DM basis), and (iii) 0.4% DSMLP (basal diet + 0.4% DSMLP, DM basis). On day one, 1 mL of mixed broth agar consisting of 2.4 x 107 cfu Salmonella enteritidis KCTC 2021 and 3.7 x 107 cfu Escherichia coli KCTC 2571 was injected orally into each chick. After one week, growth performance, immunity, mortality, internal organ weight, and cecal and fecal microbiota were investigated. Average daily gain (ADG:) and average daily feed intake (ADFI)

increased, while feed conversion ratio (FCR) (g intake/g gain per bird) decreased in response to DMLP and DSMLP supplementation ($P < 0.05$). Additionally, mortality decreased ($P < 0.05$), while IgG and IgA levels increased following DMLP and DSMLP supplementation ($P < 0.05$). Internal organs remained unaffected, except for a reduced bursa of Fabricius weight in DSMLP supplementation ($P < 0.05$). Cecal E. coli and Salmonella contents were reduced in DMLP and DSMLP supplementation ($P < 0.05$), while fecal microbiota contents and pH of cecal and fecal digesta remained unaffected. In conclusion, dietary supplementation with DMLP and DSMLP increased ADG and IgG and IgA levels, while reducing FCR, mortality and cecal E. coli and Salmonella spp. *CONTENTS*: Thus, DMLP and DSMLP can be utilized as an alternative to antibiotics in broiler diets.

Jadresin, O., Hojsak, I., Misak, Z., Kekez, A. J., Trbojevic, T., Ivkovic, L., and Kolacek, S. Lactobacillus Reuteri DSM 17938 in the Treatment of Functional Abdominal Pain in Children - RCT Study. J.Pediatr.Gastroenterol.Nutr. 11-30-2016; PM: 27906800

ABSTRACT: ***OBJECTIVES***: Beneficial therapeutic effect of probiotics has been reported in children with the irritable bowel syndrome (IBS) but not consistently in other functional abdominal pain-related disorders. The aim of this study was to investigate the effect of Lactobacillus (L.) reuteri DSM 17938 in the treatment of functional abdominal pain (FAP) and IBS in children. ***METHODS***: Children (age 4-18 years) referred to pediatric gastroenterologist at Children's Hospital Zagreb from May 2012 to December 2014, diagnosed as FAP or IBS, were randomized to receive L. reuteri DSM 17938 10 CFU daily or placebo. The study was a prospective, randomized, double-blind, placebo-controlled parallel study. Symptoms were evaluated using Wong-Baker FACES pain rating scale for pain and Bristol scale for stool shape and consistence. ***RESULTS***: Data were analyzed for 55 children (26 in the intervention group and 29 in the placebo group). Children in the intervention group had significantly more days without pain (median 89.5 vs. 51 days, $p = 0.029$). Abdominal pain was less severe in children taking probiotics during the 2 month ($p < 0.05$) and 4 month ($p < 0.01$). The two groups did not differ in the duration of abdominal pain, stool type or absence from school. Both groups experienced significant reduction in the severity of abdominal pain from 1 to 4 month, with the reduction more prominent in the intervention group ($p < 0.001$ vs. $p = 0.004$). ***CONCLUSION***: Administration of L. reuteri DSM 17938 was associated with a possible reduction of the intensity of pain and significantly more days without pain in children with FAP and IBS.

Jager, R., Purpura, M., Stone, J. D., Turner, S. M., Anzalone, A. J., Eimerbrink, M. J., Pane, M., Amoruso, A., Rowlands, D. S., and Oliver, J. M. Probiotic Streptococcus thermophilus FP4 and Bifidobacterium breve BR03 Supplementation Attenuates Performance and Range-of-Motion Decrements Following Muscle Damaging Exercise. Nutrients. 10-14-2016; 8(10); PM: 27754427

ABSTRACT: Probiotics have immunomodulatory effects. However, little is known about the potential benefit of probiotics on the inflammation subsequent to strenuous exercise. In a double-blind, randomized, placebo controlled, crossover design separated by a 21-day washout, 15 healthy resistance-trained men ingested an encapsulated probiotic Streptococcus (S.) thermophilus FP4 and Bifidobacterium (B.) breve BR03 at 5 bn live cells (AFU) concentration each, or a placebo, daily for 3 weeks prior to muscle-damaging

exercise (ClinicalTrials.gov NCT02520583). Isometric strength, muscle soreness, range of motion and girth, and blood interleukin-6 (IL-6) and creatine kinase (CK) concentrations were measured from pre- to 72 h post-exercise. Statistical analysis was via mixed models and magnitude-based inference to the standardized difference. Probiotic supplementation resulted in an overall decrease in circulating IL-6, which was sustained to 48 h post-exercise. In addition, probiotic supplementation likely enhanced isometric average peak torque production at 24 to 72 h into the recovery period following exercise (probiotic-placebo point effect +/-90% CI: 24 h, 11% +/- 7%; 48 h, 12% +/- 18%; 72 h, 8% +/- 8%). Probiotics also likely moderately increased resting arm angle at 24 h (2.4% +/- 2.0%) and 48 h (1.9% +/- 1.9%) following exercise, but effects on soreness and flexed arm angle and CK were unclear. These data suggest that dietary supplementation with probiotic strains S. thermophilus FP4 and B. breve BR03 attenuates performance decrements and muscle tension in the days following muscle-damaging exercise.

Jager, R., Shields, K. A., Lowery, R. P., De Souza, E. O., Partl, J. M., Hollmer, C., Purpura, M., and Wilson, J. M. Probiotic Bacillus coagulans GBI-30, 6086 reduces exercise-induced muscle damage and increases recovery. PeerJ. 2016; 4:e2276; PM: 27547577

ABSTRACT: ***Objective***: Probiotics have been reported to support healthy digestive and immune function, aid in protein absorption, and decrease inflammation. Further, a trend to increase vertical jump power has been observed following co-administration of protein and probiotics in resistance-trained subjects. However, to date the potential beneficial effect of probiotics on recovery from high intensity resistance exercise has yet to be explored. Therefore, this study examined the effect of co-administration of protein and probiotics on muscle damage, recovery and performance following a damaging exercise bout. ***Design***. Twenty nine (n = 29) recreationally-trained males (mean +/- SD; 21.5 +/- 2.8 years; 89.7 +/- 28.2 kg; 177.4 +/- 8.0 cm) were assigned to consume either 20 g of casein (PRO) or 20 g of casein plus probiotic (1 billion CFU Bacillus coagulans GBI-30, 6086, PROBC) in a crossover, diet-controlled design. After two weeks of supplementation, perceptional measures, athletic performance, and muscle damage were analyzed following a damaging exercise bout. ***Results***: The damaging exercise bout significantly increased muscle soreness, and reduced perceived recovery; however, PROBC significantly increased recovery at 24 and 72 h, and decreased soreness at 72 h post exercise in comparison to PRO. Perceptual measures were confirmed by increases in CK (PRO: +266.8%, p = 0.0002; PROBC: +137.7%, p = 0.01), with PROBC showing a trend towards reduced muscle damage (p = 0.08). The muscle-damaging exercise resulted in significantly increased muscle swelling and Blood Urea Nitrogen levels in both conditions with no difference between groups. The strenuous exercise significantly reduced athletic performance in PRO (Wingate Peak Power; PRO: (-39.8 watts, -5.3%, p = 0.03)), whereas PROBC maintained performance (+10.1 watts, +1.7%). ***Conclusions***: The results provide evidence that probiotic supplementation in combination with protein tended to reduce indices of muscle damage, improves recovery, and maintains physical performance subsequent to damaging exercise.

Jamali, Z., Aminabadi, N. A., Samiei, M., Sighari, Deljavan A., Shokravi, M., and Shirazi, S. Impact of Chlorhexidine Pretreatment Followed by Probiotic Streptococcus salivarius Strain K12 on Halitosis in Children: A Randomised Controlled Clinical Trial. Oral Health Prev.Dent. 2016; 14(4):305-313; PM: 27508274

ABSTRACT: The aim of this study was to examine the effect of chlorhexidine disinfection, as a chemical method of oral hygiene practice, and subsequent use of probiotics on halitosis in children. The effects of mechanical and chemical oral hygiene practice methods on the severity of halitosis were also assessed. 208 children with organoleptic test (OLT) scores of 2 or more were randomly assigned to four groups: A: conventional oral hygiene practices (COH) including toothbrushing and flossing; B: COH + tongue scraping (TS); C: COH + TS + chlorhexidine; D: COH + TS + chlorhexidine + probiotics. OLT was performed at 1-week and 3-month follow-ups. A significant and stable number of participants showed major and moderate levels of improvement in OLT scores in group D ($p < 0.001$). The improvement of OLT scores in group C was also significant ($p < 0.001$), but not stable over the follow-ups ($p = 0.44$). Neither significant nor stable improvements in the OLT scores were detected in groups A and B through follow-ups ($p > 0.05$). Probiotic therapy following oral disinfection with chlorhexidine may reduce the severity of halitosis over longer periods.

Janczarek, M., Bachanek, T., Mazur, E., and Chalas, R. The role of probiotics in prevention of oral diseases. Postepy Hig.Med.Dosw.(Online.). 1-4-2016; 70(0):850-857; PM: 27594560

ABSTRACT: The dynamic development of knowledge in the field of probiotics was commenced at the beginning of the 20th century. Since then, many ways of their possible usage in medicine have been established. In accordance with the WHO, probiotics are live microorganisms, which if applied in adequate amounts may benefit the host. Among probiotics, fungi and bacteria are distinguished, and mechanisms of action of these organisms in the oral cavity and gut are parallel. Application in dentistry, in prophylaxis and treatment of oral diseases is still not well known. Most commonly, Lactobacillus reuteri, Lactobacillus rhamnosus GG and Bifidobacterium are applied. The aim of the study was to collect and systematize the latest information about probiotics and their role in pathomechanisms of dental caries, gingivitis and periodontitis, candidiasis, and malodour. Based on the analyzed literature, it can be concluded that mechanisms of cariogenic pathogen inhibition using probiotics are still not well understood. The new research trend is based on application of probiotics which can naturally displace cariogenic bacteria in the oral cavity and influence oral health in adults and children. The results of studies also confirmed the beneficial role of probiotics in reduction of the bacterial population in periodontitis and halitosis. Long-term observation and a properly designed study protocol will allow us to answer many questions concerning substitution of one strain of bacteria by another.

Jeong, J. H., Lee, C. Y., and Chung, D. K. Probiotic Lactic Acid Bacteria and Skin Health. Crit Rev.Food Sci.Nutr. 10-25-2016; 56(14):2331-2337; PM: 26287529

ABSTRACT: Human skin is the first defense barrier against the external environment, especially microbial pathogens and physical stimulation. Many studies on skin health with Lactic acid bacteria (LAB) have been published for many years, including prevention of skin disease and improvement of skin conditions. LAB, a major group of gram-positive bacteria, are known to be beneficial to human health by acting as probiotics. Recent studies have shown that LAB and their extracts have beneficial effects on maintenance and improvement of skin health. Oral administration of Lactobacillus delbrueckii inhibits the development of atopic disease. In addition, LAB and LAB extracts are known to have

beneficial effects on intestinal diseases, with Lactobacillus plantarum having been shown to attenuate IL-10 deficient colitis. In addition to intestinal health, L. plantarum also has beneficial effects on skin. pLTA, which is lipoteichoic acid isolated from L. plantarum, has anti-photoaging effects on human skin cells by regulating the expression matrix meralloprotionase-1 (MMP-1) expression. While several studies have proposed a relationship between diseases of the skin and small intestines, there are currently no published reviews of the effects of LAB for skin health through regulation of intestinal conditions and the immune system. In this review, we discuss recent findings on the effects of LAB on skin health and its potential applications in beauty foods.

Johnson-Henry, K. C., Abrahamsson, T. R., Wu, R. Y., and Sherman, P. M. Probiotics, Prebiotics, and Synbiotics for the Prevention of Necrotizing Enterocolitis. Adv.Nutr. 2016; 7(5):928-937; PM: 27633108

ABSTRACT: Necrotizing enterocolitis (NEC) is a devastating intestinal disease in preterm infants characterized by barrier disruption, intestinal microbial dysbiosis, and persistent inflammation of the colon, which results in high mortality rates. Current strategies used to manage this disease are not sufficient, although the use of human breast milk reduces the risk of NEC. Mother's milk is regarded as a fundamental nutritional source for neonates, but pasteurization of donor breast milk affects the composition of bioactive compounds. Current research is evaluating the benefits and potential pitfalls of adding probiotics and prebiotics to pasteurized milk so as to improve the functionality of the milk and thereby reduce the burden of illness caused by NEC. Probiotics (live micro-organisms that confer health to the host) and prebiotics (nondigestible oligosaccharides that stimulate the growth of healthy bacteria) are functional foods known to mediate immune responses and modulate microbial populations in the gut. Clinical research shows strain- and compound-specific responses when probiotics or prebiotics are administered in conjunction with donor breast milk for the prevention of NEC. Despite ongoing controversy surrounding optimal treatment strategies, randomized controlled studies are now investigating the use of synbiotics to reduce the incidence and severity of NEC. Synbiotics, a combination of probiotics and prebiotics, have been proposed to enhance beneficial health effects in the intestinal tract more than either agent administered alone. This review considers the implications of using probiotic-, prebiotic-, and synbiotic-supplemented breast milk as a strategy to prevent NEC and issues that could be encountered with the preparations.

Johnston, B. C., Goldenberg, J. Z., and Parkin, P. C. Probiotics and the Prevention of Antibiotic-Associated Diarrhea in Infants and Children. JAMA. 10-11-2016; 316(14):1484-1485; PM: 27727371

ABSTRACT: In children prescribed an antibiotic, is the co-administration of a probiotic associated with lower rates of antibiotic-associated diarrhea without an increase in clinically important adverse events? Moderate-quality evidence suggests that probiotics are associated with lower rates of antibiotic-associated diarrhea in children (aged 1 month to 18 years) without an increase in adverse events.

Jones, R. M. The Influence of the Gut Microbiota on Host Physiology: In Pursuit of Mechanisms. Yale J.Biol.Med. 2016; 89(3):285-297; PM: 27698613

ABSTRACT: The results generated from the NIH funded Human Microbiome Project (HMP) are necessarily tied to the overall mission of the agency, which is to foster scientific discoveries as a basis for protecting and improving health. The investment in the HMP phase 1 accomplished many of its goals including the preliminary characterization of the human microbiome and the identification of links between microbiome diversity and disease states. Going forward, the next step in these studies must involve the identification of the functional molecular elements that mediate the positive influence of a eubiotic microbiome on health and disease. This review will focus on recent advances describing mechanistic events in the intestine elicited by the microbiome. These include symbiotic bacteria-induced activation of redox-dependent cell signaling, the bacterial production of short chain fatty acids and ensuing cellular responses, and the secretion of bacteriocins by bacteria that have anti-microbial activities against potential pathogens.

Kadirareddy, R. H., Vemuri, S. G., and Palempalli, U. M. Probiotic Conjugated Linoleic Acid Mediated Apoptosis in Breast Cancer Cells by Downregulation of NFkappaB. Asian Pac.J.Cancer Prev. 2016; 17(7):3395-3403; PM: 27509982

ABSTRACT: Conjugated linoleic acid, a functional lipid, produced from Lactobacillus plantarum (LPCLA), has been demonstrated to possess apoptotic activity. The antiproliferative and apoptotic potential of LPCLA was here evaluated in vitro using the MDAMB231 human breast cancer cell line as a model system. Proliferation of MDA MB231 cells was inhibited with increasing concentrations of LPCLA with altered morphological features like cell detachment, rounding of cells and oligonucleosomal fragmentation of DNA. Flow cytometry confirmed the apoptotic potential of LPCLA by ANNEXIN V/PI double staining. Furthermore, outcome results indicated that the apoptosis was mediated by downregulation of the NF?B pathway which in turn acted through proteasome degradation of I?Balpha, inhibition of p65 nuclear translocation, release of cytochromeC from mitochondria and finally overexpression of Bax protein. Thus, conjugated linoleic acid, a natural product derived from probiotics, could therefore be a possible potential chemotherapeutic agent due to its apoptotic activity against estrogen receptor negative breast cancer cells.

Kafshdooz, T., Akbarzadeh, A., Majdi, Seghinsara A., Pourhassan, M., Nasrabadi, H. T., and Milani, M. Role of Probiotics in Managing of Helicobacter Pylori Infection: A Review. Drug Res.(Stuttg). 11-7-2016; PM: 27824431

ABSTRACT: Background:Helicobacter pylori is a prevalent pathogen which is considered as an etiological cause for gastroduodenal ulcers, and a substantial risk factor for gastric malignancies. The vital factor to take into account is that roughly half of the world's population is infected with this bacterium. However, most subjects colonized remain asymptomatic and do not require any treatment. Several antimicrobial agents such as amoxicillin, clarithromycin and metronidazole are used to eradicate the infection. However, these drug regiments do not eradicate H. pylori in all patients because of the anti-drug resistance. Aim: In this review we aim to discuss the role and mechanisms of probiotics, as supportive medicines, in management of H. pylori infection. Methods: We have reviewed the published articles in PubMed and Medline databases. Also, abstracts presented in international conferences on the management of H. pylori infections and treatment protocol, have been thoroughly reviewed. Results: The overall trend in the literature indicates the usefulness of probiotics in controlling H. pylori infection. This

bacterium is among the most studied human pathogens regarding the efficacy of probiotics for treating its infection. Nevertheless, some studies suggest that probiotics do not efficiently eradicate H. pylori but retain the number of this bacterium at low levels inside the human stomach. Conclusion: The analyzed literature suggest that when probiotics are consumed in conjunction with antibiotics, the eradication rate may be improved through modulating the immune response and decreasing the adverse effects of routine drugs leading to gastroprotection.

Kahouli, I., Malhotra, M., Westfall, S., aoui-Jamali, M. A., and Prakash, S. Design and validation of an orally administrated active L. fermentum-L. acidophilus probiotic formulation using colorectal cancer Apc Min/+ mouse model. Appl.Microbiol.Biotechnol. 11-11-2016; PM: 27837314

ABSTRACT: Probiotics have been shown to have beneficial properties in attenuating the risk of colorectal cancer (CRC) development. However, functional evidence to support such effects for some probiotic bacteria are relatively unknown. Here, we document a significant antioxidant, anti-proliferative and pro-apoptotic activities of Lactobacillus acidophilus ATCC 314 and Lactobacillus fermentum NCIMB 5221 on CRC cells, particularly when used in combination (La-Lf). Furthermore, a superior synergistic activity on the inhibition of tumor growth and modulation of cell proliferation and epithelial markers in the Apc Min/+ CRC mouse model was explored, based on the expression levels of Ki-67, E-cadherin, beta-catenin, and cleaved caspase-3 (CC3) proteins. The anti-cancer activity of La-Lf co-culture was significantly enhanced in vitro with significant reduced proliferation (38.8 +/- 6.9 %, P = 0.009) and increased apoptosis (413 RUL, P < 0.001) towards cancer cells, as well as significant protection of normal colon cell growth from toxic treatment (18.6 +/- 9.8 %, P = 0.001). La-Lf formulation (1010cfu/animal/day) altered aspects of intestinal tumorigenesis by significantly reducing intestinal tumor multiplicity (1.7-fold, P = 0.016) and downregulating cellular proliferation markers, including beta-catenin (P = 0.041) and Ki-67 (P = 0.008). In conclusion, La-Lf showed greater protection against intestinal tumorigenesis supporting a potential use as a biotherapeutic for the prevention of CRC.

Kali, A. Psychobiotics: An emerging probiotic in psychiatric practice. Biomed.J. 2016; 39(3):223-224; PM: 27621125

ABSTRACT: Intestinal microbial flora plays critical role in maintenance of health. Probiotic organisms have been recognized as an essential therapeutic component in the treatment of intestinal dysbiosis. Current research suggests their health benefits extends beyond intestinal disorders. The neuroactive molecules produced by the gut microbiota has been found to modulate neural signals which affect neurological and psychiatric parameters like sleep, appetite, mood and cognition. Use of these novel probiotics opens up the possibility of restructuring of intestinal microbiota for effective management of various psychiatric disorders.

Kang, Y., Cai, Y., Zhang, X., Kong, X., and Su, J. Altered gut microbiota in RA: implications for treatment. Z.Rheumatol. 12-1-2016; PM: 27909795

ABSTRACT: Rheumatoid arthritis (RA) is an autoimmune disease with progressive joint disorder. The complex interplay of genetic and environmental influences is important for the development of the disease. A growing body of evidence has shed light on the

association of dysbiosis of gut microbiota with RA. Certain gut microbial strains have been shown to inhibit or attenuate immune responses in RA experimental models, suggesting that specific species among intestinal commensal bacteria may play either a pathogenic or a protective role in the development of RA. Oral intake of probiotics/prebiotics can therefore represent a therapeutic approach for RA treatment. However, the relevant scientific work has only just begun, and the available data in this field remain limited. Fortunately, utilization of new sequencing technologies allows expanded research on the association of intestinal bacterial flora and human diseases to be attempted. In this review, we summarize the role of gut microbiota in RA progression and address how specific bacterial strains regulate the immune response in disease process. Probiotics/prebiotics in the treatment of RA is also discussed.

Karakula-Juchnowicz, H., Dzikowski, M., Pelczarska, A., Dzikowska, I., and Juchnowicz, D. The brain-gut axis dysfunctions and hypersensitivity to food antigens in the etiopathogenesis of schizophrenia. Psychiatr.Pol. 2016; 50(4):747-760; PM: 27847926

ABSTRACT: Despite over 100-year history of research on schizophrenia, its etiology is still not fully understood, which might be due to the significant heterogeneity in terms of both its course, as well as the etiopathogenesis. One of the best-proven mediating mechanisms in the development of schizophrenia is the immuno-inflammatory response, the sources of which are believed to be the dysfunctions of brain-gut axis and pathological processes occurring in the intestines. This paper is a review of the literature on this subject which presents factors both involved in the functioning of brain-gut axis and important for the development of schizophrenia, i.e. 1. intestinal microbiome (intestinal microbiota), 2. permeable intestine (leaky gut syndrome), 3. hypersensitivity to food antigens, including gluten and casein of cow's milk. Research results seem to be very promising and indicate the possibility of improved clinical outcomes in some patients with schizophrenia by modifying diet, use of probiotics, and the implementation of antibiotic therapy of specific treatment groups. However, further research is needed on links between the intestinal microbiome and intestinal function as factors mediating the activation of the immune system and the development and further course of schizophrenia.

Karamese, M., Aydin, H., Sengul, E., Gelen, V., Sevim, C., Ustek, D., and Karakus, E. The Immunostimulatory Effect of Lactic Acid Bacteria in a Rat Model. Iran J.Immunol. 2016; 13(3):220-228; PM: 27671513

ABSTRACT: ***BACKGROUND***: Probiotics are "live", beneficial microbes that provide important health benefits in their hosts. There is significant interest in the modulation and regulation of the immune function by probiotics. ***OBJECTIVE***: To investigate the immunomodulatory effects of a probiotic mixture, including Lactobacillus and Bifidobacterium species, by detecting serum cytokine and immunoglobulin levels. ***METHODS***: The rats were randomly divided into 4 groups. The first group was "Control group" and other 3 groups were probiotic application groups who received different doses of probiotics. The probiotic mixture included 12 probiotic bacteria, mostly Lactobacillus and Bifidobacterium strains. Probiotic mixture was administered to rats for 12 consecutive days. TNF-alpha, TGF-beta, IL-1-beta, IL-6, and IL-10 levels as well as serum IgG and IgA concentrations were detected in the sera after 12 days. ***RESULTS***: Probiotics led to a decrease in the levels of TNF-alpha, IL-6 and TGF-beta; however, they led to increase in the serum levels of IL-10, IgG and IgA. There were significant differences

between control group and probiotic application groups (p<0.05). ***CONCLUSION***: These data suggest that the commensal microbiota are important for stimulating both proinflammatory and regulatory responses in order to rapidly clear infections and minimize inflammation-associated tissue damage.

Kato, K., Funabashi, N., Takaoka, H., Kohno, H., Kishimoto, T., Nakatani, Y., Matsumiya, G., and Kobayashi, Y. Lactobacillus paracasei endocarditis in a consumer of probiotics with advanced and severe bicuspid aortic valve stenosis complicated with diffuse left ventricular mid-layer fibrosis. Int.J.Cardiol. 12-1-2016; 224:157-161; PM: 27657466

No Abstract Available.

Kaur, R., Gulati, M., and Singh, S. K. Role of synbiotics in polysaccharide assisted colon targeted microspheres of mesalamine for the treatment of ulcerative colitis. Int.J.Biol.Macromol. 11-19-2016; 95:438-450; PM: 27876597

ABSTRACT: The present study explains the effect of concomitant administration of synbiotics with the polysaccharide based colon targeted delivery system. As the gut microflora get deranged on administration of mesalamine, drug release from such delivery systems is expected to be jeopardised because the release trigger is dependent solely on the colonic microbiota. To overcome this limitation, mesalamine was formulated as microspheres with guar gum and xanthan gum (prebiotics). These were combined with probiotics containing Lactobacillus acidophilus, L. rhamnosus, Bifidobacterium longumand Saccharomyces boulardi. Dissolution studies of the prepared formulation conducted in simulated colonic fluid clearly demonstrated its superiority over the marketed, delayed release dosage forms of mesalamine. The therapeutic benefit of the concomitant administration of synbiotic with mesalamine was evidenced by the comparative evaluation of faecal contents, weight gain trend and histopathological studies conducted in rats. The results suggest that co-administration of synbiotics with mesalamine can be used as a convenient methodology to achieve efficient and cost-effective targeting of the drug to colon.

Kelly, J. R., Allen, A. P., Temko, A., Hutch, W., Kennedy, P. J., Farid, N., Murphy, E., Boylan, G., Bienenstock, J., Cryan, J. F., Clarke, G., and Dinan, T. G. Lost in translation? The potential psychobiotic Lactobacillus rhamnosus (JB-1) fails to modulate stress or cognitive performance in healthy male subjects. Brain Behav.Immun. 11-16-2016; PM: 27865949

ABSTRACT: ***BACKGROUND***: Preclinical studies have identified certain probiotics as psychobiotics - live microorganisms with a potential mental health benefit. Lactobacillus rhamnosus (JB-1) has been shown to reduce stress-related behaviour, corticosterone release and alter central expression of GABA receptors in an anxious mouse strain. However, it is unclear if this single putative psychobiotic strain has psychotropic activity in humans. Consequently, we aimed to examine if these promising preclinical findings could be translated to healthy human volunteers. ***OBJECTIVES***: To determine the impact of L. rhamnosus on stress-related behaviours, physiology, inflammatory response, cognitive performance and brain activity patterns in healthy male participants. ***METHODS***: An 8week, randomized, placebo-controlled, cross-over design was employed. Twenty-nine healthy male volunteers participated. Participants completed self-report stress measures, cognitive assessments and resting electroencephalography (EEG).

Plasma IL10, IL1beta, IL6, IL8 and TNFalpha levels and whole blood Toll-like 4 (TLR-4) agonist-induced cytokine release were determined by multiplex ELISA. Salivary cortisol was determined by ELISA and subjective stress measures were assessed before, during and after a socially evaluated cold pressor test (SECPT). ***RESULTS***: There was no overall effect of probiotic treatment on measures of mood, anxiety, stress or sleep quality and no significant effect of probiotic over placebo on subjective stress measures, or the HPA response to the SECPT. Visuospatial memory performance, attention switching, rapid visual information processing, emotion recognition and associated EEG measures did not show improvement over placebo. No significant anti-inflammatory effects were seen as assessed by basal and stimulated cytokine levels. ***CONCLUSIONS***: L. rhamnosus was not superior to placebo in modifying stress-related measures, HPA response, inflammation or cognitive performance in healthy male participants. These findings highlight the challenges associated with moving promising preclinical studies, conducted in an anxious mouse strain, to healthy human participants. Future interventional studies investigating the effect of this psychobiotic in populations with stress-related disorders are required.

Kennedy, V., Van Laar, T. A., Aleru, O., Thomas, M., Ganci, M., and Rawat, M. Genome Sequences of Three Spore-Forming Bacteria Isolated from the Feces of Organically Raised Chickens. Genome Announc. 9-1-2016; 4(5); PM: 27587809

ABSTRACT: Antibiotic feed supplements have been implicated in the rise of multidrug-resistant bacteria. An alternative to antibiotics is probiotics. Here, we report the genome sequences of two Bacillus and one Solibacillus species, all spore-forming, Gram-positive bacteria, isolated from the feces organically raised chicken feces, with potential to serve as probiotics.

Kepert, I., Fonseca, J., Muller, C., Milger, K., Hochwind, K., Kostric, M., Fedoseeva, M., Ohnmacht, C., Dehmel, S., Nathan, P., Bartel, S., Eickelberg, O., Schloter, M., Hartmann, A., Schmitt-Kopplin, P., and Krauss-Etschmann, S. D-tryptophan from probiotic bacteria influences the gut microbiome and allergic airway disease. J.Allergy Clin.Immunol. 9-23-2016; PM: 27670239

ABSTRACT: ***BACKGROUND***: Chronic immune diseases, such as asthma, are highly prevalent. Currently available pharmaceuticals improve symptoms but cannot cure the disease. This prompted demands for alternatives to pharmaceuticals, such as probiotics, for the prevention of allergic disease. However, clinical trials have produced inconsistent results. This is at least partly explained by the highly complex crosstalk among probiotic bacteria, the host's microbiota, and immune cells. The identification of a bioactive substance from probiotic bacteria could circumvent this difficulty. ***OBJECTIVE***: We sought to identify and characterize a bioactive probiotic metabolite for potential prevention of allergic airway disease. ***METHODS***: Probiotic supernatants were screened for their ability to concordantly decrease the constitutive CCL17 secretion of a human Hodgkin lymphoma cell line and prevent upregulation of costimulatory molecules of LPS-stimulated human dendritic cells. ***RESULTS***: Supernatants from 13 of 37 tested probiotic strains showed immunoactivity. Bioassay-guided chromatographic fractionation of 2 supernatants according to polarity, followed by total ion chromatography and mass spectrometry, yielded C11H12N2O2 as the molecular formula of a bioactive substance. Proton nuclear magnetic resonance and enantiomeric separation identified D-tryptophan. In contrast, L-tryptophan and 11 other D-amino acids were inactive. Feeding

D-tryptophan to mice before experimental asthma induction increased numbers of lung and gut regulatory T cells, decreased lung TH2 responses, and ameliorated allergic airway inflammation and hyperresponsiveness. Allergic airway inflammation reduced gut microbial diversity, which was increased by D-tryptophan. ***CONCLUSIONS***: D-tryptophan is a newly identified product from probiotic bacteria. Our findings support the concept that defined bacterial products can be exploited in novel preventative strategies for chronic immune diseases.

Khailova, L., Baird, C. H., Rush, A. A., Barnes, C., and Wischmeyer, P. E. Lactobacillus rhamnosus GG treatment improves intestinal permeability and modulates inflammatory response and homeostasis of spleen and colon in experimental model of Pseudomonas aeruginosa pneumonia. Clin.Nutr. 10-1-2016; PM: 27745813

ABSTRACT: ***BACKGROUND & AIMS***: Recent clinical trials and in vivo models demonstrate probiotic administration can reduce occurrence and improve outcome of pneumonia and sepsis, both major clinical challenges worldwide. Potential probiotic benefits include maintenance of gut epithelial barrier homeostasis and prevention of downstream organ dysfunction due to systemic inflammation. However, mechanism(s) of probiotic-mediated protection against pneumonia remain poorly understood. This study evaluated potential mechanistic targets in the maintenance of gut barrier homeostasis following Lactobacillus rhamnosus GG (LGG) treatment in a mouse model of pneumonia. ***METHODS***: Studies were performed in 6-8 week old FVB/N mice treated (o.g.) with or without LGG (109 CFU/ml) and intratracheally injected with Pseudomonas aeruginosa or saline. At 4, 12, and 24 h post-bacterial treatment spleen and colonic tissue were collected for analysis. ***RESULTS***: Pneumonia significantly increased intestinal permeability and gut claudin-2. LGG significantly attenuated increased gut permeability and claudin-2 following pneumonia back to sham control levels. As mucin expression is key to gut barrier homeostasis we demonstrate that LGG can enhance goblet cell expression and mucin barrier formation versus control pneumonia animals. Further as Muc2 is a key gut mucin, we show LGG corrected deficient Muc2 expression post-pneumonia. Apoptosis increased in both colon and spleen post-pneumonia, and this increase was significantly attenuated by LGG. Concomitantly, LGG corrected pneumonia-mediated loss of cell proliferation in colon and significantly enhanced cell proliferation in spleen. Finally, LGG significantly reduced pro-inflammatory cytokine gene expression in colon and spleen post-pneumonia. ***CONCLUSIONS***: These data demonstrate LGG can maintain intestinal barrier homeostasis by enhancing gut mucin expression/barrier formation, reducing apoptosis, and improving cell proliferation. This was accompanied by reduced pro-inflammatory cytokine expression in the gut and in a downstream organ (spleen). These may serve as potential mechanistic targets to explain LGG's protection against pneumonia in the clinical and in vivo setting.

Khorasani, A. C. and Shojaosadati, S. A. Pectin-non-starch nanofibers biocomposites as novel gastrointestinal-resistant prebiotics. Int.J.Biol.Macromol. 2017; 94(Pt A):131-144; PM: 27720960

ABSTRACT: Incorporation of nanofibers of chitin (NC), lignocellulose (NLC) and bacterial cellulose (BNC) in pectin was studied to improve prebiotic activity and gastrointestinal resistance of the pectin-nanofibers biocomposites for protection of probiotics under simulated gastrointestinal conditions. The biocomposites were prepared using various compositions of pectin and nanofibers, which were designed using D-optimal mixture

method. The incorporation of the nanofibers in pectin led to a slow degradation of the pectin-nanofibers biocomposites in contrast to their rapid swelling. AFM analysis indicated the homogenous distribution of interconnected nanofibers network structure in the pectin-nanofibers biocomposite. FTIR spectra demonstrated fabrication of the biocomposites based on the inter- and intra-molecular hydrogen bonding and ionic interaction of pectin-Ca2+. XRD patterns revealed the amorphous structures of the biocomposites as compared to the crystalline structures of the nanofibers. Among the compositions, the optimal compositions were as follows: 60% pectin+40% NC, 50% pectin+50% NLC and 60% pectin+40% BNC, where the prebiotic score, probiotic survival under simulated gastric and intestinal conditions were optimum. The optimal biocomposite pectin-NC exhibited the highest survival of the entrapped probiotic bacteria under simulated gastric (97.7%) and intestinal (95.8%) conditions when compared with the corresponding to free cells (76.2 and 73.4%).

Kigerl, K. A., Hall, J. C., Wang, L., Mo, X., Yu, Z., and Popovich, P. G. Gut dysbiosis impairs recovery after spinal cord injury. J.Exp.Med. 11-14-2016; 213(12):2603-2620; PM: 27810921

ABSTRACT: The trillions of microbes that exist in the gastrointestinal tract have emerged as pivotal regulators of mammalian development and physiology. Disruption of this gut microbiome, a process known as dysbiosis, causes or exacerbates various diseases, but whether gut dysbiosis affects recovery of neurological function or lesion pathology after traumatic spinal cord injury (SCI) is unknown. Data in this study show that SCI increases intestinal permeability and bacterial translocation from the gut. These changes are associated with immune cell activation in gut-associated lymphoid tissues (GALTs) and significant changes in the composition of both major and minor gut bacterial taxa. Postinjury changes in gut microbiota persist for at least one month and predict the magnitude of locomotor impairment. Experimental induction of gut dysbiosis in naive mice before SCI (e.g., via oral delivery of broad-spectrum antibiotics) exacerbates neurological impairment and spinal cord pathology after SCI. Conversely, feeding SCI mice commercial probiotics (VSL#3) enriched with lactic acid-producing bacteria triggers a protective immune response in GALTs and confers neuroprotection with improved locomotor recovery. Our data reveal a previously unknown role for the gut microbiota in influencing recovery of neurological function and neuropathology after SCI.

Kim, S., Wang, G., Lobaton, G., Li, E., Yang, T., and Raizada, M. OS 05-10 THE MICROBIAL METABOLITE, BUTYRATE ATTENUATES ANGIOTENSIN II-INDUCED HYPERTENSION AND DYSBIOSIS. J.Hypertens. 2016; 34 Suppl 1 - ISH 2016 Abstract Book: e60-e61; PM: 27753935

*ABSTRACT: **OBJECTIVE***: Our previous studies have demonstrated that gut microbial dysbiosis is linked to high blood pressure in patients. This was associated with decreases in butyrate- and acetate- producing microbial populations. Thus, our objective in this study was to investigate the hypothesis that infusion of butyrate would impact dysbiosis, gut immunity and attenuate hypertension. ***DESIGN AND METHOD***: C57B6 mice were divided into 4 groups; Saline infused, Angiotensin II (750ng/kg/min) infused, Ang II infused and butyrate treated (0.5mg/kg) and butyrate alone treated group. 4 weeks after treatment, fecal samples were analyzed by 16S bacterial ribosomal DNA sequencing for gut microbiome identification. Intestinal leukocytes were harvested by enzyme digestion followed by percoll gradients, and the collected cells were analyzed using FACS. ***RESULTS***: Ang II caused 46mmHg increase in mean arterial pressure (MAP). Butyrate co-

administrated group showed significant reduction in MAP (148.3 +/- 8.1 mmHg, Ang II vs. 120.5 +/- 11.2 mmHg, Ang II + butyrate). Microbiota analysis demonstrated decreases of bacteroidetes (30.2 +/- 4.8%, control vs.14.0 +/- 2.6%, Ang II) and gut dysbiosis (F/B ratio 2.66, control vs. 4.38, Ang II) in Ang II infused animals. The genus level analysis showed there were marked decreases of butyrate producing bacteria (3.27 +/- 0.8%, control vs. 2.09 +/- 0.5%, Ang II) in the Ang II group. Analysis of mucosal T cells in lamina propria from these groups showed that there was a significant increase of CCR2 Th17 cells in Ang II infused mice, but not in butyrate co-treated mice. ***CONCLUSIONS***: These observations show that gut dysbiosis and the decrease of butyrate producing bacteria are associated with Ang II-induced hypertension. Supplementing butyrate in Ang II treated mice attenuated hypertension and gut dysbiosis, as well as normalizing the intestinal Th17 cells that respond to inflammation. These data suggest that gut bacteria produced butyrate plays an important role in blood pressure regulation and that butyrate producing bacteria could be considered as a novel probiotic therapy for hypertension.

Kinoshita, H., Ohuchi, S., Arakawa, K., Watanabe, M., Kitazawa, H., and Saito, T. Isolation of lactic acid bacteria bound to the porcine intestinal mucosa and an analysis of their moonlighting adhesins. Biosci.Microbiota.Food Health. 2016; 35(4):185-196; PM: 27867805

ABSTRACT: The adhesion of lactic acid bacteria (LAB) to the intestinal mucosa is one of the criteria in selecting for probiotics. Eighteen LAB were isolated from porcine intestinal mucin (PIM): ten strains of Lactobacillus, six strains of Weissella, and two strains of Streptococcus. Using sodium dodecyl sulfate polyacrylamide gel electrophoresis (SDS-PAGE) for phosphate-buffered saline (PBS) extracts from the LAB, many bands were detected in half of the samples, while a few and/or no clear bands were detected in the other half. All six of the selected LAB showed adhesion to PIM. L. johnsonii MYU 214 and MYU 221 showed adhesion at more than 10%. W. viridescens MYU 208, L. reuteri MYU 213, L. mucosae MYU 225, and L. agilis MYU 227 showed medium levels of adhesion at 5.9-8.3%. In a comprehensive analysis for the adhesins in the PBS extracts using a receptor overlay analysis, many moonlighting proteins were detected and identified as candidates for adhesins: GroEL, enolase, and elongation factor Tu in MYU 208; peptidase C1, enolase, formyl-CoA transferase, phosphoglyceromutase, triosephosphate isomerase, and phosphofructokinase in MYU 221; and DnaK, enolase, and phosphoglycerate kinase in MYU 227. These proteins in the PBS extracts, which included such things as molecular chaperones and glycolytic enzymes, may play important roles as adhesins.

Kivanc, S. A., Kivanc, M., and Yigit, T. Antibiotic susceptibility, antibacterial activity and characterisation of Enterococcus faecium strains isolated from breast milk. Exp.Ther.Med. 2016; 12(3):1732-1740; PM: 27602088

ABSTRACT: Enterococci, which have useful biotechnological applications, produce bacteriocins, including those that exert anti-Listerial activity. The present study aimed to determine the antibiotic susceptibility patterns and antimicrobial activity of Enterococcus faecium strains isolated from human breast milk. The strains were identified using carbohydrate fermentation tests and ribotyping. Subsequently, the antibacterial activity of the isolates was investigated, and the quantities of lactic acid and hydrogen peroxide produced, and the proteolytic activity of E. faecium, were determined. In addition, biofilm formation by E. faecium strains was assessed. E. faecium strains exhibited antimicrobial activity against food-borne and clinical bacterial isolates. Furthermore,

following 24 h incubation, the tested strains exhibited resistance to a pH range of 2.0-9.5 and tolerance of bile acid, lysozyme activity and phenol. Supernatants of the E. faecium TM13, TM15, TM17 and TM18 strains were shown to be effective against Listeria monocytogenes, and were also resistant to heat. Further studies are required in order to determine whether certain strains of E. faecium may be used for the development of novel antibacterial agents.

Kobyliak, N., Falalyeyeva, T., Bodnar, P., and Beregova, T. Probiotics Supplemented with Omega-3 Fatty Acids are More Effective for Hepatic Steatosis Reduction in an Animal Model of Obesity. Probiotics.Antimicrob.Proteins. 9-22-2016; PM: 27660157

ABSTRACT: Today probiotics have been suggested as a treatment for the prevention of NAFLD. Omega-3 fatty acid supplementation may have beneficial effects in regulating hepatic lipid metabolism, adipose tissue function and inflammation. The present study was designed to determine whether probiotics plus omega-3 are superior to probiotics alone on the monosodium glutamate (MSG)-induced NAFLD model in rats. We included 60 rats divided into four groups, 15 animals in each. Rats of group I were intact. Newborn rats of groups II-IV were injected with MSG. The III (Symbiter) group received 2.5 ml/kg of multiprobiotic "Symbiter" containing concentrated biomass of 14 probiotic bacteria genera. The IV (Symbiter-Omega) groups received "Symbiter-Omega" combination of probiotic biomass supplemented with flax and wheat germ oil (250 mg of each, concentration of omega-3 fatty acids 1-5 %). In both interventional groups reduction in total NAS score was observed. Supplementation of alive probiotic mixture with omega-3 fatty acids lead to 20 % higher decrease in steatosis score (0.73 +/- 0.11 vs 0.93 +/- 0.22, p = 0.848) and reduction by 16.6 % of triglycerides content in liver as compared to probiotic alone. Our study demonstrated more pronounced reduction in hepatic steatosis and hepatic lipid accumulation after treatment with combination of alive probiotics and omega-3 as compared to probiotics alone.

Koch, M. Gut Microbiota and the Liver: A Tale of 2 Cities: A Narrative View in 2 Acts. J.Clin.Gastroenterol. 2016; 50 Suppl 2, Proceedings from the 8th Probiotics, Prebiotics & New Foods for Microbiota and Human Health meeting held in Rome, Italy on September 13-15, 2015:S183-S187; PM: 27741171

ABSTRACT: Microbes are mostly important for the digestion of food, the absorption of some micronutrients, and the production of vitamins. The microbiota stimulates lymphoid structures in the gastrointestinal mucosa and decrease pathogens by competing for nutrients and space. Bacterial translocation is defined as the escape of gut bacteria and their products through the intestinal mucosa to the outside of the intestine as portovenous or systemic circulation. This is induced by a leaky gut barrier. There is evidence for a role of intestinal permeability in the pathogenesis of nonalcoholic fatty liver disease. In the liver, bacterial products can bind to their specific pathogen recognition receptors on parenchymal and nonparenchymal cells, producing an inflammatory response and enhancing disease progression. When binding, bacterial products bind to their receptors, initiating intracellular signalling and inducing an inflammatory cascade, thus accelerating liver cell damage and fibrosis. However, the liver can also increase gut permeability, producing proinflammatory cytokines, and reversing them into the blood stream. Modification of the gut microbiota could lead to benefit in patients with liver disease. Nonabsorbable antibiotics (rifaximin) prevent and

relieve overt encephalopathy. Probiotics alone are not capable of turning back overt encephalopathy, but could prevent its development. There is some evidence that probiotics could relent the progression of nonalcoholic liver disease, and possibly reverse steatosis. Antibiotics, such as fluoroquinolones, reduce the risk of development of the first episode of spontaneous bacterial peritonitis and mortality in cirrhotic patients.

Konishi, H., Fujiya, M., Tanaka, H., Ueno, N., Moriichi, K., Sasajima, J., Ikuta, K., Akutsu, H., Tanabe, H., and Kohgo, Y. Probiotic-derived ferrichrome inhibits colon cancer progression via JNK-mediated apoptosis. Nat.Commun. 8-10-2016; 7:12365; PM: 27507542

ABSTRACT: Previous reports have suggested that some probiotics inhibit tumorigenesis and cancer progression. However, the molecules involved have not yet been identified. Here, we show that the culture supernatant of Lactobacillus casei ATCC334 has a strong tumour-suppressive effect on colon cancer cells. Using mass spectrometry, we identify ferrichrome as a tumour-suppressive molecule produced by L. casei ATCC334. The tumour-suppressive effect of ferrichrome is greater than that of cisplatin and 5-fluorouracil, and ferrichrome has less of an effect on non-cancerous intestinal cells than either of those agents. A transcriptome analysis reveals that ferrichrome treatment induces apoptosis, which is mediated by the activation of c-jun N-terminal kinase (JNK). Western blotting indicates that the induction of apoptosis by ferrichrome is reduced by the inhibition of the JNK signalling pathway. This we demonstrate that probiotic-derived ferrichrome exerts a tumour-suppressive effect via the JNK signalling pathway.

Krishnan, M., Penrose, H. M., Shah, N. N., Marchelletta, R. R., and McCole, D. F. VSL#3 Probiotic Stimulates T-cell Protein Tyrosine Phosphatase-mediated Recovery of IFN-gamma-induced Intestinal Epithelial Barrier Defects. Inflamm.Bowel.Dis. 2016; 22(12):2811-2823; PM: 27824650

ABSTRACT: ***BACKGROUND***: VSL#3 is a probiotic compound that has been used in the treatment of inflammatory bowel disease. T-cell protein tyrosine phosphatase (TCPTP) is the protein product of the inflammatory bowel disease candidate gene, PTPN2, and we have previously shown that it protects epithelial barrier function. The aim of this study was to investigate whether VSL#3 improves intestinal epithelial barrier function against the effects of the inflammatory bowel disease-associated proinflammatory cytokine, interferon-gamma (IFN-gamma) through activation of TCPTP. ***METHODS***: Polarized monolayers of T84 intestinal epithelial cells were treated with increasing concentrations of VSL#3 to determine effects on TCPTP expression and enzymatic activity. Therapeutic effects of VSL#3 against barrier disruption by IFN-gamma were measured by transepithelial electrical resistance and fluorescein isothiocyanate-dextran permeability. A novel TCPTP-deficient HT-29 intestinal epithelial cell line was generated to study the role of TCPTP in mediating the effects of VSL#3. Tight junction protein distribution was assessed with confocal microscopy. ***RESULTS***: VSL#3 increased TCPTP protein levels and enzymatic activity, correlating with a VSL#3-induced decrease in IFN-gamma signaling. VSL#3 corrected the decrease in transepithelial electrical resistance and the increase in epithelial permeability induced by IFN-gamma. Moreover, the restorative effect of VSL#3 against IFN-gamma signaling, epithelial permeability defects, altered expression and localization of the tight junction proteins claudin-2, occludin, and zonula occludens-1, were not realized in stable TCPTP/(PTPN2)-deficient HT-29 intestinal epithelial cells. ***CONCLUSIONS***: VSL#3 reduces IFN-gamma signaling and IFN-gamma-induced epithelial

barrier defects in a TCPTP-dependent manner. These data point to a key role for TCPTP as a therapeutic target for restoration of barrier function using probiotics.

Ku, S., Park, M. S., Ji, G. E., and You, H. J. Review on Bifidobacterium bifidum BGN4: Functionality and Nutraceutical Applications as a Probiotic Microorganism. Int.J.Mol.Sci. 9-14-2016; 17(9); PM: 27649150

ABSTRACT: Bifidobacterium bifidum BGN4 is a probiotic strain that has been used as a major ingredient to produce nutraceutical products and as a dairy starter since 2000. The various bio-functional effects and potential for industrial application of B. bifidum BGN4 has been characterized and proven by in vitro (i.e., phytochemical bio-catalysis, cell adhesion and anti-carcinogenic effects on cell lines, and immunomodulatory effects on immune cells), in vivo (i.e., suppressed allergic responses in mouse model and anti-inflammatory bowel disease), and clinical studies (eczema in infants and adults with irritable bowel syndrome). Recently, the investigation of the genome sequencing was finished and this data potentially clarifies the biochemical characteristics of B. bifidum BGN4 that possibly illustrate its nutraceutical functionality. However, further systematic research should be continued to gain insight for academic and industrial applications so that the use of B. bifidum BGN4 could be expanded to result in greater benefit. This review deals with multiple studies on B. bifidum BGN4 to offer a greater understanding as a probiotic microorganism available in functional food ingredients. In particular, this work considers the potential for commercial application, physiological characterization and exploitation of B. bifidum BGN4 as a whole.

Kueneman, J. G., Woodhams, D. C., Harris, R., Archer, H. M., Knight, R., and McKenzie, V. J. Probiotic treatment restores protection against lethal fungal infection lost during amphibian captivity. Proc.Biol.Sci. 9-28-2016; 283(1839); PM: 27655769

ABSTRACT: Host-associated microbiomes perform many beneficial functions including resisting pathogens and training the immune system. Here, we show that amphibians developing in captivity lose substantial skin bacterial diversity, primarily due to reduced ongoing input from environmental sources. We combined studies of wild and captive amphibians with a database of over 1 000 strains that allows us to examine antifungal function of the skin microbiome. We tracked skin bacterial communities of 62 endangered boreal toads, Anaxyrus boreas, across 18 time points, four probiotic treatments, and two exposures to the lethal fungal pathogen Batrachochytrium dendrobatidis (Bd) in captivity, and compared these to 33 samples collected from wild populations at the same life stage. As the amphibians in captivity lost the Bd-inhibitory bacteria through time, the proportion of individuals exposed to Bd that became infected rose from 33% to 100% in subsequent exposures. Inoculations of the Bd-inhibitory probiotic Janthinobacterium lividum resulted in a 40% increase in survival during the second Bd challenge, indicating that the effect of microbiome depletion was reversible by restoring Bd-inhibitory bacteria. Taken together, this study highlights the functional role of ongoing environmental inputs of skin-associated bacteria in mitigating a devastating amphibian pathogen, and that long-term captivity decreases this defensive function.

Kumar, A., Anbazhagan, A. N., Coffing, H., Chatterjee, I., Priyamvada, S., Gujral, T., Saksena, S., Gill, R. K., Alrefai, W. A., Borthakur, A., and Dudeja, P. K. Lactobacillus acidophilus counteracts inhibition of NHE3 and DRA expression and alleviates diarrheal phenotype in

mice infected with Citrobacter rodentium. Am.J.Physiol Gastrointest.Liver Physiol. 11-1-2016; 311(5):G817-G826; PM: 27634011

ABSTRACT: Impaired absorption of electrolytes is a hallmark of diarrhea associated with inflammation or enteric infections. Intestinal epithelial luminal membrane NHE3 (Na+/H+ exchanger 3) and DRA (Down-Regulated in Adenoma; Cl-/.

Kumar, M., Kissoon-Singh, V., Leon, Coria A., Moreau, F., and Chadee, K. The probiotic mixture VSL#3 reduces colonic inflammation and improves intestinal barrier function in Muc2 mucin deficient mice. Am.J.Physiol Gastrointest.Liver Physiol. 11-17-2016; ajpgi; PM: 27856417

ABSTRACT: MUC2 mucin is the major glycoprotein in colonic mucus that separates intestinal microbiota from underlying host cells and serves as a food source for some eubacteria. MUC2 deficiency results in impaired epithelial barrier function, imbalance in gut microbiota and spontaneous colitis. Use of probiotics has been shown to have protective effect against colitis. In this study we tested whether the probiotic mixture VSL#3 required an intact mucin barrier to exert its beneficial effect using Muc2 mucin deficient (Muc2-/-) and Muc2+/+ littermates. VSL#3 treatment alone reduced basal colonic pro-inflammatory cytokine levels and improved epithelial barrier functions in Muc2-/- animals. Likewise, in DSS-induced colitis, VSL#3 administration dampened the pro-inflammatory chemokines KC, MCP-1 and MIP-2 and up regulated tissue regeneration growth factors TGF-beta, FGF-1 and VEGF-A that accelerated resolution of colitis symptoms in Muc2-/- animals. Importantly, improved colonic health in VSL#3 treated animals was associated with attenuated reactive oxygen species (ROS) production by peritoneal macrophages, restoration of antimicrobial peptide gene expression in the small intestine and increased abundance of bacterial commensals in the gut. The beneficial effects of VSL#3 in Muc2-/- animals were mediated by acetate, an important short chain fatty acid produced by gut bacteria. These studies provide evidence for the first time that VSL#3 can enhance epithelial barrier function by dampening pro-inflammatory cytokines and chemokine response, accelerating restitution and altering commensal microbiota in the absence of a functional mucus barrier.

Kumar, N., Kumar, V., Panwar, R., and Ram, C. Efficacy of indigenous probiotic Lactobacillus strains to reduce cadmium bioaccessibility - An in vitro digestion model. Environ.Sci.Pollut.Res.Int. 10-21-2016; PM: 27770327

ABSTRACT: The toxic heavy metal cadmium (Cd) appears as one of the major global threats to human and animal health. Human being and aquatic life are exposed to Cd by breathing, eating, or drinking when industrial effluents released into environment. The study was aimed to identify cadmium-binding Lactobacillus strain to reduce its bioaccessibility in in vitro digestion model. In this context, forty-eight lactobacilli strains isolated and characterized from fermented dairy products and human origin were screened for their Cd biosorption potential using Flame Atomic Absorption Spectroscopy (FAAS). The present study revealed that Cd biosorption potential of 48 lactobacilli strains ranged from 1.0832 +/- 0.012 to 3.562 +/- 0.03 mg Cd g-1 of cells from initial 10 mg L-1 cadmium chloride (CdCl2) aqueous solution. Lactobacillus plantarum strain HD 48 demonstrated highest biosorption of 3.562 +/- 0.03 mg Cd g-1 of cells. Lactobacilli-Cd complex stability indicated its strong stability as even after three washes with Milli-Q

water metal desorption was nonsignificant ($p < 0.05$) and further studies to delineate the influence of Cd (100 mg L-1 CdCl2) on their growth. Moreover, these strains were able to reduce Cd bioaccessibility in the in vitro digestion model in the range of 24.71 to 41.62 %. Transmission electron microscopy (TEM) investigations on Cd bioadsorption also revealed its surface associated bioadsorption phenomenon. These findings depicted that probiotic strain L. plantarum HD 48 was found to be endowed with remarkable Cd biosorption ability as well as reduction in its bioaccessibility. These results suggest that probiotic strain L. plantarum HD 48 has immense potential to sequester Cd from aqueous solution which could be further explored as a potent source to diminish body Cd burden.

Kwak, M. J., Kwon, S. K., Yoon, J. K., Song, J. Y., Seo, J. G., Chung, M. J., and Kim, J. F. Evolutionary architecture of the infant-adapted group of Bifidobacterium species associated with the probiotic function. Syst.Appl.Microbiol. 2016; 39(7):429-439; PM: 27524178

ABSTRACT: Bifidobacteria, often associated with the gastrointestinal tract of animals, are well known for their roles as probiotics. Among the dozens of Bifidobacterium species, Bifidobacterium bifidum, B. breve, and B. longum are the ones most frequently isolated from the feces of infants and known to help the digestion of human milk oligosaccharides. To investigate the correlation between the metabolic properties of bifidobacteria and their phylogeny, we performed a phylogenomic analysis based on 452 core genes of forty-four completely sequenced Bifidobacterium species. Results show that a major evolutionary event leading to the clade of the infant-adapted species is linked to carbohydrate metabolism, but it is not the only factor responsible for the adaptation of bifidobacteria to the gut. The genome of B. longum subsp. infantis, a typical bifidobacterium in the gut of breast-fed infants, encodes proteins associated with several kinds of species-specific metabolic pathways, including urea metabolism and biosynthesis of riboflavin and lantibiotics. Our results demonstrate that these metabolic features, which are associated with the probiotic function of bifidobacteria, are species-specific and highly correlate with their phylogeny.

Kwong, K. K. and Chan, S. T. Oxytocin, a missing link of the gut-brain axis between probiotics and chronic migraine?Comment on 'The effects of the multispecies probiotic mixture Ecologic(R)Barrier on migraine:results of an open-label pilot study'. Benef.Microbes. 11-30-2016; 7(5):623-624; PM: 27892710

No Abstract Available.

Lahner, E. and Annibale, B. Probiotics and Diverticular Disease: Evidence-based? J.Clin.Gastroenterol. 2016; 50 Suppl 2, Proceedings from the 8th Probiotics, Prebiotics & New Foods for Microbiota and Human Health meeting held in Rome, Italy on September 13-15, 2015:S159-S160; PM: 27741164

ABSTRACT: Diverticular disease (DD) is a common gastrointestinal condition. Clinical spectrum ranges from asymptomatic diverticulosis to symptomatic uncomplicated or complicated DD. Symptoms related to uncomplicated DD are not specific and may be indistinguishable from those of irritable bowel syndrome. Low-grade inflammation, altered intestinal microbiota, visceral hypersensitivity, and abnormal colonic motility have been identified as factors potentially contributing to symptoms. Probiotics may modify

the gut microbial balance leading to health benefits. Probiotics, due to their anti-inflammatory effects and ability to maintain an adequate bacterial colonization in the colon, are promising treatment options for DD. This review focuses on the available evidence on the efficacy of prebiotics in uncomplicated DD.

Lalles, J. P. Microbiota-host interplay at the gut epithelial level, health and nutrition. J.Anim Sci.Biotechnol. 2016; 7:66; PM: 27833747

ABSTRACT: Growing evidence suggests the implication of the gut microbiota in various facets of health and disease. In this review, the focus is put on microbiota-host molecular cross-talk at the gut epithelial level with special emphasis on two defense systems: intestinal alkaline phosphatase (IAP) and inducible heat shock proteins (iHSPs). Both IAP and iHSPs are induced by various microbial structural components (e.g. lipopolysaccharide, flagellin, CpG DNA motifs), metabolites (e.g. n-butyrate) or secreted signal molecules (e.g., toxins, various peptides, polyphosphate). IAP is produced in the small intestine and secreted into the lumen and in the interior milieu. It detoxifies microbial components by dephosphorylation and, therefore, down-regulates microbe-induced inflammation mainly by inhibiting NF-kappaB pro-inflammatory pathway in enterocytes. IAP gene expression and enzyme activity are influenced by the gut microbiota. Conversely, IAP controls gut microbiota composition both directly, and indirectly though the detoxification of pro-inflammatory free luminal adenosine triphosphate and inflammation inhibition. Inducible HSPs are expressed by gut epithelial cells in proportion to the microbial load along the gastro-intestinal tract. They are also induced by various microbial components, metabolites and secreted molecules. Whether iHSPs contribute to shape the gut microbiota is presently unknown. Both systems display strong anti-inflammatory and anti-oxidant properties that are protective to the gut and the host. Importantly, epithelial gene expressions and protein concentrations of IAP and iHSPs can be stimulated by probiotics, prebiotics and a large variety of dietary components, including macronutrients (protein and amino acids, especially L-glutamine, fat, fiber), and specific minerals (e.g. calcium) and vitamins (e.g. vitamins K1 and K2). Some food components (e.g. lectins, soybean proteins, various polyphenols) may inhibit or disturb these systems. The general cellular and molecular mechanisms involved in the microbiota-host epithelial crosstalk and subsequent gut protection through IAP and iHSPs are reviewed along with their nutritional modulation. Special emphasis is also given to the pig, an economically important species and valuable biomedical model.

Lan, R. X., Lee, S. I., and Kim, I. H. Effects of multistrain probiotics on growth performance, nutrient digestibility, blood profiles, faecal microbial shedding, faecal score and noxious gas emission in weaning pigs. J.Anim Physiol Anim Nutr.(Berl). 2016; 100(6):1130-1138; PM: 27870289

ABSTRACT: This experiment was conducted to investigate the efficacy of multistrain probiotics in weaning pigs. A total of 125 28-day-old weaning pigs [(Landrace x Yorkshire) x Duroc] with an initial average body weight (BW) of 7.26 +/- 0.76 kg were randomly allotted into 5 treatments, 5 replicate pens/treatment with 5 pigs/pen for 42-day experiment. Dietary treatments were as follows: CON, basal diet; PC1, CON + 0.01% multistrain probiotics; PC2, CON + 0.03% multistrain probiotics; PC3, CON + 0.06% multistrain probiotics; PC4, CON + 0.1% multistrain probiotics. On day 14, pigs fed the PC4 diet had higher BW gain than pigs fed the CON diet. On day 42, pigs fed multistrain probiotics supplementation diets had higher BW gain than pigs fed the CON diet. From

days 1 to 14, pigs fed the PC2, PC3 and PC4 diets had higher ($p < 0.05$) ADG than pigs fed the CON diet. From day 15 to 42, pigs fed the multistrain probiotics supplementation diets had higher ($p < 0.05$) average daily gain (ADG) and gain: feed ratio (G:F) than pigs fed the CON diet. In the overall period, pigs fed the multistrain probiotics supplementation diets had higher ($p < 0.05$) ADG and pigs fed the PC2 and PC4 diets had higher ($p < 0.05$) G:F than pigs fed the CON diet. On day 42, pigs fed the PC4 diet had higher ($p < 0.05$) apparent total tract digestibility (ATTD) of dry matter (DM), nitrogen (N) and gross energy (GE), faecal Lactobacillus counts and lower ($p < 0.05$) E. coli counts and NH3 emission than pigs fed the CON diet. Pigs fed the multistrain probiotics supplementation diets had lower ($p < 0.05$) H2 S and total mercaptans emissions than pigs fed the CON diet. Conclusions, dietary supplementation with 0.1% probiotics improved growth performance, nutrition digestibility and intestinal microflora balance and decreased faecal noxious gas emissions in weaning pigs.

Lee, C. Y., Shih, H. C., Yu, M. C., Lee, M. Y., Chang, Y. L., Lai, Y. Y., Lee, Y. C., Kuan, Y. H., and Lin, C. C. Evaluation of the potential inhibitory activity of a combination of L. Acidophilus, L. Rhamnosus and L. Sporogenes on Helicobacter Pylori: A randomized double-blind placebo-controlled clinical trial. Chin J.Integr.Med. 10-19-2016; PM: 27761791

ABSTRACT: ***OBJECTIVE***: To investigate whether three strains of probiotics, L. acidophilus, L. rhamnosus, and L. sporogenes, had significant inhibitive effects on Helicobacter pylori (H. pylori). ***METHODS***: This is a 4-week, randomly assigned, parallel-group, doubled-blind, and placebo-controlled study. Fifty patients with a positive H. pylori infection urea breath test (DeltaUBT) result > 10% and without ulcer symptoms were randomized into a treatment group and a placebo group by a computer generated allocation sheet with 1:1. These subjects took one capsule of probiotics or placebo twice daily. The primary measurement was the change in DeltaUBT values. ***RESULTS***: The DeltaUBT values during the 4-week treatment period and the 2-week follow-up period were not significantly different between the treatment group and the placebo group, indicating that the inhibitive effects on H. pylori were comparable between both groups. The monocyte count (%) was 5.77+/-1.11 in the treatment group versus 5.09+/-1.12 in the placebo group (P=0.044), and the basophile count was 0.55+/-0.32 in the treatment group versus 0.36+/-0.23 in the placebo group (P=0.024) at week 2 of the treatment period, both of which reached statistical significance. The monocyte count was 5.75+/-1.26 in the treatment group and 4.72+/-0.99 in the placebo group at the end of the follow-up period (P=0.003). ***CONCLUSIONS***: There was no significant inhibitive effects of the three probiotic strains (L. acidophilus, L. rhamnosus, and L. sporogenes) on H. pylori. Probiotics can not play the same role as antibiotics in the eradication of H. pylori, the role of probiotics is likely to be important as adjuvant to the triple or quadruple therapy for H. pylori, especially in resistance cases.

Lee, J. S., Kim, M. J., Park, S. H., Lee, S. B., Wang, T., Jung, U. S., Im, J., Kim, E. J., Lee, K. W., and Lee, H. G. Effects of dietary mixture of garlic (Allium sativum), coriander (Coriandrum sativum) and probiotics on immune responses and caecal counts in young laying hens. J.Anim Physiol Anim Nutr.(Berl). 9-28-2016; PM: 27678135

ABSTRACT: This study was conducted to evaluate the effects of a combined mixture of phytogenic extracts (garlic and coriander) and probiotics on growth performance and immune responses in laying hens based on the results of in vitro studies to screen for immunomodulatory potency of each ingredient. Several parameters of

immunomodulatory potency were estimated using lamina propria leucocytes (LPLs) isolated from rat intestinal mucosa tissue. Results show that the combined mixture enhanced LPLs proliferation, increased LPL-mediated cytotoxicity against YAC-1 tumour cells, and decreased lipopolysaccharide (LPS)-induced cytokine production including tumour necrosis factor-alpha (TNF-alpha) and interferon-gamma (IFN-gamma) in LPLs. For in vivo study, laying hens (n = 50/each diet group) were fed with control diet, a diet containing antibiotics (0.01% per kg feed) or the combined mixture (0.02% per kg feed) for 21 days. The dietary combined mixture improved egg production ($p < 0.05$) but not growth performance and carcass traits. Interestingly, the patterns of suppressing plasma IFN-gamma productions during inflammation by LPS injection and decreasing caecal E. coli counts in the combined mixture group were comparable to those in the antibiotics group. Taken together, our results suggested that the 0.02% of combined mixture of phytogenic extracts and probiotics as ingredients has potential immunomodulatory effects in laying hens.

Leelahavanichkul, A., Panpetch, W., Worasilchai, N., Somparn, P., Chancharoenthana, W., Nilgate, S., Finkelman, M., Chindamporn, A., and Tumwasorn, S. Evaluation of gastrointestinal leakage using serum (1-->3)-beta-D-glucan in a Clostridium difficile murine model. FEMS Microbiol.Lett. 2016; 363(18); PM: 27573235

ABSTRACT: Gastrointestinal (GI) leakage in Clostridium difficile-associated diarrhea (CDAD) is well known but is not routinely assessed in clinical practice. Serum (1-->3)-beta-D-glucan (BG), a fungal cell wall component used as a biomarker for invasive fungal disease, was tested in a CDAD mouse model with and without probiotics. Higher serum fluorescein isothiocyanate-dextran (FITC-dextran) and spontaneous gram-negative bacteremia, GI leakage indicators, were frequently found in CDAD mice, which died compared with those which survived. BG, serum macrophage inflammatory protein-2 and FITC-dextran but not quantitative blood bacterial count differentiated the clinical severity. Interestingly, a specific dose of Lactobacillus rhamnosus L34 attenuated CDAD and decreased serum BG and FITC-dextran, but not other parameters. BG also showed a higher area under the receiver operating characteristic curve for 7-day mortality than FITC-dextran. Fifty-five percent of CDAD mice with BG >/= 60 pg/ml (the human negative cut-off value for invasive fungal disease) at 1 day after C. difficile gavage died within 7 days. In conclusion, S: erum BG was elevated in mice with severe CDAD, an established model of GI leakage with a strong association with mortality rate. BG monitoring in patients with CDAD is of interest as both a potential prognostic tool and a therapeutic efficacy indicator.

Lei, M., Hua, L. M., and Wang, D. W. The effect of probiotic treatment on elderly patients with distal radius fracture: a prospective double-blind, placebo-controlled randomised clinical trial. Benef.Microbes. 11-30-2016; 7(5):631-637; PM: 27633174

ABSTRACT: Probiotic treatment has been shown to improve bone formation, increase bone mass density and prevent bone loss. We aimed to assess the effect of probiotic treatment on functional recovery in elderly patients with a distal radius fracture. A total of 417 elderly patients with an acute distal radius fracture were enrolled in this double-blind placebo-controlled clinical trial. They were randomised to receive skimmed milk containing either a commercial probiotic (Lactobacillus casei Shirota) or placebo daily for a period of 6 months after the fracture. Treatment outcomes were the DASH (disabilities of the arm, shoulder and hand) score, pain, complex regional pain syndrome

(CRPS) score, active range of motion and grip strength, all of which were measured on a monthly basis. Throughout the duration of the study, DASH score, pain, CRPS score, wrist flexion and grip strength of patients receiving probiotics exhibited a significantly faster pace of improvement than those on placebo, with treatment outcomes of patients receiving Lactobacillus casei Shirota at month 4 at comparable levels with those of patients receiving placebo at month 6. In elderly patients with a fracture of the distal radius, administration of the probiotic could greatly accelerating the healing process.

Lei, S., Ramesh, A., Twitchell, E., Wen, K., Bui, T., Weiss, M., Yang, X., Kocher, J., Li, G., Giri-Rachman, E., Trang, N. V., Jiang, X., Ryan, E. P., and Yuan, L. High Protective Efficacy of Probiotics and Rice Bran against Human Norovirus Infection and Diarrhea in Gnotobiotic Pigs. Front Microbiol. 2016; 7:1699; PM: 27853451

ABSTRACT: Probiotics have been recognized as vaccine adjuvants and therapeutic agents to treat acute gastroenteritis in children. We previously showed that rice bran (RB) reduced human rotavirus diarrhea in gnotobiotic pigs. Human noroviruses (HuNoVs) are the major pathogens causing non-bacterial acute gastroenteritis worldwide. In this study, Lactobacillus rhamnosus GG (LGG) and Escherichia coli Nissle 1917 (EcN) were first screened for their ability to bind HuNoV P particles and virions derived from clinical samples containing HuNoV genotype GII.3 and GII.4, then the effects of LGG+EcN and RB on HuNoV infection and diarrhea were investigated using the gnotobiotic pig model. While LGG+EcN colonization inhibited HuNoV shedding, probiotic cocktail regimens in which RB feeding started 7 days prior to or 1 day after viral inoculation in the LGG+EcN colonized gnotobiotic pigs exhibited high protection against HuNoV diarrhea and shedding, characterized by significantly reduced incidence (89 versus 20%) and shorter mean duration of diarrhea (2.2 versus 0.2 days), as well as shorter mean duration of virus shedding (3.2 versus 1.0 days). In both probiotic cocktail groups, the diarrhea reduction rates were 78% compared with the control group, and diarrhea severity was reduced as demonstrated by the significantly lower cumulative fecal scores. The high protective efficacy of the probiotic cocktail regimens was attributed to stimulation of IFN-gamma+ T cell responses, increased production of intestinal IgA and IgG, and maintenance of healthy intestinal morphology (manifested as longer villi compared with the control group). Therefore, probiotic cocktail regimens containing LGG+EcN and RB may represent highly efficacious strategies to prevent and treat HuNoV gastroenteritis, and potentially other human enteric pathogens.

Lenoir-Wijnkoop, I., Gerlier, L., Roy, D., and Reid, G. The Clinical and Economic Impact of Probiotics Consumption on Respiratory Tract Infections: Projections for Canada. PLoS.One. 2016; 11(11):e0166232; PM: 27832195

ABSTRACT: ***INTRODUCTION***: There is accumulating evidence supporting the use of probiotics, which are defined as "live micro-organisms which, when administered in adequate amounts, confer a health benefit on the host", as a preventive measure against respiratory tract infections (RTI). Two recent meta-analyses showed probiotic consumption (daily intake of 107 to 1010 CFU in any form for up to 3 months) significantly reduced RTI duration, frequency, antibiotic use and work absenteeism. ***OBJECTIVES***: The aim of this study was to assess the impact of probiotic use in terms of number of RTI episodes and days averted, and the number of antibiotic prescriptions and missed workdays averted, in the general population of Canada. In addition, the corresponding economic impact from both a healthcare payer and a productivity perspective was

estimated. ***METHODS***: A microsimulation model was developed to reproduce the Canadian population (sample rate of 1/1000 = 35 540 individuals) employing age and gender. RTI incidence was taken from FluWatch consultation rates for influenza-like illness (2013-14) and StatCan all-cause consultations statistics. The model was calibrated on a 2.1% RTI annual incidence in the general population (5.2 million RTI days) and included known risk factors (smoking status, shared living conditions and vaccination status). RTI-related antibiotic prescriptions and work absenteeism were obtained from the literature. ***RESULTS***: The results indicate that probiotic use saved 573 000-2.3 million RTI-days, according to the YHEC-Cochrane scenarios respectively. These reductions were associated with an avoidance of 52 000-84 000 antibiotic courses and 330 000-500 000 sick-leave days. A projection of corresponding costs reductions amounted to Can$1.3-8.9 million from the healthcare payer perspective and Can$61.2-99.7 million when adding productivity losses. ***CONCLUSION***: The analysis shows that the potential of probiotics to reduce RTI-related events may have a substantial clinical and economic impact in Canada.

Li, D., Wang, P., Wang, P., Hu, X., and Chen, F. The gut microbiota: A treasure for human health. Biotechnol.Adv. 11-15-2016; 34(7):1210-1224; PM: 27592384

ABSTRACT: The interplay between the host and host-associated gut microbiota is an area of increasing interest during the recent decade. From young infants to elderly people, from primitive tribes to modern societies, accumulating evidence has suggested the association of critical physiological roles of gut microbiota in the pathogenesis of a variety of human metabolic, immunological and neurological diseases. Importantly, it appears that the relationship between the gut microbiota and disease is bidirectional, instead of causal or consequential. Personalized nutritional and therapeutic strategies targeting the gut microbiota such as prebiotics, probiotics, drugs and fecal microbiota transplantation may create a new era in the human health.

Li, X., Xu, Q., Jiang, T., Fang, S., Wang, G., Zhao, J., Zhang, H., and Chen, W. A comparative study of the antidiabetic effects exerted by live and dead multi-strain probiotics in the type 2 diabetes model of mice. Food Funct. 12-7-2016; 7(12):4851-4860; PM: 27812581

ABSTRACT: Type 2 diabetes is a metabolic syndrome characterized by insulin resistance and relative insulin deficiency. In this study, the anti-diabetic effects of live and dead multi-strain probiotics were explored and compared in a high-fat and streptozotocin-induced model of type 2 diabetes in mice. Either live or dead probiotics were daily administered orally to the mice over 10 weeks. Both live and dead multi-strain probiotics reduced HbA1C and leptin levels, improved glucose tolerance, and protected against the impairment of the pancreas, while the live probiotic showed a greater ability to reduce fasting and postprandial blood glucose levels. In addition, the live multi-strain probiotic exerted the beneficial effect of ameliorating insulin resistance. This effect was associated with the gut microbiota, butyrate production, and the inflammatory response, and was more effective for the live probiotic than the dead probiotic. These findings showed that the viable probiotic more effectively relieved hypoglycemic symptoms in the host by ameliorating insulin resistance. Furthermore, a pathway related to insulin resistance, i.e., the gut microbiota-butyrate-inflammatory axis, provided a promising rationale for further research into preventing or treating type 2 diabetes.

Lim, F. T., Lim, S. M., and Ramasamy, K. Pediococcus acidilactici LAB4 and Lactobacillus plantarum LAB12 assimilate cholesterol and modulate ABCA1, CD36, NPC1L1 and SCARB1 in vitro. Benef.Microbes. 12-1-2016; 1-14; PM: 27903090

ABSTRACT: There is growing interest in the use of probiotic lactic acid bacteria (LAB) for prevention of hypercholesterolaemia. This study assessed the cholesterol lowering ability of Pediococcus acidilactici LAB4 and Lactobacillus plantarum LAB12 in growth media. Both LAB yielded >98% (39.2 mug/ml) cholesterol lowering in growth media. Nile Red staining indicated direct assimilation of cholesterol by the LAB. The LAB were then explored for their prophylactic (pre-treatment of HT29 cells with LAB prior to cholesterol exposure) and biotherapeutic (treatment of HT29 cells with LAB after exposure to cholesterol) use against short and prolonged exposure of HT29 cells to cholesterol, respectively. For HT29 cells pre-treated with LAB, cholesterol lowering was accompanied by down-regulation of ATP-binding cassette family transporter-type A1 (ABCA1), cluster of differentiation 36 (CD36) and scavenger receptor class B member 1 (SCARB1). HT29 cells treated with LAB after prolonged exposure to cholesterol source, on the other hand, was associated with up-regulation of ABCA1, restoration of CD36 to basal level and down-regulation of Neimann-Pick C1-Like 1 (NPC1L1). The present findings implied the potential use of LAB4 and LAB12 as part of the strategies in prevention and management of hypercholesterolaemia.

Lin, H. L., Shiu, Y. L., Chiu, C. S., Huang, S. L., and Liu, C. H. Screening probiotic candidates for a mixture of probiotics to enhance the growth performance, immunity, and disease resistance of Asian seabass, Lates calcarifer (Bloch), against Aeromonas hydrophila. Fish.Shellfish.Immunol. 11-9-2016; PM: 27838565

ABSTRACT: Six bacteria, including, Lactobacillus casei M15, Lac. plantarum D8, Lac. pentosus BD6, Lac. fermentum LW2, Enterococcus faecium 10-10, and Bacillus subtilis E20, and one yeast, Saccharomyces cerevisiae P13 were selected as probiotics for Asian seabass, Lates calcarifer, by tracking the growth performance and disease resistance of fish against Aeromonas hydrophila in the first trial. The probiotic efficiency screening results showed that B. subtilis E20 and Lac. pentosus BD6, and S. cerevisiae P13 and Lac. fermentum LW2 respectively improved either the growth performance or disease resistance. Therefore, these four probiotics were then selected to prepare a probiotics mixture, and this was incorporated in equal proportions into diets for Asian seabass at levels of 0 (control), and 106 (MD6), 107 (MD7), 108 (MD8), and 109 (MD9) colony-forming units (cfu) (kg diet)-1. A synergistic effect of the combined probiotics was investigated in this study, and the probiotics mixture was able to improve both the growth performance and health status of fish. After 56 days of feeding, fish fed the MD9 diet had a higher final weight and percentage of weight gain. In addition, protein contents in the dorsal muscle of fish fed the MD8 and MD9 diets were significantly higher compared to the control. For the pathogen challenge test, fish fed the MD7, MD8, and MD9 diets had significantly lower cumulative mortalities after A. hydrophila infection compared to those of fish fed the control and MD6 diets, which might have been due to increased respiratory bursts, decreased superoxide dismutase activity in leucocytes, and increased phagocytic activity. Therefore, we considered that the probiotics mixture could adequately provide probiotic efficiency for Asian seabass, and the diet containing 109 cfu (kg diet)-1 probiotic mixture is recommended to improve the growth and health status of Asian seabass.

Lin, S. H., Chou, L. M., Chien, Y. W., Chang, J. S., and Lin, C. I. Prebiotic Effects of Xylooligosaccharides on the Improvement of Microbiota Balance in Human Subjects. Gastroenterol.Res.Pract. 2016; 2016:5789232; PM: 27651791

ABSTRACT: It has been indicated that probiotics can be nourished by consuming prebiotics in order to function more efficiently, allowing the bacteria to stay within a healthy balance. In this study, we investigated the effects of xylooligosaccharides- (XOS-) enriched rice porridge consumption on the ecosystem in the intestinal tract of human subjects. Twenty healthy subjects participated in this 6-week trial, in which 10 subjects received XOS-enriched rice porridge while the others received placebo rice porridge. Fecal samples were collected at the end of weeks 0, 1, 3, 4, 6, and 7 for microorganism examination. The results showed that 6-week daily ingestion of the XOS-enriched rice porridge induced significant increases in fecal bacterial counts of Lactobacillus spp. and Bifidobacterium spp., as well as decreases in Clostridium perfringens without changing the total anaerobic bacterial counts, compared to that of placebo rice porridge. However, fluctuations in the counts of coliforms were observed in both groups during the 6-week intervention. In conclusion, the intestinal microbiota balance was improved after daily consumption of 150 g of rice porridge containing XOS for 6 weeks, demonstrating the prebiotic potential of XOS incorporated into foods. This also indicates the effectiveness of XOS as a functional ingredient in relation to its role as a prebiotic compound.

Lin, Y., Wang, Z. N., Ma, C. C., Liu, C. K., Yang, J. R., Shen, Z. W., and Wu, R. H. [Proton nuclear magnetic resonance spectroscopy recognition of metabolic patterns in fecal extracts for early diagnosis of colorectal cancer]. Zhonghua Yu Fang Yi.Xue.Za Zhi. 9-6-2016; 50(9):788-793; PM: 27655598

ABSTRACT: ***Objective***: To characterize the metabolic " fingerprint" of fecal extracts for diagnosis of early-stage colorectal cancer(CRC)using proton nuclear magnetic resonance spectroscopy(1H-NMR)-based metabolomics coupled with pattern recognition. ***Methods***: From January 2014 to December 2014, we collected fecal samples at the Second Affiliated Hospital of Shantou University Medical College, from 25 patients with colorectal adenomas(CR-Ad), 20 with stage / CRC, and 32 healthy controls(HCs). The patients were diagnosed by histopathology. No subjects had any complicating diseases. HCs showed no abnormalities from blood tests, endoscopic examination, diagnostic imaging, and/or medical interviews. We excluded participants who used antibiotics, NSAIDS, statins, or probiotics within two months of study participation, and any patients who underwent chemotherapy or radiation treatments prior to surgery. We used orthogonal partial least-squares-discriminant analysis(OPLS-DA)for pattern recognition(dimension reduction)on 1H-NMR processed data(1H frequency of 400.13 MHz), to find metabolic differences among CR-Ad, carcinoma and HC fecal samples; and receiver operating characteristic(ROC)analysis to determine the diagnostic value of the fecal metabolic biomarkers. ***Results***: Fecal samples were collected from 20 patients with Stage / CRC(11 M, 9 F, median age(52+/-13)years), 25 with CR-Ad(14 M, 11 F, median age(53 +/- 11)years)and 32 HCs(15 M, 17 F, median age(53 +/- 14)years). OPLS-DA clearly distinguished CR-Ad and stage / CRC from HC samples, based on their metabolomic profiles. Relative signal intensities in HCs were significantly lower than in the cancer patients for butyrate(HC: 23.0+/-6.0; CR-Ad: 18.0+/-5.0; CRC: 14.0+/-6.0; Z=-2.07, P=0.008), acetate(HC: 45.0+/-11.0; CR-Ad: 31.0+/-11.0; CRC: 24.0+/-8.0; Z=- 2.32, P=0.011), propionate(HC: 26.0 +/- 7.0; CR-Ad: 22.0 +/- 6.0; CRC: 19.0 +/- 5.0; Z=- 2.43, P=0.032), glucose(HC: 37.0+/-7.0; CR-Ad: 31.0+/-7.0; CRC: 26.0+/-8.0; Z=-2.07, P=0.044)and

glutamine(HC: 4.5+/-2.0; CR-Ad: 4.9 +/- 1.0; CRC: 5.4 +/- 1.0; Z=2.21, P=0.044). However, relative signal intensities in HCs were significantly higher than in patients for lactate(HC: 4.8+/-1.0; CR-Ad: 6.9+/-2.0; CRC: 4.8+/- 1.0; Z=2.02, P= 0.038), glutamate(HC: 3.2 +/- 2.0; CR-Ad: 4.9 +/- 1.0; CRC: 3.2 +/- 2.0; Z=2.21, P=0.044)and succinate(HC: 12.0+/-2.0; CR-Ad: 15.0+/-3.0; CRC: 12.0+/- 2.0; Z=2.25, P=0.011). Among the potential biomarkers, acetate at 1.92 ppm, and succinate at 2.41 ppm displayed relatively high area under ROC, with sensitivity and specificity both >90%, to distinguish early-stage CRC patients from HCs. ***Conclusion***: Fecal metabolic profiles distinguish of HCs from patients with CRC patients, even in the early stages(stage /), highlighting the potential of NMR-based fecal metabolomic fingerprinting as tools for early CRC diagnosis.

Liu, X. H. [Research advances in breastfeeding]. Zhongguo Dang.Dai Er.Ke.Za Zhi. 2016; 18(10):921-925; PM: 27751204

ABSTRACT: Human breast milk is the most natural and ideal food for the baby. Breastfeeding provides benefits for maternal and child health, child immune function, growth and development, and society. The operation of human milk bank and the use of donor human milk undoubtedly provides a new way of nutrition support for the preterm infants without their own mother's milk and a new kind of treatment for other diseases. Present research on the composition of breast milk focuses on the variety and quantity of proteins, bioactive substances, probiotics and cell population.Future research may focus on the bioactive substances, the mechanism of regulation and effect of cell population, the application of probiotics and the clinical application of donor human milk.

Liu, Y., Gibson, G. R., and Walton, G. E. An In Vitro Approach to Study Effects of Prebiotics and Probiotics on the Faecal Microbiota and Selected Immune Parameters Relevant to the Elderly. PLoS.One. 2016; 11(9):e0162604; PM: 27612304

ABSTRACT: The aging process leads to alterations of gut microbiota and modifications to the immune response, such changes may be associated with increased disease risk. Prebiotics and probiotics can modulate microbiome changes induced by aging; however, their effects have not been directly compared. The aim of this study was to use anaerobic batch culture fermenters to assess the impact of various fermentable carbohydrates and microorganisms on the gut microbiota and selected immune markers. Elderly volunteers were used as donors for these experiments to enable relevance to an aging population. The impact of fermentation supernatants on immune markers relevant to the elderly were assessed in vitro. Levels of IL-1beta, IL-6, IL-8, IL-10 and TNF-alpha in peripheral blood mononuclear cell culture supernatants were measured using flow cytometry. Trans-galactooligosaccharides (B-GOS) and inulin both stimulated bifidobacteria compared to other treatments ($p<0.05$). Fermentation supernatants taken from faecal batch cultures supplemented with B-GOS, inulin, B. bifidum, L. acidophilus and Ba. coagulans inhibited LPS induced TNF-alpha ($p<0.05$). IL-10 production, induced by LPS, was enhanced by fermentation supernatants from faecal batch cultures supplemented with B-GOS, inulin, B. bifidum, L. acidophilus, Ba. coagulans and Bac. thetaiotaomicron ($p<0.05$). To conclude, prebiotics and probiotics could lead to potentially beneficial effects to host health by targeting specific bacterial groups, increasing saccharolytic fermentation and decreasing inflammation associated with aging. Compared to probiotics, prebiotics led to greater microbiota modulation at the genus level within the fermenters.

Lloyd-Lavery, A., Rogers, N. K., Hatfield, S. J., Grindlay, D., Barnett, R., and Thomas, K. S. What's new in atopic eczema? An analysis of systematic reviews published in 2014. Part 2. Treatment and prevention. Clin.Exp.Dermatol. 11-21-2016; PM: 27868221

ABSTRACT: This review forms part of a series of annual updates that summarize the evidence base for atopic eczema (AE). It provides a summary of key findings from 12 systematic reviews (SRs) that were published or indexed during 2014, and focuses on the treatment and prevention of AE. For an update of SRs on the epidemiology, mechanisms of disease and methodological issues, see Part 1 of this update. Although phototherapy and various systemic medications (including ciclosporin, azathioprine and methotrexate) are commonly used to treat AE, many of these have not been robustly assessed in head-to-head randomized controlled trials. Educational interventions may improve AE severity and quality of life for children and their families. Intake of probiotics prenatally and postnatally may help prevent AE, but there is little evidence to suggest a role in the treatment of AE. Although no benefit was found for allergen avoidance in preventing AE, the use of immunotherapy to treat AE-associated aeroallergen sensitivity requires further evaluation. There is insufficient evidence for Vitamin D supplementation for the treatment of AE This overview of reviews provides a succinct guide for clinicians and patients wishing to remain up to date with the most recent evidence for the treatment and prevention of AE.

Lopes, E. G., Moreira, D. A., Gullon, P., Gullon, B., Cardelle-Cobas, A., and Tavaria, F. K. Topical application of probiotics in skin: adhesion, antimicrobial and antibiofilm in vitro assays. J.Appl.Microbiol. 11-12-2016; PM: 27862685

ABSTRACT: ***AIMS***: When skin dysbiosis occurs as a result of skin disorders, probiotics can act as modulators, restoring microbial balance. Several properties of selected probiotics were evaluated so that their topical application could be considered. ***METHODS AND RESULTS***: Adhesion, antimicrobial, quorum sensing and antibiofilm assays were carried out with several probiotic strains and tested against selected skin pathogens. All tested strains displayed significant adhesion to keratin. All lactobacilli with the exception of L. delbrueckii, showed antimicrobial activity against skin pathogens, mainly due to organic acid production. Most of them also prevented biofilm formation, but only P. innocua was able to break down mature biofilms. ***CONCLUSIONS***: This study demonstrates that although all tested probiotics adhered to human keratin, they showed limited ability to prevent adhesion of some potential skin pathogens. Most of the tested probiotics successfully prevented biofilm formation, suggesting that they may be successfully used in the future as a complement to conventional therapies in the treatment of a range of skin disorders. ***SIGNIFICANCE AND IMPACT OF STUDY***: The topical use probiotics may be a natural, targeted treatment approach to several skin disorders and a complement to conventional therapies which present many undesirable side effects. This article is protected by copyright. All rights reserved.

Loren, V., Manye, J., Fuentes, M. C., Cabre, E., Ojanguren, I., and Espadaler, J. Comparative Effect of the I3.1 Probiotic Formula in Two Animal Models of Colitis. Probiotics.Antimicrob.Proteins. 11-10-2016; PM: 27832441

ABSTRACT: Use of probiotic therapy is an active area of investigation to treat intestinal disorders. The clinical benefits of the I3.1 probiotic formula (Lactobacillus plantarum

(CECT7484, CECT7485) and P. acidilactici (CECT7483)) were demonstrated in irritable bowel syndrome (IBS) patients in a randomized, double-blind, placebo-controlled clinical trial. The aim of this study was to evaluate the therapeutic effects of I3.1 in two experimental models of colitis, a dextran sulfate sodium (DSS)-induced colitis model and an interleukin (IL)-10-deficient mice model. Colitis was induced in 32 8-week-old Balb/c mice by administering 3% (w/v) DSS in drinking water for 5 days. Probiotics were administered orally (I3.1 or VSL#3, 1 x 109 CFU daily) for 10 days before the administration of DSS. Also, probiotics (I3.1 or VSL#3, 1 x 109 CFU daily) were administered orally to 36 6-week-old C57B6J IL-10(-/-) mice for 10 weeks. Body weight was recorded daily. Colon samples were harvested for histological examination and cytokine measurements. Body weight after DSS administration did not change in the I3.1 group, whereas the VSL#3 group had weight loss. Also, I3.1 normalized IL-6 to levels similar to that of healthy controls and significantly increased the reparative histologic score. In the IL-10-deficient model, both VSL#3 and I3.1 reduced the severity of colitis compared to untreated controls, and I3.1 significantly reduced the levels of IFN-gamma compared to the other two groups. In conclusion, I3.1 displays a protective effect on two murine models of experimental colitis. Results suggest that the mechanism of action could be different from VSL#3.

Lu, M., Yu, S., Deng, J., Yan, Q., Yang, C., Xia, G., and Zhou, X. Efficacy of Probiotic Supplementation Therapy for Helicobacter pylori Eradication: A Meta-Analysis of Randomized Controlled Trials. PLoS.One. 2016; 11(10):e0163743; PM: 27723762

ABSTRACT: ***BACKGROUND***: Traditional Helicobacter pylori (H. pylori) eradication therapies have shown efficacies below 80% in several studies, and their use has been accompanied by antibiotic-related side effects. Some recent studies have reported that supplementing standard therapies with probiotics can improve the efficacy and tolerability of Helicobacter pylori eradication therapy. ***OBJECTIVE***: To assess the effects of probiotic supplementation on the eradication rates and therapy-related adverse event rates of anti-Helicobacter pylori regimens. ***METHODS***: We searched PubMed, Medline, the Cochrane Central Registry of Controlled Trials and the Chinese Biomedical Database for eligible randomized controlled trials published through July, 2015. Review Manager 5.3 was used for all statistical analyses. ***RESULTS***: Thirteen randomized controlled trials involving a total of 2306 patients were included in our analysis. Intent-to-treat (ITT) analysis performed using a fixed-effects model (test for heterogeneity I2 = 45%) showed that the pooled relative risk (RR) of eradication was significantly higher in the probiotic supplementation group than in the control group [RR 1.15, 95% confidence interval (CI): 1.10-1.20, P<0.00001]. The incidence of total antibiotic-related side effects was lower in the probiotic supplementation group than in the control group, and the pooled RR (studies n = 9) was 0.71 (95% CI: 0.54-0.94, P = 0.02), as determined using a random-effects model (heterogeneity test I2 = 59%). Certain adverse events, such as nausea and vomiting (RR = 0.58, 95% CI 0.35-0.95, P = 0.03), diarrhea (RR = 0.51, 95% CI: 0.31-0.84, P = 0.008) and constipation (RR = 0.47, 95% CI: 0.28-0.80, P = 0.005), were reported at lower rates in the probiotic supplementation group than in the control group. Subgroup analysis showed that eradication rates were significantly improved in both adults (RR = 1.14, 95% CI: 1.09-1.19, P<0.00001) and children (RR = 1.24, 95% CI: 1.05-1.47, P = 0.01) in the probiotic supplementation group and that no regional differences between Europe (RR = 1.17, 95% CI: 1.09-1.24, P<0.00001) and Asia were present (RR = 1.14, 95% CI: 1.06-1.22, P = 0.0002). However, the total adverse event rate was not decreased in the adult group (RR = 0.80, 95% CI: 0.61-1.04, P = 0.1) or the Asian group (RR = 0.68, 95% CI: 0.39-1.18, P = 0.17). Subgroup analyses examining therapy regimens and treatment durations showed that probiotic supplementation increased eradication rates in the triple-therapy

(RR = 1.18, 95% CI: 1.12-1.25, P<0.00001), seven-day treatment (RR = 1.21, 95% CI: 1.12-1.31, P<0.00001) and fourteen-day treatment (RR = 1.13, 95% CI: 1.06-1.20, P = 0.0002) groups. The incidence of antibiotic-related side effects was significantly reduced in all groups, with the exception of the quadruple-therapy subgroup (RR = 1.13, 95% CI: 0.60-2.13, P = 0.07) and the fourteen-day therapy subgroup (RR = 0.96, 95% CI 0.61-1.51, P = 0.86). Supplementation with Lactobacillus alone (RR = 1.24, 95% CI: 1.12-1.38, P<0.0001) or multi-strain probiotics (RR = 1.12, 95% CI 1.07-1.18, P<0.00001) was effective at improving H. pylori eradication rates. However, supplementation with Lactobacillus alone did not significantly decrease the overall incidence of side effects (RR = 0.61, 95% CI: 0.11-3.51, P = 0.58). Our study also showed that probiotic supplementation before, during or after H. pylori eradication therapy improved eradication rates, regardless of supplementation duration. Furthermore, probiotic supplementation during H. pylori treatment reduced the incidence of side effects. ***CONCLUSION***: Probiotic supplementation during anti-Helicobacter pylori treatment may be effective for improving H. pylori eradication rates, minimizing the incidence of therapy-related adverse events and alleviating most disease-related clinical symptoms. However, our results should be interpreted with caution because of the presence of heterogeneity across the trials included in this analysis.

Lundelin, K., Poussa, T., Salminen, S., and Isolauri, E. Long-term safety and efficacy of perinatal probiotic intervention: evidence from a follow-up study of four randomized, double-blind, placebo-controlled trials. Pediatr.Allergy Immunol. 10-25-2016; PM: 27779809

ABSTRACT: ***BACKGROUND***: Societies worldwide are faced with a progressive increase in immune-mediated health problems such as allergic, autoimmune and inflammatory diseases, as well as obesity. Perinatal administration of specific probiotic bacteria is an attractive approach in reducing the risk of these conditions, but long-term efficacy and safety data are lacking. The aim here was to evaluate the clinical benefit and long-term safety of specific probiotics administered during the perinatal period. ***METHODS***: The probiotic strains used were Lactobacillus rhamnosus GG, Bifidobacterium lactis Bb-12, Lactobacillus paracasei ST11 and Bifidobacterium longum BL999. The children involved have subsequently undergone prospective long-term follow-up. In addition to physical examination, data were collected by structured questionnaires on non-communicable diseases and continued probiotic use, and growth data from welfare clinics and school nurses. ***RESULTS***: Altogether 303 mother-infant pairs were included in the analysis. Seventy-six out of 163 (47%) children receiving perinatal probiotics had developed allergic disease compared with 79 out of 140 (56%) receiving placebo (OR 0.67, 95% CI 0.43-1.06, p=0.09). Fifty-nine out of 133 (44%) children receiving Lactobacillus rhamnosus GG perinatally had developed allergic disease, OR 0.62, 95% CI 0.38-0.99, p=0.047, as compared to placebo. We found no differences in growth or non-communicable disease prevalence between children receiving perinatally probiotics or placebo. ***CONCLUSIONS***: Perinatal probiotic administration is safe in long-term follow-up. Children receiving Lactobacillus rhamnosus GG perinatally tended to have decreased allergy prevalence. This article is protected by copyright. All rights reserved.

Luo, J., Ranadheera, C. S., King, S., Evans, C. A., and Baines, S. K. Potential influence of dairy propionibacteria on the growth and acid metabolism of Streptococcus bovis and Megasphaera elsdenii. Benef.Microbes. 11-8-2016; 1-10; PM: 27824275

ABSTRACT: Ruminal acidosis is a prevalent disorder among dairy cows and feedlot cattle, which can significantly impair their health and productivity. This study, involving seven

different strains of dairy propionibacteria, represents an in vitro investigation of the feasibility of using these organisms as direct-fed microbials to control lactic acid acumulation in the rumen. Interactions between the propionibacteria, Streptococcus bovis and Megasphaera elsdenii were evaluated in terms of effects on lactic, acetic and propionic acid metabolism, following co-incubation. Spot resistance tests showed slight but varying degrees of growth inhibition by S. bovis among the propionibacteria, while no inhibition was observed between M. elsdenii and the different strains of dairy propionibacteria. In the co-culture experiments comprising S. bovis in nutrient broth, significant differences in pH and the levels of production of lactic, acetic and propionic acid, were observed between treatments following inoculation with various propionibacteria and/or M. elsdenii. In general, lactic acid concentrations at the end of the incubation were significantly lower in the cultures containing propionibacteria compared with cultures comprising either S. bovis only or S. bovis + M. elsdenii, although efficacy of lactate metabolism varied between species and strains. Moreover,the accumulation of acetic and propionic acid in the combined cultures, but not in the solo S. bovis culture, indicated that these compounds were produced as a result of the metabolism of lactic acid by the propionibacteria and M. elsdenii.

Mai, V., Waugh, S., Byrd, D., Simpson, D., and Ukhanova, M. Novel encapsulation improves recovery of probiotic strains in fecal samples of human volunteers. Appl.Microbiol.Biotechnol. 10-28-2016; PM: 27796434

ABSTRACT: Probiotic supplements can contribute to maintaining health and ameliorating various disease symptoms. Probiotics can be delivered in many forms with crucial differences in their survival during gastrointestinal (GI) passage. Previously, a novel encapsulation, Probiotic Pearls Acidophilus, Integrative Therapeutics, LLC, USA (Pearls), was shown to increase survival in vitro after exposure to gastric conditions. Here, we compare fecal recovery in human volunteers consuming Pearls or a conventional hard-shelled gelatin capsule. We performed a randomized double-blinded, two-armed trial, with six healthy subjects in each 12-day study arm. In fecal samples collected at baseline, twice during the intervention period, and after washout, we compared colony counts between the two encapsulation methods. The identity of the colonies was confirmed by colony morphology, strain-specific PCR, and 16S rRNA gene sequencing. We further performed a comprehensive 16S rRNA gene sequencing-based analysis to identify differential effects on overall microbiota composition. We detected an average log increase in bifidobacteria of 0.152 cfu/g with gelatin and 0.651 cfu/g with Pearls capsules ($p > 0.05$). Total lactobacilli counts increased in both groups with no difference between the groups. However, the supplemented Lactobacillus acidophilus NCFM decreased to baseline levels within 7 days after end of supplementation with gelatin capsules while 3.11 log cfu/g higher counts compared to baseline ($p = 0.05$) remained for Pearls. Targeted qPCR largely confirmed the trends observed by viable plate counts. Protecting the probiotic strains by Pearls encapsulation results in higher recovery rates of the supplemented lactobacilli and bifidobacteria in fecal samples and increased persistence, suggesting an improved survival and viability that might increase efficacy towards achieving desired health benefits.

Majidi-Mosleh, A., Sadeghi, A. A., Mousavi, S. N., Chamani, M., and Zarei, A. Ileal MUC2 gene expression and microbial population, but not growth performance and immune response, are influenced by in ovo injection of probiotics in broiler chickens. Br.Poult.Sci. 11-15-2016; PM: 27844495

ABSTRACT: 1. The objective of present study was to evaluate the effects of intra-amniotic injection of different probiotic strains (Bacillus subtilis, Enterococcus faecium and Pediococcus acidilactici) on the intestinal MUC2 gene expression, microbial population, growth performance and immune response in broiler chicken. 2. In a completely randomised design, different probiotic strains was injected into the amniotic fluid of the 480 live embryos (d 18 of incubation), with 4 treatments and 5 replicates. Ileal MUC2 gene expression, microbial profile, growth performance and immune response were determined. 3. Injection of probiotic strains, especially Bacillus subtilis, had significant effect on expression of the MUC2 on d 21 of incubation and d 3 post-hatch, but not on d 19 of incubation. 4. Injection of the probiotic strains decreased significantly the E. coli population and increased the lactic acid bacteria population during the first week post-hatch. 5. Inoculation of probiotics had no significant effect on antibody titres against Newcastle disease virus, antibody titres against sheep red blood cell and cell-mediated immune response of chickens compared to control. 6. In ovo injection of the probiotic strains had no significant effect on growth performance of broiler chickens. 7. It was concluded that injection of probiotic bacteria especially Bacillus subtilis into the amniotic fluid has a beneficial effect on ileal MUC2 gene expression and bacteria population during the first week post-hatch, but has no effect on growth performance and immune response in broiler chickens.

Maldonado-Gomez, M. X., Martinez, I., Bottacini, F., O'Callaghan, A., Ventura, M., van, Sinderen D., Hillmann, B., Vangay, P., Knights, D., Hutkins, R. W., and Walter, J. Stable Engraftment of Bifidobacterium longum AH1206 in the Human Gut Depends on Individualized Features of the Resident Microbiome. Cell Host.Microbe. 10-12-2016; 20(4):515-526; PM: 27693307

ABSTRACT: Live bacteria (such as probiotics) have long been used to modulate gut microbiota and human physiology, but their colonization is mostly transient. Conceptual understanding of the ecological principles as they apply to exogenously introduced microbes in gut ecosystems is lacking. We find that, when orally administered to humans, Bifidobacterium longum AH1206 stably persists in the gut of 30% of individuals for at least 6 months without causing gastrointestinal symptoms or impacting the composition of the resident gut microbiota. AH1206 engraftment was associated with low abundance of resident B. longum and underrepresentation of specific carbohydrate utilization genes in the pre-treatment microbiome. Thus, phylogenetic limiting and resource availability are two factors that control the niche opportunity for AH1206 colonization. These findings suggest that bacterial species and functional genes absent in the gut microbiome of individual humans can be reestablished, providing opportunities for precise and personalized microbiome reconstitution.

Maldonado, N. C., Silva de, Ruiz C., and Nader-Macias, M. E. Design of a beneficial product for newborn calves by combining Lactobacilli, minerals, and vitamins. Prep.Biochem.Biotechnol. 10-2-2016; 46(7):648-656; PM: 26675304

ABSTRACT: Diarrhea is one of the most frequent diseases affecting newborn calves in intensive systems. Several strategies were proposed to protect and improve health, such as probiotics. This work was directed to design a product containing freeze-dried bacteria, vitamins, and minerals, as well as to optimize conditions with lyoprotectors, combine strains and add vitamins, minerals, and inulin to the product. The lyoprotectors were milk, milk-whey, and actose, and products were stored for 6 months at 4 degrees C.

Combined bacteria were freeze-dried in milk and the final products were added with minerals, vitamins, and insulin. The viable cells were determined by the plate count assay and antibiotic profiles to differentiate strains. Lactobacillus johnsonii CRL1693, L. murinus CRL1695, L. mucosae CRL1696, L. salivarius CRL1702, L. amylovorus CRL1697, and Enterococcus faecium CRL1703 were evaluated. The optimal conditions were different for each strain. Milk and milk whey maintained the viability during the process and storage after 6 months for most of the strains, except for L. johnsonii. Lactose did not improve cell's recovery. L. murinus was viable for 6 months in all the conditions, with similar results in enterococci. In strains combined before freeze-dried, the viability decreased deeply, showing that one-step process with bacteria mixtures, vitamins, and minerals were not adequate. Freeze-dried resistance depends on each strain and must be lyophilized individually.

Manning, J., Dunne, E. M., Wescombe, P. A., Hale, J. D., Mulholland, E. K., Tagg, J. R., Robins-Browne, R. M., and Satzke, C. Investigation of Streptococcus salivarius-mediated inhibition of pneumococcal adherence to pharyngeal epithelial cells. BMC.Microbiol. 9-29-2016; 16(1):225; PM: 27681377

ABSTRACT: ***BACKGROUND***: Pneumococcal adherence to the nasopharyngeal epithelium is a critical step in colonisation and disease. The probiotic bacterium, Streptococcus salivarius, can inhibit pneumococcal adherence to epithelial cells in vitro. We investigated the mechanism(s) of inhibition using a human pharyngeal epithelial cell line (Detroit 562) following pre-administration of two different strains of S. salivarius. **RESULTS**: Whilst the bacteriocin-encoding megaplasmids of S. salivarius strains K12 and M18 were essential to prevent pneumococcal growth on solid media, they were not required to inhibit pneumococcal adherence. Experiments testing S. salivarius K12 and two pneumococcal isolates (serotypes 19F and 6A) showed that inhibition of 19F may involve S. salivarius-mediated blocking of pneumococcal binding sites: a negative correlation was observed between adherence of K12 and 19F, and no inhibition occurred when K12 was prevented from contacting epithelial cells. K12-mediated inhibition of adherence by 6A may involve additional mechanisms, since no correlation was observed between adherence of K12 and 6A, and K12 could inhibit 6A adherence in the absence of cell contact. ***CONCLUSIONS***: These results suggest that S. salivarius employs several mechanisms, including blocking pneumococcal binding sites, to reduce pneumococcal adherence to pharyngeal epithelial cells. These findings extend our understanding of how probiotics may inhibit pneumococcal adherence and could assist with the development of novel strategies to prevent pneumococcal colonisation in the future.

Manzanares, W., Lemieux, M., Langlois, P. L., and Wischmeyer, P. E. Probiotic and synbiotic therapy in critical illness: a systematic review and meta-analysis. Crit Care. 8-19-2016; 19:262; PM: 27538711

ABSTRACT: ***BACKGROUND***: Critical illness is characterized by a loss of commensal flora and an overgrowth of potentially pathogenic bacteria, leading to a high susceptibility to nosocomial infections. Probiotics are living non-pathogenic microorganisms, which may protect the gut barrier, attenuate pathogen overgrowth, decrease bacterial translocation and prevent infection. The purpose of this updated systematic review is to evaluate the overall efficacy of probiotics and synbiotic mixtures on clinical outcomes in critical illness. ***METHODS***: Computerized databases from 1980 to 2016 were searched.

Randomized controlled trials (RCT) evaluating clinical outcomes associated with probiotic therapy as a single strategy or in combination with prebiotic fiber (synbiotics). Overall number of new infections was the primary outcome; secondary outcomes included mortality, ICU and hospital length of stay (LOS), and diarrhea. Subgroup analyses were performed to elucidate the role of other key factors such as probiotic type and patient mortality risk on the effect of probiotics on outcomes. ***RESULTS***: Thirty trials that enrolled 2972 patients were identified for analysis. Probiotics were associated with a significant reduction in infections (risk ratio 0.80, 95 % confidence interval (CI) 0.68, 0.95, P = 0.009; heterogeneity I (2) = 36 %, P = 0.09). Further, a significant reduction in the incidence of ventilator-associated pneumonia (VAP) was found (risk ratio 0.74, 95 % CI 0.61, 0. 90, P = 0.002; I (2) = 19 %). No effect on mortality, LOS or diarrhea was observed. Subgroup analysis indicated that the greatest improvement in the outcome of infections was in critically ill patients receiving probiotics alone versus synbiotic mixtures, although limited synbiotic trial data currently exists. ***CONCLUSION***: Probiotics show promise in reducing infections, including VAP in critical illness. Currently, clinical heterogeneity and potential publication bias reduce strong clinical recommendations and indicate further high quality clinical trials are needed to conclusively prove these benefits.

Maqsood, R. and Stone, T. W. The Gut-Brain Axis, BDNF, NMDA and CNS Disorders. Neurochem.Res. 2016; 41(11):2819-2835; PM: 27553784

ABSTRACT: Gastro-intestinal (GI) microbiota and the 'gut-brain axis' are proving to be increasingly relevant to early brain development and the emergence of psychiatric disorders. This review focuses on the influence of the GI tract on Brain-Derived Neurotrophic Factor (BDNF) and its relationship with receptors for N-methyl-D-aspartate (NMDAR), as these are believed to be involved in synaptic plasticity and cognitive function. NMDAR may be associated with the development of schizophrenia and a range of other psychopathologies including neurodegenerative disorders, depression and dementias. An analysis of the routes and mechanisms by which the GI microbiota contribute to the pathophysiology of BDNF-induced NMDAR dysfunction could yield new insights relevant to developing novel therapeutics for schizophrenia and related disorders. In the absence of GI microbes, central BDNF levels are reduced and this inhibits the maintenance of NMDAR production. A reduction of NMDAR input onto GABA inhibitory interneurons causes disinhibition of glutamatergic output which disrupts the central signal-to-noise ratio and leads to aberrant synaptic behaviour and cognitive deficits. Gut microbiota can modulate BDNF function in the CNS, via changes in neurotransmitter function by affecting modulatory mechanisms such as the kynurenine pathway, or by changes in the availability and actions of short chain fatty acids (SCFAs) in the brain. Interrupting these cycles by inducing changes in the gut microbiota using probiotics, prebiotics or antimicrobial drugs has been found promising as a preventative or therapeutic measure to counteract behavioural deficits and these may be useful to supplement the actions of drugs in the treatment of CNS disorders.

Maraolo, A. E. Reassessing the death risk related to probiotics in critically ill patients. Crit Care. 11-29-2016; 20(1):388; PM: 27894315

No Abstract Available.

Marcondes, M. I., Pereira, T. R., Chagas, J. C., Filgueiras, E. A., Castro, M. M., Costa, G. P., Sguizzato, A. L., and Sainz, R. D. Performance and health of Holstein calves fed different

levels of milk fortified with symbiotic complex containing pre- and probiotics. Trop.Anim Health Prod. 2016; 48(8):1555-1560; PM: 27557699

ABSTRACT: The objective of this study was to evaluate the performance and health of Holstein calves fed low or high milk supply (MSP) with or without symbiotic complex (SYM) supplementation, consisting of prebiotics, probiotics, and fibrolytic enzymes. Thirty-two Holstein calves with body weight (BW) of 34 +/- 7 kg were distributed in a randomized block design in a 2 x 2 factorial arrangement. Treatments consisted of low and high MSP: 10 % of BW from 1st to 8th weeks after birth (low) and 20 % BW from 1st and 2nd weeks after birth, 15 % BW for the 3rd and 4th weeks after birth, and 10 % BW from 5th and 8th weeks after birth (high). Solid ration was supplied in addition to milk. Intake, ADG, diet digestibility, and fecal consistency index were evaluated. Low and high MSP groups tended ($P < 0.10$) to differ in calf growth, final BW (69 vs. 73 kg), post-weaning average weight gain (548 vs. 788 g/day), and final average weight gain (549 vs. 646 g/day) in low and high MSP calves, respectively. There was an interaction between MSP level and SYM on the digestibilities of dry matter (DM) and neutral detergent fiber (NDF) ($P < 0.10$). In the low MSP group, inclusion of SYM increased digestibility of DM (0.720 to 0.736 g/kg) and NDF (0.758 to 0.783 g/kg). The inclusion of SYM improved calf health ($P < 0.10$) with a fecal score of 0.31 compared to 0.42 without SYM. Milk-feeding level was an important factor in calf performance, while SYM supplementation improved diet digestibility and animal health.

Markus, J., Mathiyalagan, R., Kim, Y. J., Abbai, R., Singh, P., Ahn, S., Perez, Z. E., Hurh, J., and Yang, D. C. Intracellular synthesis of gold nanoparticles with antioxidant activity by probiotic Lactobacillus kimchicus DCY51T isolated from Korean kimchi. Enzyme Microb.Technol. 2016; 95:85-93; PM: 27866630

ABSTRACT: A straightforward synthesis of gold nanoparticles (AuNps) is achieved by novel probiotic Lactobacillus kimchicus DCY51T isolated from Korean kimchi via an intracellular membrane-bound mechanism. The bioreduction of HAuCl4 into AuNps was verified by ultraviolet-visible spectrophotometry at approximately 540nm. AuNps were spherical with varying sizes of 5-30nm. AuNps maintained an average crystallite size of 13nm and demonstrated long-term stability in physiological buffer and biological media. Furthermore, the protective capping layer consisted of amino acid residues and surface-bound proteins rendered them non-toxic to murine macrophage (RAW264.7) and human colorectal adenocarcinoma (HT29) cell lines. Finally, biosynthesized AuNps served as superior free radical scavengers against 2,2-diphenyl-1-picrylhydrazyl (DPPH) in contrast to their corresponding gold salt. In short, this green synthesis is cost-effective and advantageous for the development of a new class of probiotics mediated and non-toxic carriers in targeted drug delivery systems, cancer diagnostic, photothermal therapy, biosensing, and medical imaging.

Martin, V. J., Leonard, M. M., Fiechtner, L., and Fasano, A. Transitioning From Descriptive to Mechanistic Understanding of the Microbiome: The Need for a Prospective Longitudinal Approach to Predicting Disease. J.Pediatr. 2016; 179:240-248; PM: 27634626

No Abstract Available.

Martinez, K. B., Pierre, J. F., and Chang, E. B. The Gut Microbiota: The Gateway to Improved Metabolism. Gastroenterol.Clin.North Am. 2016; 45(4):601-614; PM: 27837775

ABSTRACT: Obesity is an emerging global epidemic with profound challenges to world health care economies and societies. Traditional approaches to fighting obesity have not shown promise in promoting a decline in obesity prevalence. The gut microbiota is becoming widely appreciated for its role in regulating metabolism and thus represents a target for new therapies to combat obesity and associated comorbidities. This article provides an overview of altered microbial community structure in obesity, dietary impact on the gut microbiota, host-microbe interactions contributing to the disease, and improvements in microbial assemblage after bariatric surgery and with therapies targeting the gut microbiome.

Marzorati, M. and Van de, Wiele T. An Advanced In Vitro Technology Platform to Study the Mechanism of Action of Prebiotics and Probiotics in the Gastrointestinal Tract. J.Clin.Gastroenterol. 2016; 50 Suppl 2, Proceedings from the 8th Probiotics, Prebiotics & New Foods for Microbiota and Human Health meeting held in Rome, Italy on September 13-15, 2015:S124-S125; PM: 27741154

ABSTRACT: The gastrointestinal tract (GIT) hosts the most complex microbial community in the human body. Given the extensive metabolic potential which is present in this community, this additional organ is of key importance to maintain a healthy status and several diseases are frequently correlated with an alteration of the composition/functionality of the gut microbiota. Consequently, there is a great interest in identifying potential approaches that could modulate the microbiota and its metabolism to bring about a positive health effect. A classical approach to reach this goal is the use of prebiotics and/or probiotics. How to study the potential effect of new prebiotics/probiotics and how to localize this effect along the full GIT? Human intervention trials are the golden standard to validate functional properties of food products. Yet, most studies on gut microbiota are based on the analysis of fecal samples because they are easily collected in a non-invasive manner. A complementary option is represented by well-designed in vitro simulation technologies. Among all the available systems, the Simulator of Human Intestinal Microbial Ecosystem has already been shown to be a useful model for nutrition studies in terms of analysis of the intestinal microbial community composition and activity. The Simulator of Human Intestinal Microbial Ecosystem is a scientifically validated platform representing the physiology and microbiology of the adult human GIT. Furthermore, recent advances in in vitro modelling also allow to combine the study of bacteria-host interactions, such as mucosal adhesion and interaction with the immune system, thereby further increasing the value of the scientific output.

Matsusaki, T., Takeda, S., Takeshita, M., Arima, Y., Tsend-Ayush, C., Oyunsuren, T., Sugita, C., Yoshida, H., Watanabe, W., and Kurokawa, M. Augmentation of T helper type 1 immune response through intestinal immunity in murine cutaneous herpes simplex virus type 1 infection by probiotic Lactobacillus plantarum strain 06CC2. Int.Immunopharmacol. 2016; 39:320-327; PM: 27517518

ABSTRACT: We previously found that Lactobacillus plantarum strain 06CC2 showed probiotic potential, and its oral administration effectively induced Th1 cytokine production and activated the Th1 immune response associated with intestinal immunity in mice. In this study, to evaluate its potential as a versatile oral adjuvant for treatment of viral infection, we assessed the immunomodulatory activity of 06CC2 on murine

cutaneous herpes simplex virus type 1 (HSV-1) infection, in which a major immune defense system is a delayed-type hypersensitivity (DTH) reaction based on activation of the Th1 immune response, in relation to its oral efficacy for alleviation of herpetic symptoms. In the HSV-1 infection model, oral administration of 06CC2 (20mg/mouse) twice daily for seven days starting two days before infection was significantly effective in delaying the development of skin lesions in the early phase of infection and reducing virus yields in the brain on day 4 after infection. In addition, 06CC2 significantly augmented the DTH reaction to inactivated HSV-1 antigen and elevated interferon (IFN)-gamma production by HSV-1 antigen from splenocytes. On day 2, natural killer (NK) cell activity was significantly elevated, and the elevation was still observed on day 4. Furthermore, gene expressions of interleukin-12 receptor beta2 and IFN-gamma in Peyer's patches were augmented on day 4 by 06CC2 administration. Thus, 06CC2 was suggested to alleviate herpetic symptoms in mice in correlation with augmentation of the Th1 immune responses associated with NK cell activity through intestinal immunity. Strain 06CC2 may be a versatile oral adjuvant to activate Th1 immune response.

McDonald, D., Ackermann, G., Khailova, L., Baird, C., Heyland, D., Kozar, R., Lemieux, M., Derenski, K., King, J., Vis-Kampen, C., Knight, R., and Wischmeyer, P. E. Extreme Dysbiosis of the Microbiome in Critical Illness. mSphere. 2016; 1(4); PM: 27602409

ABSTRACT: Critical illness is hypothesized to associate with loss of "health-promoting" commensal microbes and overgrowth of pathogenic bacteria (dysbiosis). This dysbiosis is believed to increase susceptibility to nosocomial infections, sepsis, and organ failure. A trial with prospective monitoring of the intensive care unit (ICU) patient microbiome using culture-independent techniques to confirm and characterize this dysbiosis is thus urgently needed. Characterizing ICU patient microbiome changes may provide first steps toward the development of diagnostic and therapeutic interventions using microbiome signatures. To characterize the ICU patient microbiome, we collected fecal, oral, and skin samples from 115 mixed ICU patients across four centers in the United States and Canada. Samples were collected at two time points: within 48 h of ICU admission, and at ICU discharge or on ICU day 10. Sample collection and processing were performed according to Earth Microbiome Project protocols. We applied SourceTracker to assess the source composition of ICU patient samples by using Qiita, including samples from the American Gut Project (AGP), mammalian corpse decomposition samples, childhood (Global Gut study), and house surfaces. Our results demonstrate that critical illness leads to significant and rapid dysbiosis. Many taxons significantly depleted from ICU patients versus AGP healthy controls are key "health-promoting" organisms, and overgrowth of known pathogens was frequent. Source compositions of ICU patient samples are largely uncharacteristic of the expected community type. Between time points and within a patient, the source composition changed dramatically. Our initial results show great promise for microbiome signatures as diagnostic markers and guides to therapeutic interventions in the ICU to repopulate the normal, "health-promoting" microbiome and thereby improve patient outcomes. IMPORTANCE Critical illness may be associated with the loss of normal, "health promoting" bacteria, allowing overgrowth of disease-promoting pathogenic bacteria (dysbiosis), which, in turn, makes patients susceptible to hospital-acquired infections, sepsis, and organ failure. This has significant world health implications, because sepsis is becoming a leading cause of death worldwide, and hospital-acquired infections contribute to significant illness and increased costs. Thus, a trial that monitors the ICU patient microbiome to confirm and characterize this hypothesis is urgently needed. Our study analyzed the microbiomes of 115 critically ill subjects and demonstrated rapid dysbiosis from unexpected environmental sources after ICU

admission. These data may provide the first steps toward defining targeted therapies that correct potentially "illness-promoting" dysbiosis with probiotics or with targeted, multimicrobe synthetic "stool pills" that restore a healthy microbiome in the ICU setting to improve patient outcomes.

McFarland, L. V., Huang, Y., Wang, L., and Malfertheiner, P. Systematic review and meta-analysis: Multi-strain probiotics as adjunct therapy for Helicobacter pylori eradication and prevention of adverse events. United.European.Gastroenterol.J. 2016; 4(4):546-561; PM: 27536365

ABSTRACT: ***BACKGROUND***: Eradication rates with triple therapy for Helicobacter pylori infections have currently declined to unacceptable levels worldwide. Newer quadruple therapies are burdened with a high rate of adverse events. Whether multi-strain probiotics can improve eradication rates or diminish adverse events remains uncertain. ***METHODS***: Relevant publications in which patients with H. pylori infections were randomized to a multi-strain probiotic or control were identified in PubMed, Cochrane Databases, and other sources from 1 January 1960-3 June 2015. Primary outcomes included eradication rates, incidence of any adverse event and the incidence of antibiotic-associated diarrhea. As probiotic efficacy is strain-specific, pooled relative risks and 95% confidence intervals were calculated using meta-analysis stratified by similar multi-strain probiotic mixtures. ***RESULTS***: A total of 19 randomized controlled trials (20 treatment arms, n = 2730) assessing one of six mixtures of strains of probiotics were included. Four multi-strain probiotics significantly improved H. pylori eradication rates, five significantly prevented any adverse reactions and three significantly reduced antibiotic-associated diarrhea. Only two probiotic mixtures (Lactobacillus acidophilus/Bifidobacterium animalis and an eight-strain mixture) had significant efficacy for all three outcomes. ***CONCLUSIONS***: Our meta-analysis found adjunctive use of some multi-strain probiotics may improve H. pylori eradication rates and prevent the development of adverse events and antibiotic-associated diarrhea, but not all mixtures were effective.

McKean, J., Naug, H., Nikbakht, E., Amiet, B., and Colson, N. Probiotics and Subclinical Psychological Symptoms in Healthy Participants: A Systematic Review and Meta-Analysis. J.Altern.Complement Med. 11-14-2016; PM: 27841940

ABSTRACT: ***INTRODUCTION/AIM***: Interest in the gut-brain axis and emerging evidence that the intestinal microbiota can influence central nervous system function has led to the hypothesis that probiotic supplementation can have a positive effect on mood and psychological symptoms such as depression and anxiety. Although several human clinical trials have investigated this, results have been inconsistent. Therefore, a systematic review and meta-analytic approach was chosen to examine if probiotic consumption has an effect on psychological symptoms. ***METHODS***: The online databases PubMed, Scopus, and the Cochrane Library were searched for relevant studies up to July 2016. Those that were randomized and placebo controlled and measured preclinical psychological symptoms of depression, anxiety, and perceived stress in healthy volunteers pre and post supplementation with a probiotic were included. To control for differences in scales of measurement, data were converted to percentage change, and the standardized mean difference between the probiotic and control groups was investigated using Revman software. A random effects model was used for analysis. Heterogeneity was assessed using the I2 statistic. Quality assessment was undertaken using the Rosendal scale. ***RESULTS***: Seven studies met the inclusion criteria and provided

data for nine comparisons. All studies passed the quality analysis. The meta-analysis showed that supplementation with probiotics resulted in a statistically significant improvement in psychological symptoms (standardized mean difference 0.34; 95% confidence interval 0.07-0.61, Z = 2.49) compared with placebo. ***CONCLUSION***: These results show that probiotic consumption may have a positive effect on psychological symptoms of depression, anxiety, and perceived stress in healthy human volunteers.

McKinlay, C. J., Rebello, C., and Tarnow-Mordi, W. Probiotics in very preterm infants: the PiPS trial. Lancet. 8-13-2016; 388(10045):655; PM: 27533429

No Abstract Available.

Mehta, M., Goldfarb, D. S., and Nazzal, L. The role of the microbiome in kidney stone formation. Int.J.Surg. 11-12-2016; PM: 27847292

ABSTRACT: Nephrolithiasis is a complex disease of worldwide prevalence that is influenced by both genetic and environmental factors. About 75% of kidney stones are predominantly composed of calcium oxalate and urinary oxalate is considered a crucial risk factor. Microorganisms may have a role in the pathogenesis and prevention of kidney stones and the involvement of the intestinal microbiome in this renal disease has been a recent area of interest. Oxalobacter formigenes is a gram negative bacteria that degrades oxalate in the gut decreasing urinary oxalate excretion. In this review, we examine the data studying the role of Oxalobacter formigenes in kidney stone disease in humans and animals, the effect of antibiotics on its colonization, and the potential role of probiotics and whole microbial communities as therapeutic interventions.

Melendez-Illanes, L., Gonzalez-Diaz, C., Chilet-Rosell, E., and varez-Dardet, C. Does the scientific evidence support the advertising claims made for products containing Lactobacillus casei and Bifidobacterium lactis? A systematic review. J.Public Health (Oxf). 2016; 38(3):e375-e383; PM: 26515088

ABSTRACT: ***BACKGROUND***: To analyse the scientific evidence that exists for the advertising claims made for two products containing Lactobacillus casei and Bifidobacterium lactis and to conduct a comparison between the published literature and what is presented in the corporate website. ***METHODS***: Systematic review, using Medline through Pubmed and Embase. We included human clinical trials that exclusively measured the effect of Lactobacillus casei or Bifidobacterium lactis on a healthy population, and where the objective was related to the health claims made for certain products in advertising. We assessed the levels of evidence and the strength of the recommendation according to the classification criteria established by the Oxford Centre for Evidence Based Medicine (CEBM). We also assessed the outcomes of the studies published on the website that did not appear in the search. ***RESULTS***: Of the 440 articles identified, 16 met the inclusion criteria. Only four (25%) of these presented a level of evidence of 1b and a recommendation grade of A, all corresponding to studies on product containing Bifidobacterium lactis, and only 12 of the 16 studies were published on the corporate website (47). ***CONCLUSIONS***: There is insufficient scientific evidence to support the health claims made for these products, especially in the case of product containing Lactobacillus casei.

Meng, D., Zhu, W., Ganguli, K., Shi, H. N., and Walker, W. A. Anti-inflammatory effects of Bifidobacterium longum subsp infantis secretions on fetal human enterocytes are mediated by TLR-4 receptors. Am.J.Physiol Gastrointest.Liver Physiol. 10-1-2016; 311(4):G744-G753; PM: 27562058

ABSTRACT: The therapeutic and preventive application of probiotics for necrotizing enterocolitis (NEC) has been supported by more and more experimental and clinical evidence in which Toll-like receptor 4 (TLR-4) exerts a significant role. In immune cells, probiotics not only regulate the expression of TLR-4 but also use the TLR-4 to modulate the immune response. Probiotics may also use the TLR-4 in immature enterocytes for anti-inflammation. Here we demonstrate that probiotic conditioned media (PCM) from Bifidobacterium longum supp infantis but not isolated organisms attenuates interleukin-6 (IL-6) induction in response to IL-1beta by using TLR-4 in a human fetal small intestinal epithelial cell line (H4 cells), human fetal small intestinal xenografts, mouse fetal small intestinal organ culture tissues, and primary NEC enterocytes. Furthermore, we show that PCM, using TLR-4, downregulates the mRNA expression of interleukin-1 receptor-associated kinase 2 (IRAK-2), a common adapter protein shared by IL-1beta and TLR-4 signaling. PCM also reduces the phosphorylation of the activator-protein 1 (AP-1) transcription factors c-Jun and c-Fos in response to IL-1beta stimulation in a TLR-4-dependent manner. This study suggests that PCM may use TLR-4 through IRAK-2 and via AP-1 to prevent IL-1beta-induced IL-6 induction in immature enterocytes. Based on these observations, the combined use of probiotics and anti-TLR-4 therapy to prevent NEC may not be a good strategy.

Mezzasalma, V., Manfrini, E., Ferri, E., Sandionigi, A., La, Ferla B., Schiano, I., Michelotti, A., Nobile, V., Labra, M., and Di, Gennaro P. A Randomized, Double-Blind, Placebo-Controlled Trial: The Efficacy of Multispecies Probiotic Supplementation in Alleviating Symptoms of Irritable Bowel Syndrome Associated with Constipation. Biomed.Res.Int. 2016; 2016:4740907; PM: 27595104

ABSTRACT: ***Background and Aim***: The efficacy of supplementation treatment with two multispecies probiotic formulates on subjects diagnosed with IBS-C and the assessment of their gut microbiota were investigated. ***Methods***: A randomized, double-blind, three-arm parallel group trial was carried out on 150 IBS-C subjects divided into three groups (F_1, F_2, and F_3). Each group received a daily oral administration of probiotic mixtures (for 60 days) F_1 or F_2 or placebo F_3, respectively. Fecal microbiological analyses were performed by species-specific qPCR to assess the different amount of probiotics. ***Results***: The percentage of responders for each symptom was higher in the probiotic groups when compared to placebo group during the treatment period (t60) and was maintained quite similar during the follow-up period (t90). Fecal analysis demonstrated that probiotics of the formulations increased during the times of treatment only in fecal DNA from subjects treated with F_1 and F_2 and not with F_3, and the same level was maintained during the follow-up period. ***Conclusions***: Multispecies probiotic supplementations are effective in IBS-C subjects and induce a different assessment in the composition of intestinal microbiota. This clinical study is registered with the clinical study registration number ISRCTN15032219.

Mezzasalma, V., Manfrini, E., Ferri, E., Boccarusso, M., Di, Gennaro P., Schiano, I., Michelotti, A., and Labra, M. Orally administered multispecies probiotic formulations to prevent uro-genital infections: a randomized placebo-controlled pilot study. Arch.Gynecol.Obstet. 11-9-2016; PM: 27826653

ABSTRACT: ***PURPOSE***: The aim of this study was to evaluate in the vagina of 60 pre-menopausal women the detection of orally administered multispecies probiotic formulations showing anti-microbial properties in test in vitro. ***METHODS***: A randomized, double-blind, three-arm parallel pilot study was carried out on 60 pre-menopausal women. Subjects were randomly divided in three groups (F_1, F_2, F_3). Each group received a daily oral administration of probiotic mixtures (for 14 days and at the day 21, 7 days after the wash-out) containing: Lactobacillus acidophilus and Lactobacillus reuteri (F_1), or Lactobacillus plantarum, Lactobacillus rhamnosus and Bifidobacterium animalis subsp. lactis (F_2), or placebo (F_3), respectively. Vaginal swabs were collected at four experimental times, at t0 and at t7, t14 and t21 days, and analyzed by qPCR. At the same time, the anti-microbial activity of the probiotic formulations was verified by assays in vitro against microorganisms as Escherichia coli and Candida albicans. ***RESULTS***: L. acidophilus and L. reuteri as well as L. plantarum, L. rhamnosus and B. lactis were significantly increased on 7 days in the groups administered with F_1 and F_2, respectively, compared to group F_3. A similar significant trend was observed on 21 days, 7 days after the wash-out. F_1 and F_2 showed coherent anti-microbial properties. ***CONCLUSION***: Both probiotic formulations F_1 and F_2, chosen because of their anti-microbial activity against pathogens responsible for vaginal dysbiosis and infections, led to vaginal detection and enhancement of the amount of species of formulates when orally administered. This work provides the basis for further clinical investigations of the F_1 and F_2 capacity to prevent or treat uro-genital infections.

Mika, A., Rumian, N., Loughridge, A. B., and Fleshner, M. Exercise and Prebiotics Produce Stress Resistance: Converging Impacts on Stress-Protective and Butyrate-Producing Gut Bacteria. Int.Rev.Neurobiol. 2016; 131:165-191; PM: 27793217

ABSTRACT: The gut microbial ecosystem can mediate the negative health impacts of stress on the host. Stressor-induced disruptions in microbial ecology (dysbiosis) can lead to maladaptive health effects, while certain probiotic organisms and their metabolites can protect against these negative impacts. Prebiotic diets and exercise are feasible and cost-effective strategies that can increase stress-protective bacteria and produce resistance against the detrimental behavioral and neurobiological impacts of stress. The goal of this review is to describe research demonstrating that both prebiotic diets and exercise produce adaptations in gut ecology and the brain that arm the organism against inescapable stress-induced learned helplessness. The results of this research support the novel hypothesis that some of the stress-protective effects of prebiotics and exercise are due to increases in stress-protective gut microbial species and their metabolites. In addition, new evidence also suggests that prebiotic diet or exercise interventions are most effective if given early in life (juvenile-adolescence) when both the gut microbial ecosystem and the brain are plastic. Based on our new understanding of the mechanistic convergence of these interventions, it is feasible to propose that in adults, both interventions delivered in combination may elevate their efficacy to promote a stress-resistant phenotype.

Mitra, A., MacIntyre, D. A., Marchesi, J. R., Lee, Y. S., Bennett, P. R., and Kyrgiou, M. The vaginal microbiota, human papillomavirus infection and cervical intraepithelial neoplasia: what do we know and where are we going next? Microbiome. 11-1-2016; 4(1):58; PM: 27802830

ABSTRACT: The vaginal microbiota plays a significant role in health and disease of the female reproductive tract. Next-generation sequencing techniques based upon the analysis of bacterial 16S rRNA genes permit in-depth study of vaginal microbial community structure to a level of detail not possible with standard culture-based microbiological techniques. The human papillomavirus (HPV) causes both cervical intraepithelial neoplasia (CIN) and cervical cancer. Although the virus is highly prevalent, only a small number of women have a persistent HPV infection and subsequently develop clinically significant disease. There is emerging evidence which leads us to conclude that increased diversity of vaginal microbiota combined with reduced relative abundance of Lactobacillus spp. is involved in HPV acquisition and persistence and the development of cervical precancer and cancer. In this review, we summarise the current literature and discuss potential mechanisms for the involvement of vaginal microbiota in the evolution of CIN and cervical cancer. The concept of manipulation of vaginal bacterial communities using pre- and probiotics is also discussed as an exciting prospect for the field of cervical pathology.

Mladineo, I., Buselic, I., Hrabar, J., Radonic, I., Vrbatovic, A., Jozic, S., and Trumbic, Z. Autochthonous Bacterial Isolates Successfully Stimulate In vitro Peripheral Blood Leukocytes of the European Sea Bass (Dicentrarchus labrax). Front Microbiol. 2016; 7:1244; PM: 27551281

ABSTRACT: Commercially available probiotics are routinely administered as feed supplements in aquaculture important species. Among them, the European sea bass (Dicentrarchus labrax) is the most widely reared fish in the Mediterranean, whose rearing systems are highly variable between countries, affecting at some level the sustainability of production. After random isolation of autochthonous gut bacteria of the sea bass, their identification and pathogenicity testing, we have selected three potentially probiotic isolates; Pseudoalteromonas sp., Alteromonas sp., and Enterovibrio coralii. Selected isolates were tested and their immunostimulative efficiency was compared with a commercially available Lactobacillus casei isolate, inferring inflammatory, apoptotic and anti-pathogen response of sea bass' peripheral blood leukocytes. Phagocytic activity, respiratory burst, and expression of lysozyme, Mx protein, caspase 3, TNF-alpha, IL-10 genes was measured 1, 3, 5, and 12 h post-stimulation by four bacterial isolates to evaluate early kinetics of the responses. Best immunostimulative properties were observed in Pseudoalteromonas-stimulated leukocytes, followed by Alteromonas sp. and L. casei, while Enterovibrio coralii failed to induce significant stimulation. Based on such in vitro assay intestinal autochthonous bacterial isolates showed to have better immunostimulative effect in sea bass compared to aquaculture-widely used L. casei, and further steps need to engage tank and field feeding trials to evaluate long-term prophylactic suitability of the chosen isolates. A panel of biomarkers that represent pro-/anti-inflammatory, pro-/anti-apoptotic, and anti-bacteria/viral responses of the fish should be taken into consideration when evaluating the usefulness of the potential probiotic in aquaculture.

Mohamed, A. H., Osman, G. Y., Zowail, M. E., and El-Esawy, H. M. Effect of Lactobacillus sporogenes (probiotic) on certain parasitological and molecular aspects in Schistosoma mansoni infected mice. J.Parasit.Dis. 2016; 40(3):823-832; PM: 27605791

ABSTRACT: The prospective effects of Lactobacillus sporogenes (probiotics) and/or praziquantel (PZQ) treatment on some parasitological, histological and molecular aspects in Schistosoma mansoni infected mice were studied. The present data recorded that combination between PZQ (300 mg/Kg one dose 7 weeks post infection) and L. sporogenes (12.5 million spore/mice/week for 8 weeks from the first day of infection) reduced worm and ova count. Also, oogram patterns in liver and intestine recorded that treatment with L. sporogenes alone increase number of dead eggs especially in intestine. Histological observations showed significant ($P < 0.05$) reduction in the mean values of granuloma diameters in liver and intestine in infected mice groups treated with PZQ and/or L. sporogenes. Single strand breaks (comet assay) showed increase in number of damaged and strong damaged lymphocyte cells in mice infected with S. mansoni and infected treated with PZQ while L. sporogenes administration reduced DNA damage. Flow cytometry also confirmed role of L. sporogenes in reducing significantly DNA damage according to determination of cell cycle analysis apoptosis. It can be concluded that administration of L. sporogenes accompanied with PZQ treatment ameliorates the hepatic and intestinal damage caused by S. mansoni infection.

Mohammadi, A. A., Jazayeri, S., Khosravi-Darani, K., Solati, Z., Mohammadpour, N., Asemi, Z., Adab, Z., Djalali, M., Tehrani-Doost, M., Hosseini, M., and Eghtesadi, S. The effects of probiotics on mental health and hypothalamic-pituitary-adrenal axis: A randomized, double-blind, placebo-controlled trial in petrochemical workers. Nutr.Neurosci. 2016; 19(9):387-395; PM: 25879690

ABSTRACT: ***OBJECTIVE***: The aim of this study was to determine effects of probiotic yogurt and multispecies probiotic capsule supplementation on mental health and hypothalamic-pituitary-adrenal axis in petrochemical workers. ***METHODS***: The present randomized double-blind, placebo-controlled trial was conducted on 70 petrochemical workers. Subjects were randomly divided into three groups to receive 100 g/day probiotic yogurt + one placebo capsule (n = 25) or one probiotic capsule daily + 100 g/day conventional yogurt (n = 25) or 100 g/day conventional yogurt + one placebo capsule (n = 20) for 6 weeks. Mental health parameters including general health questionnaire (GHQ) and depression anxiety and stress scale (DASS) scores were measured. Fasting blood samples were obtained at the beginning and 6 weeks after the intervention to quantify hypothalamic-pituitary-adrenal axis. ***RESULTS***: After 6 weeks of intervention, a significant improvement of GHQ was observed in the probiotic yogurt (18.0 +/- 1.5 vs. 13.5 +/- 1.9, P = 0.007) and in the probiotic capsule group (16.9 +/- 1.8 vs. 9.8 +/- 1.9, P = 0.001), as well as a significant improvement in DASS scores in the probiotic yogurt (23.3 +/- 3.7 vs. 13.0 +/- 3.7, P = 0.02) and the probiotic capsule group (18.9 +/- 3.2 vs. 9.4 +/- 4.0, P = 0.006). However, there was no significant improvement in the conventional yogurt group (P = 0.05 for GHQ and P = 0.08 for DASS). ***DISCUSSION***: The consumption of probiotic yogurt or a multispecies probiotic capsule had beneficial effects on mental health parameters in petrochemical workers.

Montoro, B. P., Benomar, N., Lavilla, Lerma L., Castillo, Gutierrez S., Galvez, A., and Abriouel, H. Fermented Alorena Table Olives as a Source of Potential Probiotic Lactobacillus pentosus Strains. Front Microbiol. 2016; 7:1583; PM: 27774088

ABSTRACT: A collection of 31 Lactobacillus pentosus strains isolated from naturally fermented Alorena green table olives were screened in depth in the present study for their probiotic potential. Several strains could be considered promising probiotic candidates since they showed good growth capacity and survival under simulated gastro-intestinal conditions (acidic pH of 1.5, up to 4% of bile salts and 5 mM of nitrate), good ability to auto-aggregate which may facilitate their adhesion to host cells as multiple aggregates and the subsequent displacement of pathogens. Moreover, co-aggregation of lactobacilli with pathogenic bacteria was shown with Listeria innocua, Staphylococcus aureus, Escherichia coli, and Salmonella Enteritidis as good defense strategy against gut and food pathogens. Furthermore, they exhibited adherence to intestinal and vaginal cell lines, such property could be reinforced by their capacity of biofilm formation which is also important in food matrices such as the olive surface. Their antagonistic activity against pathogenic bacteria by means of acids and plantaricins, and also their different functional properties may determine their efficacy not only in the gastro-intestinal tract but also in food matrices. Besides their ability to ferment several prebiotics, the new evidence in the present study was their capacity to ferment lactose which reinforces their use in different food matrices including dairy as a dietary adjunct to improve lactose digestibility. Lactobacillus pentosus CF2-10N was selected to have the best probiotic profile being of great interest in further studies. In conclusion, spontaneous fermented Alorena table olives are considered a natural source of potential probiotic L. pentosus to be included as adjunct functional cultures in different fermented foods.

Morales-Barrera, E., Calhoun, N., Lobato-Tapia, J. L., Lucca, V., Prado-Rebolledo, O., Hernandez-Velasco, X., Merino-Guzman, R., Petrone-Garcia, V. M., Latorre, J. D., Mahaffey, B. D., Teague, K. D., Graham, L. E., Wolfenden, A. D., Baxter, M. F., Hargis, B. M., and Tellez, G. Risks Involved in the Use of Enrofloxacin for Salmonella Enteritidis or Salmonella Heidelberg in Commercial Poultry. Front Vet.Sci. 2016; 3:72; PM: 27630995

ABSTRACT: The objectives of the present study were to evaluate the risks involved in the use of Enrofloxacin for Salmonella Enteritidis (SE) or Salmonella Heidelberg (SH) in commercial poultry and determine the effects of a probiotic as an antibiotic alternative. Two experiments were conducted to evaluate the risks involved in the use of Enrofloxacin for SE or SH in commercial poultry. Experiment 1 consisted of two trials. In each trial, chickens were assigned to one of three groups; control + SE challenged; Enrofloxacin 25 mg/kg + SE; and Enrofloxacin 50 mg/kg + SE. Chickens received Enrofloxacin in the drinking water from days 1 to 5 of age. On day 6, all groups received fresh water without any treatment. All chickens were orally gavaged with 10(7) cfu/chick of SE at 7 days of age and euthanized on 8 days of age. In Experiment 2, turkey poults were assigned to one of the three groups; control + SH; probiotic + SH; and Enrofloxacin 50 mg/kg + SH. Poults received probiotic or Enrofloxacin in the drinking water from days 1 to 5 of age. On day 6, poults received fresh water without any treatment. Poults were orally gavaged with 10(7) cfu/poult of SH at 7 days of age. Poults were weighed and humanely killed 24 h post-SH challenge to evaluate serum concentration of fluorescein isothiocyanate-dextran to evaluate intestinal permeability, metagenomics, and SH infection. In both trials of Experiment 1, chickens treated with Enrofloxacin were more susceptible to SE organ invasion and intestinal colonization when compared with control non-treated chickens ($P < 0.05$). In Experiment 2, poults treated with 50 mg/kg of Enrofloxacin showed an increase in body weight, however, this group also showed an increase in SH susceptibility, intestinal permeability, and lower proportion of Firmicutes and Bacteroidetes, but with control group had the highest proportion of Proteobacteria. By

contrast, poults that received the probiotic had the highest proportion of Firmicutes and Bacteroidetes, but lowest Proteobacteria. The results of the present study suggest that prophylactic utilization of Enrofloxacin at five times the recommended dose in poultry increases the susceptibility to salmonellae infections, and confirms that probiotics may be an effective tool in salmonellae infections.

Moreno Algarra, M. C., Fernandez, Romero, V, Sanchez, Tamayo T., Espinosa Fernandez, M. G., and Salguero, Garcia E. [Variability in enteral feeding practices of preterm infants among hospitals in the SEN1500 Spanish neonatal network]. An.Pediatr.(Barc.). 11-9-2016; PM: 27838353

ABSTRACT: ***INTRODUCTION***: Proper nutrition is one of the primary objectives in the management of preterm infants. However, lack of evidence on the best strategy to achieve this objective has led to a great variability in feeding practices. This variability may be related to the differences in the incidence of complications, such as necrotising enterocolitis (NEC). ***OBJECTIVE***: The aim of this study is to assess the variability in clinical practice regarding enteral feeding in SEN-1500 Spanish network. ***METHOD***: An observational study was conducted using a questionnaire sent out in 2013 requesting information about feeding very low birth weight (VLBW) neonates (bank milk, start time, trophic feeding, increases, fortifiers and probiotics). ***RESULTS***: Responses were received from 60 of the 98 hospitals. The response rate was higher in centres with more than 50VLBW/year (30/31). Just over two-thirds (67%) have feeding protocols, and 52% refer to variability within their unit. A milk bank is available in 25% of the units. First feeding occurs fairly evenly throughout first 48hours, although it is delayed in lower gestational ages, even when there is no haemodynamic failure. In addition to hemodynamic instability there are other situations when the start is delayed (absence of breast milk, CIR, altered umbilical flow, asphyxia), while it is rarely delayed by absence of meconium or maintain an umbilical catheter.Half of those under 25 weeks begin directly with progressive increases instead of trophic feeding. Increases rarely reach 30ml/kg/day. Almost all use fortification and vitamins. There was a significant use of probiotics at the time of the survey. ***CONCLUSIONS***: There is great variability in enteral nutrition policies in VLBW in Spain. Although some differences are justified by the lack of evidence, there are other interventions that have proven to be effective, such as evidence-based protocols or access to donor milk. Implementation in all the units could reduce the incidence of NEC and improve the nutritional status.

Mori, N., Kano, M., Masuoka, N., Konno, T., Suzuki, Y., Miyazaki, K., and Ueki, Y. Effect of probiotic and prebiotic fermented milk on skin and intestinal conditions in healthy young female students. Biosci.Microbiota.Food Health. 2016; 35(3):105-112; PM: 27508111

ABSTRACT: Although persistent constipation is considered to be associated with skin problems, the supporting evidence is limited. Hence, this study investigated the effects of probiotic and prebiotic fermented milk on skin and intestinal conditions in an open-label trial. Among the 101 Japanese healthy young female students that participated, the 81 subjects in the intake group consumed a bottle of probiotic and prebiotic fermented milk containing Bifidobacterium breve strain Yakult and galactooligosaccharides daily for 4 weeks after a pre-intake period of 4 weeks, while the 20 subjects in the non-intake group did not consume the test beverage. Defecation patterns, skin conditions including hydration levels in the stratum corneum, and urinary phenol and p-cresol levels were evaluated before (baseline) and after intake. All subjects completed the study. No differences in dietary intake and body mass index were observed between both groups

during the trial. In the intake group, hydration levels of the stratum corneum, defecation frequency, and feces quantity significantly increased, and urinary phenol and p-cresol levels significantly decreased after intake compared with the corresponding baseline values. However, they did not significantly change in the non-intake group. A significant difference was observed between the intake group and non-intake group in regard to clearness of the skin as assessed by visual analogue scale. Therefore, consecutive intake of probiotic and prebiotic fermented milk might have beneficial effects on the skin that prevent dryness and beneficial effects on the intestinal conditions that stimulate defecation and decrease phenol production by gut bacteria in healthy young women.

Morovic, W., Hibberd, A. A., Zabel, B., Barrangou, R., and Stahl, B. Genotyping by PCR and High-Throughput Sequencing of Commercial Probiotic Products Reveals Composition Biases. Front Microbiol. 2016; 7:1747; PM: 27857709

ABSTRACT: Recent advances in microbiome research have brought renewed focus on beneficial bacteria, many of which are available in food and dietary supplements. Although probiotics have historically been defined as microorganisms that convey health benefits when ingested in sufficient viable amounts, this description now includes the stipulation "well defined strains," encompassing definitive taxonomy for consumer consideration and regulatory oversight. Here, we evaluated 52 commercial dietary supplements covering a range of labeled species using plate counting and targeted genotyping. Strain identities were assessed using methods recently published by the United States Pharmacopeial Convention. We also determined the relative abundance of individual bacteria by high-throughput sequencing (HTS) of the 16S rRNA sequence using paired-end 2 x 250 bp Illumina MiSeq technology. Using these methods, we tested the hypothesis that products do contain the quantitative and qualitative list of labeled microbial species. We found that 17 samples (33%) were below label claim for CFU prior to their expiration dates. A multiplexed-PCR scheme showed that only 30/52 (58%) of the products contained a correctly labeled classification, with issues encompassing incorrect taxonomy, missing species, and un-labeled species. The HTS revealed that many blended products consisted predominantly of Lactobacillus acidophilus and Bifidobacterium animalis subsp. lactis. These results highlight the need for reliable methods to determine the correct taxonomy and quantify the relative amounts of mixed microbial populations in commercial probiotic products.

Morrow, L. E. and Wischmeyer, P. Blurred Lines: Dysbiosis and Probiotics in the Intensive Care Unit. Chest. 10-19-2016; PM: 27771302

ABSTRACT: Clinicians have traditionally dichotomized bacteria as friendly commensals or harmful pathogens. However, the line separating the two has become blurred with the recognition that the intestinal microbiome is a complex entity wherein species can shift sides - from friend to foe and back again - based on crucial factors in their local environment. Significant disruptions in the homeostasis of the microbiome, a phenomenon called 'dysbiosis,' is increasingly associated with a host of untoward effects. Intensive care unit patients are at high risk for dysbiosis given high rates of antibiotic use, acute changes in diet, and the stress of critical illness. Probiotics are living microbes of human origin that, when ingested in sufficient quantities, can colonize sites such as the oropharynx and gastrointestinal tract and provide benefits to the host. In recent years we have increasingly explored the utility of using probiotics to reverse the intestinal dysbiosis associated with critical illness, thereby reducing select intensive care unit complications associated with increased morbidity and mortality. While these preliminary efforts have

demonstrated varying degrees of success, our present studies suffer from a host of limitations that hinder the strength of their conclusions and/or the generalizability of their results. Probiotic investigations have been further hobbled by current regulatory requirements, which were designed to serve as the framework for pharmaceutical research. While such measures are intended to ensure patient safety, they inadvertently impose barriers that stifle innovation regarding nutraceuticals. This review strives to summarize the current evidence regarding the efficacy and safety of probiotics in the intensive care unit as well as to provide an overview of the obstacles probiotic researchers face going forward.

Mourand, G., Paboeuf, F., Fleury, M. A., Jouy, E., Bougeard, S., Denamur, E., and Kempf, I. Escherichia coli probiotic strain ED1a in pigs has a limited impact on the gut carriage of extended-spectrum beta-lactamase-producing E. coli. Antimicrob.Agents Chemother. 10-17-2016; PM: 27795372

ABSTRACT: Four trials were conducted to evaluate the impact of Escherichia coli probiotic strain ED1a (EcED1a) administration to pigs on the gut carriage or survival in manure of extended-spectrum beta-lactamase-producing E. coli Groups of pigs were orally inoculated with strain E. coli M63 (EcM63) carrying the blaCTX-M-1 gene (n=84) or used as control (n=26). In the first two trials, eight days later, 24 of 40 EcM63 inoculated pigs were given EcED1a orally for six days. In the third trial, 10 EcM63 inoculated pigs were given either EcED1a or probiotic E. coli Nissle 1917 (EcN) for five days. In the fourth trial, EcED1a was given to a sow and its 12 piglets, the latter, as well as 12 piglets which did not received EcED1a, then being inoculated with EcM63. Fecal shedding of cefotaxime-resistant Enterobacteriaceae (CTX-RE) was studied by culture and blaCTX-M-1 genes were quantified by PCR. The persistence of CTX-RE in manure samples from inoculated pigs, or inoculated in vitro with EcM63 with or without probiotics was studied. Results showed that EcM63and EcED1a were good gut colonizers. The reduction in fecal excretion of CTX-RE in EcED1a-treated pigs compared to non-treated pigs was usually inferior to 1 log10 CFU and mainly observed during the probiotic administration period. The results obtained with EcN did not differ significantly from those with EcED1a. CTX-RE survival did not differ significantly in manures with or without probiotics. In conclusion, in our experimental conditions, EcED1a and EcN could not durably prevent CTX-RE colonization of the pig gut.

Mousavi, E., Makvandi, M., Teimoori, A., Ataei, A., Ghafari, S., Najafian, M., Ourang, Z., and Samarbaf-Zadeh, A. In vitro adherence of Lactobacillus strains isolated from the vaginas of healthy Iranian women. J.Chin Med.Assoc. 2016; 79(12):665-671; PM: 27562422

ABSTRACT: ***BACKGROUND***: The lactobacilli are a part of the bacterial flora of the human vagina. Detection of normal Lactobacillus species in the vaginas of healthy women in different geographical locations, and evaluation of their specific properties, can aid in the selection of the best species for preventing sexually transmitted diseases in the future. This study was performed to isolate and identify the Lactobacillus species in the vaginas of healthy women and to evaluate the adherence of these lactobacilli to Vero and HeLa cell lines. ***METHODS***: The study included 100 women. Bacteria were isolated from healthy women and purified. Phenotypic and biochemical tests were performed to identify the lactobacilli. The Lactobacillus species were detected by molecular methods using polymerase chain reaction amplification of the full length of the 16S rDNA of the isolated bacteria. Several isolates of each species were then selected to study their

adherence to Vero and HeLa cell lines. ***RESULTS***: Among the 50 samples taken from healthy women meeting the inclusion criteria, Lactobacillus species were identified in 33 (66%) samples. Of these lactobacilli, 14 isolates were Lactobacillus crispatus, six (18.2%) were Lactobacillus gasseri, nine (27%) were Lactobacillus rhamnosus, and the rest were either Lactobacillus salivarius (6%) or Lactobacillus plantarum (6%). L. rhamnosus showed the greatest adhesion to the cells when compared to the other tested species. All the lactobacilli isolated in this study showed a smaller capacity for cell adherence when compared with control species. ***CONCLUSION***: L. crispatus, L. rhamnosus, and L. gasseri were the dominant Lactobacillus species in the vaginas of healthy women in Iran. L. rhamnosus attached more readily to the cells than did the other species; therefore, this isolate is a good candidate for further studies on the potential health benefits and application of lactobacilli as probiotics.

Mudd, J. C. and Brenchley, J. M. Gut Mucosal Barrier Dysfunction, Microbial Dysbiosis, and Their Role in HIV-1 Disease Progression. J.Infect.Dis. 10-1-2016; 214 Suppl 2:S58-S66; PM: 27625432

ABSTRACT: Distinct pathological events occur within the gastrointestinal (GI) tract of Asian macaques with progressive simian immunodeficiency virus (SIV) infection and humans with human immunodeficiency virus type 1 (HIV-1) infection that are critical in shaping disease course. These events include depletion and functional alteration of GI-resident CD4(+) T cells, loss of antigen-presenting cells, loss of innate lymphocytes, and possible alterations to the composition of the gut microbiota. These contribute to structural damage to the GI tract and systemic translocation of GI tract microbial products. These translocated microbial products directly stimulate the immune system, and there is now overwhelming evidence that this drives chronic immune activation in HIV-1 and SIV infection. While combined antiretroviral therapy (cART) in HIV-1-infected subjects generally allows for immune reconstitution in peripheral blood, reconstitution of the GI tract occurs at a much slower pace, and both immunological and structural abnormalities persist in the GI tract. Importantly, studies of large cohorts of individuals have linked suboptimal GI reconstitution to residual inflammation and heightened morbidities in HIV-1-infected cART recipients. As a result, current era treatments aimed at augmenting restoration of the GI tract hold promise in returning cART recipients to full health.

Mukherjee, A., Dutta, D., Banerjee, S., Ringo, E., Breines, E. M., Hareide, E., Chandra, G., and Ghosh, K. Potential probiotics from Indian major carp, Cirrhinus mrigala. Characterization, pathogen inhibitory activity, partial characterization of bacteriocin and production of exoenzymes. Res.Vet.Sci. 2016; 108:76-84; PM: 27663374

ABSTRACT: The study explored antagonistic activity of the cellular components of potential probiotic bacteria from mrigal (Cirrhinus mrigala) against fish pathogens with a basic insight of the chemical nature of the antagonistic compound. Totally 208 autochthonous gut bacteria were isolated, of which 22 strains revealed antagonism towards >/=2 of the six common fish pathogens. Zones of inhibition (halo diameter) were presented as score and the four most promising strains were selected as putative probiotics based on the cumulative score assigned. Further, evaluation of different cellular components exhibited bactericidal activity against the fish pathogens. Verification of other probiotic properties revealed that each of the selected strains produced diverse extra-cellular enzymes. The selected strains grew better in intestinal mucus than skin mucus, were resistant to diluted bile juice (2-20%) and safe for the target

fish. The extracellular product used as crude bacteriocin revealed thermostability (up to 90 degrees C) and activity over wide pH range (4-9). Partial loss of activity through treatment with proteinase-K and trypsin indicated proteinaceous nature of the antibacterial compound produced by the probiotic strains. 16S rRNA partial gene sequencing revealed that the four strains CM1FG7, CM1HG5, CM3FG19 and CM3HG10 were similar to Bacillus stratosphericus (KM277362), Bacillus aerophilus (KM277363), Bacillus licheniformis (KM277364) and Solibacillus silvestris (KM277365), respectively.

Mukherjee, K., Kavalukas, S. L., and Barbul, A. Nutritional Aspects of Gastrointestinal Wound Healing. Adv.Wound.Care (New Rochelle.). 11-1-2016; 5(11):507-515; PM: 27867755

ABSTRACT: Significance: Although the wound healing cascade is similar in many tissues, in the gastrointestinal tract mucosal healing is critical for processes such as inflammatory bowel disease and ulcers and healing of the mucosa, submucosa, and serosal layers is needed for surgical anastomoses and for enterocutaneous fistula. Failure of wound healing can result in complications including infection, prolonged hospitalization, critical illness, organ failure, readmission, new or worsening enterocutaneous fistula, and even death. Recent Advances: Recent advances are relevant for the role of specific micronutrients, such as vitamin D, trace elements, and the interplay between molecules with pro- and antioxidant properties. Our understanding of the role of other small molecules, genes, proteins, and macronutrients is also rapidly changing. Recent work has elucidated relationships between oxidative stress, nutritional supplementation, and glucose metabolism. Thresholds have also been established to define adequate preoperative nutritional status. Critical Issues: Further work is needed to establish standards and definitions for measuring the extent of wound healing, particularly for inflammatory bowel disease and ulcers. In addition, a mounting body of evidence has determined the need for adequate preoperative nutritional supplementation for elective surgical procedures. Future Directions: A large portion of current work is restricted to model systems in rodents. Therefore, additional clinical and translational research is needed in this area to promote gastrointestinal wound healing in humans, particularly those suffering from critical illness, patients with enterocutaneous fistula, inflammatory bowel disease, and ulcers, and those undergoing surgical procedures.

Munir, M. B., Hashim, R., bdul Manaf, M. S., and Nor, S. A. Dietary Prebiotics and Probiotics Influence the Growth Performance, Feed Utilisation, and Body Indices of Snakehead (Channa striata) Fingerlings. Trop.Life Sci.Res. 2016; 27(2):111-125; PM: 27688855

ABSTRACT: This study used a two-phase feeding trial to determine the influence of selected dietary prebiotics and probiotics on growth performance, feed utilisation, and morphological changes in snakehead (Channa striata) fingerlings as well as the duration of these effects over a post-experimental period without supplementation. Triplicate groups of fish (22.46 +/-0.17 g) were raised on six different treatment diets: three prebiotics (0.2% beta-glucan, 1% galacto-oligosaccharides [GOS], 0.5% mannan-oligosaccharides [MOS]), two probiotics (1% live yeast [Saccharomyces cerevisiae] and 0.01% Lactobacillus acidophilus [LBA] powder) and a control (unsupplemented) diet; there were three replicates for each treatment. All diets contained 40% crude protein and 12% crude lipid. Fish were fed to satiation three times daily. No mortalities were recorded during Phase 1; however, 14% mortality was documented in the control and prebiotic-amended fish during Phase 2. At the end of Phase 1, growth performance and feed utilisation were significantly higher ($p<0.05$) in the LBA-treated fish, followed by live

yeast treatment, compared with all other diets tested. The performance of fish on the three prebiotic diets were not significantly different from one another but was significantly higher than the control diet. During Phase 2 (the post-feeding phase), fish growth continued until the 6th week for the probiotic-based diets but levelled off after four weeks for the fish fed the prebiotic diets. The feed conversion ratio (FCR) was higher in all treatments during the post-feeding period. The hepatosomatic index (HSI) did not differ significantly among the tested diets. The visceral somatic index (VSI) and intraperitoneal fat (IPF) were highest in the LBA-based diet and the control diet, respectively. The body indices were significantly different ($p<0.05$) between Phases 1 and 2. This study demonstrates that probiotic-based diets have a more positive influence on the growth, feed utilisation, and survival of C. striata fingerlings compared with supplementation with prebiotics.

Mupsilonller, M. J., Paul, T., and Seeliger, S. Necrotizing enterocolitis in premature infants and newborns. J.Neonatal Perinatal.Med. 9-16-2016; 9(3):233-242; PM: 27589549

ABSTRACT: Necrotizing enterocolitis (NEC) is the most common acquired disease of the gastrointestinal tract (GIT) in premature infants and newborns. It is defined as an ulcerative inflammation of the intestinal wall. The clinical signs of incipient NEC are often very discrete, and range from localized intestinal symptoms to generalized signs of sepsis. NEC is classified depending on its severity into disease states according to the modified Bell's Classification. Treatment of NEC ranges, depending on its severity, from a conservative therapeutic approach to surgery with resection of the affected parts of the intestine. Mortality is considerably high in extremely small preterm infants reaching up to 42% of the affected children. Measures such as breastfeeding or alternatively nutrition with pasteurized human donor milk from a milk bank, administration of probiotics, avoidance of histamine type II receptor antagonists, and restrictive antibiotic treatment should be considered early on for prevention of NEC.

Mur Perez, A. M., Mateo, Segura Z., Ramirez, Dominguez N., and Vela, Condon P. [Use of probiotics in bacterial vaginosis]. Semergen. 11-14-2016; PM: 27856152

ABSTRACT: ***INTRODUCTION***: Bacterial vaginosis (BV) is a change in the vaginal microbiota due to bacterial overgrowth of normal microorganisms, which produces symptoms of vaginal discomfort, increased malodorous vaginal discharge, etc. Although they been seen to improve recurrence rates in clinical practice, the role of probiotics is questioned in these situations. A literature review on BV is presented in order to evaluate the effectiveness of using probiotics as adjuvant therapy in BV. ***METHOD***: PubMed, Cochrane and SCOPUS review of the last 5 years publications using probiotics to treat women diagnosed with BV. ***RESULTS***: An analysis was performed on the 10 items that refer to the use of probiotics in addition to antibiotic treatments. ***CONCLUSIONS***: All items conclude that the use of probiotics is beneficial in cases of BV in addition to conventional treatments. However, the conclusions are not categorical due to there being a wide variety of studies and probiotics used.

Murad, H. A. Does Helicobacter pylori eradication therapy trigger or protect against Crohn's disease? Acta Gastroenterol.Belg. 2016; 79(3):349-354; PM: 27821031

ABSTRACT: Helicobacter pylori (H. pylori) infection is involved in multiple gastrointestinal and extra-gastrointestinal disorders. This review focuses on possible link between H. pylori eradication and Crohn's disease (CD) which is a chronic inflammatory bowel disease (IBD). Fecal calprotectin and; to lesser extent; fecal lactoferrin are sensitive and specific markers for monitoring CD activity. Data about link between H. pylori eradication and CD are limited and inconclusive. The infection likely shifts equilibrium between T helper 1 (Th1) and Th2 immune responses to the Th2 pattern. In subjects genetically predisposed to CD (a Th1-related disease), H. pylori eradication increases Th1 proinflammatory cytokines causing development of CD. In contrast, clarithromycin and/or proton pump inhibitors that are used to eradicate H. pylori can suppress Th1 factors, and theoretically can protect against CD, but there are no data to support this supposition. This Th1/Th2 approach seems very simplistic. Another theory is that alterations in gut microbiota form "continuous antigenic stimulation" predisposing to IBD. H. pylori infection can inhibit such stimulation through activation of regulatory T cells, and thus eradication may predispose to CD. Probiotics weren't found useful in treatment of CD. The reported data about link between H. pylori eradication and CD are currently limited. Case reports, suggesting a positive association between both conditions, provide a very little evidence. On eradicating H. pylori in CD patients and/or patients with high risk for CD, patient counseling and follow-up in addition to measuring fecal calprotectin may help monitor CD activity. (Acta gastro-enterol. belg., 2016, 79, 349-354).

Nakae, H., Tsuda, A., Matsuoka, T., Mine, T., and Koga, Y. Gastric microbiota in the functional dyspepsia patients treated with probiotic yogurt. BMJ Open.Gastroenterol. 2016; 3(1):e000109; PM: 27752337

*ABSTRACT: **OBJECTIVE***: To investigate the structure of the gastric microbiota in functional dyspepsia (FD) and its role in the pathophysiology. ***DESIGN***: We compared the basic physiological properties of the gastric fluid (GF) and the structure of the microbiota in the GF of 44 healthy control (HC) participants and 44 patients with FD. We then treated the patients with FD with a yogurt containing a probiotic strain of Lactobacillus gasseri OLL2716 (LG21 yogurt) and investigated the effects on the bacteriological parameters and symptoms to examine the relationship between them. ***RESULTS***: The volume of GF recovered from the stomach after overnight fasting was greater in the patients with FD than in the HCs, and decreased in the patients with FD whose symptoms were improved by the LG21 yogurt treatment. An analysis using a terminal restriction fragment polymorphism method demonstrated that the overall structure of the bacterial community and the abundance of genus Prevotella in the GF of the patients in the FD group were significantly different from those in the HC group. In the patients with FD, this bacteriological change was restored by treatment with LG21 yogurt. A significant inverse correlation was found between the abundance of Prevotella and the severity of postprandial distress-like symptoms in patients with FD who received LG21 yogurt. ***CONCLUSIONS***: Significant dysbiosis was found in the GF microbiota of patients with FD and considered to be involved in the pathogenesis. The abundance of genus Prevotella in the GF may be used as a biomarker of the efficacy of the treatment of FD. ***TRIAL REGISTRATION NUMBER***: UMINCTR000022026.

Nakao, H., Saito, K., Tomita, S., Magariyama, Y., Kaizuka, Y., and Takeda, Y. Direct Imaging of Carboxymethyl Cellulose-mediated Aggregation of Lactic Acid Bacteria Using Dark-field Microscopy. Anal.Sci. 2016; 32(10):1047-1051; PM: 27725602

ABSTRACT: The ecological functions of lactic acid bacteria (LAB) have been utilized in human life for food processing and probiotic therapy. Understanding the interaction mechanisms between LAB and food ingredients may help to clarify the fermentation process and physiological functions of LAB in the production of fermented foods made from plant materials and dairy products. However, the interaction mechanisms have yet to be fully clarified. Although laser diffraction was used for measuring the size changes of aggregates caused by the interaction between LAB and food ingredients, aggregate sizes could not be determined because of the precipitation of aggregates and its disruption from stirring. Therefore, a microscopy-based method for directly visualizing their interactions is required. We directly observed aggregation processes of LAB cells mediated by water-soluble polysaccharides, carboxymethyl cellulose (CMC), by dark-filed microscopy (DFM). DFM could visualize CMC-mediated cell aggregation with high contrast in real time, and revealed that the aggregates were formed by repeated collisions of LAB cells in a suspension. This suggests that our method can be used as a useful assay to directly visualize grain formation caused by interactions between LAB cells and various polysaccharides in food ingredients.

Nandi, A., Dan, S. K., Banerjee, G., Ghosh, P., Ghosh, K., Ringo, E., and Ray, A. K. Probiotic Potential of Autochthonous Bacteria Isolated from the Gastrointestinal Tract of Four Freshwater Teleosts. Probiotics.Antimicrob.Proteins. 8-24-2016; PM: 27557836

ABSTRACT: In this study, a total of 121 bacterial strains were isolated from the gastrointestinal tract of four teleostean species, namely striped snakehead (Channa striatus), striped dwarf catfish (Mystus vittatus), orangefin labeo (Labeo calbasu) and mrigal carp (Cirrhinus mrigala), among which 8 isolates showed promising antibacterial activity against four potential fish pathogens, Aeromonas hydrophila, Aeromonas salmonicida, Aeromonas sobria and Pseudomonas fluorescens and were non-hemolytic. The isolates were further screened in response to fish bile tolerance and extracellular digestive enzyme activity. Two bacterial strains MVF1 and MVH7 showed highest tolerance and extracellular enzymes activities, and selected for further studies. Antagonistic activity of these two isolates was further confirmed by in vitro growth inhibition assay against four selected fish pathogens in liquid medium. Finally, these two bacterial strains MVF1 and MVH7 were selected as potential probiotic candidates and thus identification by partial 16S rRNA gene sequence analysis. The bacterial isolates MVF1 and MVH7 were identified as two strains of Bacillus sp.

Neffe-Skocinska, K., Okon, A., Kolozyn-Krajewska, D., and Dolatowski, Z. Amino acid profile and sensory characteristics of dry fermented pork loins produced with a mixture of probiotic starter cultures. J.Sci.Food Agric. 11-9-2016; PM: 27859340

*ABSTRACT: **BACKGROUND***: Proteolysis is a biochemical process in dry-aged meat products where proteins are metabolized and broken down to polypeptides, peptides, and free amino acids. In the literature it is reported that an appropriate choice of probiotic starter culture limits proteolytic changes in dry-fermented meat products. In this study the combined effect of a mixture of probiotic starter cultures on the free amino

acid profile, total count of lactic acid bacteria, and the sensory quality of dry-aged pork loins after fermentation and after storing the vacuum-packed samples was evaluated. ***RESULTS***: LOCK900 and BB12 probiotic strains were the technologically best two-species mixture of starter cultures for the production of probiotic dry-aged pork loins. They allowed us to obtain products with high and stable bacterial count and acceptable sensory quality, both after 21 days of fermentation and after 2 months of cold storage. Changes in the free amino acid profile and increased intensity of the selected sensory attributes result from a significant share of probiotics in meat proteolysis occurring during fermentation and storage. ***CONCLUSION***: The results suggest the relevance of using probiotic bacteria as a two-species starter culture for the production of dry-aged products. (c) 2016 Society of Chemical Industry.

Nettis, E., Di, Leo E., Pastore, A., Distaso, M., Zaza, I., Vacca, M., Macchia, L., and Vacca, A. Probiotics and refractory chronic spontaneous urticaria. Eur.Ann.Allergy Clin.Immunol. 2016; 48(5):182-187; PM: 27608474

ABSTRACT: ***Background***: In chronic spontaneous urticaria (CSU) first-line therapy with an antihistamine-based regimen may not achieve satisfactory control in patients. Thus, a continuing need exists for effective and safe treatments for refractory CSU. **Aim**: To evaluate the clinical efficacy and safety of an intake of a combination of 2 probiotics (Lactobacillus salivarius LS01 and Bifidobacterium breve BR03) in patients with CSU who remain symptomatic despite concomitant H1-antihistamine therapy. ***Methods***: This report analyzes the effects of therapy with two probiotic strains on the clinical progress of 52 unselected patients with difficulty to treat CSU underwent to medical examination in two Italian specialist urticaria Clinics between September 2013 and September 2014. A mixture of Lactobacillus LS01 and Bifidobacterium BR03 were administered in each patient twice daily for 8 weeks. To evaluate patients' improvement with probiotics, urticaria activity score over 7 days (UAS7) was used at baseline and at week 8 in addition to a 5-question urticaria quality of life questionnaire. ***Results***: Fifty-two patients with CSU were included in this study (10 male and 42 female, age range 19-72 years). Mean disease duration was 1.5 years. Fourteen patients discontinued treatment, so evaluable population consisted of 38 patients. Nine of the 38 patients experienced mild clinical improvement during probiotic treatment (23.7%); one patient reported significant clinical improvement (2.6%) and one patient had complete remission of urticaria (2.6%). Twenty-seven patients did not have improvement in symptoms (71.1%). No side effects during the course of therapy were reported. ***Conclusions***: A combination of Lactobacillus salivarius LS01 and Bifidobacterium breve BR03 administered twice daily for 8 weeks might reduce the symptoms scores and improve quality of life scores in a part of patients with CSU who remained symptomatic despite treatment with H1 antihistamine mostly in subjects with allergic rhinitis.

Nicola, S., Amoruso, A., Deidda, F., Pane, M., Allesina, S., Mogna, L., Del, Piano M., and Mogna, G. Searching for the Perfect Homeostasis: Five Strains of Bifidobacterium longum From Centenarians Have a Similar Behavior in the Production of Cytokines. J.Clin.Gastroenterol. 2016; 50 Suppl 2, Proceedings from the 8th Probiotics, Prebiotics & New Foods for Microbiota and Human Health meeting held in Rome, Italy on September 13-15, 2015:S126-S130; PM: 27741155

ABSTRACT: ***GOALS***: To investigate the modulation of human cytokines by Bifidobacterium longum strains isolated from Centenarians. In particular, we measured the production of

interleukin (IL)-12p70, interferon-gamma, IL-17A, and IL-4 from human peripheral blood mononuclear cells after stimulation with live bacteria. ***BACKGROUND***: Probiotics may inhibit pathogens and modulate the immune system, bringing a beneficial effect on human health. Among the probiotic strains, bifidobacteria play a key role in the maturation of the host's immune system. At present, only a few comparative data are available on the effects of bifidobacteria associations on cytokine production. **STUDY**: Peripheral blood mononuclear cells were isolated, cultured, and stimulated (ratio 1:1) with B. longum DLBL07, B. longum DLBL08, B. longum DLBL09, B. longum DLBL10, or B. longum DLBL11, either alone or in association. Cytokine production was determined by an enzyme-linked immunosorbent assay. ***RESULTS***: Both the B. longum DLBL mixture and the individual B. longum DLBL strains induced similar levels of IL-4, interferon-gamma, and IL-17A. Under all conditions tested, no IL-12p70 release was detected. ***CONCLUSIONS***: The fact that B. longum strains were obtained from Centenarians suggests a perfect homeostasis between this specific species and the host. Moreover all the B. longum strains from Centenarians used in our study share some biological similarities.

Nikbakht, E., Khalesi, S., Singh, I., Williams, L. T., West, N. P., and Colson, N. Effect of probiotics and synbiotics on blood glucose: a systematic review and meta-analysis of controlled trials. Eur.J.Nutr. 9-3-2016; PM: 27590729

ABSTRACT: ***PURPOSE***: High fasting blood glucose (FBG) can lead to chronic diseases such as diabetes mellitus, cardiovascular and kidney diseases. Consuming probiotics or synbiotics may improve FBG. A systematic review and meta-analysis of controlled trials was conducted to clarify the effect of probiotic and synbiotic consumption on FBG levels. ***METHODS***: PubMed, Scopus, Cochrane Library, and Cumulative Index to Nursing and Allied Health Literature databases were searched for relevant studies based on eligibility criteria. Randomized or non-randomized controlled trials which investigated the efficacy of probiotics or synbiotics on the FBG of adults were included. Studies were excluded if they were review articles and study protocols, or if the supplement dosage was not clearly mentioned. ***RESULTS***: A total of fourteen studies (eighteen trials) were included in the analysis. Random-effects meta-analyses were conducted for the mean difference in FBG. Overall reduction in FBG observed from consumption of probiotics and synbiotics was borderline statistically significant (-0.18 mmol/L 95 % CI -0.37, 0.00; p = 0.05). Neither probiotic nor synbiotic subgroup analysis revealed a significant reduction in FBG. The result of subgroup analysis for baseline FBG level >/=7 mmol/L showed a reduction in FBG of 0.68 mmol/L (-1.07, -0.29; rho < 0.01), while trials with multiple species of probiotics showed a more pronounced reduction of 0.31 mmol/L (-0.58, -0.03; rho = 0.03) compared to single species trials. ***CONCLUSION***: This meta-analysis suggests that probiotic and synbiotic supplementation may be beneficial in lowering FBG in adults with high baseline FBG (>/=7 mmol/L) and that multispecies probiotics may have more impact on FBG than single species.

Nobili, V. and Cucchiara, S. The Use of Probiotics in Pediatric Nonalcoholic Fatty Liver Disease: Teachable Moment or Missed Opportunity? J.Pediatr.Gastroenterol.Nutr. 10-4-2016; PM: 27749611

No Abstract Available.

Nobutani, K., Sawada, D., Fujiwara, S., Kuwano, Y., Nishida, K., Nakayama, J., Kutsumi, H., Azuma, T., and Rokutan, K. The effects of administration of the Lactobacillus gasseri strain CP2305 on quality of life, clinical symptoms and changes in gene expression in patients with irritable bowel syndrome. J.Appl.Microbiol. 10-20-2016; PM: 27761980

ABSTRACT: ***AIMS***: To clarify the effects of Lactobacillus gasseri CP2305 (CP2305) on quality of life and clinical symptoms and its functional mechanisms in patients with irritable bowel syndrome (IBS). ***METHODS AND RESULTS***: After the patients were administered CP2305 daily for 4 weeks, the IBS-severity index score was significantly improved compared with that of the placebo group, and this improvement was accompanied by a reduction of health-related worry and changes in intestinal microbiota. The gene expression profiling of the peripheral blood leukocytes showed that CP2305 treatment significantly up-regulated genes related to eukaryotic initiation factor 2 (EIF2) signaling. Eighty-two genes were down-regulated in IBS patients compared with healthy controls. The expression of 23 of these genes exhibited a CP2305-dependent increase associated with an improvement in IBS severity. The majority of the restored genes were related to EIF2 signaling. ***CONCLUSION***: CP2305 administration is a potential candidate therapeutic option for patients with IBS. This article is protected by copyright. All rights reserved.

Nouri, Z., Karami, F., Neyazi, N., Modarressi, M. H., Karimi, R., Khorramizadeh, M. R., Taheri, B., and Motevaseli, E. Dual Anti-Metastatic and Anti-Proliferative Activity Assessment of Two Probiotics on HeLa and HT-29 Cell Lines. Cell J. 2016; 18(2):127-134; PM: 27551673

ABSTRACT: ***OBJECTIVE***: Lactobacilli are a group of probiotics with beneficial effects on prevention of cancer. However, there is scant data in relation with the impacts of probiotics in late-stage cancer progration, especially metastasis. The present original work was aimed to evaluate the anti-metastatic and anti-proliferative activity of lactobacillus rhamnosus supernatant (LRS) and lactobacillus crispatus supernatant (LCS) on the human cervical and colon adenocarcinoma cell lines (HeLa and HT-29, respectively). ***MATERIALS AND METHODS***: In this experimental study, the anti-proliferative activities of LRS and LCS were determined through MTT assay. MRC-5 was used as a normal cell line. Expression analysis of CASP3, MMP2, MMP9, TIMP1 and TIMP2 genes was performed by quantitative reverse transcriptase-polymerase chain reaction (qRT-PCR), following the cell synchronization. ***RESULTS***: Supernatants of these two lactobacilli had cytotoxic effect on HeLa, however LRS treatment was only effective on HT-29 cell line. In addition, LRS had no side-effect on normal cells. It was shown that CASP3 gene expression has been reduced after treatment with supernatants of two studied lactobacilli. According to our study, LRS and LCS are efficacious in the prevention of metastasis potency in HeLa cells with decreased expression of MMP2, MMP9 and increased expression of their inhibitors. In the case of HT-29 cells, only LRS showed this effect. ***CONCLUSION***: Herein, we have demonstrated two probiotics which have anti-metastatic effects on malignant cells and they can be administrated to postpone late-stage of cancer disease. LRS and LCS are effective on HeLa cell lines while only the effect of LRS is significant on HT-29, through cytotoxic and anti-metastatic mechanisms. Further assessments are required to evaluate our results on the other cancer cell lines, in advance to use these probiotics in other extensive trial studies.

Nozari, S., Mohammadzadeh, M., Faridvand, Y., Tockmechi, A., Movassaghpour, A., and Abdolalizadeh, J. The Study of Extracellular Protein Fractions of Probiotic Candidate Bacteria on Cancerous Cell Line. Arch.Iran Med. 2016; 19(11):779-785; PM: 27845547

ABSTRACT: ***INTRODUCTION***: Probiotics are live microorganisms, habituated in the human intestine, which have a beneficial effect on our health. In spite of many reports about the anticancer effect of these bacteria in in-vivo and in-vitro, their mechanisms of action are not completely understood. The goal of this study was to compare the extracellular fractions of Lactobacillus casei and L. paracasei on the anti proliferation and apoptosis induction in K562 cell line. ***MATERIALS AND METHODS***: L. casei and L. paracasei were cultured in MRS broth medium. Then extracellular secretions were collected and after enrichment, analyzed by electrophoresis. Fractionation were determined by gel filtration chromatography using sephadex G100 column, and the anticancer properties were evaluated. ***RESULTS***: The results of SDS-PAGE showed various molecular weight of fractionated proteins of L. casei and L. paracasei. Bioactivity assessment illustrated that anti proliferative effects on K562 cells is dose and time dependent and the cytotoxic effects was parallel with protein concentration and the increase of time from 36 to 72 hours. ***CONCLUSION***: Regarding the cell cytotoxicity results, the fractionated extracellular proteins of L. casei and L. paracasei have significant effects in inhibition of cancer cell proliferation. However, more study is needed to better elucidate the mechanisms of extracted proteins, and its effect on other human cancer cell lines.

Nwokoro, E., Leach, R., Ardal, C., Baraldi, E., Ryan, K., and Plahte, J. An assessment of the future impact of alternative technologies on antibiotics markets. J.Pharm.Policy Pract. 2016; 9:34PM: 27800166

ABSTRACT: ***BACKGROUND***: The increasing threat of antimicrobial resistance combined with the paucity of new classes of antibiotics represents a serious public health challenge. New treatment technologies could, in theory, have a significant impact on the future use of traditional antibiotics, be it by facilitating rational and responsible use or by product substitution in the existing antibiotics markets, including by reducing the incidence of bacterial infections through preventative approaches. The aim of this paper is to assess the potential of alternative technologies in reducing clinical use of and demand for antibiotics, and to briefly indicate which segments of the antibiotics market that might be impacted by these technologies. ***METHODS***: An initial mapping exercise to identify the alternative technologies was followed by a review of relevant published and grey literature (n = 52). We also carried out stakeholder engagement activities by a round-table discussion with infectious disease specialists and a multi-criteria decision analysis exercise with pharmaceutical industry experts. ***RESULTS***: Ten alternative technologies were identified and analyzed for their potential impact on the antibiotics market. Of these, rapid point-of-care diagnostics, vaccines, fecal microbiota transplantation, and probiotics were considered to have a "high" or "medium" potential impact over a 10-20 year horizon. Therapeutic antibodies, antibiotic biomaterials, bacteriophages, antimicrobial nanoparticles, antimicrobial peptides, and anti-virulence materials were rated as having "low" potential impact. ***CONCLUSION***: Despite the apparent potential of the most promising alternative technologies to reduce demand, that reduction will likely only happen in limited segments of the antibiotics market or, in the case of preventing community acquired streptococcal infections by vaccination, in a low-price generics market segment. Thus, alternative technologies are not expected to represent any disincentive to antibiotics developers. Finally, it is unlikely that alternative

technologies will displace the need for new classes, and sub-classes, of antibiotics in the short and medium terms.

Ogbogu, U. and Necyk, C. Community Pharmacists' Views and Practices Regarding Natural Health Products Sold in Community Pharmacies. PLoS.One. 2016; 11(9):e0163450; PM: 27661618

*ABSTRACT: **BACKGROUND**: Reports of regulatory and evidentiary gaps have raised concerns about the marketing and use of natural health products (NHPs). The majority of NHPs offered for sale are purchased at a community pharmacy and pharmacists are "front-line" health professionals involved in the marketing and provision of NHPs. To date, the involvement of pharmacists in pharmacy care involving NHPs and the degree to which concerns over the safety, efficacy, marketing and regulation of NHPs are addressed in pharmacy care in Canada have not been studied. **METHODS**: Using Qualtrics, a web-based data collection and analysis software, and a study instrument made up of fifteen (15) open-ended, closed and rating scale questions, we surveyed the attitudes and practices of 403 community pharmacists in the Canadian province of Alberta regarding NHPs offered for sale in community pharmacies. **RESULTS**: The majority of pharmacists surveyed (276; 68%) recommend NHPs to clients sometimes to very often. Vitamin D, calcium, multivitamins, prenatal vitamins, probiotics and fish oil and omega-3 fatty acids were the most frequently recommended NHPs. The most common indications for which NHPs are recommended include bone and musculoskeletal disorders, maintenance of general health, gastrointestinal disorders and pregnancy. Review articles published in the Pharmacist's Letter and Canadian Pharmacists Journal were the primary basis for recommending NHPs. The majority of pharmacists surveyed (339; 84%) recommend the use of NHPs concurrently with conventional drugs, while a significant number and proportion (125; 31%) recommend alternative use. Pharmacists in the study overwhelmingly reported providing counselling on NHPs to clients based on information obtained mainly from the Natural Medicines Comprehensive Database. **CONCLUSIONS**: The study findings indicate a high prevalence of pharmacy care relating to NHPs among study participants. Although pharmacists' practices around NHPs are consistent with the existing licensing framework, we found some involvement in problematic practices that necessitate further research and potential policy scrutiny. The study also uncovered patterns of recommendations, including sources relied on in recommending NHPs and in providing counselling to patients, that raise concerns about the quality and credibility of NHP-related care provided to pharmacy patrons.*

Oh, N. S., Joung, J. Y., Lee, J. Y., Kim, S. H., and Kim, Y. Characterization of the Microbial Diversity and Chemical Composition of Gouda Cheese Made by Potential Probiotic Strains as an Adjunct Starter Culture. J.Agric.Food Chem. 10-5-2016; 64(39):7357-7366; PM: 27606488

ABSTRACT: This study characterized the microbial diversity and chemical properties of Gouda cheese made by probiotics during ripening periods. Lactobacillus plantarum H4 (H4) and Lactobacillus fermentum H9 (H9), which demonstrate probiotic properties and bioactivity, were used as adjunct starter cultures. Gouda cheese made with H4 (GCP1) and H9 (GCP2) demonstrated the highest production of formic acid and propionic acid, respectively. Moreover, the bacterial diversity, including richness and evenness of nonstarter lactic acid bacteria (NSLAB), increased in probiotic cheeses. Specifically, Lactobacillus, Leuconostoc, and Streptococcaceae were present at higher

concentrations in probiotic cheeses than in control Gouda cheese (GCC). The proportion of H4 in GCP1 increased and culminated at 1.76%, whereas H9 in GCP2 decreased during ripening. Peptide profiles were altered by the addition of probiotics and included various bioactive peptides. In particular, three peptide fragments are newly detected. Therefore, Gouda cheese could be used as an effective probiotic carrier for H4 and H9.

Okesene-Gafa, K., Li, M., Taylor, R. S., Thompson, J. M., Crowther, C. A., McKinlay, C. J., and McCowan, L. M. A randomised controlled demonstration trial of multifaceted nutritional intervention and or probiotics: the healthy mums and babies (HUMBA) trial. BMC.Pregnancy.Childbirth. 11-24-2016; 16(1):373; PM: 27884128

ABSTRACT: ***BACKGROUND***: Maternal obesity is associated with adverse pregnancy outcomes and has lifelong negative implications for offspring health. The Institute of Medicine recommends limited gestational weight gain (GWG) in obese women for optimal maternal and infant outcomes. However, there is a gap regarding an effective and sustainable intervention strategy to achieve this goal. The aim of the healthy mums and babies (HUMBA) demonstration trial is to assess whether a multifaceted nutritional intervention and/or an oral probiotic treatment in obese pregnant women can reduce excessive GWG and optimise pregnancy outcomes. ***METHODS AND DESIGN***: The study is a two by two factorial randomised controlled demonstration trial conducted in Counties Manukau health region, New Zealand, a multi-ethnic region with a high prevalence of obesity. A total of 220 non-diabetic obese women with a singleton pregnancy will be recruited between 120 and 176 weeks. At recruitment, women are randomised to receive either a culturally tailored multifaceted dietary intervention or routine dietary advice, and either an oral probiotic or placebo capsule. Randomisation is undertaken via a web-based protocol, randomize.net, with a 1:1 ratio using stratification by body mass index (BMI) category (BMI of 30-34.9 or BMI >/=35 kg/m2). The dietary intervention includes 4 customised nutrition education visits by a trained community health worker combined with motivational text messaging. Probiotic capsules consist of Lactobacillus rhamnosus GG and Bifidobacterium lactis BB12 at a dose of 7 x 109 colony-forming units one per day until birth. Probiotic and placebo capsules are identically pre-packed and labelled by a third party, and are prescribed in a double blinded fashion. Research assessments are conducted at enrolment, 28 weeks, 36 weeks, at birth and at 5 months post-delivery. The primary outcomes for the study are proportion of women with excessive GWG and infant birthweight. ***DISCUSSION***: The HUMBA demonstration trial will assess the efficacy of a culturally tailored multifaceted dietary intervention and probiotic treatment in limiting excessive GWG and optimising birthweight in a multiethnic sample of obese pregnant women. If successful, either one or both of the interventions may be incorporated into future studies powered to investigate important pregnancy outcomes. ***TRIAL REGISTRATION***: Australian New Zealand Clinical Trials Registry registration number: ACTRN12615000400561, Universal Trial Number: U1111-1155-0409. Date registered: 29th April 2015.

Oliveira, L. F., Salvador, S. L., Silva, P. H., Furlaneto, F. A., Figueiredo, L., Casarin, R., Ervolino, E., Palioto, D. B., Souza, S. L., Taba, M., Jr., Novaes, A. B., Jr., and Messora, M. R. Benefits of Bifidobacterium Animalis Subsp Lactis Probiotic in Experimental Periodontitis. J.Periodontol. 9-23-2016; 1-20; PM: 27660886

ABSTRACT: ***BACKGROUND***: This study evaluated the effects of topical administration of probiotic bacteria (PROB) of the genus Bifidobacterium on experimental periodontitis (EP) in rats. ***METHODS***: 32 rats were divided into groups C (control; without EP), EP (EP only), C-HN019 (control+PROB) and EP-HN019 (EP+PROB). On day 0 of the experiment, animals of groups EP and EP-HN019 received cotton ligatures around mandibular first molars (MFM). In groups C-HN019 and EP-HN019, 1mL of suspensions containing Bifidobacterium animalis subsp lactis (B. lactis) HN019 was topically administered in the subgingival region of MFM on days 0, 3 and 7. In groups C and EP, topical administrations were performed using a sham suspension (without PROB). All animals were euthanized at day 14. Gingival tissue, hemi-mandibles and oral biofilm were collected. Data were statistically analyzed (P<0.05). ***RESULTS***: Group EP presented greater bone porosity, trabecular separation and connective tissue attachment loss as well as reduced bone volume than all the other groups (P<0.05). In group EP-HN019, there were greater proportions of Actinomyces and Streptococcus-like species and lower proportions of Veillonella parvula, Capnocytophaga sputigena, Eikenella corrodens, Prevotella intermedia-like species than group EP. Besides, group EP-HN019 presented greater expressions of Osteoprotegerin (OPG) and beta-defensins than group EP (P<0.05). Group EP presented greater levels of Interleukin (IL)-1beta and Receptor Activator of Nuclear Factor-Kappa B ligand (RANKL) than group EP-HN019 (P<0.05). ***CONCLUSION***: Topical use of B. lactis HN019 promotes a protective effect against alveolar bone and connective tissue attachment losses attributable to EP in rats, modifying immunoinflammatory and microbiological parameters.

Oliveira, V. M., Santos, S. S., Silva, C. R., Jorge, A. O., and Leao, M. V. Lactobacillus is able to alter the virulence and the sensitivity profile of Candida albicans. J.Appl.Microbiol. 2016; 121(6):1737-1744; PM: 27606962

ABSTRACT: ***AIMS***: The study investigated whether the interaction with Lactobacillus rhamnosus (ATCC7469) interfere with the expression of virulence factors by Candida albicans (ATCC18804). ***METHODS AND RESULTS***: These micro-organisms were grown in biofilms for 24, 48 and 72 h, Candida was isolated and the expression of the major virulence factors were investigated. The production of phospholipase, protease and haemolysin were observed in appropriate media; observation of germ tubes formation in serum; biofilm formation, after growth in microtitre plates and reading in spectrophotometer. Candida was also tested for antifungal sensitivity to amphotericin B, fluconazole and ketoconazole. The results were compared with the cells of Candida grown in the absence of lactobacilli (control group). Candida cells, which interacted with Lact. rhamnosus (test group), showed significantly lower proteinase and haemolysin activity, when compared with control group. The germ tube formation and biofilm formation capacity also decreased in tested groups, which demonstrated alterations in susceptibility to antifungal drugs. ***CONCLUSIONS***: The results suggest that Lact. rhamnosus is able to influence the expression of virulence factors by C. albicans and can alter its antifungal sensitivity profile. ***SIGNIFICANCE AND IMPACT OF THE STUDY***: These results suggest reduction in the pathogenicity of Candida and improvement in candidiasis therapy and control.

Olveira, G. and Gonzalez-Molero, I. An update on probiotics, prebiotics and symbiotics in clinical nutrition. Endocrinol.Nutr. 2016; 63(9):482-494; PM: 27633133

ABSTRACT: The concept of prebiotics, probiotics, and symbiotics and their use in different situations of daily clinical practice related to clinical nutrition is reviewed, as well as their role in the treatment/prevention of diarrhea (acute, induced by antibiotics, secondary to radiotherapy), inflammatory bowel disease (ulcerative colitis and pouchitis), in colonic health (constipation, irritable bowel), in liver disease (steatosis and minimum encephalopathy), and in intensive care, surgical, and liver transplantation. While their effectiveness for preventing antibiotic-induced diarrhea and pouchitis in ulcerative colitis appears to be shown, additional studies are needed to establish recommendations in most clinical settings. The risk of infection associated to use of probiotics is relatively low; however, there are selected groups of patients in whom they should be used with caution (as jejunum infusion).

Oo, K. M., Lwin, A. A., Kyaw, Y. Y., Tun, W. M., Fukada, K., Goshima, A., Shimada, T., and Okada, S. Safety and long-term effect of the probiotic FK-23 in patients with hepatitis C virus infection. Biosci.Microbiota.Food Health. 2016; 35(3):123-128; PM: 27508113

ABSTRACT: A clinical trial was conducted on 39 adult HCV-positive subjects to determine the safety and long-term effect of the probiotic FK-23 (heat-treated Enterococcus faecalis strain FK-23). Asymptomatic anti-HCV positive adults who fulfilled the selection criteria and gave voluntary consent were recruited from attendees of the Hepatitis Carrier Clinic, Department of Medical Research (Lower Myanmar). Each subject was given 2,700 mg of FK-23 per day by oral route. Blood samples were taken at enrollment and every 3 months and tested for alanine aminotransferase (ALT) and aspartate transaminase (AST). Viral load, urea, total protein, hemoglobin and platelet count were determined every 6 months. Among the subjects, 23 completed 36 months, 31 completed 24 months, 35 completed 12 months and 37 completed 6 months of probiotic therapy. Significant decreases in mean ALT levels were observed at 3 months (34. 9 +/- 15.1 IU/l) as compared with the initial level (64.8 +/- 17.5 IU/l) and persisted up to 36 months (43.7 +/- 25.2 IU/l). Decrease of AST was detected after 9 months (46.2 +/- 21.7 IU/l) of probiotic therapy as compared with the initial level (64.3 +/- 28.7 IU/l). FK-23 was safe based on the stable levels of biochemical and hematological parameters and the absence of untoward side effects. The FK-23 preparation was well tolerated and accepted by the subjects.

Ostlund-Lagerstrom, L., Kihlgren, A., Repsilber, D., Bjorksten, B., Brummer, R. J., and Schoultz, I. Probiotic administration among free-living older adults: a double blinded, randomized, placebo-controlled clinical trial. Nutr.J. 9-10-2016; 15(1):80; PM: 27612653

*ABSTRACT: **BACKGROUND***: Diseases of the digestive system have been found to contribute to a higher symptom burden in older adults. Thus, therapeutic strategies able to treat gastrointestinal discomfort might impact the overall health status and help older adults to increase their overall health status and optimal functionality. *OBJECTIVE*: The aim of this double-blinded, randomized, placebo-controlled clinical trial was to evaluate the effect of the probiotic strain Lactobacillus reuteri on digestive health and wellbeing in older adults. ***METHODS***: The study enrolled general older adults (>65 years). After eligibility screening qualified subjects (n = 290) participated in a 2-arm study design, with each arm consisting of 12 weeks of intervention of either active or placebo product. Primary

outcome measure was set to changes in gastrointestinal symptoms and secondary outcome measures were changes in level of wellbeing, anxiety and stress. Follow up was performed at 8 and 12 weeks. ***RESULTS***: No persistent significant effects were observed on the primary or secondary outcome parameters of the study. A modest effect was observed in the probiotic arm, were levels of stress decreased at week 8 and 12. Similarly, we found that subjects suffering from indigestion and abdominal pain, respectively, showed a significant decrease of anxiety at week 8 after probiotic treatment, but not at week 12. ***CONCLUSION***: The RCT failed to show any improvement in digestive health after daily intake of a probiotic supplement containing L. reuteri. Neither was any significant improvement in wellbeing, stress or anxiety observed. Even though the RCT had a negative outcome, the study highlights issues important to take into consideration when designing trials among older adults. *TRIAL REGISTRATION*: Clinicaltrials.gov/ NCT01837940.

Pagnini, C., Martorelli, M., Lanini, C., and Delle, Fave G. Development of an Ex Vivo Organ Culture Technique to Evaluate Probiotic Utilization in IBD. J.Clin.Gastroenterol. 2016; 50 Suppl 2, Proceedings from the 8th Probiotics, Prebiotics & New Foods for Microbiota and Human Health meeting held in Rome, Italy on September 13-15, 2015:S179-S182; PM: 27741170

ABSTRACT: The consistent technical and conceptual progress in the study of the microbiota has led novel impulse to the research for therapeutical application of probiotic bacteria in human pathologies, such as inflammatory bowel disease (IBD). Considering the heterogenous results of probiotics in clinical studies, the model of translational medicine may lead to a more specific and efficacious utilization of probiotic bacteria in IBD. In this regard, the selection and utilization of appropriate experimental models may drive the transition from pure in vitro systems to practical clinical application. We developed a simple and reproducible ex vivo organ culture method with potential utilization for the evaluation of probiotic bacteria efficacy in IBD patients.

Palumbo, V. D., Romeo, M., Marino, Gammazza A., Carini, F., Damiani, P., Damiano, G., Buscemi, S., Lo Monte, A. I., Gerges-Geagea, A., Jurjus, A., and Tomasello, G. The long-term effects of probiotics in the therapy of ulcerative colitis: A clinical study. Biomed.Pap.Med.Fac.Univ Palacky.Olomouc.Czech.Repub. 2016; 160(3):372-377; PM: 27623957

ABSTRACT: ***AIM***: Intestinal dysbiosis seems to be the leading cause of inflammatory bowel diseases, and probiotics seems to represent the proper support against their occurrence. Actually, probiotic blends and anti-inflammatory drugs represent a weapon against inflammatory bowel diseases. The present study evaluates the long-term (2 years) effects of combination therapy (mesalazine plus a probiotic blend of Lactobacillus salivarius, Lactobacillus acidophilus and Bifidobacterium bifidus strain BGN4) on ulcerative colitis activity. ***METHOD***: Sixty patients with moderate-to-severe ulcerative colitis were enrolled: 30 of them were treated with a single daily oral administration of mesalazine 1200 mg; 30 patients received a single daily oral administration of mesalazine 1200 mg and a double daily administration of a probiotic blend of Lactobacillus salivarius, Lactobacillus acidophilus and Bifidobacterium bifidus strain BGN4. The treatment was carried out for two years and the clinical response evaluated according to the Modified Mayo Disease Activity Index. ***RESULTS***: All patients treated with combination therapy showed better improvement compared to the controls. In particular, the beneficial effects of probiotics were evident even after two years of treatment. ***CONCLUSIONS***: A long-term treatment

modality of anti-inflammatory drugs and probiotics is viable and could be an alternative to corticosteroids in mild-to moderate ulcerative colitis.

Pan, Y., Wang, F., Sun, D. W., and Li, Q. Intestinal Lactobacillus community structure and its correlation with diet of Southern Chinese elderly subjects. J.Microbiol. 2016; 54(9):594-601; PM: 27572508

ABSTRACT: This study aimed to investigate the relationship between the intestinal Lactobacillus species and diet of elderly subjects in a longevity area in Southern China. Healthy elderly subjects ranging from 80 to 99 years old were respectively selected from the regions of Bama and Nanning, Guangxi, China. The nested polymerase chain reaction and denaturing gradient gel electrophoresis (PCR-DGGE) technology was used to analyze the intestinal Lactobacillus community structure. Results showed that Weissella confusa, L. mucosae, L. crispatus, L. salivarius, and L. delbrueckii were the representative Lactobacillus of elderly subjects. Among them, L. crispatus and L. delbrueckii were the dominant Lactobacillus of all species. In comparison to Nanning elderly subjects, the detection frequencies of W. confusa and L. salivarius were significantly increased in Bama elderly subjects ($P < 0.01$), whereas L. mucosae was significantly decreased ($P < 0.01$). Interestingly, it was also found that there were 4 kinds of representative Lactobacillus, which were significantly correlated with dietary fiber. W. confusa ($P < 0.01$) and L. salivarius ($P < 0.05$) were significantly positively correlated with the intake of dietary fiber, while L. mucosae ($P < 0.01$) and L. crispatus ($P < 0.05$) were significantly negatively correlated with the intake of dietary fiber, respectively. Results confirmed that different diets had obvious effects on the intestinal Lactobacillus community structure of elderly subjects in Southern China, which may provide a certain theoretical basis for the elderly's healthy food strategic design and probiotics product development.

Pandey, K. R., Joshi, C., and Vakil, B. V. Statistical optimization for enhanced yields of probiotic Bacillus coagulans and its phage resistant mutants followed by kinetic modelling of the process. Springerplus. 2016; 5(1):1654; PM: 27722071

ABSTRACT: Probiotics are microorganisms which when administered in adequate amounts confer health benefits to the host. A leading pharmaceutical company producing Bacillus coagulans as a probiotic was facing the problem of recurring phage attacks. Two mutants viz. B. co PIII and B. co MIII that were isolated as phage resistant mutants after UV irradiation and MMS treatment of phage sensitive B. coagulans parental culture were characterized at functional and molecular level and were noted to have undergone interesting genetic changes. The non-specific genetic alterations induced by mutagenesis can also lead to alterations in cell performance. Hence, in the current study the parental strain and the two mutants were selected for shake flask optimization. Plackett-Burman design was used to select the significant culture variables affecting biomass production. Evolutionary operation method was applied for further optimization. The study showed wide variations in the nutritional requirements of phage resistant mutants, post exposure to mutagens. An increment of 150, 134 and 152 % was observed in the biomass productions of B. coagulans (parental type) and mutants B.co PIII and B.co MIII respectively, compared to the yield from one-factor-at-a-time technique. Using Logistic and modified Leudeking-Piret equations, biomass accumulation and substrate utilization efficiency of the bioprocess were determined. The experimental data was in agreement with the results predicted by statistical analysis and modelling. The developed model may be useful for controlling the growth and substrate consumption kinetics in large scale fermentation using B. coagulans.

Park, Y., Moniruzzaman, M., Lee, S., Hong, J., Won, S., Lee, J. M., Yun, H., Kim, K. W., Ko, D., and Bai, S. C. Comparison of the effects of dietary single and multi-probiotics on growth, non-specific immune responses and disease resistance in starry flounder, Platichthys stellatus. Fish.Shellfish.Immunol. 2016; 59:351-357; PM: 27818339

ABSTRACT: An 8-week feeding trial was conducted to evaluate the effects of dietary probiotics on growth performance and non-specific immune responses in starry flounder, Platichthys stellatus. Fish averaging 46.5 +/- 0.65 g (mean +/- SD) were fed one of the six experimental diets; one control (Cont), and five other diets were prepared by supplementing single-probiotics 1 (Bacillus subtilis; SP1, 2 x 109 CFU kg-1 diet), single-probiotics 2 (Bacillus licheniformis; SP2, 2 x 109 CFU kg-1 diet), multi-probiotics 1 (Bacillus subtilis + Bacillus licheniformis; MP1, 2 x 109 CFU kg-1 diet), multi-probiotics 2 (commercial probiotics; Bacillus subtills + Bacillus licheniformis + Paenibacillus polymyxa + Aspergillus oryzae + Saccharomyces cerevisiae; MP2, 2 x 109 CFU kg-1 diet) and oxytetracycline (OTC) at 5 g OTC kg-1 diet. At the end of 8 weeks feeding trial, weight gain (WG) and specific growth rate (SGR) of fish fed SP1, MP1 and MP2 diets were significantly higher than those of fish fed control diet ($P < 0.05$). Superoxide dismutase (SOD) activity of fish fed MP2 diet was significantly higher than those of fish fed OTC diet ($P < 0.05$). Nitro blue tetrazolium (NBT) activity and lysozyme activity of fish fed SP1, MP1 and MP2 diets were significantly higher than those of fish fed OTC diet ($P < 0.05$). However, there was no significant difference among fish fed SP1, SP2, MP1 and MP2 diets. During the Edwardsiella tarda challenge test, the first mortality occurred on day 2. After the 14 days challenge test, cumulative survival rate of fish fed MP1 and MP2 diets were significantly higher than those of fish fed control diet ($P < 0.05$). However, there was no significant difference among fish fed SP1, SP2, MP1, MP2 and OTC diets in survival rate at the termination of the challenge test. Although there was little advantage in immunological parameters with fish fed MP diets, single and multi-probiotics were equally effective statistically. These results demonstrated that single or multi-probiotics had equal beneficial effect as an antibiotic replacer in terms of growth performance, non-specific immune responses and disease resistance in starry flounder.

Park, Y. H., Hamidon, F., Rajangan, C., Soh, K. P., Gan, C. Y., Lim, T. S., Abdullah, W. N., and Liong, M. T. Application of Probiotics for the Production of Safe and High-quality Poultry Meat. Korean J.Food Sci.Anim Resour. 10-31-2016; 36(5):567-576; PM: 27857531

ABSTRACT: Poultry industry has always been a dynamic and integral part of national economies in many countries. Economic losses incur especially in large-scale rearing facilities, often attributed to the deterioration of environmental conditions, poultry exposure to stressors and development of diseases. While antibiotics have been commonly used for prophylactic purposes and as growth stimulants, extensive documentation of antimicrobial resistance among pathogenic bacteria due to indiscriminate utilization of antibiotic in the industry has led to public and governmental outcries. Elimination of antibiotics from poultry production has thus encouraged intensive search for alternatives. In this review, we discuss the immense potential of probiotics to fill the gap as alternative growth promoters and evidences of beneficial effects of probiotic application in poultry production.

Parnell, J. A., Wiens, K. P., and Erdman, K. A. Dietary Intakes and Supplement Use in Pre-Adolescent and Adolescent Canadian Athletes. Nutrients. 8-26-2016; 8(9); PM: 27571101

ABSTRACT: Young athletes experience numerous dietary challenges including growth, training/competition, unhealthy food environments, and travel. The objective was to determine nutrient intakes and supplement use in pre-adolescent and adolescent Canadian athletes. Athletes (n = 187) aged 11-18 years completed an on-line 24-h food recall and dietary supplement questionnaire. Median energy intake (interquartile range) varied from 2159 kcal/day (1717-2437) in 11-13 years old females to 2905 kcal/day (2291-3483) in 14-18 years old males. Carbohydrate and protein intakes were 8.1 (6.1-10.5); 2.4 (1.6-3.4) in males 11-13 years, 5.7 (4.5-7.9); 2.0 (1.4-2.6) in females 11-13 years, 5.3 (4.3-7.4); 2.0 (1.5-2.4) in males 14-18 y and 4.9 (4.4-6.2); 1.7 (1.3-2.0) in females 14-18 years g/kg of body weight respectively. Median vitamin D intakes were below the recommended dietary allowance (RDA) and potassium was below the adequate intake (AI) for all athlete groups. Females 14-18 years had intakes below the RDA for iron 91% (72-112), folate 89% (61-114) and calcium 84% (48-106). Multivitamin-multiminerals, vitamin C, vitamin D, vitamin-enriched water, protein powder, sport foods, fatty acids, probiotics, and plant extracts were popular supplements. Canadian pre-adolescent and adolescent athletes could improve their dietary intakes by focusing on food sources of calcium, vitamin D, potassium, iron, and folate. With the exceptions of vitamin D and carbohydrates during long exercise sessions, supplementation is generally unnecessary.

Patrone, V., Molinari, P., and Morelli, L. Microbiological and molecular characterization of commercially available probiotics containing Bacillus clausii from India and Pakistan. Int.J.Food Microbiol. 11-21-2016; 237:92-97; PM: 27543819

ABSTRACT: Probiotics are actively used for treatment of diarrhoea, respiratory infections, and prevention of infectious gastrointestinal diseases. The efficacy of probiotics is due to strain-specific features and the number of viable cells; however, several reports of deviations from the label in the actual content of strains in probiotic products are a matter of concern. Most of the available data on quality focuses on probiotic products containing lactobacilli and/or bifidobacteria, while very few data are available on spore-forming probiotics. The present study evaluates the label claims for spore count and species identification in five commercial probiotic products marketed in India and Pakistan that claim to contain Bacillus clausii: Tufpro, Ecogro, Enterogermina, Entromax, and Ospor. Bacterial enumeration from three batches was done by microbiological plating methods by two independent operators. Species identification was done using PCR amplification and sequence analysis of the 16S rRNA gene, and determination of the total amount of species present in the products was done using PCR-denaturing gradient gel electrophoresis (PCR-DGGE) analysis followed by DNA sequencing of the excised bands. Plate count methods demonstrated poor correlations between quantitative label indications and bacteria recovered from plates for Tufpro, Ecogro, and Ospor. The 16S rRNA analysis performed on bacteria isolated from plate counts showed that only Enterogermina and Ospor contained homogenous B. clausii. PCR-DGGE analysis revealed that only Enterogermina had a homogenous B. clausii population while other products had mixed bacterial populations. In conclusion, the current analysis clearly demonstrates that of the five analysed commercial probiotics, only Enterogermina followed the label claims.

Pender, C. M., Kim, S., Potter, T. D., Ritzi, M. M., Young, M., and Dalloul, R. A. Effects of in ovo supplementation of probiotics on performance and immunocompetence of broiler chicks to an Eimeria challenge. Benef.Microbes. 11-30-2016; 7(5):699-705; PM: 27726419

ABSTRACT: Coccidiosis is regarded as the parasitic disease with the greatest economic impact on the poultry industry due to reduced performance and increased mortality. A study was conducted to investigate the effects of in ovo administration of probiotics on hatchability, performance, immune organ weights, and lesion scores in broiler chicks during a mixed Eimeria infection. At embryonic day 18, 210 eggs were injected with either sterile water or 1x106 cfu probiotic bacteria. On day 3 post-hatch, half of the chicks from each treatment group were challenged with a mixed inoculum of Eimeria acervulina, Eimeria maxima and Eimeria tenella. Measurements and tissue samples were taken on day of hatch (DOH) and days 3, 9 and 15. On day 9, 24 birds per treatment were scored for intestinal Eimeria lesions. No differences were seen among groups for hatchability as well as for body weight (BW), BW gain (BWG), or immune organ weights prior to the Eimeria challenge. On day 9, the non-challenged birds with probiotic supplementation had higher BW and BWG than the non-supplemented controls while no differences were seen among the challenged groups. On day 15, probiotic supplemented birds had improved BW compared to the non-supplemented birds as well as increased BWG from day 9 to 15. Bursa weight was not affected by treatment at any time point while spleen weight was greater in supplemented birds on day 15. Birds receiving the probiotic had significantly lower mortality than non-treated birds. Additionally, gross lesion severity was reduced due to probiotic supplementation in all intestinal segments evaluated. These results suggest that in ovo supplementation of probiotics may improve early performance and provide protection against a mixed Eimeria infection.

Perbal, B. To flush or not to flush ... that is a question. J.Cell Commun.Signal. 11-7-2016; PM: 27822878

ABSTRACT: The human gut microflora has drawn a lot of attention as a potent therapeutic tool for many decades. More recently, efforts have been developed to devise efficient ways of complementing or replacing deficient intestinal microflora associated with intestinal diseases that are resistant to conventional medical treatments. Aside from the medical and industrial applications that emerged from the use of gut microbiota, the complex constitution of this ecosystem raises fascinating questions regarding host-cell communication and host response mechanisms to the ever changing environment. This brief comment also points to questions raised by some unexpected applications that have recently emerged from this field.

Pereira-Caro, G., Fernandez-Quiros, B., Ludwig, I. A., Pradas, I., Crozier, A., and Moreno-Rojas, J. M. Catabolism of citrus flavanones by the probiotics Bifidobacterium longum and Lactobacillus rhamnosus. Eur.J.Nutr. 10-8-2016; PM: 27722779

ABSTRACT: PURPOSE: Orange juice (OJ) flavanones undergo limited absorption in the upper gastrointestinal tract and reach the colon where they are transformed by the microbiota prior to absorption. This study investigated the ability of two probiotic bacteria, Bifidobacterium longum R0175 and Lactobacillus rhamnosus subsp. Rhamnosus NCTC 10302 to catabolise OJ flavanones. *METHODS*: The bacteria were incubated with hesperetin-7-O-rutinoside, naringenin-7-O-rutinoside, hesperetin and naringenin, and the

culture medium and intracellular cell extracts were collected at intervals over a 48 h of incubation period. The flavanones and their phenolic acid catabolites were identified and quantified by HPLC-HR-MS. *RESULTS*: Both probiotics were able to subject hesperetin to ring fission yielding 3-(3'-hydroxy-4'-methoxyphenyl)propionic acid which was subsequently demethylated producing 3-(3',4'-dihydroxyphenyl)propionic acid and then via successive dehydroxylations converted to 3-(3'-hydroxyphenyl)propionic acid and 3-(phenyl)propionic acid. Incubation of both bacteria with naringenin resulted in its conversion to 3-(4'-hydroxyphenyl)propionic acid which underwent dehydroxylation yielding 3-(phenyl)propionic acid. In addition, only L. rhamnosus exhibited rhamnosidase and glucosidase activity and unlike B. longum, which was able to convert hesperetin-7-O-rutinoside and naringenin-7-O-rutinoside to their respective aglycones. The aglycones were then subjected to ring fission and further catabolised in a similar manner to that described above. The flavanones and their catabolites were found in the culture medium but not accumulated in the bacterial cells. *CONCLUSIONS*: These findings demonstrate the enzymatic potential of single strains of bifidobacterium and lactobacillus which may be involved in the colonic catabolism of OJ flavanones in vivo.

Pericolini, E., Gabrielli, E., Ballet, N., Sabbatini, S., Roselletti, E., Cayzeele, Decherf A., Pelerin, F., Luciano, E., Perito, S., Justen, P., and Vecchiarelli, A. Therapeutic activity of a Saccharomyces cerevisiae-based probiotic and inactivated whole yeast on vaginal candidiasis. Virulence. 7-19-2016; 1-17; PM: 27435998

ABSTRACT: Vulvovaginal candidiasis is the most prevalent vaginal infection worldwide and Candida albicans is its major agent. Vulvovaginal candidiasis is characterized by disruption of the vaginal microbiota composition, as happens following large spectrum antibiotic usage. Recent studies support the effectiveness of oral and local probiotic treatment for prevention of recurrent vulvovaginal candidiasis. Saccharomyces cerevisiae is a safe yeast used as, or for, the production of ingredients for human nutrition and health. Here, we demonstrate that vaginal administration of probiotic Saccharomyces cerevisiae live yeast (GI) and, in part, inactivated whole yeast Saccharomyces cerevisiae (IY), used as post-challenge therapeutics, was able to positively influence the course of vaginal candidiasis by accelerating the clearance of the fungus. This effect was likely due to multiple interactions of Saccharomyces cerevisiae with Candida albicans. Both live and inactivated yeasts induced coaggregation of Candida and consequently inhibited its adherence to epithelial cells. However, only the probiotic yeast was able to suppress some major virulence factors of Candida albicans such as the ability to switch from yeast to mycelial form and the capacity to express several aspartyl proteases. The effectiveness of live yeast was higher than that of inactivated whole yeast suggesting that the synergy between mechanical effects and biological effects were dominant over purely mechanical effects. The protection of epithelial cells to Candida-induced damage was also observed. Overall, our data show for the first time that Saccharomyces cerevisiae-based ingredients, particularly the living cells, can exert beneficial therapeutic effects on a widespread vaginal mucosal infection.

Pinto, J. M. and Petrova, A. Lactobacillus acidophilus Mixture in Treatment of Children Hospitalized With Acute Diarrhea. Clin.Pediatr.(Phila). 2016; 55(13):1202-1209; PM: 26581358

ABSTRACT: Despite unproven effectiveness, Lactobacillus acidophilus is a widely used

probiotic in the treatment of pediatric diarrhea. In this report, we evaluated the association between length of stay (LOS) for 290 young children hospitalized with acute diarrhea and adjuvant therapy with a probiotic mixture containing 80% L acidophilus that was included in treatment for 22.4% of them. Overall, no association between LOS and use of L acidophilus was recorded after controlling for age, length of diarrhea symptoms, duration of intravenous fluids, and prior exposure to antibiotic. However, LOS was directly associated with use of L acidophilus in children with negative stool studies, and no such association was recorded in children with positive stool for rotavirus or other infections. We concluded that adjuvant therapy with L acidophilus mixture is not beneficial for young children hospitalized with acute diarrhea.

Pirbaglou, M., Katz, J., de Souza, R. J., Stearns, J. C., Motamed, M., and Ritvo, P. Probiotic supplementation can positively affect anxiety and depressive symptoms: a systematic review of randomized controlled trials. Nutr.Res. 2016; 36(9):889-898; PM: 27632908

ABSTRACT: Gastrointestinal microbiota, consisting of microbial communities in the gastrointestinal tract, play an important role in digestive, metabolic, and immune functioning. Preclinical studies on rodents have linked behavioral and neurochemical changes in the central nervous system with deficits or alterations in these bacterial communities. Moreover, probiotic supplementation in rodents has been shown to markedly change behavior, with correlated changes in central neurochemistry. While such studies have documented behavioral and mood-related supplementation effects, the significance of these effects in humans, especially in relation to anxiety and depression symptoms, are relatively unknown. Thus, the purpose of this paper was to systematically evaluate current literature on the impact of probiotic supplementation on anxiety and depression symptoms in humans. To this end, multiple databases, including Medline, PsycINFO, PubMed, Scopus, and Web of Science were searched for randomized controlled trials published between January 1990 and January 2016. Search results led to a total of 10 randomized controlled trials (4 in clinically diagnosed and 6 in non-clinical samples) that provided limited support for the use of some probiotics in reducing human anxiety and depression. Despite methodological limitations of the included trials and the complex nature of gut-brain interactions, results suggest the detection of apparent psychological benefits from probiotic supplementation. Nevertheless a better understanding of developmental, modulatory, and metagenomic influences on the GI microbiota, specifically as they relate to mood and mental health, represent strong priorities for future research in this area.

Popova, B., Mitev, D., and Nikolov, A. [THE ROLE OF MOTHER'S MILK AND BREAST FEEDING. MEDICAL PROBLEMS DURING THE LACTATION PERIOD LACTOBACILUS FERMENTUM--A NEW APPROACH TOWARDS THE PREVENTION AND THE TREATMENT OF ACUTE AND SUBACUTE MASTITIS]. Akush.Ginekol.(Sofiia). 2016; 55(3):47-52; PM: 27514147

ABSTRACT: Breast feeding provides a lot of short and long-term benefits for the mother and the baby. It prevents the baby of gastrointestinal, urinary and respiratory infections, atopical conditions and assures long-term protection of cardiovascular and metabolic diseases. The breast feeding decreases the risk for the mother of ovary and breast carcinoma and creates a positive emotional bond between the mother and the baby Mother's milk is a species specific; its content is relatively stable regardless of mother's age race, way and place of living. Mother's milk is not sterile. There is a 10 year international trial held in Spanish and Finnish universities. It has identified and count all

microorganisms in mother's milk (more than 700) and proved that their content and quantity varies according the age of the baby. Mother's milk is a source of lactobacillus for baby's intestines and most of them have probiotic potential. Lactobacillus fermentum Lc40 (hereditum) is isolated from mother's milk. It has a good viability in gastrointestinal system, high level of adhesion to intestinal epithelium cells, produces glutation--strong antioxidant, good antibacterial activity to entero-pathogens and potential of increasing the immunologic response. Clinical trials reveal that Lactobacillus fermentum plays important role to microflora balance of mother's milk in mastitis during lactation. Many trials estimating the efficiency of lactobacillus fermentum in prevention and treatment of acute and subacute mastitis have been carried out. The results of them open a new door in front of us in the treatment of these conditions--treatment with probiotics instead of antibiotics.

Principi, N. and Esposito, S. Gut microbiota and central nervous system development. J.Infect. 2016; 73(6):536-546; PM: 27725185

ABSTRACT: ***OBJECTIVES***: Gut dysbiosis has been associated with several clinically relevant conditions, including alterations of central nervous system (CNS) structure and function development. This review discussed aspects of the relationship between gut microbiota and the CNS during development. ***METHODS***: PubMed was used to search for all of the studies published over the last 15 years using the key word "microbiota" and "gut" or "intestinal" and "nervous system". More than 350 articles were found, and only those published in English and providing data on aspects related to neurologic diseases were included in the evaluation. ***RESULTS***: The data indicate that the gut microbiota influences CNS development and function and that gut dysbiosis is associated with significant neurological problems. However, most of these data have been collected in experimental animals and cannot be transferred to humans. Moreover, it is not definitively established whether neurologic diseases depend on a generic modification of the gut microbiota or whether a single bacterial phylum or species plays a specific role for any single condition. Furthermore, limited information exists regarding protective bacteria. ***CONCLUSIONS***: Both probiotics and prebiotics can have different impacts on CNS according to the microbial species or oligosaccharides that are administered. In humans, particularly in children, several factors may be important in conditioning gut microbiota modifications; unfortunately, most of these factors act simultaneously. More efforts are required to fully define both the array of complex behaviors that are influenced by the gut microbiota at the CNS level and the mechanisms involved.

Qiao, Y. Q., Cai, C. W., and Ran, Z. H. Therapeutic Modulation of the Gut Microbiota in. J.Dig.Dis. 10-15-2016; PM: 27743467

ABSTRACT: Inflammatory bowel disease (IBD) patients exhibit impaired control of the microbiome in the gut. "Dysbiosis" is commonly observed. A Western diet is a risk factor for the development of IBD but may have different effects on the gut microbiota in IBD and non-IBD individuals. Exclusive enteral nutrition (EEN) can induce remission in pediatric Crohn's disease (CD) with a decrease in gut microbiome diversity after EEN treatment. Although there are some theoretical benefits, the actual treatment effects of prebiotics and probiotics in IBD patients can vary. Many high-quality studies have shown that probiotics VSL#3 exhibit benefits in ulcerative colitis (UC) therapy, and animal models have shown that gut microbial diversity is reduced after treatment with VSL#3. Fecal microbiome transplant (FMT) is controversial in IBD. Increasing microbiome diversity vs.

impaired handling of bacteria presents a dilemma. Antibiotics are the strongest factors in the reduction of microbiome ecological diversity. Some antibiotics may have benefits by inducing remission of IBD. Recent studies suggested that microbiome alteration might be an intrinsic property of IBD and a potential predictor in diagnosis and prognosis. However, the effects of therapeutic modulation are variable; thus, there are more questions to be answered.

Qin, Y., Hou, J., Deng, M., Liu, Q., Wu, C., Ji, Y., and He, X. Bacterial abundance and diversity in pond water supplied with different feeds. Sci.Rep. 10-19-2016; 6:35232;PM: 27759010

ABSTRACT: The abundance and diversity of bacteria in two types of ponds were investigated by quantitative PCR and Illumina MiSeq sequencing. The results revealed that the abundance of bacterial 16S rRNA genes in D ponds (with grass carp fed sudan grass) was significantly lower than that in E ponds (with grass carp fed commercial feed). The microbial communities were dominated by Proteobacteria, Cyanobacteria, Bacteroidetes, and Actinobacteria in both E and D ponds, while the abundance of some genera was significantly different between the two types of ponds. Specifically, some potential pathogens such as Acinetobacter and Aeromonas were found to be significantly decreased, while some probiotics such as Comamonadaceae unclassified and Bacillales unclassified were significantly increased in D ponds. In addition, water quality of D ponds was better than that of E ponds. Temperature, dissolved oxygen and nutrients had significant influence on bacterial communities. The differences in bacterial community compositions between the two types of ponds could be partially explained by the different water conditions.

Rachid, R. and Chatila, T. A. The role of the gut microbiota in food allergy. Curr.Opin.Pediatr. 2016; 28(6):748-753; PM: 27749359

ABSTRACT: ***PURPOSE OF REVIEW***: The rise in the prevalence of food allergy over the past decades has focused attention of factors that may impact disease development, most notably the gut microbiota. The gut microbial communities play a crucial role in promoting oral tolerance. Their alteration by such factors as Cesarean section delivery, diet and antibiotics may influence disease development. This review highlights recent progress in our understanding of the role of the gut microbiota in the development of food allergy. ***RECENT FINDINGS***: Food allergy is associated with alterations in the gut microbiota or dysbiosis early in life that may be predictive of disease persistence versus tolerance acquisition. Evidence for the benefits of adjunct therapy with probiotics for the prevention of food allergies and for potentiating oral immunotherapy remains circumstantial, with further studies needed to validate its use. Studies in murine models of food allergy suggest that microbial therapy with protolerogenic bacteria such as certain Clostridial species holds promise in future applications for prevention or therapy of food allergy. ***SUMMARY***: Progress in understanding the role of dysbiosis in food allergy and the factors that promote its development, such as antibiotic therapy, diet, modes of infant delivery, and environmental exposures, offer windows of opportunity for both preventive and therapeutic interventions to stem the rising tide of the food allergy epidemic.

Rad, A. H., Sahhaf, F., Hassanalilou, T., Ejtahed, H. S., Motayagheni, N., Soroush, A. R., Javadi, M., Mortazavian, A. M., and Khalili, L. Diabetes management by probiotics: Current knowledge and future perspectives. Curr.Diabetes Rev. 10-14-2016; PM: 27758705

ABSTRACT: ***BACKGROUND***: Diabetes mellitus, a condition of multifactorial origin, is related to the intestinal microbiota by numerous molecular mechanisms. Controlling the vast increase in the prevalence of diabetes needs a natural and safe solution. Probiotics, known as live microorganisms that exert health benefits to the host, have anti-diabetic property. ***OBJECTIVE***: This review will highlight the current evidences in probiotic effectiveness and future prospects for exploring probiotic therapy in the prevention and control of diabetes. ***METHOD***: We interrogated Pub Med and Science Direct by using "Probiotics" and "Diabetes" for searching the studies aiming the application of probiotics and the beneficial effects of probiotics in diabetes prevention and control. ***RESULTS***: It has been shown that probiotics can increase insulin sensitivity and reduce autoimmune responses by modulating intestinal microbiota and decreasing the inflammatory reactions and oxidative stress. Recent evidences show that probiotics influences the host through modulating intestinal permeability and mucosal immune response, manipulating eating behaviors by appetite-regulating hormones and controlling gut endocannabinoid (eCB) system that is believed to be associated with inflammation and diabetes. Moreover, modulating the intestinal microbiota by probiotics controls host metabolism by affecting energy extraction from food and by biochemically converting molecules derived from the host or from gut microbes themselves. ***CONCLUSION***: Experimental and clinical evidences support the hypothesis that the modulation of the gut microbiota by probiotics could be effective in prevention and management of diabetes.

Raghunathan, V. M., Sheng, I., and Lim, S. H. Intestinal dysbiosis and allogeneic hematopoietic progenitor cell transplantation. J.Transl.Med. 12-3-2016; 14(1):335; PM: 27912759

ABSTRACT: The intestinal microbiota is a diverse and dynamic ecosystem that is increasingly understood to play a vital role in human health. Hematopoietic stem cell transplant recipients undergo prolonged exposure to antimicrobials, chemotherapeutic agents, and immunosuppressants, resulting in profound shifts in the gut microbiome. A growing body of research has revealed the ways in which these microbiologic shifts shape immune modulation, affecting susceptibility to infections and graft-versus-host disease, the two major post-transplant complications in this population. As transplant medicine becomes increasingly personalized, the potential for microbiome-modulating treatments holds immense potential. Strategies to preserve the intestinal microbiota, including targeted antibiotics, prebiotics and probiotics, and fecal microbiota transplant could mitigate some of the microbiologic shifts in stem cell transplant recipients, and reduce the incidence of peri-transplant morbidity and mortality.

Rangan, K. J., Pedicord, V. A., Wang, Y. C., Kim, B., Lu, Y., Shaham, S., Mucida, D., and Hang, H. C. A secreted bacterial peptidoglycan hydrolase enhances tolerance to enteric pathogens. Science. 9-23-2016; 353(6306):1434-1437; PM: 27708039

ABSTRACT: The intestinal microbiome modulates host susceptibility to enteric pathogens, but the specific protective factors and mechanisms of individual bacterial species are not fully characterized. We show that secreted antigen A (SagA) from Enterococcus faecium is sufficient to protect Caenorhabditis elegans against Salmonella pathogenesis by promoting pathogen tolerance. The NlpC/p60 peptidoglycan hydrolase activity of SagA is required and generates muramyl-peptide fragments that are sufficient to protect C. elegans against Salmonella pathogenesis in a tol-1-dependent manner. SagA can also be heterologously expressed and secreted to improve the protective activity of

probiotics against Salmonella pathogenesis in C. elegans and mice. Our study highlights how protective intestinal bacteria can modify microbial-associated molecular patterns to enhance pathogen tolerance.

Ribeiro, F. C., de Barros, P. P., Rossoni, R. D., Junqueira, J. C., and Jorge, A. O. Lactobacillus rhamnosus inhibits Candida albicans virulence factors in vitro and modulates immune system in Galleria mellonella. J.Appl.Microbiol. 10-11-2016; PM: 27727499

ABSTRACT: ***AIM***: The aim of this study was to evaluate the potential anti-Candida effects of Lactobacillus rhamnosus ATCC 9595 on Candida albicans ATCC 18804 using in vitro and in vivo models. ***METHODS AND RESULTS***: The in vitro analysis evaluated the effects of L. rhamnosus on C. albicans's biofilm formation by CFU count and metabolic activity, filamentation capacity, and adhesion (ALS3 and HWP1) and transcriptional regulatory gene (BCR1 and CPH1) expression. The in vitro results showed that both the L. rhamnosus cells and supernatant reduced C. albicans biofilm formation, filamentation and gene expression. In the in vivo study, the treatment with L. rhamnosus supernatant increased 80% the survival of Galleria mellonella larvae infected with C. albicans. Furthermore, the supernatant of L. rhamnosus recruited haemocytes into the haemolymph (2.1-fold increase). ***CONCLUSIONS***: Lactobacillus rhamnosus reduced the biofilm formation and filamentation of C. albicans in vitro by negatively regulating all studied C. albicans genes. Lactobacillus rhamnosus protected G. mellonella against experimental candidiasis in vivo. ***SIGNIFICANCE AND IMPACT OF THE STUDY***: This study is the first study to report the anti-Candida properties of L. rhamnosus ATCC 9595. The supernatant of this strain has immunomodulatory effects on the G. mellonella model and protects the larvae against pathogens.

Rios, A. C., Moutinho, C. G., Pinto, F. C., Del Fiol, F. S., Jozala, A., Chaud, M. V., Vila, M. M., Teixeira, J. A., and Balcao, V. M. Alternatives to overcoming bacterial resistances: State-of-the-art. Microbiol.Res. 2016; 191:51-80; PM: 27524653

ABSTRACT: Worldwide, bacterial resistance to chemical antibiotics has reached such a high level that endangers public health. Presently, the adoption of alternative strategies that promote the elimination of resistant microbial strains from the environment is of utmost importance. This review discusses and analyses several (potential) alternative strategies to current chemical antibiotics. Bacteriophage (or phage) therapy, although not new, makes use of strictly lytic phage particles as an alternative, or a complement, in the antimicrobial treatment of bacterial infections. It is being rediscovered as a safe method, because these biological entities devoid of any metabolic machinery do not possess any affinity whatsoever to eukaryotic cells. Lysin therapy is also recognized as an innovative antimicrobial therapeutic option, since the topical administration of preparations containing purified recombinant lysins with amounts in the order of nanograms, in infections caused by Gram-positive bacteria, demonstrated a high therapeutic potential by causing immediate lysis of the target bacterial cells. Additionally, this therapy exhibits the potential to act synergistically when combined with certain chemical antibiotics already available on the market. Another potential alternative antimicrobial therapy is based on the use of antimicrobial peptides (AMPs), amphiphilic polypeptides that cause disruption of the bacterial membrane and can be used in the treatment of bacterial, fungal and viral infections, in the prevention of biofilm formation, and as antitumoral agents. Interestingly, bacteriocins are a common strategy of bacterial defense against other bacterial agents, eliminating the potential opponents

of the former and increasing the number of available nutrients in the environment for their own growth. They can be applied in the food industry as biopreservatives and as probiotics, and also in fighting multi-resistant bacterial strains. The use of antibacterial antibodies promises to be extremely safe and effective. Additionally, vaccination emerges as one of the most promising preventive strategies. All these will be tackled in detail in this review paper.

Ritzi, M. M., Abdelrahman, W., van-Heerden, K., Mohnl, M., Barrett, N. W., and Dalloul, R. A. Combination of probiotics and coccidiosis vaccine enhances protection against an Eimeria challenge. Vet.Res. 11-8-2016; 47(1):111; PM: 27825377

ABSTRACT: Coccidiosis is endemic in the commercial broiler industry capable of inflicting devastating economic losses to poultry operations. Vaccines are relatively effective in controlling the disease; their efficacy could potentially be improved with concurrent use of probiotics as evaluated in this study using an Eimeria challenge. Day of hatch 400 Cobb-500 male broilers were assigned to one of four treatment groups including control (CON), vaccine-only gel application (VNC), probiotic-only gel application (NPC), and vaccine-plus-probiotic gel application (VPC). Birds were placed in floor pens (6 replicate pens/treatment, 16-17 birds/pen). NPC and VPC birds received the probiotics in the water on days 2-4, 8, 14-20, 22, 29, and 34-36. On day 15, birds were mildly challenged with 0.5 mL of a mixed oral inoculum of Eimeria sp. prepared with the coccidiosis vaccine at 10x the vaccination dose. Performance measurements were recorded on first day and weekly afterwards, and lesion scores were evaluated 6 days post-challenge. Overall, the probiotics and coccidiosis vaccine resulted in an enhanced protective effect against the challenge, with VPC birds exhibiting lower lesion scores in the duodenum than VNC or NPC birds. Birds in the VPC treatment also demonstrated higher weight gains during days 1-15, days 7-15, and days 21-28 when compared to the VNC birds. These results suggest that the combination of probiotics and coccidiosis vaccines could enhance performance and provide an additional protective effect against a mixed Eimeria challenge.

Roberts, J. D., Suckling, C. A., Peedle, G. Y., Murphy, J. A., Dawkins, T. G., and Roberts, M. G. An Exploratory Investigation of Endotoxin Levels in Novice Long Distance Triathletes, and the Effects of a Multi-Strain Probiotic/Prebiotic, Antioxidant Intervention. Nutrients. 11-17-2016; 8(11); PM: 27869661

ABSTRACT: Gastrointestinal (GI) ischemia during exercise is associated with luminal permeability and increased systemic lipopolysaccharides (LPS). This study aimed to assess the impact of a multistrain pro/prebiotic/antioxidant intervention on endotoxin unit levels and GI permeability in recreational athletes. Thirty healthy participants (25 males, 5 females) were randomly assigned either a multistrain pro/prebiotic/antioxidant (LAB(4)ANTI; 30 billion CFU.day-1 containing 10 billion CFU.day-1Lactobacillus acidophilus CUL-60 (NCIMB 30157), 10 billion CFU.day-1Lactobacillus acidophillus CUL-21 (NCIMB 30156), 9.5 billion CFU.day-1Bifidobacterium bifidum CUL-20 (NCIMB 30172) and 0.5 billion CFU.day-1Bifidobacterium animalis subspecies lactis CUL-34 (NCIMB 30153)/55.8 mg.day-1 fructooligosaccharides/ 400 mg.day-1 alpha-lipoic acid, 600 mg.day-1N-acetyl-carnitine); matched pro/prebiotic (LAB(4)) or placebo (PL) for 12 weeks preceding a long-distance triathlon. Plasma endotoxin units (via Limulus amebocyte lysate chromogenic quantification) and GI permeability (via 5 h urinary lactulose (L): mannitol (M) recovery) were assessed at baseline, pre-race and six days post-race. Endotoxin unit

levels were not significantly different between groups at baseline (LAB(4)ANTI: 8.20 +/- 1.60 pg.mL-1; LAB(4): 8.92 +/- 1.20 pg.mL-1; PL: 9.72 +/- 2.42 pg.mL-1). The use of a 12-week LAB(4)ANTI intervention significantly reduced endotoxin units both pre-race (4.37 +/- 0.51 pg.mL-1) and six days post-race (5.18 +/- 0.57 pg.mL-1; p = 0.03, etap(2) = 0.35), but only six days post-race with LAB(4) (5.01 +/- 0.28 pg.mL-1; p = 0.01, etap(2) = 0.43). In contrast, endotoxin units remained unchanged with PL. L:M significantly increased from 0.01 +/- 0.01 at baseline to 0.06 +/- 0.01 with PL only (p = 0.004, etap(2) = 0.51). Mean race times (h:min:s) were not statistically different between groups despite faster times with both pro/prebiotoic groups (LAB(4)ANTI: 13:17:07 +/- 0:34:48; LAB(4): 12:47:13 +/- 0:25:06; PL: 14:12:51 +/- 0:29:54; p > 0.05). Combined multistrain pro/prebiotic use may reduce endotoxin unit levels, with LAB(4)ANTI potentially conferring an additive effect via combined GI modulation and antioxidant protection.

Rosenberg, H. F., Masterson, J. C., and Furuta, G. T. Eosinophils, probiotics, and the microbiome. J.Leukoc.Biol. 2016; 100(5):881-888; PM: 27549754

ABSTRACT: There is currently substantial interest in the therapeutic properties of probiotic microorganisms as recent research suggests that oral administration of specific bacterial strains may reduce inflammation and alter the nature of endogenous microflora in the gastrointestinal tract. Eosinophils are multifunctional tissue leukocytes, prominent among the resident cells of the gastrointestinal mucosa that promote local immunity. Recent studies with genetically altered mice indicate that eosinophils not only participate in maintaining gut homeostasis, but that the absence of eosinophils may have significant impact on the nature of the endogenous gut microflora and responses to gut pathogens, notably Clostridium difficile Furthermore, in human subjects, there is an intriguing relationship between eosinophils, allergic inflammation, and the nature of the lung microflora, notably a distinct association between eosinophil infiltration and detection of bacteria of the phylum Actinobacteria. Among topics for future research, it will be important to determine whether homeostatic mechanisms involve direct interactions between eosinophils and bacteria or whether they involve primarily eosinophil-mediated responses to cytokine signaling in the local microenvironment. Likewise, although is it clear that eosinophils can and do interact with bacteria in vivo, their ability to discern between pathogenic and probiotic species in various settings remains to be explored.

Rosenfeld, C. S. Homage to the 'H' in developmental origins of health and disease. J.Dev.Orig.Health Dis. 8-31-2016; 1-22; PM: 27577791

ABSTRACT: Abundant evidence exists linking maternal and paternal environments from pericopconception through the postnatal period to later risk to offspring diseases. This concept was first articulated by the late Sir David Barker and as such coined the Barker Hypothesis. The term was then mutated to Fetal Origins of Adult Disease and finally broadened to developmental origins of adult health and disease (DOHaD) in recognition that the perinatal environment can shape both health and disease in resulting offspring. Developmental exposure to various factors, including stress, obesity, caloric-rich diets and environmental chemicals can lead to detrimental offspring health outcomes. However, less attention has been paid to date on measures that parents can take to promote the long-term health of their offspring. In essence, have we neglected to consider the 'H' in DOHaD? It is the 'H' component that should be of primary concern to expecting mothers and fathers and those seeking to have children. While it may not be

possible to eliminate exposure to all pernicious factors, prevention/remediation strategies may tip the scale to health rather than disease. By understanding disruptive DOHaD mechanisms, it may also illuminate behavioral modifications that parents can adapt before fertilization and throughout the neonatal period to promote the lifelong health of their male and female offspring. Three possibilities will be explored in the current review: parental exercise, probiotic supplementation and breastfeeding in the case of mothers. The 'H' paradigm should be the focus going forward as a healthy start can indeed last a lifetime.

Rossi, R. E., Whyand, T., Murray, C. D., Hamilton, M. I., Conte, D., and Caplin, M. E. The role of dietary supplements in inflammatory bowel disease: a systematic review. Eur.J.Gastroenterol.Hepatol. 2016; 28(12):1357-1364; PM: 27769076

ABSTRACT: Inflammatory bowel diseases (IBD) are chronic immune disorders of unclear aetiology. Dietary deficiencies may be a potential pathogenic factor in their development. Patients often take food supplements without knowledge of any evidence base. We have therefore assessed the evidence for food supplementation in the management of IBD. A PubMed search was performed for the terms Inflammatory bowel disease; nutritional deficiencies; dietary supplements; curcumin; green tea; vitamin D/other vitamins; folic acid; iron; zinc; probiotics; andrographis paniculata; and boswellia serrate. PubMed was used to search for all relevant articles published between January 1975 and September 2015. Curcumin supplementation has been reported to be effective in reducing the symptoms and the inflammatory indices in IBD patients. Similar results have been observed for green tea; however, pertinent studies are limited. Vitamin D supplementation may help to increase bone mineral density in IBD patients and to reduce disease activity. IBD patients with ileal resections higher than 20 cm may develop vitamin B12 deficiency that requires parenteral supplementation. There is no current evidence to support fat-soluble vitamin supplementation in IBD patients. Zinc and iron should be supplemented in selected cases. Probiotics (VSL#3) may reduce disease activity in IBD patients with pouchitis. Complementary and alternative medicines are used by IBD patients and some studies have shown promising results. In summary, attention to dietary factors such as curcumin, green tea and vitamins, including vitamins D and B12, appears to be beneficial and, if necessary, supplementation may be appropriate.

Rutella, G. S., Tagliazucchi, D., and Solieri, L. Survival and bioactivities of selected probiotic lactobacilli in yogurt fermentation and cold storage: New insights for developing a bi-functional dairy food. Food Microbiol. 2016; 60:54-61; PM: 27554146

ABSTRACT: In previous work, we demonstrated that two probiotic strains, namely Lactobacillus casei PRA205 and Lactobacillus rhamnosus PRA331, produce fermented milks with potent angiotensin-converting enzyme (ACE)-inhibitory and antioxidant activities. Here, we tested these strains for the survivability and the release of antihypertensive and antioxidant peptides in yogurt fermentation and cold storage. For these purposes three yogurt batches were compared: one prepared using yogurt starters alone (Lactobacillus delbrueckii subspecies bulgaricus 1932 and Streptococcus thermophilus 99), and the remaining two containing either PRA205 or PRA331 in addition to yogurt starters. Despite the lower viable counts at the fermentation end compared to PRA331, PRA205 overcame PRA331 in survivability during refrigerated storage for 28 days, leading to viable counts (>10(8) CFU/g) higher than the minimum therapeutic threshold

(10(6) CFU/g). Analyses of in vitro ACE-inhibitory and antioxidant activities of peptide fractions revealed that yogurt supplemented with PRA205 displays higher amounts of antihypertensive and antioxidant peptides than that produced with PRA331 at the end of fermentation and over storage. Two ACE-inhibitory peptides, Valine-Proline-Proline (VPP) and Isoleucine-Proline-Proline (IPP), were identified and quantified. This study demonstrated that L. casei PRA205 could be used as adjunct culture for producing bi-functional yogurt enriched in bioactive peptides and in viable cells, which bring health benefits to the host as probiotics.

Saber, A., Alipour, B., Faghfoori, Z., and Yari, Khosroushahi A. Cellular and molecular effects of yeast probiotics on cancer. Crit Rev.Microbiol. 8-25-2016; 1-20; PM: 27561003

ABSTRACT: The cancer is one of the main causes of human deaths worldwide. The exact mechanisms of initiation and progression of malignancies are not clear yet, but there is a common agreement about the role of colonic microbiota in the etiology of different cancers. Probiotics have been examined for their anti-cancer effects, and different mechanisms have been suggested about their antitumor functions. Nonpathogenic yeasts, as members of probiotics family, can be effective on gut microbiota dysbiosis. Generally safe yeasts have shown so many beneficial effects on human health. Probiotic yeasts influence physiology, metabolism, and immune homeostasis in the colon and contribute to cancer treatment due to possessing anti-inflammatory, anti-proliferative and anti-cancer properties. This study reviews some of the health-beneficial effects of probiotic yeasts and their biological substances like folic acid and beta-glucan on cancer and focuses on the possible cellular and molecular mechanisms of probiotic yeasts such as influencing pathogenic bacteria, inactivation of carcinogenic compounds, especially those derived from food, improvement of intestinal barrier function, modulation of immune responses, antitoxic function, apoptosis, and anti-proliferative effects.

Sadeghzadeh, J., Vakili, A., Sameni, H. R., Shadnoush, M., Bandegi, A. R., and Zahedi, Khorasani M. The Effect of Oral Consumption of Probiotics in Prevention of Heart Injury in a Rat Myocardial Infarction Model: a Histopathological, Hemodynamic and Biochemical Evaluation. Iran Biomed.J. 11-22-2016; PM: 27874107

ABSTRACT: ***BACKGROUND***: Despite the emerging evidence on beneficial effects of probiotics on the cardiovascular system, their impact on the management of ischemic heart diseases and its possible mechanism have not been elucidated. ***METHODS***: Four viable probiotics bacterial strains, including Bifidobacterium breve, Lactobacillus casei, Lactobacillus bulgaricus and Lactobacillus acidophilus, at the concentrations of 2x106 colony-forming units/ml were orally administered to the rats daily for 14 days before the induction of infarct-like myocardial injury using isoproterenol. Subsequently, 24 h after myocardial injury, the right carotid artery and the left ventricle were catheterized for recording blood pressure and cardiac parameters. At the end of the experiment, the heart was removed for the evaluation of histopathological and biochemical parameters, as well as tumor necrosis factor-alpha (TNF-alpha) assay. ***RESULTS***: The induction of acute myocardial injury resulted in significant (P</=0.01) left ventricular (LV) dysfunction, as shown by an increase in LV end-diastolic pressure and a decrease in LV dp/dt max, LV dp/dt min, LV systolic pressure, and blood pressure, as compared with normal rats. Pretreatment with viable probiotics significantly reduced lipid peroxidation and TNF-alpha level and improved cardiac function (P<0.01). ***CONCLUSION***: This study shows that

viable probiotics have a cardioprotective effect on infarct-like myocardial injury through suppressing TNF-alpha and oxidative stress damage in a rat model. Probiotic supplements may be used as a new option for prophylaxis in patients at the risk of ischemic heart disease in future.

Sah, B. N., Vasiljevic, T., McKechnie, S., and Donkor, O. N. Effect of pineapple waste powder on probiotic growth, antioxidant and antimutagenic activities of yogurt. J.Food Sci.Technol. 2016; 53(3):1698-1708; PM: 27570295

ABSTRACT: Although many fruit by-products are good sources of nutrients, little is known about their prebiotic potential. This research was aimed at establishing the prebiotic effect of pineapple wastes on probiotics including Lactobacillus (L.) acidophilus (ATCC(R) 4356), L. casei (ATCC(R) 393) and L. paracasei spp. paracasei (ATCC(R) BAA52) and the subsequent release of antioxidant and antimutagenic peptides in yogurt during their growth. Oven- and freeze- dried peel and pomace were milled separately into powders and tested for prebiotic activities. The net probiotic growth (1.28-2.14 log cfu/g) in customized MRS broth containing the pineapple powders as a direct carbohydrate source was comparable to MRS broth containing glucose. The powders were also separately added to milk during the manufacturing of yogurt with or without probiotics. An increase (by 0.3-1.4 log cycle) in probiotic populations was observed in the yogurts as a consequence of pineapple powder supplementation. Crude water-soluble peptide extracts, prepared by high-speed centrifugation of the yogurts, displayed remarkable antioxidant activities assessed through in vitro assays, namely scavenging activity of 1,1-diphenyl-2-picrylhydrazyl radicals (IC50 = 0.37-0.19 mg/ml) and hydroxyl radicals (58.52-73.55 %). The peptide extracts also exhibited antimutagenic activities (18.60-32.72 %) as sodium azide inhibitor in the Salmonella mutagenicity test. Together, these results suggest that pineapple by-products exhibited prebiotic properties and could possibly be commercially applied in new functional food formulations.

Saha, C., Mukherjee, G., Agarwal, Banka P., and Seal, A. A consortium of non-rhizobial endophytic microbes from Typha angustifolia functions as probiotics in rice and improves nitrogen metabolism. Plant Biol.(Stuttg). 7-25-2016; PM: 27453497

ABSTRACT: 1.Endophytic microbes isolated from plants growing in nutrient poor environments often possess properties that improve nutrition when applied to agriculturally important plants. 2.Ten endophytic bacteria isolated from Typha angustifolia growing in the marginal wetlands associated with a Uranium mine in India were characterized for their plant growth promoting mechanisms and were found to have biological nitrogen (N) fixing properties. These were used as a consortium for their ability to function as bio-fertilizer and their effect on N-metabolism in rice was tested. 3.Evidence of recent horizontal gene transfer events of dinitrogen reductase gene from a Pseudomonas stutzeri strain within the consortium was found among these microbes. The consortium behaved as plant probiotic and showed substantial growth benefits to Typha, their natural host as well as to rice. Plant biomass, shoot length and chlorophyll content were increased in rice plants in response to microbe treatment under N-sufficient as well as deficient conditions. At least some of these microbes inhabited the rice endosphere indicating their broad host range. 4.Up-regulation of nitrate and ammonium uptake genes together with the N-assimilation genes, increased N-content and relative nitrogen use efficiency suggested that the endophytic consortium triggered

a long term nitrogen deficiency response in the treated plants improving both uptake and assimilation of nitrogen. This article is protected by copyright. All rights reserved.

Salas-Jara, M. J., Ilabaca, A., Vega, M., and Garcia, A. Biofilm Forming Lactobacillus: New Challenges for the Development of Probiotics. Microorganisms. 9-20-2016; 4(3); PM: 27681929

ABSTRACT: Probiotics are live bacteria, generally administered in food, conferring beneficial effects to the host because they help to prevent or treat diseases, the majority of which are gastrointestinal. Numerous investigations have verified the beneficial effect of probiotic strains in biofilm form, including increased resistance to temperature, gastric pH and mechanical forces to that of their planktonic counterparts. In addition, the development of new encapsulation technologies, which have exploited the properties of biofilms in the creation of double coated capsules, has given origin to fourth generation probiotics. Up to now, reviews have focused on the detrimental effects of biofilms associated with pathogenic bacteria. Therefore, this work aims to amalgamate information describing the biofilms of Lactobacillus strains which are used as probiotics, particularly L. rhamnosus, L. plantarum, L. reuteri, and L. fermentum. Additionally, we have reviewed the development of probiotics using technology inspired by biofilms.

Salazar, N., Valdes-Varela, L., Gonzalez, S., Gueimonde, M., and de Los Reyes-Gavilan CG. Nutrition and the gut microbiome in the elderly. Gut Microbes. 11-3-2016; 1-16; PM: 27808595

ABSTRACT: The gut microbiota is the assembly of microorganisms living in our intestine and their genomes are known as the microbiome. The correct composition and functionality of this microbiome is essential for maintaining a "healthy status." Aging is related to changes in the gut microbiota which are frequently associated with physiological modifications of the gastrointestinal tract, as well as, to changes in dietary patterns, together with a concomitant decline in cognitive and immune function, all together contributing to frailty. Therefore, nutritional strategies directed at restoring the microbiota in the elderly have to be addressed from a global perspective, considering not only the microbiota but also other extra-intestinal targets of action. The present review aims at summarizing the current knowledge on intestinal microbiota alterations and other functions impaired in the elderly and to analyze tools for implementing nutritional strategies, through the use of probiotics, prebiotics or specific nutrients in order to counterbalance such alterations.

Samuels, N., van de, Graaf R., Been, J. V., de Jonge, R. C., Hanff, L. M., Wijnen, R. M., Kornelisse, R. F., Reiss, I. K., and Vermeulen, M. J. Necrotising enterocolitis and mortality in preterm infants after introduction of probiotics: a quasi-experimental study. Sci.Rep. 8-22-2016; 6:31643; PM: 27545195

ABSTRACT: Evidence on the clinical effectiveness of probiotics in the prevention of necrotising enterocolitis (NEC) in preterm infants is conflicting and cohort studies lacked adjustment for time trend and feeding type. This study investigated the association between the introduction of routine probiotics (Lactobacillus acidophilus and Bifidobacterium bifidum; Infloran((R))) on the primary outcome 'NEC or death'. Preterm infants (gestational age <32 weeks or birth weight <1500 gram) admitted before (Jan 2008-Sep 2012; n = 1288) and after (Oct 2012-Dec 2014; n = 673) introduction of

probiotics were compared. Interrupted time series logistic regression models were adjusted for confounders, effect modification by feeding type, seasonality and underlying temporal trends. Unadjusted and adjusted analyses showed no difference in 'NEC or death' between the two periods. The overall incidence of NEC declined from 7.8% to 5.1% (OR 0.63, 95% CI 0.42-0.93, p = 0.02), which was not statistically significant in the adjusted models. Introduction of probiotics was associated with a reduced adjusted odds for 'NEC or sepsis or death' in exclusively breastmilk-fed infants (OR 0.43, 95% CI 0.21-0.93, p = 0.03) only. We conclude that introduction of probiotics was not associated with a reduction in 'NEC or death' and that type of feeding seems to modify the effects of probiotics.

Sanadgol, N., Zahedani, S. S., Sharifzadeh, M., Khalseh, R., Barbari, G. R., and Abdollahi, M. Recent updates in imperative natural compounds for healthy brain and nerve function: A systematic review of implications for multiple sclerosis. Curr.Drug Targets. 11-8-2016; PM: 27829351

ABSTRACT: ***BACKGROUND***: The evolution of the 'Green' movement in western society has changed attitudes in the general population who now perceive natural compounds as being inherently harmless and more desirable than artificial chemical products. ***OBJECTIVE***: Considering the growing interest towards introducing naturally emerged medicines, the purpose of this review was to overview the ongoing research into prevention and treatment of multiple sclerosis (MS) lesions. ***METHOD***: This review was carried out by searching bibliographic databases such as PubMed and Scopus for studies reported between 1th January 2008 to 30rd January 2016 on MS patients or animal models of MS, investigating the beneficial effects of natural compounds in MS treatment. In this updated systematic review, the search terms were "multiple sclerosis" or "neurodegeneration" and ("natural compounds" or "medicinal plants", "traditional medicine" or "native medicine"). ***RESULTS***: Studies with vitamins (A, B12, D, H), minerals (selenium and lithium), n-3 PUFAs, lipoic acid, statins, resveratrol, marijuana, EGCG and some probiotics have shown significant helpful effects in MS by preventing or delaying the onset of disease. Other natural compounds such as xanthines, anthocyanins, glucosinolates, isoflavones, organosulfurs, steroid glycosides, and alkaloids have also shown protective effects in the treatment of MS in animal models. Adverse effects were also reported in some of the experiments. ***CONCLUSION***: Further studies with a focus on the molecular mechanisms of the protective natural compounds are needed to decrease possible side effects and to develop new medicines for MS. Apigenin, chrysin, baicalein, cyanidin, flavone glycoside, daidzein, coumestrol, sulforaphane, bee venom and huperzine A are the candidates for more prospective investigations.

Sanchez-Ortiz, A. C., Angulo, C., Luna-Gonzalez, A., varez-Ruiz, P., Mazon-Suastegui, J. M., and Campa-Cordova, A. I. Effect of mixed-Bacillus spp isolated from pustulose ark Anadara tuberculosa on growth, survival, viral prevalence and immune-related gene expression in shrimp Litopenaeus vannamei. Fish.Shellfish.Immunol. 2016; 59:95-102; PM: 27744059

ABSTRACT: The widespread overuse of antibiotics in aquaculture has led to the emergence of antibiotic-resistance shrimp pathogens, the negative impact on shrimp gut microbiota, and the presence of antimicrobial residues in aquaculture products, with negative consequences on human health. Alternatively, probiotics have positive effects on immunological responses and productive performance of aquatic animals. In this

study, three probiotic bacteria, (Bacillus licheniformis MAt32, B. subtilis MAt43 and B. subtilis subsp. subtilis GAtB1), isolated from the Anadara tuberculosa were included in diets for juvenile shrimp, Litopenaeus vannamei, to evaluate their effects on growth, survival, disease prevalence, and immune-related gene expression. Shrimp naturally infected with WSSV and IHHNV were fed with the basal diet (control, T1) and diets supplemented with four levels of bacilli probiotic mix (1:1:1) at final concentration of (T2) 1 x 106, (T3) 2 x 106, (T4) 4 x 106, and (T5) 6 x 106 CFU g-1 of feed. The specific growth rate of shrimp was significantly higher in T2 than in T1 (control) treatment, and the final growth as well as the survival were similar among treated groups. The prevalence of WSSV and IHHNV infected shrimp was reduced in T2 and T4 treatments, respectively, compared with control. The mRNA expression of proPO gene was higher in treatment T4 than control. The LvToll1 gene was significantly up-regulated in treatments T4 and T5 compared to control. The SOD gene was up-regulated in treatment T5 compared to control. In contrast, the mRNA expression of the Hsp70 gene was down-regulated in treatments T4 and T5 respect to control, and the TGase gene remained unaffected by the level of bacillus probiotic mix. As conclusion, the bacilli probiotic mix (Bacillus spp.) enhanced immune-related gene expression in WSSV and IHHNV naturally infected shrimp. This is the first report of probiotic potential of bacteria isolated from A. tuberculosa on the immune response and viral prevalence in shrimp Litopenaeus vannamei.

Sanders, K., Moran, Z., Shi, Z., Paul, R., and Greenlee, H. Natural Products for Cancer Prevention: Clinical Update 2016. Semin.Oncol.Nurs. 2016; 32(3):215-240; PM: 27539278

ABSTRACT: ***OBJECTIVES***: To present a clinical update of natural products for cancer prevention and provide oncology nurses with an evidence-based review of natural products for patient counseling and education. ***DATA SOURCES***: Clinical trials published in PubMed. ***CONCLUSION***: In the past 4 years since the publication of the original review there have been minimal changes in the conclusions of the published literature on the use of natural products for cancer prevention. To date, clinical trials have not demonstrated conclusive benefit of using natural products for cancer prevention, and current guidelines do not recommend their use. This review provides an update on published and ongoing trials and can serve as an updated resource for nurses. Evidence-based natural products databases can help nurses stay current with the scientific literature and be effective educators and health coaches for their patients, who can be influenced by marketing of unregulated products. ***IMPLICATIONS FOR NURSING PRACTICE***: Patients often discuss the use of natural products with nurses. Nurses have an opportunity to educate and coach patients in effective preventive lifestyle practices.

Sanders, M. E., Merenstein, D. J., Ouwehand, A. C., Reid, G., Salminen, S., Cabana, M. D., Paraskevakos, G., and Leyer, G. Probiotic use in at-risk populations. J.Am.Pharm.Assoc.(2003.). 2016; 56(6):680-686; PM: 27836128

ABSTRACT: ***OBJECTIVE***: To inform health care providers about quality standards for manufacture of probiotic products being recommended for at-risk patient populations. ***SUMMARY***: Probiotics are used in a variety of clinical settings, sometimes in at-risk populations for therapeutic endpoints. Although probiotics might not be approved as drugs, they are sometimes used for the prevention or treatment of disease. In the United States, and many regions of the world, probiotic products are marketed as dietary supplements (not drugs) and are therefore subject to different manufacturing and quality control standards than approved drugs are. Health care providers need to be

assured that probiotic products used in at-risk populations are safe for this use. Pharmacists should require certificates of analysis, which document quality standards, from manufacturers of products stocked in hospital formularies or other pharmacies dispensing to at-risk people. Although responsible manufacturers use stringent quality standards on their processes and finished products, using a third party to verify compliance with manufacturing and accuracy of product labeling adds assurance to end users that the product is of high quality. ***CONCLUSION***: It is in patients' best interest to use probiotics in the prevention and treatment of conditions when the evidence is convincing. To protect high-risk patients, probiotic products should meet stringent microbiological standards. Product testing results should be available for review before recommending probiotic products to at-risk individuals. For products used in at-risk populations, manufacturers should provide this information or participate in a third-party verification program that certifies compliance.

Santisteban, M. M., Qi, Y., Zubcevic, J., Kim, S., Yang, T., Shenoy, V., Cole-Jeffrey, C. T., Lobaton, G. O., Stewart, D. C., Rubiano, A., Simmons, C. S., Garcia-Pereira, F., Johnson, R. D., Pepine, C. J., and Raizada, M. K. Hypertension-Linked Pathophysiological Alterations in the Gut. Circ.Res. 10-31-2016; PM: 27799253

ABSTRACT: ***RATIONALE***: Sympathetic nervous system control of inflammation plays a central role in hypertension. The gut receives significant sympathetic innervation, is densely populated with a diverse microbial ecosystem, and contains immune cells that greatly impact overall inflammatory homeostasis. Despite this uniqueness, little is known about the involvement of the gut in hypertension. ***OBJECTIVE***: Test the hypothesis that increased sympathetic drive to the gut is associated with increased gut wall permeability, increased inflammatory status, and microbial dysbiosis and that these gut pathological changes are linked to hypertension. ***METHODS AND RESULTS***: Gut epithelial integrity and wall pathology were examined in spontaneously hypertensive rat (SHR) and chronic Angiotensin II infusion rat models. The increase in blood pressure in SHR was associated with gut pathology that included increased intestinal permeability and decreased tight junction proteins. These changes in gut pathology in hypertension were associated with alterations in microbial communities relevant in blood pressure control. We also observed enhanced gut-neuronal communication in hypertension originating from paraventricular nucleus of the hypothalamus and presenting as increased sympathetic drive to the gut. Finally, angiotensin converting enzyme inhibition (captopril) normalized blood pressure and was associated with reversal of gut pathology. ***CONCLUSIONS***: A dysfunctional sympathetic-gut communication is associated with gut pathology, dysbiosis, and inflammation, and plays a key role in hypertension. Thus, targeting of gut microbiota by innovative probiotics, antibiotics, and fecal transplant, in combination with current pharmacotherapy, may be a novel strategy for hypertension treatment.

Sarjapuram, N., Mekala, N., Singh, M., and Tatu, U. The Potential of Lactobacillus casei and Entercoccus faecium Combination as a Preventive Probiotic Against Entamoeba. Probiotics.Antimicrob.Proteins. 10-5-2016; PM: 27709564

ABSTRACT: Travellers' diarrhoea caused by enteric protozoa like Entamoeba histolytica is among the most common protozoan diseases in developing countries. In developing countries, amoebiasis is the second most prevalent protozoan disease. This protozoan parasite is often known to coexist as a part of the normal gut microbiota. It is estimated

that around 50-60 % of population in developing countries might be harbouring Entamoeba in an asymptomatic manner. Due to physiological perturbation or upon immuno-compromise, it can become virulent and then cause diarrhoea, bloody stools and may invade other organs if left untreated. Nitroimidazole drugs, namely metronidazole and tinidazole, are widely used to treat protozoan infections. These drugs often show dose-dependent side effects. With emerging antibiotic resistance, novel therapeutics to prevent parasitic infections is required. This study aims to study effect of probiotics on prevention of Amoebiasis. In this study, we have investigated the effect of selected probiotics on the growth of Entamoeba. From the list of probiotics being currently used, five bacterial strains were selected for testing. These probiotic strains were co-cultured with Entamoeba, and their effect on Entamoeba proliferation was checked. Of the five probiotics chosen, individual treatments of Lactobacillus casei and Enterococcus faecium showed a significant reduction of up to 71 % in parasite survival only at higher CFUs. When the two probiotics were used in combination, the percentage of survival reduced gradually further to 80 % at a total CFU of 109 cells/ml of bacteria. The study lays the foundation for providing cost-effective prophylactic treatment for amoebiasis without the overuse of antibiotics.

Sarkar, A. and Mandal, S. Bifidobacteria-Insight into clinical outcomes and mechanisms of its probiotic action. Microbiol.Res. 2016; 192:159-171; PM: 27664734

ABSTRACT: The invasion of pathogens causes a disruption of the gut homeostasis. Innate immune responses and those triggered by endogenous microbiota form the first line of defence in our body. Pathogens often successfully overcome the resistances offered, calling for therapeutic intervention. Conventional strategy involving antibiotics might eradicate pathogens, but often leave the gut uncolonised and susceptible to recurrences. Probiotic supplements are useful alternatives. Bifidobacterium is one of widely studied probiotic genus, effective in restoring gut homeostasis. Mechanisms of probiotic action of bifidobacteria are several, often with strain-specificity. Analysis of streamlined literature reports reveal that although most studies report the probiotic aspect of bifidobacteria, sporadic documented contradictory results exist, challenging its therapeutic application and prompting studies to unambiguously establish the strain-associated probiotic activity and negate adverse effects prior to its clinical administration. Multi-strain/combinatorial therapy possibly relies on a combination of underlying operating mechanisms, each contributing towards enhanced probiotic efficacy, understanding which could help in developing customised formulations against targeted pathogens. Bifidogenic activity is also mediated by surface-associated structural components such as exopolysaccharides, lipoteichoic acids along with metabolites and bifidocins. This highlights scope for developing advanced structural therapeutic strategy which might be pivotal in replacing intact cell probiotics therapy.

Sarkar, A., Lehto, S. M., Harty, S., Dinan, T. G., Cryan, J. F., and Burnet, P. W. Psychobiotics and the Manipulation of Bacteria-Gut-Brain Signals. Trends Neurosci. 2016; 39(11):763-781; PM: 27793434

ABSTRACT: Psychobiotics were previously defined as live bacteria (probiotics) which, when ingested, confer mental health benefits through interactions with commensal gut bacteria. We expand this definition to encompass prebiotics, which enhance the growth

of beneficial gut bacteria. We review probiotic and prebiotic effects on emotional, cognitive, systemic, and neural variables relevant to health and disease. We discuss gut-brain signalling mechanisms enabling psychobiotic effects, such as metabolite production. Overall, knowledge of how the microbiome responds to exogenous influence remains limited. We tabulate several important research questions and issues, exploration of which will generate both mechanistic insights and facilitate future psychobiotic development. We suggest the definition of psychobiotics be expanded beyond probiotics and prebiotics to include other means of influencing the microbiome.

Satish, L., Gallo, P. H., Johnson, S., Yates, C. C., and Kathju, S. Local Probiotic Therapy with Lactobacillus plantarum Mitigates Scar Formation in Rabbits after Burn Injury and Infection. Surg.Infect.(Larchmt.). 10-27-2016; PM: 27788042

ABSTRACT: ***BACKGROUND***: Infection is the most common complication in burn-injured patients and is believed to contribute to the hypertrophic scarring frequently observed in such injury. Pseudomonas aeruginosa is a common pathogen in burn wound infection. We examined the effect of local probiotic therapy with Lactobacillus plantarum on the severity of the scarring following burn wounding and infection with P. aeruginosa in a rabbit model. ***METHODS***: Full-thickness burn wounds were inoculated with control vehicle or L. plantarum; wounds were then challenged with bioluminescent P. aeruginosa. The time course of the ensuing infection was monitored by quantification of the emitted light. After allowing wounds to contract to near completion, they were harvested and analyzed for markers of scar formation. ***RESULTS***: Application of L. plantarum curtailed both the severity and the length of the pseudomonal infection. Probiotic therapy significantly reduced both Type I collagen mRNA concentrations and total collagen protein accumulation in infected wounds, consistent with reduced scarring. Surprisingly, the probiotic showed a nearly equivalent effect in uninfected wounds. Masson's trichrome staining confirmed these findings histologically. ***CONCLUSIONS***: Lactobacillus plantarum shows exciting potential as a therapeutic agent to both counteract burn wound infection and to alleviate scarring even in the absence of infection.

Savilahti, E. M., Makitie, O., Kukkonen, A. K., Andersson, S., Viljakainen, H., Savilahti, E., and Kuitunen, M. Serum 25-Hydroxyvitamin D in Early Childhood Is Nonlinearly Associated with Allergy. Int.Arch.Allergy Immunol. 2016; 170(3):141-148; PM: 27533066

ABSTRACT: ***BACKGROUND***: Vitamin D has several immunological functions. Data on the relation of vitamin D status and allergy are controversial. ***METHODS***: We investigated the association between serum concentrations of 25-hydroxyvitamin D (25-OHD) and allergy in childhood. The study population (n = 819) was part of a randomized, double-blind, placebo-controlled trial where the mothers of offspring with a high risk for allergy received a mixture of probiotics (or placebo) for the last 4 weeks of pregnancy, and the child received this from birth to 6 months. Study subjects were followed for the emergence of sensitization and allergic symptoms for a period of 5 years, with medical examinations at the ages of 3 and 6 months, 2 and 5 years and also in the event of allergic symptoms. Levels of 25-OHD were measured in umbilical cord blood (UCB) samples (n = 724) and serum samples drawn at the age of 2 years (n = 369); the data were categorized into tertiles (T1-T3) and quartiles (Q1-Q4). The relation between 25-OHD levels and sensitization and allergy was analyzed with multivariable logistic regression analysis. ***RESULTS***: 25-OHD levels in T2 in UCB were associated with a higher risk for sensitization by the age of 2 years and allergic disorders by the age of 5 years. In the

serum samples, at the age of 2 years, 25-OHD levels in Q3 were associated with a higher risk of sensitization and IgE-mediated allergies by the age of 5 years. ***CONCLUSIONS***: The 25-OHD levels in early childhood are associated with the emergence of allergy, but the association appears to be nonlinear.

Sawh, S. C., Deshpande, S., Jansen, S., Reynaert, C. J., and Jones, P. M. Prevention of necrotizing enterocolitis with probiotics: a systematic review and meta-analysis. PeerJ. 2016; 4:e2429; PM: 27761306

ABSTRACT: ***CONTEXT***: Necrotizing enterocolitis (NEC) is the most frequent gastrointestinal emergency in neonates. The microbiome of the preterm gut may regulate the integrity of the intestinal mucosa. Probiotics may positively contribute to mucosal integrity, potentially reducing the risk of NEC in neonates. ***OBJECTIVE***: To perform an updated systematic review and meta-analysis on the efficacy and safety of probiotics for the prevention of NEC in premature infants. ***DATA SOURCES***: Structured searches were performed in: Medline, Embase, and the Cochrane Central Register of Controlled Trials (all via Ovid, from 2013 to January 2015). Clinical trial registries and electronically available conference materials were also searched. An updated search was conducted June 3, 2016. ***STUDY SELECTION***: Randomized trials including infants less than 37 weeks gestational age or less than 2,500 g on probiotic vs. standard therapy. ***DATA EXTRACTION***: Data extraction of the newly-identified trials with a double check of the previously-identified trials was performed using a standardized data collection tool. ***RESULTS***: Thirteen additional trials (n = 5,033) were found. The incidence of severe NEC (RR 0.53 95% CI [0.42-0.66]) and all-cause mortality (RR 0.79 95% CI [0.68-0.93]) were reduced. No difference was shown in culture-proven sepsis RR 0.88 95% CI [0.77-1.00]. ***LIMITATIONS***: Heterogeneity of organisms and dosing regimens studied prevent a species-specific treatment recommendation from being made. ***CONCLUSIONS***: Preterm infants benefit from probiotics to prevent severe NEC and death.

Scagnolari, C., Corano, Scheri G., Selvaggi, C., Schietroma, I., Najafi, Fard S., Mastrangelo, A., Giustini, N., Serafino, S., Pinacchio, C., Pavone, P., Fanello, G., Ceccarelli, G., Vullo, V., and d'Ettorre, G. Probiotics Differently Affect Gut-Associated Lymphoid Tissue Indolamine-2,3-Dioxygenase mRNA and Cerebrospinal Fluid Neopterin Levels in Antiretroviral-Treated HIV-1 Infected Patients: A Pilot Study. Int.J.Mol.Sci. 9-27-2016; 17(10); PM: 27689995

ABSTRACT: Recently the tryptophan pathway has been considered an important determinant of HIV-1 infected patients' quality of life, due to the toxic effects of its metabolites on the central nervous system (CNS). Since the dysbiosis described in HIV-1 patients might be responsible for the microbial translocation, the chronic immune activation, and the altered utilization of tryptophan observed in these individuals, we speculated a correlation between high levels of immune activation markers in the cerebrospinal fluid (CSF) of HIV-1 infected patients and the over-expression of indolamine-2,3-dioxygenase (IDO) at the gut mucosal surface. In order to evaluate this issue, we measured the levels of neopterin in CSF, and the expression of IDO mRNA in gut-associated lymphoid tissue (GALT), in HIV-1-infected patients on effective combined antiretroviral therapy (cART), at baseline and after six months of probiotic dietary management. We found a significant reduction of neopterin and IDO mRNA levels after the supplementation with probiotic. Since the results for the use of adjunctive therapies to reduce the levels of immune activation markers in CSF have been disappointing so far,

our pilot study showing the efficacy of this specific probiotic product should be followed by a larger confirmatory trial.

Scarpignato, C., Bertele, A., and Tursi, A. Probiotics for the Treatment of Symptomatic Uncomplicated Diverticular Disease: Rationale and Current Evidence. J.Clin.Gastroenterol. 2016; 50 Suppl 1:S70-S73; PM: 27622371

ABSTRACT: Changes in the colonic microbiota are critical to the pathogenesis of diverticular complications such as diverticulitis and peridiverticular abscesses. However, more subtle changes in microbiota composition may well be important to the more chronic manifestations of diverticulosis. Some studies have shown the presence of bacterial overgrowth in subgroups of patients with diverticular disease and recent studies, using molecular biology techniques, found an increase of proteobacteria and actinobacteria in patients with symptomatic uncomplicated diverticular disease (SUDD), compared with healthy controls. The use of probiotics to modulate intestinal microecology in SUDD appears therefore rational. Although several investigations evaluating the clinical efficacy of probiotics have been performed, no definitive results have yet been achieved, mainly due to the heterogeneity of the available studies. Most of the studies used probiotics in combination with poorly absorbed antimicrobials or anti-inflammatory drugs. In only 4 studies, there was a harm using probiotics alone, but only 1 was a placebo-controlled, double-blind trial. The analysis of the available evidence reveals a poor quality of the published studies, whose design was heterogeneous, with only 2 out of 11 trials being double-blind and randomized. Therefore, available data can only suggest a benefit of probiotics in SUDD, but do not allow any evidence-based definite conclusion. As a consequence, current guidelines state that there is insufficient evidence to recommend probiotics for symptom relief in patients with diverticular disease.

Schlichte, M. J., Vandersall, A., and Katta, R. Diet and eczema: a review of dietary supplements for the treatment of atopic dermatitis. Dermatol.Pract.Concept. 2016; 6(3):23-29; PM: 27648380

ABSTRACT: In the context of increasing popularity of "natural" alternatives to conventional medicine, several dietary supplements have gained the attention of researchers and consumers alike in the treatment of atopic dermatitis (AD). Readily available without a prescription and frequently perceived to have fewer side effects than traditional medications, these "natural" remedies may be featured in discussions with patients, and clinicians should therefore be familiar with their efficacy and safety. Based on trials to date, no dietary supplements can be recommended for routine use in the treatment of AD. However, some promising results have been noted from the use of probiotics and prebiotics taken in combination. Given significant differences in study design to date, however, further studies would be needed to clarify dose and strains of probiotics. Studies of vitamin D have been limited and have produced conflicting results, although further trials in selected subsets of patients may be indicated. Very limited data is available on fish oil supplements, while future studies on Chinese herbal medicine would require evaluation of comparable herbs and formulations. Finally, multiple trials of evening primrose oil and borage seed oil have shown improvement similar to placebo, and neither is currently recommended in eczema therapy.

Schmidt, K. and Engel, P. Probiotic Treatment with a Gut Symbiont Leads to Parasite Susceptibility in Honey Bees. Trends Parasitol. 2016; 32(12):914-916; PM: 27697444

ABSTRACT: Gut symbionts are critical for host health and as such might be used as probiotics. In a recent study, Schwarz et al. showed that pretreatment of honey bees with a dominant gut bacterium causes dysbiosis and increases pathogen susceptibility, showing that probiotic applications for animal health can have unwanted effects.

Schoster, A., Guardabassi, L., Staempfli, H. R., Abrahams, M., Jalali, M., and Weese, J. S. The longitudinal effect of a multi-strain probiotic on the intestinal bacterial microbiota of neonatal foals. Equine Vet.J. 2016; 48(6):689-696; PM: 26509834

ABSTRACT: ***REASONS FOR PERFORMING THE STUDY***: The microbiota plays a key role in health and disease. Probiotics are a potential way to therapeutically modify the intestinal microbiota and prevent disease. *OBJECTIVES*: The aim of this study was to investigate the effects of probiotics on the bacterial microbiota of foals during and after administration. ***STUDY DESIGN***: Randomised placebo controlled field trial. ***METHODS***: Thirty-eight healthy neonatal foals enrolled in a previous study were selected. The foals had received a multi-strain probiotic (four Lactobacillus spp. 3-4 x 103 colony-forming units (cfu)/g each, Bifidobacterium animalis spp. lactis, 1 x 103-4 cfu/g) or placebo once daily for 3 weeks. A total of 3 faecal samples were collected from each foal at 2-week intervals and assessed via metagenomic sequencing. The Wilcoxon test was used to compare data between treatment groups. ***RESULTS***: There were no changes on the phylum, order or class level between treatment groups at any age (all P>0.05) but some significant changes in relative abundance of families. Probiotic administration did not result in an increased relative abundance of lactobacilli or bifidobacteria at any age (Lactobacillus: P = 0.9, P = 0.1 and P = 0.2, Bifidobacterium: P = 0.3, P = 0.6 and P = 0.1 for Weeks 2, 4 and 6, respectively). Lactobacillus was enriched in the probiotic group at Week 6 on LEfSe analysis (linear discriminant analysis score 0.34, P = 0 .02). There was no effect on alpha diversity (all P>0.2) or community structure when parsimony and unifrac analysis were applied (all P>0.6). ***CONCLUSIONS***: There were limited effects of probiotic treatment on the bacterial microbiota of foals. The studied probiotic based on lactobacilli and bifidobacteria has a limited potential for therapeutic modification of the gastrointestinal microbiota.

Sepideh, A., Karim, P., Hossein, A., Leila, R., Hamdollah, M., Mohammad, E. G., Mojtaba, S., Mohammad, S., Ghader, G., and Seyed, Moayed A. Effects of Multistrain Probiotic Supplementation on Glycemic and Inflammatory Indices in Patients with Nonalcoholic Fatty Liver Disease: A Double-Blind Randomized Clinical Trial. J.Am.Coll.Nutr. 2016; 35(6):500-505; PM: 26430826

ABSTRACT: ***OBJECTIVE***: Nonalcoholic fatty liver disease (NAFLD) is a condition defined by exceeding triglycerides accumulation in the liver. The condition can develop into fibrosis, cirrhosis, and hepatocellular carcinoma. Considering the ever-increasing prevalence of NAFLD, the aim of the present study was to investigate the effects of probiotic supplementation on glycemic and inflammatory indices in patients with NAFLD. ***METHODS***: This randomized clinical trial was conducted on 42 patients with NAFLD who had been referred to a gastroenterology clinic. Subjects in the intervention and control groups consumed 2 capsules/day probiotic or placebo, respectively, for 8 weeks. Fasting blood sugar (FBS), insulin, insulin resistance, tumor necrosis factor alpha (TNF-alpha), and

interleukin 6 (IL-6) were measured at baseline and at the end of the study. ***RESULTS***: Means of FBS, insulin, insulin resistance, and IL-6 were significantly different between groups after intervention ($p < 0.05$), whereas TNF-alpha was not significantly modified ($p > 0.05$). In the probiotic group, insulin, insulin resistance, TNF-alpha, and IL-6 decreased significantly at the end of the study compared to the beginning of study. ***CONCLUSION***: Considering the effects of probiotic supplementation on the reduction of glycemic and inflammatory indices in patients with NAFLD, consumption of probiotics is recommended as a complementary therapy in these patients.

Severance, E. G., Gressitt, K. L., Stallings, C. R., Katsafanas, E., Schweinfurth, L. A., Savage, C. L., Adamos, M. B., Sweeney, K. M., Origoni, A. E., Khushalani, S., Dickerson, F. B., and Yolken, R. H. Probiotic normalization of Candida albicans in schizophrenia: A randomized, placebo-controlled, longitudinal pilot study. Brain Behav.Immun. 11-18-2016; PM: 27871802

ABSTRACT: The molecules and pathways of the gut-brain axis represent new targets for developing methods to diagnose and treat psychiatric disorders. Manipulation of the gut microbiome with probiotics may be a therapeutic strategy with the potential to relieve gastrointestinal (GI) comorbidities and improve psychiatric symptoms. Candida albicans and Saccharomyces cerevisiae, commensal yeast species, can be imbalanced in the unhealthy human microbiome, and these fungal exposures were previously found elevated in schizophrenia. In a longitudinal, double-blind, placebo-controlled, pilot investigation of 56 outpatients with schizophrenia, we examined the impact of probiotic treatment on yeast antibody levels, and the relationship between treatment and antibody levels on bowel discomfort and psychiatric symptoms. We found that probiotic treatment significantly reduced C. albicans antibodies over the 14-week study period in males, but not in females. Antibody levels of S. cerevisiae were not altered in either treatment group. The highest levels of bowel discomfort over time occurred in C. albicans-seropositive males receiving the placebo. We observed trends towards improvement in positive psychiatric symptoms in males treated with probiotics who were seronegative for C. albicans. Results from this pilot study hint at an association of C. albicans seropositivity with worse positive psychiatric symptoms, which was confirmed in a larger cohort of 384 males with schizophrenia. In conclusion, the administration of probiotics may help normalize C. albicans antibody levels and C. albicans-associated gut discomfort in many male individuals. Studies with larger sample sizes are warranted to address the role of probiotics in correcting C. albicans-associated psychiatric symptoms.

Seyedain-Ardabili, M., Sharifan, A., and Tarzi, B. G. The Production of Synbiotic Bread by Microencapsulation. Food Technol.Biotechnol. 2016; 54(1):52-59; PM: 27904393

ABSTRACT: Bread is a global staple food. Despite attempts to develop functional breads containing viable microorganisms, this has not been done yet because of the high temperature during baking. The aim of this study is to obtain synbiotic bread, hence hamburger bun and white pan bread were selected. Lactobacillus acidophilus LA-5 and L. casei 431 were encapsulated with calcium alginate and Hi-maize resistant starch via emulsion technique and coated with chitosan. The morphology and size of microcapsules were measured by scanning electron microscopy and particle size analyser. Inulin was added at 5% wheat flour mass basis for prebiotic effect. The

encapsulated probiotics were inoculated into the bread dough and bread loaves were baked. The survival of encapsulated probiotics was determined after baking; also sensory evaluation was performed. Both types of bread met the standard criteria for probiotic products. The probiotic survival was higher in hamburger bun. L. casei 431 was more resistant to high temperature than L. acidophilus LA-5. A significant increase in probiotic survival was observed when the protective coating of chitosan was used in addition to calcium alginate and Hi-maize resistant starch. Storage for 4 days did not have any effect on the viability of encapsulated bacteria. The addition of encapsulated bacteria did not have any effect on flavour and texture; however, 5% inulin improved the texture of bread significantly. Results show that microencapsulation used in the production of synbiotic bread can enhance the viability and thermal resistance of the probiotic bacteria.

Sgibnev, A. and Kremleva, E. Influence of Hydrogen Peroxide, Lactic Acid, and Surfactants from Vaginal Lactobacilli on the Antibiotic Sensitivity of Opportunistic Bacteria. Probiotics.Antimicrob.Proteins. 11-10-2016; PM: 27832440

ABSTRACT: We studied as hydrogen peroxide, lactic acid, or surfactants from clinical isolates of vaginal lactobacilli and cell-free supernatants from probiotic strain LCR35 can influence on the sensitivity of opportunistic bacteria to antibiotics. We found that the most effective in increasing sensitivity to antibiotics were hydrogen peroxide and surfactants or their combination but no lactic acid. In some cases, the effect of the composition of hydrogen peroxide and surfactants was clearly higher than the sum of effects of these substances alone. With using of the supernatant of LCR35 was shown that the combination of surfactant and lactate has greater effect compared with surfactants alone. In concluding, metabolites of vaginal lactobacilli are suitable for the role of "antibiotic assistants" and it can help solve the problems the antibiotic resistance.

Shang, M. and Sun, J. Vitamin D/VDR, probiotics, and gastrointestinal diseases. Curr.Med.Chem. 12-2-2016; PM: 27915988

ABSTRACT: Vitamin D is an important factor in regulating inflammation, immune responses, and carcinoma inhibition via action of its receptor, vitamin D receptor (VDR). Recent studies have demonstrated the role of vitamin D/VDR in regulating host-bacterial interactions. Probiotics are beneficial bacteria with the power of supporting or favoring life on the host. In the current review, we will discuss the recent progress on the roles of vitamin D/VDR in gut microbiome and inflammation. We will summarize evidence of probiotics in modulating vitamin D/VDR and balancing gut microbiota in health and gastrointestinal diseases. Moreover, we will review the clinical application of probiotics in prevention and therapy of IBD or colon cancer. Despite the gains, there remain several barriers to advocate broad use of probiotics in clinical therapy. We will also discuss the limits and future direction in scientific understanding of probiotics, vitamin D/VDR, and host responses.

Shehata, A. A., Tarabees, R., Basiouni, S., Gamil, M., Kamal, A. S., and Kruger, M. Phenotypic and Genotypic Characterization of Bacteriocinogenic Enterococci Against Clostridium botulinum. Probiotics.Antimicrob.Proteins. 12-2-2016; PM: 27914002

ABSTRACT: The present study aimed to characterize Enterococcus faecalis (n = -6) and

Enterococcus faecium (n = 1) isolated from healthy chickens to find a novel perspective probiotic candidate that antagonize Clostridium botulinum types A, B, D, and E. The isolated enterococci were characterized based on phenotypic properties, PCR, and matrix-assisted laser desorption/ionization time of flight (MALDI-TOF). The virulence determinants including hemolytic activity on blood agar, gelatinase activity, sensitivity to vancomycin, and presence of gelatinase (gelE) and enterococcal surface protein (esp) virulence genes were investigated. Also, the presence of enterocin structural genes enterocin A, enterocin B, enterocin P, enterocin L50A/B, bacteriocin 31, enterocin AS48, enterocin 1071A/1071B, and enterocin 96 were assessed using PCR. Lastly, the antagonistic effect of the selected Enterococcus spp. on the growth of C. botulinum types A, B, D, and E was studied. The obtained results showed that four out of six E. faecalis and one E. faecium proved to be free from the tested virulence markers. All tested enterococci strains exhibited more than one of the tested enterocin. Interestingly, E. faecalis and E. faecium significantly restrained the growth of C. botulinum types A, B, D, and E. In conclusion, although, the data presented showed that bacteriocinogenic Enterococcus strains lacking of virulence determinants could be potentially used as a probiotic candidate against C. botulinum in vitro; however, further investigations are still urgently required to verify the beneficial effects of the tested Enterococcus spp. in vivo.

Sheng, Y., Yang, X., Lian, Y., Zhang, B., He, X., Xu, W., and Huang, K. Characterization of a cadmium resistance Lactococcus lactis subsp. lactis strain by antioxidant assays and proteome profiles methods. Environ.Toxicol.Pharmacol. 2016; 46:286-291; PM: 27522548

ABSTRACT: Heavy metal contamination poses a major threat to the environment and human health for their potential toxicity and non-biodegradable properties. At present, some probiotics bacteria are reported to have great potential to eliminate heavy metals from food and water. In this study, resistance properties of a newly isolated Lactococcus lactis subsp. lactis for cadmium were studied by antioxidant assays and proteomics analysis. Antioxidant capacity of this strain was significantly activated under cadmium stress indicated by Fenton reaction, DPPH assay, SOD assay and GSH assay. Intracellular antioxidant enzyme systems, such as superoxide dismutase, glutathione reductase and catalase were suggested to play vital roles in the activated antioxidant capacity. The up-regulated cadA was associated with the activated P-type ATPases that plays an important role in cadmium resistance. Proteomics analysis identified 12 over-expressed proteins under 50mg/L cadmium stress and these proteins are abundant in oxidative stress response and energy metabolism regulation, which were considered as consequences as cadmium resistance of the strain. Thus, the probiotics Lactococcus lactis subsp. lactis may resist cadmium stress through antioxidant approach and enhanced energy metabolism. The food grade lactis strain may be applied in metal decontamination in environment and food/feed.

Sherid, M., Samo, S., Sulaiman, S., Husein, H., Sifuentes, H., and Sridhar, S. Liver abscess and bacteremia caused by lactobacillus: role of probiotics? Case report and review of the literature. BMC.Gastroenterol. 11-18-2016; 16(1):138; PM: 27863462

ABSTRACT: ***BACKGROUND***: Lactobacilli are non-spore forming, lactic acid producing, gram-positive rods. They are a part of the normal gastrointestinal and genitourinary microbiota and have rarely been reported to be the cause of infections. Lactobacilli species are considered non-pathogenic organisms and have been used as probiotics to prevent antibiotic associated diarrhea. There are sporadic reported cases of infections related to lactobacilli containing probiotics. ***CASE PRESENTATION***: In this paper we discuss

a case of an 82 year old female with liver abscess and bacteremia from lactobacillus after using probiotics containing lactobacilli in the course of her treatment of Clostridium difficile colitis. The Lactobacillus strain identification was not performed and therefore, both commensal microbiota and the probiotic product should be considered as possible sources of the strain. ***CONCLUSION***: Lactobacilli can lead to bacteremia and liver abscesses in some susceptible persons and greater awareness of this potential side effect is warranted with the increasing use of probiotics containing lactobacilli.

Shi, L. H., Balakrishnan, K., Thiagarajah, K., Mohd Ismail, N. I., and Yin, O. S. Beneficial Properties of Probiotics. Trop.Life Sci.Res. 2016; 27(2):73-90; PM: 27688852

ABSTRACT: Probiotics are live microorganisms that can be found in fermented foods and cultured milk, and are widely used for the preparation of infant food. They are well-known as "health friendly bacteria", which exhibit various health beneficial properties such as prevention of bowel diseases, improving the immune system, for lactose intolerance and intestinal microbial balance, exhibiting antihypercholesterolemic and antihypertensive effects, alleviation of postmenopausal disorders, and reducing traveller's diarrhoea. Recent studies have also been focused on their uses in treating skin and oral diseases. In addition to that, modulation of the gut-brain by probiotics has been suggested as a novel therapeutic solution for anxiety and depression. Thus, this review discusses on the current probiotics-based products in Malaysia, criteria for selection of probiotics, and evidences obtained from past studies on how probiotics have been used in preventing intestinal disorders via improving the immune system, acting as an antihypercholesterolemic factor, improving oral and dermal health, and performing as anti-anxiety and anti-depressive agents.

Shin, J. H., Chung, M. J., and Seo, J. G. A multistrain probiotic formulation attenuates skin symptoms of atopic dermatitis in a mouse model through the generation of CD4+Foxp3+ T cells. Food Nutr.Res. 2016; 60:32550; PM: 27802847

ABSTRACT: ***BACKGROUND***: Atopic dermatitis (AD) is characterized by chronic inflammation of the skin. AD develops mainly in infants and young children. It induces skin disorders and signals the initiation of the allergic march including allergic asthma and rhinitis. Probiotics modify intestinal microbial populations in a beneficial way for human and animal hosts by reducing inflammatory cytokines. ***OBJECTIVE***: As a result of their immunomodulatory properties, probiotics have been considered a promising therapeutic option for the prevention and treatment of AD. ***DESIGN***: In this study, we examined the effects of GI7, a potential probiotic mixture consisting of seven strains of bifidobacteria and lactic acid bacteria, on AD in a mouse model. ***RESULTS***: Administration of GI7 for 8 weeks reduced AD-like skin lesions and induced changes in the levels of serum markers such as immunoglobulin E and cytokines related to T helper (Th)1 and Th2 cells, and in skin barrier genes. Alleviation of AD seems to be associated with GI7-induced generation of CD4+Foxp3+ regulatory T cells. ***CONCLUSIONS***: The probiotic mixture may have potential to improve symptoms of AD.

Shiraishi, T., Yokota, S., Fukiya, S., and Yokota, A. Structural diversity and biological significance of lipoteichoic acid in Gram-positive bacteria: focusing on beneficial probiotic lactic acid bacteria. Biosci.Microbiota.Food Health. 2016; 35(4):147-161; PM: 27867802

ABSTRACT: Bacterial cell surface molecules are at the forefront of host-bacterium interactions. Teichoic acids are observed only in Gram-positive bacteria, and they are one of the main cell surface components. Teichoic acids play important physiological roles and contribute to the bacterial interaction with their host. In particular, lipoteichoic acid (LTA) anchored to the cell membrane has attracted attention as a host immunomodulator. Chemical and biological characteristics of LTA from various bacteria have been described. However, most of the information concerns pathogenic bacteria, and information on beneficial bacteria, including probiotic lactic acid bacteria, is insufficient. LTA is structurally diverse. Strain-level structural diversity of LTA is suggested to underpin its immunomodulatory activities. Thus, the structural information on LTA in probiotics, in particular strain-associated diversity, is important for understanding its beneficial roles associated with the modulation of immune response. Continued accumulation of structural information is necessary to elucidate the detailed physiological roles and significance of LTA. In this review article, we summarize the current state of knowledge on LTA structure, in particular the structure of LTA from lactic acid bacteria. We also describe the significance of structural diversity and biological roles of LTA.

Shokryazdan, P., Faseleh, Jahromi M., Navidshad, B., and Liang, J. B. Effects of prebiotics on immune system and cytokine expression. Med.Microbiol.Immunol. 10-4-2016; PM: 27704207

ABSTRACT: Nowadays, use of prebiotics as feed and food additives has received increasing interest because of the beneficial effects of prebiotics on the health of animals and humans. One of the beneficial effects of prebiotics is stimulation of immune system, which can be direct or indirect through increasing population of beneficial microbes or probiotics, especially lactic acid bacteria and bifidobacteria, in the gut. An important mechanism of action of probiotics and prebiotics, by which they can affect the immune system, is changing the expression of cytokines. The present review tried to summarize the findings of studies that investigated the effects of prebiotics on immune system with focusing on their effects on cytokine expression. Generally, most of reviewed studies indicated beneficial effects for prebiotics in terms of improving immune system, by increasing the expression of anti-inflammatory cytokines, while reducing the expressions of proinflammatory cytokines. However, most of studies mainly considered the indirect effects of prebiotics on the immune system (through changing the composition and population of gut microbiota), and their direct effects still need to be further studied using prebiotics with different degree of polymerization in different hosts.

Silva, B. C., Sandes, S. H., Alvim, L. B., Bomfim, M. R., Nicoli, J. R., Neumann, E., and Nunes, A. C. Selection of a candidate probiotic strain of Pediococcus pentosaceus from the faecal microbiota of horses by in vitro testing and health claims in a mouse model of Salmonella infection. J.Appl.Microbiol. 11-4-2016; PM: 27813217

ABSTRACT: ***AIMS***: The aim of this study was to verify the suitable use of candidate 'probiotics' selected by in vitro tests and the importance of in vivo assays to nominate the microorganisms as probiotics and alternatives prophylactic treatments for Salmonella Typhimurium infection. ***METHODS AND RESULTS***: Thirty-three lactic acid bacteria (LAB) isolated from foal's faeces were assessed based on the main desirable functional in vitro criteria. Based on these results, we choosed Pediococcus pentosaceus strain #40, to

evaluate its putative probiotic features in a mouse model of Salmonella infection. Daily intragastric doses of Ped. pentosaceus #40 for 10 days before and 10 days after Salmonella challenge (106 CFU of Salmonella Typhimurium/mouse) led to significant aggravation in mouse health by increasing weight loss, worsening clinical symptoms, and anticipating the time and number of deaths by Salmonella. Ped. pentosaceus modulated cell-mediated immune responses by up-regulation of the gene expression of the proinflammatory cytokines IFN-gamma and TNF-alpha in the small intestine. ***CONCLUSION***: We used the usual criteria for in vitro screening of a large number of LAB for desirable probiotic functional properties. However, the best candidate probiotic strain identified, Ped. pentosaceus #40, aggravated the experimental disease in mice. This article is protected by copyright. All rights reserved.

Singh, S., Jing, E., and Stollman, N. Self-Limited Sepsis Syndrome Following Fecal Microbiota Therapy for Refractory C. difficile Infection. Dig.Dis.Sci. 2016; 61(9):2488-2491; PM: 26541990

No Abstract Available.

Slade, E., Dale, A., and Mahtani, K. Probiotics in very preterm infants: the PiPS trial. Lancet. 8-13-2016; 388(10045):656; PM: 27533431

No Abstract Available.

Slattery, J., MacFabe, D. F., and Frye, R. E. The Significance of the Enteric Microbiome on the Development of Childhood Disease: A Review of Prebiotic and Probiotic Therapies in Disorders of Childhood. Clin.Med.Insights.Pediatr. 2016; 10:91-107; PM: 27774001

ABSTRACT: Recent studies have highlighted the fact that the enteric microbiome, the trillions of microbes that inhabit the human digestive tract, has a significant effect on health and disease. Methods for manipulating the enteric microbiome, particularly through probiotics and microbial ecosystem transplantation, have undergone some study in clinical trials. We review some of the evidence for microbiome alteration in relation to childhood disease and discuss the clinical trials that have examined the manipulation of the microbiome in an effort to prevent or treat childhood disease with a primary focus on probiotics, prebiotics, and/or synbiotics (ie, probiotics + prebiotics). Studies show that alterations in the microbiome may be a consequence of events occurring during infancy and/or childhood such as prematurity, C-sections, and nosocomial infections. In addition, certain childhood diseases have been associated with microbiome alterations, namely necrotizing enterocolitis, infantile colic, asthma, atopic disease, gastrointestinal disease, diabetes, malnutrition, mood/anxiety disorders, and autism spectrum disorders. Treatment studies suggest that probiotics are potentially protective against the development of some of these diseases. Timing and duration of treatment, the optimal probiotic strain(s), and factors that may alter the composition and function of the microbiome are still in need of further research. Other treatments such as prebiotics, fecal microbial transplantation, and antibiotics have limited evidence. Future translational work, in vitro models, long-term and follow-up studies, and guidelines for the

composition and viability of probiotic and microbial therapies need to be developed. Overall, there is promising evidence that manipulating the microbiome with probiotics early in life can help prevent or reduce the severity of some childhood diseases, but further research is needed to elucidate biological mechanisms and determine optimal treatments.

Smith, G. D. Is there a role for probiotics in older people? Br.J.Community Nurs. 10-2-2016; 21(10):501-503; PM: 27715260

No Abstract Available.

Smith, T. D., Watt, H., Gunn, L., Car, J., and Boyle, R. J. Recommending Oral Probiotics to Reduce Winter Antibiotic Prescriptions in People With Asthma: A Pragmatic Randomized Controlled Trial. Ann.Fam.Med. 2016; 14(5):422-430; PM: 27621158

ABSTRACT: ***PURPOSE***: Evidence from studies mainly in children has shown that orally administered probiotics may prevent respiratory tract infections and associated antibiotic use. We evaluated whether advice to take daily probiotics can reduce antibiotic prescribing for winter respiratory tract infections in people with asthma. ***METHODS***: We conducted a randomized controlled, parallel-group pragmatic study for participants aged 5 years and older with asthma in a UK primary care setting. The intervention was a postal leaflet with advice to take daily probiotics from October 2013 to March 2014, compared with a standard winter advice leaflet. Primary outcome was the proportion of participants prescribed antibiotics for respiratory tract infections. ***RESULTS***: There were 1,302 participants randomly assigned to a control group (n = 650) or intervention group (n = 652). There was no significant difference in the primary outcome measure, with 27.7% receiving antibiotics in the intervention group and 26.9% receiving antibiotics in the control group (odds ratio = 1.04; 95% CI, 0.82-1.34). Uptake of probiotics was low, but outcomes were similar in those who accessed probiotics (adjusted odds ratio = 1.08; 95% CI, 0.69-1.69, compared with controls). We also found no evidence of an effect on respiratory tract infections or asthma exacerbations. ***CONCLUSIONS***: In this pragmatic community-based trial in people with asthma, we found no evidence that advising use of winter probiotics reduces antibiotic prescribing.

Soltan Dallal, M. M., Davoodabadi, A., Abdi, M., Hajiabdolbaghi, M., Sharifi Yazdi, M. K., Douraghi, M., and Tabatabaei Bafghi, S. M. Inhibitory effect of Lactobacillus plantarum and Lb. fermentum isolated from the faeces of healthy infants against nonfermentative bacteria causing nosocomial infections. New Microbes.New Infect. 2017; 15:9-13; PM: 27830081

ABSTRACT: Nosocomial infection constitutes a major public health problem worldwide. Increasing antibiotic resistance of pathogens associated with nosocomial infections has also become a major therapeutic challenge for physicians. Thus, development of alternative treatment protocols, such as the use of probiotics, matters. The aim of this research was to determine the antagonistic properties of Lactobacillus plantarum and Lb. fermentum isolated from the faeces of healthy infants against nonfermentative bacteria causing nosocomial infections. One hundred five samples of nosocomial infections were collected and processed for bacterial isolation and antimicrobial susceptibility testing following standard bacteriologic techniques. The antibiotic susceptibility test was performed by the disk diffusion method, and antagonistic effect of

Lactobacillus strains was investigated by well diffusion method. Of 105 samples, a total of 29 bacterial strains were identified as nonfermentative bacteria, including 17 Acinetobacter baumannii and 12 Pseudomonas aeruginosa. A. baumannii showed high resistance to tested antibiotics except ampicillin/sulbactam, and P. aeruginosa showed resistance to ampicillin/sulbactam and gentamicin and sensitive to amikacin and meropenem. Lb. plantarum had antagonistic properties against both A. baumannii and P. aeruginosa strains. Lb. plantarum had considerable effects on preventing the growth of A. baumannii and P. aeruginosa strains. However, further research is needed to better understanding of these effects on P. aeruginosa.

Song, S., Lee, S. J., Park, D. J., Oh, S., and Lim, K. T. The anti-allergic activity of Lactobacillus plantarum L67 and its application to yogurt. J.Dairy Sci. 2016; 99(12):9372-9382; PM: 27743673

ABSTRACT: Recently, interest in the beneficial role of probiotics in the protection and management of allergic diseases caused by immune disorders has been increasing. This study investigated the inhibitory effect of Lactobacillus plantarum L67 on induced allergic inflammatory response in bisphenol A-treated rat basophilic leukemia 2H3 (RBL-2H3) cells and mouse splenocytes. We also evaluated the applicability of L. plantarum L67 as a yogurt starter culture. We measured the ability of Lactobacillus strains to induce the production of IL-12 and IFN- gamma in cultured splenocytes by ELISA. Bisphenol A (50muM)-treated RBL-2H3 cells were cotreated with a glycoprotein (18kDa) isolated from L. plantarum L67 (5-100microg/mL) for 30min. We measured the expression of mitogen-activated protein kinase (ERK and p38), AP-1 (c-Fos and c-Jun), T-bet, and GATA-binding protein 3 (GATA-3) using Western blotting to examine the differentiation of T helper cells. Furthermore, we evaluated the gene expression of IL-1beta, IL-6, and IL-10 using real-time quantitative PCR. Finally, we evaluated the applicability of L. plantarum L67 as a yogurt starter by measuring pH, enumeration of bacteria, and sensory scores. Our results showed that L67 protein inhibited the phosphorylation of ERK and p38 mitogen-activated protein kinase through the transcriptional activation of AP-1 in bisphenol A-treated RBL-2H3 cells. During differentiation of T helper cells, the expression of transcription factor GATA-3 was significantly suppressed by L67 protein (100microg/mL) treatment, whereas expression of transcription factor T-bet was increased. In addition, the L67 protein significantly attenuated the expression of T helper 2-linked cytokines IL-1beta, IL-6, and IL-10. These results indicate that L. plantarum L67, made available as yogurt starters and dietary supplements, has the potential to prevent allergy-related immune disorders.

Soreide, K., Boermeester, M. A., Humes, D. J., and Velmahos, G. C. Acute colonic diverticulitis: modern understanding of pathomechanisms, risk factors, disease burden and severity. Scand.J.Gastroenterol. 2016; 51(12):1416-1422; PM: 27539786

ABSTRACT: ***INTRODUCTION***: Conservative, non-antibiotic and non-surgical management of acute diverticulitis is currently being investigated. To better inform clinical decisions, better understanding of disease mechanisms, disease burden and severity is needed. ***METHODS***: Literature search of risk factors, pathophysiology, epidemiology and disease burden/severity reported over the last decade. ***RESULTS***: Acute diverticulitis is a common disease and has a high disease burden. Incidence of hospital admissions is reported around 71 per 100,000 population, with reported increase in several subpopulations over the last decades. The incidence is likely to increase further with the aging populations. Risk factors for left-sided acute diverticulitis include dietary, anthropometric and lifestyle factors. Disease mechanisms are still poorly understood, but a distinction between

inflammation and infection is emerging. The integrative and complex role of the gut microbiota has become an interesting factor for both understanding the disease as well as a potential target for intervention using probiotics. Mild, self-limiting events are increasingly reported from studies of successful non-antibiotic management in a considerable number of cases. Risk markers of progression to or presence of severe, complicated disease are needed for better disease stratification. Current risk stratification by clinical, imaging or endoscopic means is imperfect and needs validation. Long-term results from minimal-invasive and comparative surgical trials may better help inform clinicians and patients. ***CONCLUSIONS***: Over- and under-treatment as well as over- and under-diagnosis of severity is likely to continue in clinical practice due to lack of reliable, robust and universal severity and classification systems. Better understanding of pathophysiology is needed.

Starn, E. S., Hampe, H., and Cline, T. The Cost-efficiency and Care Effectiveness of Probiotic Administration with Antibiotics to Prevent Hospital-Acquired Clostridium difficile Infection. Qual.Manag.Health Care. 2016; 25(4):238-243; PM: 27749722

ABSTRACT: Health care facility-acquired Clostridium difficile infections (HCFA-CDI) have increased over the last several decades despite facilities developing protocols for prescribing probiotics with antibiotics to prevent HCFA-CDI. The literature does not consistently support this. A retrospective medical record review evaluated the care effectiveness of this practice. Care effectiveness was not found; patients receiving probiotics with antibiotics were twice as likely to develop HCFA-CDI (P = .004). Except with glycopeptides, patients were 1.88 times less likely to experience HCFA-CDI (P = .05).

Strasser, B., Geiger, D., Schauer, M., Gostner, J. M., Gatterer, H., Burtscher, M., and Fuchs, D. Probiotic Supplements Beneficially Affect Tryptophan-Kynurenine Metabolism and Reduce the Incidence of Upper Respiratory Tract Infections in Trained Athletes: A Randomized, Double-Blinded, Placebo-Controlled Trial. Nutrients. 11-23-2016; 8(11); PM: 27886064

ABSTRACT: ***BACKGROUND***: Prolonged intense exercise has been associated with transient suppression of immune function and an increased risk of infections. In this context, the catabolism of amino acid tryptophan via kynurenine may play an important role. The present study examined the effect of a probiotic supplement on the incidence of upper respiratory tract infections (URTI) and the metabolism of aromatic amino acids after exhaustive aerobic exercise in trained athletes during three months of winter training. ***METHODS***: Thirty-three highly trained individuals were randomly assigned to probiotic (PRO, n = 17) or placebo (PLA, n = 16) groups using double blind procedures, receiving either 1 x 1010 colony forming units (CFU) of a multi-species probiotic (Bifidobacterium bifidum W23, Bifidobacterium lactis W51, Enterococcus faecium W54, Lactobacillus acidophilus W22, Lactobacillus brevis W63, and Lactococcus lactis W58) or placebo once per day for 12 weeks. The serum concentrations of tryptophan, phenylalanine and their primary catabolites kynurenine and tyrosine, as well as the concentration of the immune activation marker neopterin were determined at baseline and after 12 weeks, both at rest and immediately after exercise. Participants completed a daily diary to identify any infectious symptoms. ***RESULTS***: After 12 weeks of treatment, post-exercise tryptophan levels were lowered by 11% (a significant change) in the PLA group compared to the concentrations measured before the intervention (p = 0.02), but remained unchanged in the PRO group. The ratio of subjects taking the placebo who

experienced one or more URTI symptoms was increased 2.2-fold compared to those on probiotics (PLA 0.79, PRO 0.35; p = 0.02). ***CONCLUSION***: Data indicate reduced exercise-induced tryptophan degradation rates in the PRO group. Daily supplementation with probiotics limited exercise-induced drops in tryptophan levels and reduced the incidence of URTI, however, did not benefit athletic performance.

Sung, H., Kim, S. W., Hong, M., and Suk, K. T. Microbiota-based treatments in alcoholic liver disease. World J.Gastroenterol. 8-7-2016; 22(29):6673-6682; PM: 27547010

ABSTRACT: Gut microbiota plays a key role in the pathogenesis of alcoholic liver disease (ALD). Consumption of alcohol leads to increased gut permeability, small intestinal bacterial overgrowth, and enteric dysbiosis. These factors contribute to the increased translocation of microbial products to the liver via the portal tract. Subsequently, bacterial endotoxins such as lipopolysaccharide, in association with the Toll-like receptor 4 signaling pathway, induce a gamut of damaging immune responses in the hepatic milieu. Because of the close association between deleterious inflammation and ALD-induced microbiota imbalance, therapeutic approaches that seek to reestablish gut homeostasis should be considered in the treatment of alcoholic patients. To this end, a number of preliminary studies on probiotics have confirmed their effectiveness in suppressing proinflammatory cytokines and improving liver function in the context of ALD. In addition, there have been few studies linking the administration of prebiotics and antibiotics with reduction of alcohol-induced liver damage. Because these preliminary results are promising, large-scale randomized studies are warranted to elucidate the impact of these microbiota-based treatments on the gut flora and associated immune responses, in addition to exploring questions about optimal delivery. Finally, fecal microbiota transplant has been shown to be an effective method of modulating gut microbiota and deserve further investigation as a potential therapeutic option for ALD.

Suzuki, K., Ohshimo, S., and Shime, N. Are probiotics actually useful in decreasing VAP rate? Intensive Care Med. 2016; 42(10):1643-1644; PM: 27620299

No Abstract Available.

Takahashi, S., Anzawa, D., Takami, K., Ishizuka, A., Mawatari, T., Kamikado, K., Sugimura, H., and Nishijima, T. Effect of Bifidobacterium animalis ssp. lactis GCL2505 on visceral fat accumulation in healthy Japanese adults: a randomized controlled trial. Biosci.Microbiota.Food Health. 2016; 35(4):163-171; PM: 27867803

ABSTRACT: Bifidobacterium animalis ssp. lactis GCL2505 (B. lactis GCL2505) is able to survive passage through the intestine and then proliferate, leading to an increase in the amount of gut bifidobacteria. In the present study, we evaluated the impact of B. lactis GCL2505 on abdominal visceral fat storage in overweight and mildly obese Japanese adults. This clinical study was a double-blind, randomized, placebo-controlled, parallel-group comparative trial performed for 12 weeks. Healthy Japanese subjects (N=137) with body mass indices ranging from 23 to 30 kg/m2 consumed either fermented milk containing B. lactis GCL2505 or a placebo every day, and then visceral and subcutaneous abdominal fat areas were measured by computed tomography as the

primary endpoints. The number of fecal bifidobacteria was also measured. Visceral fat area, but not subcutaneous fat area, was significantly reduced from baseline at 8 and 12 weeks in the GCL2505 group, compared with the placebo group. The total number of fecal bifidobacteria was significantly increased in the GCL2505 group. These results indicate that B. lactis GCL2505 reduces abdominal visceral fat, a key factor associated with metabolic disorders. This finding suggests that this probiotic strain can potentially serve as a specific functional food to achieve visceral fat reduction in overweight or mildly obese individuals.

Tang, Z., Xu, L., Shi, B., Deng, H., Lai, X., Liu, J., and Sun, Z. Oral administration of synthetic porcine beta-defensin-2 improves growth performance and cecal microbial flora and down-regulates the expression of intestinal toll-like receptor-4 and inflammatory cytokines in weaned piglets challenged with enterotoxigenic Escherichia coli. Anim Sci.J. 2016; 87(10):1258-1266; PM: 26707031

ABSTRACT: Synthetic porcine beta-defensin-2 (pBD-2) was tested as an alternative to antimicrobial growth-promoters in pig production. Thirty 21-day weaned piglets were challenged with enterotoxigenic Escherichia coli, and orally dosed with either sterile water (CON), pBD-2 (BD) or neomycin sulphate (NS) twice daily for 21 days. pBD-2 and NS led to higher growth performance, jejunum villus height and increased expression of insulin-like growth factor-I compared with the CON group ($P < 0.05$). Hemolytic E. coli scores from rectal swabs, and copy numbers of E. coli, Bacteroides fragilis and Streptococcus in the cecal digesta of the BD- or NS-treated piglets were lower than those in the CON group ($P < 0.05$). Messenger RNA levels of toll-like receptor 4, tumor necrosis factor-alpha, interleukin (IL)-1beta, and IL-8 in the jejunum mucosa of the BD and NS groups were lower than those in the CON group ($P < 0.05$). Copy numbers of Lactobacilli and Bifidobacteria in the cecal digesta of the BD group were higher than those of the CON and NS groups ($P < 0.05$). Therefore, pBD-2 has antimicrobial activity in piglets, and it can improve growth performance, reduce inflammatory cytokine expression and affect intestinal morphological indices in the same way as probiotics. (c) 2015 Japanese Society of Animal Science.

Tilg, H., Cani, P. D., and Mayer, E. A. Gut microbiome and liver diseases. Gut. 10-8-2016; PM: 27802157

ABSTRACT: The gut microbiota has recently evolved as a new important player in the pathophysiology of many intestinal and extraintestinal diseases. The liver is the organ which is in closest contact with the intestinal tract, and is exposed to a substantial amount of bacterial components and metabolites. Various liver disorders such as alcoholic liver disease, non-alcoholic liver disease and primary sclerosing cholangitis have been associated with an altered microbiome. This dysbiosis may influence the degree of hepatic steatosis, inflammation and fibrosis through multiple interactions with the host's immune system and other cell types. Whereas few results from clinical metagenomic studies in liver disease are available, evidence is accumulating that in liver cirrhosis an oral microbiome is overrepresented in the lower intestinal tract, potentially contributing to disease process and severity. A major role for the gut microbiota in liver disorders is also supported by the accumulating evidence that several complications of severe liver disease such as hepatic encephalopathy are efficiently treated by various prebiotics, probiotics and antibiotics. A better understanding of the gut microbiota and

its components in liver diseases might provide a more complete picture of these complex disorders and also form the basis for novel therapies.

Travers, M. A., Sow, C., Zirah, S., Deregnaucourt, C., Chaouch, S., Queiroz, R. M., Charneau, S., Allain, T., Florent, I., and Grellier, P. Deconjugated Bile Salts Produced by Extracellular Bile-Salt Hydrolase-Like Activities from the Probiotic Lactobacillus johnsonii La1 Inhibit Giardia duodenalis In vitro Growth. Front Microbiol. 2016; 7:1453; PM: 27729900

ABSTRACT: Giardiasis, currently considered a neglected disease, is caused by the intestinal protozoan parasite Giardia duodenalis and is widely spread in human as well as domestic and wild animals. The lack of appropriate medications and the spread of resistant parasite strains urgently call for the development of novel therapeutic strategies. Host microbiota or certain probiotic strains have the capacity to provide some protection against giardiasis. By combining biological and biochemical approaches, we have been able to decipher a molecular mechanism used by the probiotic strain Lactobacillus johnsonii La1 to prevent Giardia growth in vitro. We provide evidence that the supernatant of this strain contains active principle(s) not directly toxic to Giardia but able to convert non-toxic components of bile into components highly toxic to Giardia. By using bile acid profiling, these components were identified as deconjugated bile-salts. A bacterial bile-salt-hydrolase of commercial origin was able to mimic the properties of the supernatant. Mass spectrometric analysis of the bacterial supernatant identified two of the three bile-salt-hydrolases encoded in the genome of this probiotic strain. These observations document a possible mechanism by which L. johnsonii La1, by secreting, or releasing BSH-like activity(ies) in the vicinity of replicating Giardia in an environment where bile is present and abundant, can fight this parasite. This discovery has both fundamental and applied outcomes to fight giardiasis, based on local delivery of deconjugated bile salts, enzyme deconjugation of bile components, or natural or recombinant probiotic strains that secrete or release such deconjugating activities in a compartment where both bile salts and Giardia are present.

Trimarco, V., Rozza, F., Izzo, R., De, Leo, V, Cappelli, V., Riccardi, C., and Di, Carlo C. Effects of a new combination of nutraceuticals on postmenopausal symptoms and metabolic profile: a crossover, randomized, double-blind trial. Int.J.Womens Health. 2016; 8:581-587; PM: 27785106

ABSTRACT: ***OBJECTIVES***: This study was designed to measure the beneficial effects of a combination of nutraceutics (NUT; AkP04, Morestril(R), Akademy Pharma) containing soy isoflavones (80 mg), dry extract of Angelica sinensis (50 mg), dry extract of Morus alba leaf (200 mg) and magnesium (56.25 mg) in the relief of somatic, psychological, and urogenital symptoms in postmenopausal patients, using the validated Menopause Rating Scale (MRS) and cardiovascular risk factors. ***MATERIALS AND METHODS***: A total of 43 symptomatic postmenopausal women (MRS >/=20) were enrolled in a crossover trial. After a 2-week run-in period, patients were randomized into two arms. One arm received probiotics plus placebo over 4 weeks, followed by a 4-week treatment with probiotics plus NUT. The second arm received probiotics plus NUT for 4 weeks, followed by a 4-week treatment with probiotics plus placebo. ***RESULTS***: After the NUT period, participants showed a significant reduction in MRS score (18.4+/-5.4) in comparison to baseline (28.4+/-5) and the placebo period (28+/-5.2) (P<0.0001 for both comparisons). Furthermore, at the end of the active-treatment period, we observed a significant reduction in triglycerides, total and low-density lipoprotein cholesterol plasma levels and

an increase in high-density lipoprotein cholesterol plasma concentration versus baseline and versus placebo (all P<0.04). Moreover, during the NUT period, we detected a significant reduction in diastolic blood pressure in comparison to baseline, but not in comparison to the placebo period. ***CONCLUSION***: This NUT combination was effective for the relief of menopause symptoms in postmenopausal patients and improved their cardiovascular risk profile.

Truong Thy, H. T., Tri, N. N., Quy, O. M., Kannika, K., Unajak, S., and Areechon, N. Effects of the dietary supplementation of mixed probiotic spores of Bacillus amyloliquefaciens 54A, and Bacillus pumilus 47B on growth, innate immunity and stress responses of striped catfish (Pangasianodon hypophthalmus). Fish.Shellfish.Immunol. 11-8-2016; PM: 27836719

ABSTRACT: The study used the mixed probiotics of Bacillus amyloliquefaciens 54A and B. pumilus 47B isolated from striped catfish (Pangasianodon hypophthalmus) intestine aiming to stimulate growth performance, innate immunity, stress tolerance of striped catfish. The average weight gain (AWG), specific growth rate (SGR), and feed conversion ratio (FCR) were analyzed after fish were fed the mixture of probiotics (B. amyloliquefaciens 54A and B. pumilus 47B) at concentrations of 1 x 108, 3 x 108, and 5 x 108 CFU g-1 feed for 90 days. Immunity parameters, survival rate of fish challenged with Edwardsiella ictaluri and ammonia tolerance were also investigated. The amounts of B. amyloliquefaciens and B. pumilus were counted and identified by specific primer pairs of Ba1-F/Ba1-R, and 16-F/Bpu-R to confirm the presence of probiotics in fish intestine. The AWG (476.6 +/- 7.81 g fish-1) of fish fed probiotics at 5 x 108 CFU g-1 was significant higher than the control (390 +/- 25.7 g fish-1) after 90 days of feeding, but there was no significant (P > 0.05) effect of probiotics on FCR and SGR. Fish fed diet containing probiotics at 5 x 108 CFU g-1 also expressed resistance to E. ictaluri infection and higher immune parameters such as phagocytic activity, respiratory bursts, and lysozyme activity than the control. Stress response with ammonia showed significantly lower mortality rate (25%, 20% and 27%) of fish fed probiotics at all three levels of 1, 3 and 5 x 108 CFU g-1 than the fish fed control diet (75%). The study also demonstrated that the probiotics survived in the intestine of striped catfish after 90 days of feeding. Therefore, the dietary supplementation of a mixture of B. amyloliquefaciens and B. pumilus at 5 x 108 CFU g-1 can be used to improve the health and growth rate of striped catfish.

Tursi, A., Picchio, M., Elisei, W., Di, Mario F., Scarpignato, C., and Brandimarte, G. Current Management of Patients With Diverticulosis and Diverticular Disease: A Survey From the 2nd International Symposium on Diverticular Disease. J.Clin.Gastroenterol. 2016; 50 Suppl 1:S97-S100; PM: 27622380

ABSTRACT: ***BACKGROUND***: Management of diverticular disease (DD) remains a point of debate. *GOALS*: To investigate the current opinion of participants of the 2nd International Symposium on Diverticular Disease, on real-life management of patients with DD of the colon. ***STUDY***: Twelve questions were aimed at the diagnosis, treatment, and management options for diverticulosis and symptomatic DD. ***RESULTS***: In total, 115 surveys from 8 European Countries were filled out. High fiber diet was widely prescribed in diverticulosis (59.1%). Probiotics (25%) were the most frequent prescribed drug, whereas 29.8% of participants did not prescribe any treatment in diverticulosis. Colonoscopy was frequently prescribed in symptomatic patients (69.3%), whereas 72.9% of participants did not prescribe any instrumental tool in their follow-up. Rifaximin, probiotics, and mesalazine were the most frequent prescribed drugs both in symptomatic patients (28.1,

14.9%, and 11.4%, respectively) and to prevent recurrence of the disease (42.5%, 12.4%, and 28.2%, respectively). With respect to laboratory exams, 57.9% of participants prescribed them during follow-up. The majority of participants (64.9%) managed suspected acute diverticulitis at home. Rifaximin, probiotics, and mesalazine were the most frequent prescribed drugs to prevent recurrence of the disease (32.2%, 13.2%, and 11.4%, respectively), whereas 25.4% of participants did not prescribe any drugs. Finally, no differences were found among gastroenterologists, surgeons, and general practitioners in managing this disease. ***CONCLUSIONS***: This surveys shows that current management of DD is similar between different medical specialities, generally in line with current literature.

Tyler, H. L., Haron, M. H., Pugh, N. D., Zhang, J., Jackson, C. R., and Pasco, D. S. Bacterial components are the major contributors to the macrophage stimulating activity exhibited by extracts of common edible mushrooms. Food Funct. 10-12-2016; 7(10):4213-4221; PM: 27711863

ABSTRACT: Recent studies have indicated that a major contributor to the innate immune enhancing properties of some medicinal plants is derived from the cell wall components of bacteria colonizing these plants. The purpose of the current study was to assess if the bacteria present within edible and medicinal mushrooms substantially contribute to the innate immune stimulating potential of these mushrooms. Whole mushrooms from thirteen types of edible fungi and individual parts from Agaricus bisporus were analyzed for in vitro macrophage activation as well as bacterial lipopolysaccharides (LPS) content, cell load, and community composition. Substantial variation between samples was observed in macrophage activation (over 500-fold), total bacterial load (over 200-fold), and LPS content (over 10 million-fold). Both LPS content ($rho = 0.832$, $p < 0.0001$) and total bacterial load ($rho = 0.701$, $p < 0.0001$) correlated significantly with macrophage activation in the whole mushroom extracts. Extract activity was negated by treatment with NaOH, conditions that inactivate LPS and other bacterial components. Significant correlations between macrophage activation and total bacterial load ($rho = 0.723$, $p = 0.0001$) and LPS content ($rho = 0.951$, $p < 0.0001$) were also observed between different tissues of Agaricus bisporus. Pseudomonas and Flavobacterium were the most prevalent genera identified in the different tissue parts and these taxa were significantly correlated with in vitro macrophage activation ($rho = 0.697$, $p < 0.0001$ and $rho = 0.659$, $p = 0.0001$, respectively). These results indicate that components derived from mushroom associated bacteria contribute substantially to the innate immune enhancing activity exhibited by mushrooms and may result in similar therapeutic actions as reported for ingestion of bacterial preparations such as probiotics.

Ulbricht, K., Layer, P., and Andresen, V. [Chronic, non-infectious diarrhea: diagnostics and therapy]. Dtsch.Med.Wochenschr. 2016; 141(19):1395-1402; PM: 27642742

ABSTRACT: Chronic, non-infectious diarrhea can be caused by a variety of gastrointestinal diseases. In anamnesis, it is important to take accompanying warning symptoms and specific triggers into account. The fecal inflammatory marker calprotectin may help differentiating between organic and functional gastrointestinal disorders, but it is not specific. Among other options, gelling fibres, Loperamide and Cholestyramine as well as probiotics are available for the symptomatic treatment of chronic diarrhea. For long-term treatment of chronic diarrhea with the enkephalinase inhibitor racecadotril, which is approved for acute diarrhea, only limited data are available. Eluxadolin

presents a new therapeutic option. It can alleviate abdominal pain and diarrhea by modulation of opioid receptors in the enteric nervous system. Additional approaches in intractable irritable bowel syndrome with diarrhea (IBS-D) include 5-HT3 receptor antagonists, the antibiotic Rifaximin as well as low-dose tricyclic antidepressants. Specific diets such as the low-FODMAP diet can also relieve symptoms in IBS.

Umbrello, G. and Esposito, S. Microbiota and neurologic diseases: potential effects of probiotics. J.Transl.Med. 10-19-2016; 14(1):298; PM: 27756430

ABSTRACT: ***BACKGROUND***: The microbiota colonizing the gastrointestinal tract have been associated with both gastrointestinal and extra-gastrointestinal diseases. In recent years, considerable interest has been devoted to their role in the development of neurologic diseases, as many studies have described bidirectional communication between the central nervous system and the gut, the so-called "microbiota-gut-brain axis". Considering the ability of probiotics (i.e., live non-pathogenic microorganisms) to restore the normal microbial population and produce benefits for the host, their potential effects have been investigated in the context of neurologic diseases. The main aims of this review are to analyse the relationship between the gut microbiota and brain disorders and to evaluate the current evidence for the use of probiotics in the treatment and prevention of neurologic conditions. ***DISCUSSION***: Overall, trials involving animal models and adults have reported encouraging results, suggesting that the administration of probiotic strains may exert some prophylactic and therapeutic effects in a wide range of neurologic conditions. Studies involving children have mainly focused on autism spectrum disorder and have shown that probiotics seem to improve neuro behavioural symptoms. However, the available data are incomplete and far from conclusive. ***CONCLUSIONS***: The potential usefulness of probiotics in preventing or treating neurologic diseases is becoming a topic of great interest. However, deeper studies are needed to understand which formulation, dosage and timing might represent the optimal regimen for each specific neurologic disease and what populations can benefit. Moreover, future trials should also consider the tolerability and safety of probiotics in patients with neurologic diseases.

Underwood, M. A. Impact of probiotics on necrotizing enterocolitis. Semin.Perinatol. 11-8-2016; PM: 27836423

ABSTRACT: A large number of randomized placebo-controlled clinical trials and cohort studies have demonstrated a decrease in the incidence of necrotizing enterocolitis with administration of probiotic microbes. These studies have prompted many neonatologists to adopt routine prophylactic administration of probiotics while others await more definitive studies and/or probiotic products with demonstrated purity and stable numbers of live organisms. Cross-contamination and inadequate sample size limit the value of further traditional placebo-controlled randomized controlled trials. Key areas for future research include mechanisms of protection, optimum probiotic species or strains (or combinations thereof) and duration of treatment, interactions between diet and the administered probiotic, and the influence of genetic polymorphisms in the mother and infant on probiotic response. Next generation probiotics selected based on bacterial genetics rather than ease of production and large cluster-randomized clinical trials hold great promise for NEC prevention.

Unterweger, C., Kahler, A., Gerlach, G. F., Viehmann, M., von, Altrock A., and Hennig-Pauka, I. Administration of non-pathogenic isolates of Escherichia coli and Clostridium perfringens type A to piglets in a herd affected with a high incidence of neonatal diarrhoea. Animal. 8-30-2016; 1-7; PM: 27574018

ABSTRACT: A bacterial cocktail of living strains of Clostridium perfringens type A (CPA) without beta2-toxin gene and non-pathogenic Escherichia coli was administered orally to newborn piglets before first colostrum intake and on 2 consecutive days on a farm with a high incidence of diarrhoea and antibiotic treatment in suckling piglets associated with E. coli and CPA. This clinical field study was driven by the hypothetic principle of competitive exclusion of pathogenic bacteria due to prior colonization of the gut mucosal surface by non-pathogenic strains of the same bacterial species with the aim of preventing disease. Although CPA strains used in this study did not produce toxins in vitro, their lack of pathogenicity cannot be conclusively confirmed. The health status of the herd was impaired by a high incidence of postpartum dysgalactia syndrome in sows (70%) and a high incidence of neonatal diarrhoea caused by enterotoxigenic E. coli and CPA during the study. No obvious adverse effect of the bacterial treatment occurred. On average, more piglets were weaned in litters treated (P=0.009). Visual pathological alterations in the small intestinal wall were more frequent in dead piglets of the control group (P=0.004) and necrotizing enteritis was only found in that group. A higher average daily weight gain of piglets in the control group (P<0.001) may be due to an increased milk uptake due to less competition in the smaller litters. The bacterial cocktail was tested under field conditions for its potential to stabilize gut health status in suckling piglets before disease development due to colibacillosis and clostridial infections; however, the gut flora stabilizing effect of the bacterial cocktail was not clearly discernible in this study. Further basic research is needed to confirm the positive effects of the bacterial treatment used and to identify additional potential bacterial candidates for competitive exclusion.

Valdes-Varela, L., Ruas-Madiedo, P., and Gueimonde, M. In vitro fermentation of different fructo-oligosaccharides by Bifidobacterium strains for the selection of synbiotic combinations. Int.J.Food Microbiol. 11-12-2016; 242:19-23; PM: 27866040

ABSTRACT: The use of selected probiotics, prebiotics and/or synbiotics, constitute an interesting dietary strategy for intestinal microbiota modulation in case of dysbiosis. Species of the genus Bifidobacterium are among the most currently used probiotics for human consumption since they have shown beneficial effects in the prevention and treatment of some disorders. Bifidobacteria are saccharolytic microorganisms, but their ability to use different carbohydrates varies among strains. In this study, we investigate the utilization of three prebiotic substrates (two different short-chain fructo-oligosaccharides [scFOS] and inulin) by strains of Bifidobacterium, in order to determine the synbiotic potential of the different probiotic/prebiotic combinations. Batch culture fermentations from six Bifidobacterium strains (Bifidobacterium longum IPLA20021, B. longum IPLA20022, Bifidobacterium animalis IPLA20031, B. animalis IPLA20032, B. animalis IPLA20020 and B. animalis Bb12) were carried out in the presence of inulin or scFOS (Synergy or Actilight), or glucose, as carbon source. Bifidobacteria levels were quantified by plate counting. The pH and production of organic acids in the different batch-culture fermentations were also determined. Our results showed that all the studied strains of B. animalis and B. longum were able to utilize scFOS but not inulin. The use of scFOS as carbon source affected the pattern of metabolite's production, when compared with cultures carried out in glucose, particularly in the case of B. longum. The results indicated

that the scFOS are well suited to be used in combination with B. animalis or B. longum strains for the development of synbiotic foods or food supplements.

Valeriano, V. D., Balolong, P., and Kang, D. K. Probiotic Roles of Lactobacillus spp. in Swine: Insights from Gut Microbiota. J.Appl.Microbiol. 12-3-2016; PM: 27914202

ABSTRACT: The use of lactobacilli as probiotics in swine has been gaining attention due to their ability to improve growth performance and carcass quality, prevent gastrointestinal infection, and most importantly, their 'generally recognised as safe' status. Previous studies support the potential of lactobacilli to regulate host immune systems, enhance gut metabolic capacities, and maintain balance in the gut microbiota. Research on swine gut microbiota has revealed complex gut microbial community structure and showed the importance of Lactobacillus to the host's health. However, the species- and strain-specific characteristics of lactobacilli that confer probiotic benefits are still not well understood. The diversity of probiotic traits in a complex gut ecosystem makes it challenging to infer the relationships between specific functions of Lactobacillus spp. and host health. In this review, we provide an overview of how lactobacilli play a pivotal role in the swine gut ecosystem and identify key characteristics that influence gut microbial community structure and the health of pigs. In addition, based on recent and on-going meta-omics and omics research on the gut microbiota of pigs, we suggest a workflow combining culture-dependent and culture-independent approaches for more effective selection of probiotic lactobacilli. This article is protected by copyright. All rights reserved.

Van Beek, A. A., Sovran, B., Hugenholtz, F., Meijer, B., Hoogerland, J. A., Mihailova, V., van der, Ploeg C., Belzer, C., Boekschoten, M. V., Hoeijmakers, J. H., Vermeij, W. P., De, Vos P., Wells, J. M., Leenen, P. J., Nicoletti, C., Hendriks, R. W., and Savelkoul, H. F. Supplementation with Lactobacillus plantarum WCFS1 Prevents Decline of Mucus Barrier in Colon of Accelerated Aging Ercc1-/Delta7 Mice. Front Immunol. 2016; 7:408; PM: 27774093

ABSTRACT: Although it is clear that probiotics improve intestinal barrier function, little is known about the effects of probiotics on the aging intestine. We investigated effects of 10-week bacterial supplementation of Lactobacillus plantarum WCFS1, Lactobacillus casei BL23, or Bifidobacterium breve DSM20213 on gut barrier and immunity in 16-week-old accelerated aging Ercc1-/Delta7 mice, which have a median lifespan of ~20 weeks, and their wild-type littermates. The colonic barrier in Ercc1-/Delta7 mice was characterized by a thin (< 10 mum) mucus layer. L. plantarum prevented this decline in mucus integrity in Ercc1-/Delta7 mice, whereas B. breve exacerbated it. Bacterial supplementations affected the expression of immune-related genes, including Toll-like receptor 4. Regulatory T cell frequencies were increased in the mesenteric lymph nodes of L. plantarum- and L. casei-treated Ercc1-/Delta7 mice. L. plantarum- and L. casei-treated Ercc1-/Delta7 mice showed increased specific antibody production in a T cell-dependent immune response in vivo. By contrast, the effects of bacterial supplementation on wild-type control mice were negligible. Thus, supplementation with L. plantarum - but not with L. casei and B. breve - prevented the decline in the mucus barrier in Ercc1-/Delta7 mice. Our data indicate that age is an important factor influencing beneficial or detrimental effects of candidate probiotics. These findings also highlight the need for caution in translating beneficial effects of probiotics observed in young animals or humans to the elderly.

van der Meulen, T. A., Harmsen, H., Bootsma, H., Spijkervet, F., Kroese, F., and Vissink, A. The microbiome-systemic diseases connection. Oral Dis. 2016; 22(8):719-734; PM: 26953630

ABSTRACT: The human microbiome consists of all microorganisms occupying the skin, mucous membranes and intestinal tract of the human body. The contact of the mucosal immune system with the human microbiome is a balanced interplay between defence mechanisms of the immune system and symbiotic or pathogenic microbial factors, such as microbial antigens and metabolites. In systemic autoimmune diseases (SADs) such as rheumatoid arthritis, systemic lupus erythematosus and Sjogren's syndrome, the immune system is deranged to a chronic inflammatory state and autoantibodies are an important hallmark. Specific bacteria and/or a dysbiosis in the human microbiome can lead to local mucosal inflammation and increased intestinal permeability. Proinflammatory lymphocytes and cytokines can spread to the systemic circulation and increase the risk of inflammation at distant anatomical sites, such as the joints or salivary glands. Increased intestinal permeability increases antigen exposure and the risk of autoantibody production. If the human microbiome indeed plays such a critical role in SADs, this finding holds a great promise for new therapeutic strategies, such as diet interventions and probiotics and prebiotics. This review provides a background on the human microbiome and mucosal immunity in the gut and oral cavity and gives a summary of the current knowledge on the microbiome-SADs connection.

Vandenplas, Y., Alturaiki, M. A., Al-Qabandi, W., AlRefaee, F., Bassil, Z., Eid, B., El, Beleidy A., Almehaidib, A. I., Mouawad, P., and Sokhn, M. Middle East Consensus Statement on the Diagnosis and Management of Functional Gastrointestinal Disorders in <12 Months Old Infants. Pediatr.Gastroenterol.Hepatol.Nutr. 2016; 19(3):153-161; PM: 27738596

ABSTRACT: This paper covers algorithms for the management of regurgitation, constipation and infantile colic in infants. Anti-regurgitation formula may be considered in infants with troublesome regurgitation, while diagnostic investigations or drug therapy are not indicated in the absence of warning signs. Although probiotics have shown some positive evidence for the management of functional gastrointestinal disorders (FGIDs), the evidence is not strong enough to make a recommendation. A partially hydrolyzed infant formula with prebiotics and beta-palmitate may be considered as a dietary intervention for functional constipation in formula fed infants. Lactulose has been shown to be effective and safe in infants younger than 6 months that are constipated. Macrogol (polyethylene glycol, PEG) is not approved for use in infants less than 6 months of age. However, PEG is preferred over lactulose in infants >6 months of age. Limited data suggests that infant formula with a partial hydrolysate, galacto-oligosaccharides/fructo-oligosaccharides, added beta-palmitate may be of benefit in reducing infantile colic in formula fed infants in cases where cow's milk protein allergy (CMPA) is not suspected. Evidence suggests that the use of extensively hydrolyzed infant formula for a formula-fed baby and a cow's milk free diet for a breastfeeding mother may be beneficial to decrease infantile colic if CMPA is suspected. None of the FGIDs is a reason to stop breastfeeding.

Vidal, S., Tapia-Paniagua, S. T., Morinigo, J. M., Lobo, C., Garcia de, la Banda, I, Balebona, M. D., and Morinigo, M. A. Effects on intestinal microbiota and immune genes of Solea senegalensis after suspension of the administration of Shewanella putrefaciens Pdp11. Fish.Shellfish.Immunol. 9-10-2016; 58:274-283; PM: 27623340

ABSTRACT: The interaction host-intestinal microbiota is essential for the immunological homeostasis of the host. Probiotics, prebiotics and synbiotics are promising tools for the manipulation of the intestinal microbiota towards beneficial effects to the host. The objective of this study was to evaluate the modulation effect on the intestinal microbiota and the transcription of genes involved in the immune response in head kidney of Solea senegalensis after administration of diet supplemented with the prebiotic alginate and the probiotic Shewanella putrefaciens Pdp11 CECT 7627 (SpPdp11). The results showed higher adaptability to dietary changes in the intestinal microbiota of fish fed diet with alginate and SpPdp11 together compared to those fish that received an alginate-supplemented diet. The alginate-supplemented diet produced up-regulation of genes encoding proteins involved in immunological responses, such as complement, lysozyme G and transferrin, and oxidative stress, such as NADPH oxidase and glutation peroxidase. On the other hand, the administration of alginate combined with SpPdp11 resulted in a significant increase of the transcription of genes encoding for glutation peroxidase and HSP70, indicating a potential protective effect of SpPdp11 against oxidative stress. In addition, these effects were maintained after the suspension of the probiotic treatment. The relationship between the modulation of the intestinal microbiota and the expression of genes with protective effect against the oxidative stress was demonstrated by the Principal Components Analysis.

Vinogradov, E., Sadovskaya, I., Grard, T., and Chapot-Chartier, M. P. Structural studies of the rhamnose-rich cell wall polysaccharide of Lactobacillus casei BL23. Carbohydr.Res. 11-29-2016; 435:156-161; PM: 27756016

ABSTRACT: Lactobacillus casei is a Gram positive lactic acid bacterium used in dairy fermentations and present in the normal human gut microbiota. Certain strains are recognized as probiotics with beneficial effects on human and animal health. L. casei BL23 is a potential probiotic strain endowed with anti-inflammatory properties and a model strain widely used in genetic, physiological and biochemical studies. A number of bacterial cell surface polysaccharides have been shown to play a role in the immune modulation activities observed for probiotic lactic acid bacteria. In the present work, we purified the most abundant carbohydrate polymer of L. casei BL23 cell wall, a neutral wall polysaccharide (WPS) and established its chemical structure by periodate oxidation, methylation analysis and 2D NMR spectroscopy. The WPS of L. casei BL23 was shown to contain alpha-Rha, alpha-Glc, beta-GlcNAc and beta-GalNAc forming a branched heptasaccharide repeating unit (variant 1) with an additional partial substitution with alpha-Glc (variant 2). A modified non-reducing end octasaccharide, corresponding to a terminal unit of the WPS (variant 3), was also identified and allowed to define the biological repeating unit of the WPS. To our knowledge, this is the first report of the identification of a biological repeating unit based on a chemical evidence, in a cell wall polysaccharide of a Gram positive bacterial species.

Viswanathan, S., Lau, C., Akbari, H., Hoyen, C., and Walsh, M. C. Survey and evidence based review of probiotics used in very low birth weight preterm infants within the United States. J.Perinatol. 2016; 36(12):1106-1111; PM: 27583387

ABSTRACT: ***OBJECTIVE***: Data from multiple clinical trials, mostly conducted outside the US, indicate that probiotic prophylaxis is an effective intervention for prevention of necrotizing enterocolitis (NEC) in preterm infants. Probiotics are routinely used in many countries. However, in the US, probiotic use in preterm infants is limited (6.7% of very low

birth weight (VLBW) infants in the US were exposed to probiotics in 2014, Vermont Oxford Network (VON)). Probiotic products are often considered in 'generic' terms, but considerable variation exists between commercially available probiotics in terms of their quantity and quality. The study objectives were to identify the probiotic products used in VLBW infants within the US, and to determine whether their use was supported by reliable evidence. ***STUDY DESIGN***: A phone survey of all neonatal intensive care units (NICU) participating in VON within the US was conducted between May and September 2015 to identify NICUs that are using probiotics in VLBW infants. Data was collected regarding specific probiotic brands, timing, dose and duration of probiotic use. An evidence based literature search, limited to randomized controlled trials in VLBW infants, was conducted to determine whether the use of identified probiotics were supported by reliable evidence. ***RESULTS***: There was a 70.3% (500/711) response rate to the phone survey. During the survey period, 14.0% of NICUs were using probiotics in VLBW infants (70/500). Probiotics were routinely given to all VLBW infants in 8.8% (44/500) NICUs, while it was given in selected VLBW infants in 5.2% (26/500) of NICUs. The common indications for selective use of probiotics were feeding intolerance and antibiotic use. Sixteen commercial probiotics products were identified through the phone survey. Probiotic products most commonly used were Culturelle (27.1%), Biogaia (14.3%), Gerber Soothe (14.3%) and Florababy (8.6%). The literature search identified evidence that evaluated 4/16 probiotic products identified (Culturelle, Align, Biogaia and ABC Dophilus). Only ABC Dophilus was reported to have a protective effect against NEC, but is used sparingly in US NICUs (2.9%). ***CONCLUSIONS***: The probiotics use in VLBW infants within the US is increasing, but is still limited. There was no evidence for safety or efficacy of 90% of the probiotics currently used in US NICUs, and therefore, caution is warranted.

Vitetta, L., Saltzman, E. T., Nikov, T., Ibrahim, I., and Hall, S. Modulating the Gut Micro-Environment in the Treatment of Intestinal Parasites. J.Clin.Med. 11-16-2016; 5(11); PM: 27854317

ABSTRACT: The interactions of micro-organisms cohabitating with Homo sapiens spans millennia, with microbial communities living in a symbiotic relationship with the host. Interacting to regulate and maintain physiological functions and immunological tolerance, the microbial community is able to exert an influence on host health. An example of micro-organisms contributing to an intestinal disease state is exhibited by a biodiverse range of protozoan and bacterial species that damage the intestinal epithelia and are therefore implicated in the symptoms of diarrhea. As a contentious exemplar, Blastocystis hominis is a ubiquitous enteric protist that can adversely affect the intestines. The symptoms experienced are a consequence of the responses of the innate immune system triggered by the disruption of the intestinal barrier. The infiltration of the intestinal epithelial barrier involves a host of immune receptors, including toll like receptors and IgM/IgG/IgA antibodies as well as CD8+ T cells, macrophages, and neutrophils. Whilst the mechanisms of interactions between the intestinal microbiome and protozoan parasites remain incompletely understood, it is acknowledged that the intestinal microbiota is a key factor in the pathophysiology of parasitic infections. Modulating the intestinal environment through the administration of probiotics has been postulated as a possible therapeutic agent to control the proliferation of intestinal microbes through their capacity to induce competition for occupation of a common biotype. The ultimate goal of this mechanism is to prevent infections of the like of giardiasis and eliminate its symptoms. The differing types of probiotics (i.e., bacteria and yeast) modulate immunity by stimulating the host immune system. Early animal studies support the potential benefits of probiotic administration to prevent intestinal infections, with human clinical studies showing probiotics can reduce the number of parasites and the severity of symptoms. The early clinical indications endorse probiotics as adjuncts in the pharmaceutical

treatment of protozoan infections. Currently, the bar is set low for the conduct of well-designed clinical studies that will translate the use of probiotics to ameliorate protozoan infections, therefore the requisite is for further clinical research.

von, Muller L. [New aspects on Clostridium difficile infection]. Dtsch.Med.Wochenschr. 2016; 141(16):1144-1147; PM: 27509341

ABSTRACT: Clostridium difficile infection (CDI) is a frequent and complex disease which is influenced by the repertoire of bacterial virulence factors, by host immunity and by the intestinal microbiome. These complex interaction opens a number of options which may be used for treatment in the future. One example for new treatment options is fecal microbiota transplantation (FMT). Driven by C. difficile related research activities the knowledge of protective microorganism is increasing and it may be assumed that bacteriotherapy by next-generation probiotics may be used very soon also for other diseases. Very often, CDI reflects to the clinician that antibiotic therapy is associated with side effects. Therefore, C. difficile is the guilty conscience which helps to implement targeted and restrictive antibiotic use in the daily practice.

Wan, L. Y., Chen, Z. J., Shah, N. P., and El-Nezami, H. Modulation of Intestinal Epithelial Defense Responses by Probiotic Bacteria. Crit Rev.Food Sci.Nutr. 12-9-2016; 56(16):2628-2641; PM: 25629818

ABSTRACT: Probiotics are live microorganisms, which when administered in food confer numerous health benefits. In previous studies about beneficial effects of probiotic bacteria to health, particularly in the fields of intestinal mucosa defense responses, specific probiotics, in a strain-dependent manner, show certain degree of potential to reinforce the integrity of intestinal epithelium and/or regulate some immune components. The mechanism of probiotic action is an area of interest. Among all possible routes of modulation by probiotics of intestinal epithelial cell-mediated defense responses, modulations of intestinal barrier function, innate, and adaptive mucosal immune responses, as well as signaling pathways are considered to play important role in the intestinal defense responses against pathogenic bacteria. This review summarizes the beneficial effects of probiotic bacteria to intestinal health together with the mechanisms affected by probiotic bacteria: barrier function, innate, and adaptive defense responses such as secretion of mucins, defensins, trefoil factors, immunoglobulin A (IgA), Toll-like receptors (TLRs), cytokines, gut associated lymphoid tissues, and signaling pathways.

Wang, P., Tao, J. H., and Pan, H. F. Probiotic bacteria: a viable adjuvant therapy for relieving symptoms of rheumatoid arthritis. Inflammopharmacology. 2016; 24(5):189-196; PM: 27581587

ABSTRACT: The burgeoning use of probiotics has proliferated during the past two decades. However, the effect of probiotic administration for either the prevention or treatment of rheumatoid arthritis (RA) has been investigated in a limited number of studies. Randomized controlled clinical trials have provided evidences that specific probiotics supplementation exhibit anti-inflammatory effects, help to increase daily activities and alleviate symptoms in patients with RA. Therefore, using probiotic bacteria as an adjuvant therapy may be considered as a promising treatment option for RA. This review summarizes the available data about the therapeutic and preventive effect of

probiotics in RA, together with probiotic supplement as a possible therapy in clinical treatment.

Wang, T. and Su, J. Bacillus subtilis from soybean food shows antimicrobial activity for multidrug-resistant Acinetobacter baumannii by affecting AdeS gene. J.Microbiol.Biotechnol. 9-2-2016; PM: 27586533

ABSTRACT: Exploring novel antibiotics is necessary for multidrug-resistant pathogenic bacteria while the probiotics in soybean food have antimicrobial activities. Thus, we investigated the effects of the probiotics from soybean food on multidrug-resistant Acinetobacter baumannii. Nineteen multidrug-resistant A. baumannii were clinically isolated as an experimental group and 11 multidrug-sensitive strains as controls. The growth rates of all bacteria were determined by using the analysis for xCELLigence Real-Time Cell. The combination of antibiotics showed synergistic effects on the strains in a control group but no effect on the strains in an experimental group. Efflux pump gene adeS is absent in all the strains from the control group, whereas it exists in all the strains from experimental group. Furthermore, all the strains lost multidrug resistance when adeS inhibitor was used. One strain of probiotics isolated from soybean food showed high antimicrobial activity for multidrug-resistant A. baumannii. The isolated strain belongs to Bacillus subtilis according to 16S RNA analysis. Furthermore, E. coli showed multidrug resistance when it was transformed with AdeS gene from A. baumannii while the resistant bacteria could be inhibited completely by isolated Bacillus subtilis. Thus, probiotics from soybean food provides potential antibiotics for multidrug-resistant pathogenic bacteria.

Wassenaar, T. M. Insights from 100 Years of Research with Probiotic E. Coli. Eur.J.Microbiol.Immunol.(Bp). 9-29-2016; 6(3):147-161; PM: 27766164

ABSTRACT: A century ago, Alfred Nissle discovered that intentional intake of particular strains of Escherichia coli could treat patients suffering from infectious diseases. Since then, one of these strains became the most frequently used probiotic E. coli in research and was applied to a variety of human conditions. Here, properties of that E. coli Nissle 1917 strain are compared with other commercially available E. coli probiotic strains, with emphasis on their human applications. A literature search formed the basis of a summary of research findings reported for the probiotics Mutaflor, Symbioflor 2, and Colinfant. The closest relatives of the strains in these products are presented, and their genetic content, including the presence of virulence, genes is discussed. A similarity to pathogenic strains causing urinary tract infections is noticeable. Historic trends in research of probiotics treatment for particular human conditions are identified. The future of probiotic E. coli may lay in what Alfred Nissle originally discovered: to treat gastrointestinal infections, which nowadays are often caused by antibiotic-resistant pathogens.

Watters, C., Fleming, D., Bishop, D., and Rumbaugh, K. P. Host Responses to Biofilm. Prog.Mol.Biol.Transl.Sci. 2016; 142:193-239; PM: 27571696

ABSTRACT: From birth to death the human host immune system interacts with bacterial

cells. Biofilms are communities of microbes embedded in matrices composed of extracellular polymeric substance (EPS), and have been implicated in both the healthy microbiome and disease states. The immune system recognizes many different bacterial patterns, molecules, and antigens, but these components can be camouflaged in the biofilm mode of growth. Instead, immune cells come into contact with components of the EPS matrix, a diverse, hydrated mixture of extracellular DNA (bacterial and host), proteins, polysaccharides, and lipids. As bacterial cells transition from planktonic to biofilm-associated they produce small molecules, which can increase inflammation, induce cell death, and even cause necrosis. To survive, invading bacteria must overcome the epithelial barrier, host microbiome, complement, and a variety of leukocytes. If bacteria can evade these initial cell populations they have an increased chance at surviving and causing ongoing disease in the host. Planktonic cells are readily cleared, but biofilms reduce the effectiveness of both polymorphonuclear neutrophils and macrophages. In addition, in the presence of these cells, biofilm formation is actively enhanced, and components of host immune cells are assimilated into the EPS matrix. While pathogenic biofilms contribute to states of chronic inflammation, probiotic Lactobacillus biofilms cause a negligible immune response and, in states of inflammation, exhibit robust antiinflammatory properties. These probiotic biofilms colonize and protect the gut and vagina, and have been implicated in improved healing of damaged skin. Overall, biofilms stimulate a unique immune response that we are only beginning to understand.

Watts, A. M., West, N. P., Smith, P. K., Cripps, A. W., and Cox, A. J. Probiotics and Allergic Rhinitis: A Simon Two-Stage Design to Determine Effectiveness. J.Altern.Complement Med. 10-12-2016; PM: 27732056

ABSTRACT: ***BACKGROUND***: Allergic rhinitis (AR) is a chronic upper respiratory disease affecting 10-30% of the population worldwide. It associated with significant economic and medical burden. Probiotics have received attention in recent years as a novel strategy to treat infectious/immune conditions, including AR. However, substantiation of these health claims by regulatory bodies has been rejected due, in part, to inadequate clinical trial design. While randomized controlled trials are considered the gold standard for assessing clinical efficacy, such trials require a priori preclinical data on effect size, which may be a reason for the conflicting results in the probiotic and AR literature. Progressive clinical trial designs, such as the Simon Two-Stage Design, are showing promise within the area of integrative and alternative medicine, particularly in relation to probiotic supplementation, to obtain empirical data for the design of clinical trials that meet regulatory requirements. ***METHODS***: This Phase II study uses a Simon Two-Stage Design to determine the response rate of patients with AR to a probiotic supplement. Patients will consume a multispecies probiotic twice daily for 8 weeks, and will attend an allergy clinic at the beginning and end of the intervention period for assessment. Symptom improvement following probiotic supplementation will be measured by the mini-Rhinoconjunctivitis Quality of Life Questionnaire. Secondary outcomes include twice-weekly symptom and medication diaries, objective determination of nasal congestion via Nasal Rhinomanometry, and change in frequency of medication usage. ***DISCUSSION***: This study provides an exemplar of the value of using a progressive study design in the complementary and alternative medicine setting. A Simon Two-Stage Design was adopted to investigate whether a multispecies probiotic supplement, not yet trialed in the context of AR, has promise as a therapeutic intervention and warrants the design of larger placebo-controlled studies.

Weiner, J. P., Wong, A. T., Schwartz, D., Martinez, M., Aytaman, A., and Schreiber, D. Endoscopic and non-endoscopic approaches for the management of radiation-induced rectal bleeding. World J.Gastroenterol. 8-21-2016; 22(31):6972-6986; PM: 27610010

ABSTRACT: Pelvic radiation is a commonly utilized treatment for malignancy of the genitourinary and lower gastrointestinal tract. Radiation proctitis and the resultant clinical picture varies from asymptomatic to potentially life threatening. Similarly, treatment options also vary greatly, from medical therapy to surgical intervention. Commonly utilized medical therapy includes sucralfate enemas, antibiotics, 5-aminosalicylic acid derivatives, probiotics, antioxidants, short-chain fatty acids, formalin instillation and fractionated hyperbaric oxygen. More invasive treatments include endoscopic-based, focally ablative interventions such as dilation, heater and bipolar cautery, neodymium/yttrium aluminum garnet argon laser, radiofrequency ablation or argon plasma coagulation. Despite its relatively common frequency, there is a dearth of existing literature reporting head-to-head comparisons of the various treatment options via a randomized controlled approach. The purpose of our review was to present the reader a consolidation of the existing evidence-based literature with the goal of highlighting the comparative effectiveness and risks of the various treatment approaches. Finally, we outline a pragmatic approach to the treatment of radiation proctitis. In light of the lack of randomized data, our goal is to pursue as least invasive an approach as possible, with escalation of care tailored to the severity of the patient's symptoms. For those cases that are clinically asymptomatic or only mildly symptomatic, observation or medical management can be considered. Once a patient fails such management or symptoms become more severe, invasive procedures such as endoscopically based focal ablation or surgical intervention can be considered. Although not all recommendations are supported by level I evidence, reported case series and single-institutional studies in the literature suggest that successful treatment with cessation of symptoms can be obtained in the majority of cases.

Wu, C. T., Chen, P. J., Lee, Y. T., Ko, J. L., and Lue, K. H. Effects of immunomodulatory supplementation with Lactobacillus rhamnosus on airway inflammation in a mouse asthma model. J.Microbiol.Immunol.Infect. 2016; 49(5):625-635; PM: 25440975

ABSTRACT: ***BACKGROUND***: Asthma is a common allergic disease. In previous studies, probiotics improved the balance of intestinal microbes, reduced inflammation, and promoted mucosal tolerance. This study investigated whether oral administrations of Lactobacillus rhamnosus GG (LGG) inhibited allergen (ovalbumin or OVA)-induced airway inflammation in a mouse asthma model. **METHODS**: The allergy/asthma animal model in this study was sensitization with OVA. After intranasal challenge with OVA, the airway inflammation and hyper-responsiveness were determined by a Buxco system, bronchoalveolar lavage fluid analysis with Liu stain, and enzyme-linked immunosorbent assay. Histopathologic changes in the lung were detected by hematoxylin and eosin staining and immunohistochemistry staining. **RESULTS**: Both pre- and post-treatment with LGG suppressed the airway hyper-responsiveness to methacholine and significantly decreased the number of infiltrating inflammatory cells and Th2 cytokines in bronchoalveolar lavage fluid and serum compared with the OVA-sensitized mice. In addition, LGG reduced OVA-specific IgE levels in serum. Oral LGG decreased matrix metalloproteinase 9 expression in lung tissue and inhibited inflammatory cell infiltration. **CONCLUSION**: LGG had an anti-inflammatory effect on OVA-induced airway

inflammation and might be an additional or supplementary therapy for allergic airway diseases.

Wu, M., Wu, Y., Deng, B., Li, J., Cao, H., Qu, Y., Qian, X., and Zhong, G. Isoliquiritigenin decreases the incidence of colitis-associated colorectal cancer by modulating the intestinal microbiota. Oncotarget. 11-15-2016; PM: 27863401

ABSTRACT: Imbalances in intestinal bacteria correlate with colitis-associated colorectal cancer (CAC). Traditional Chinese medicines have been used to adjust the gut microbiota, and isoliquiritigenin (ISL), a flavonoid extracted from licorice, has shown antitumor efficacy. In this study, the effects of ISL on CAC development and the gut microbiota were evaluated using an azoxymethane and dextran sulphate sodium (AOM/DSS)-induced mouse model of CAC (CACM). Histopathological analysis suggested that ISL reduced tumor incidence in vivo. Moreover, high-throughput sequencing and terminal restriction fragment length polymorphism (T-RFLP) studies of the bacterial 16S rRNA gene revealed that the structure of the gut microbial community shifted significantly following AOM/DSS treatment, and that effect was alleviated by treatment with high-dose ISL (150 mg/kg). Compared to the microbiota in the control mice (CK), the levels of Bacteroidetes decreased and the levels of Firmicutes increased during CAC development. ISL reversed the imbalance at the phylum level and altered the familial constituents of the gut microbiota. Specifically, the abundance of Helicobacteraceae increased after treatment with high-dose ISL, while the abundance of Lachnospiraceae and Rikenellaceae decreased. At the genus level, ISL reduced the abundance of opportunistic pathogens (Escherichia and Enterococcus), and increased the levels of probiotics, particularly butyrate-producing bacteria (Butyricicoccus, Clostridium, and Ruminococcus). Thus, ISL protects mice from AOM/DSS-induced CAC, and ISL and the gut microbiota may have synergistic anti-cancer effects.

Wu, X. D., Liu, M. M., Liang, X., Hu, N., and Huang, W. Effects of perioperative supplementation with pro-/synbiotics on clinical outcomes in surgical patients: A meta-analysis with trial sequential analysis of randomized controlled trials. Clin.Nutr. 10-26-2016; PM: 27836310

ABSTRACT: ***BACKGROUND & AIMS***: The Potential benefits and possible risks of perioperative supplementation with probiotics/synbiotics in surgical patients are not fully understood. Recent evidence has rapidly evolved and conveys conflicting results. Thus, we undertook a meta-analysis of randomized controlled trials (RCTs) to valuate the effectiveness, safety, cost-effectiveness and quality of life of perioperative supplementation with pro-/synbiotics. ***METHODS***: We systematically searched PubMed, Embase and the Cochrane Library through October 2015 to identify RCTs that assessed the effects of perioperative supplementation with pro-/synbiotics in surgical patients. The predefined primary efficacy outcome was surgical site infection (SSI). Random-effects model was applied to pool outcome data accounting for clinical heterogeneity. ***RESULTS***: Our meta-analysis included data from 34 trials comprising 2634 participants, of whom 1300 received perioperative pro-/synbiotics intervention and 1334 received valid control treatment. Compared with the control group, patients in the pro-/synbiotics group had a lower risk of SSI (relative risk: 0.65; 95% confidence interval: 0.51, 0.84; P = 0.0007). Trial sequential analysis confirmed the evidence was sufficient and conclusive. Subgroup analyses indicated the findings were consistent in all subgroup analyses except for the probiotics, enteral feeding, pre-/postoperative and live transplantation subgroups. Pro-/synbiotics also reduced the incidence of other infectious complications

(including any infection, pneumonia, urinary tract infection, wound infection and sepsis); shortened antibiotic therapy, intensive care unit stay and hospital stay; and promoted earlier first defecation and first bowel movement. Pro-/synbiotics further reduced the incidence of abdominal side effects, lowered hospital costs and improved the Gastro-Intestinal Quality of Life. ***CONCLUSIONS***: For surgical patients, perioperative supplementation with pro-/synbiotics is effective in preventing or controlling SSI and other infectious complications. Perioperative pro-/synbiotics might also be associated with fewer side effects, lower hospital cost and better quality of life. Current evidence indicated that perioperative synbiotics supplementation is preferred and recommended as an adjunct in surgical patients.

Yadav, R., Puniya, A. K., and Shukla, P. Probiotic Properties of Lactobacillus plantarum RYPR1 from an Indigenous Fermented Beverage Raabadi. Front Microbiol. 2016; 7:1683; PM: 27818658

ABSTRACT: Present study documents the potential probiotic Lactobacillus isolated from indigenous fermented beverage Raabadi, consumed during summers in Haryana and Rajasthan regions of India. A total of five Raabadi samples were collected aseptically and 54 isolates were purified using MRS medium. All the isolates were assessed for tolerance to low pH and bile salts. It was observed that out of 54 only 24 isolates could survive the simulated gastric conditions. These isolates were further evaluated in vitro for cell surface hydrophobicity, cell surface hydrophobicity, hypocholesteramic activity, anti-oxidative potential, BSH activity, antagonistic activity, and antibiotic resistance profile. In addition, the confirmation of phenol resistance was also done. On the basis of results obtained, the survival rate of isolates was noted and six isolates were finally selected for further studies. Among them Lactobacillus plantarum RYPR1 and RYPC7 showed good survival at pH 2 which shows good acid tolerance. Moreover, L. plantarum RYPR1 showed the highest hydrophobicity (79.13%) and represented the deconjugation of bile salts, which help in their adhesion to epithelial cells and colonization. Furthermore, RYPR1 also exhibited highest cholesterol reduction (59%) and subsequent analysis of results revealed that the above mentioned isolates further exhibit a good hypocholesterolemic effect and could be possibly used to prevent hypercholesterolemia. The present study divulges that L. plantarum RYPR1 has an excellent probiotic potential.

Yadav, R., Singh, P. K., and Shukla, P. Metabolic engineering for probiotics and their genome-wide expression profiling. Curr.Protein Pept.Sci. 11-11-2016; PM: 27834122

ABSTRACT: Probiotic supplements in food industry have attracted a lot of attention and shown a remarkable growth in this field. Metabolic engineering (ME) approaches enable understanding their mechanism of action and increases possibility of designing probiotic strains with desired functions. Probiotic microorganisms generally referred as industrially important lactic acid bacteria (LAB) which are involved in fermenting dairy products, food, beverages and produces lactic acid as final product. A number of illustrations of metabolic engineering approaches in industrial probiotic bacteria have been described in this review including Transcriptomic studies of Lactobacillus reuteri and improvement in exopolysaccharide (EPS) biosynthesis yield in Lactobacillus casei LC2W. This review summaries various metabolic engineering approaches for exploring metabolic pathways. These approaches enable evaluation of cellular metabolic state and effective editing of microbial genome or introduction of novel enzymes to redirect the

carbon fluxes. In addition, various system biology tools such as in silico design commonly used for improving strain performance is also discussed. Finally, we discuss the integration of metabolic engineering and genome profiling which offers a new way to explore metabolic interactions, fluxomics and probiogenomics using probiotic bacteria like Bifidobacterium spp and Lactobacillus spp.

Yan, X., Feng, B., Li, P., Tang, Z., and Wang, L. Microflora Disturbance during Progression of Glucose Intolerance and Effect of Sitagliptin: An Animal Study. J.Diabetes Res. 2016; 2016:2093171; PM: 27631013

ABSTRACT: ***Background***: Emerging evidences have shown a close interplay between obesity, diabetes, and intestinal flora disturbance. Dipeptidyl peptidase-4 inhibitor, exemplified by sitagliptin, is highly efficacious in treating type 2 diabetes (T2DM), yet little is known if sitagliptin exerts beneficial effects on microbiota associated with obesity and T2DM. We evaluated changes of gut microbiota following the induction of obesity and T2DM in a streptozotocin treated high fat/high carbohydrate fed (HF/HC-STZ) rat model and explored the effect of sitagliptin on gut microbiota for HF/HC-STZ rats. *Methods*: Sitagliptin was administered via oral gavage to diabetic rats. Fecal DNA extraction and 454 pyrosequencing based on analysis of 16S rRNA genes was utilized to determine the overall structure of microbiota in fecal DNA samples. ***Results***: Results showed that, at the level of phylum, there was higher abundance of Firmicutes and Tenericutes and less abundance of Bacteroidetes in obese rats compared to their lean counterparts. At the level of genus, short-chain fatty acid- (SCFA-) producing bacteria, Blautia, Roseburia, and Clostridium, and probiotics Lactobacillus, Bifidobacterium, and so forth were identified significantly different from each other among conditions. ***Conclusion***: Marked shifts of the gut microbiota structure were observed in the rats during development of glucose intolerance. Intestinal flora changed in the process of glucose intolerance, and treatment of sitagliptin moderately corrected the dysbiosis of microbiota in T2DM.

Yap, W. B., Ahmad, F. M., Lim, Y. C., and Zainalabidin, S. Lactobacillus casei strain C1 attenuates vascular changes in spontaneously hypertensive rats. Korean J.Physiol Pharmacol. 2016; 20(6):621-628; PM: 27847439

ABSTRACT: Hypertension can be caused by various factors while the predominant causes include increase in body fluid volume and resistance in the circulatory system that elevate the blood pressure. Consumption of probiotics has been proven to attenuate hypertension; however, the effect is much strain-dependent. In this study, a newly isolated Lactobacillus casei (Lb. casei) strain C1 was investigated for its antihypertensive properties in spontaneously hypertensive rats (SHR). Lactic acid bacteria (LAB) suspension of 11 log colony-forming unit (CFU) was given to SHR (SHR+LAB, n=8), and phosphate buffer saline (PBS) was given as a control in SHR (SHR, n=8) and in Wistar rats as sham (WIS, n=8). The treatment was given via oral gavage for 8 weeks. The results showed that the weekly systolic blood pressure (SBP), mean arterial pressure (MAP), diastolic blood pressure (DBP) and aortic reactivity function were remarkably improved after 8 weeks of bacterial administration in SHR+LAB. These effects were mostly attributed by restoration of wall tension and tensile stress following the bacterial treatment. Although not statistically significant, the level of malondialdehye (MDA) in SHR+LAB serum was found declining. Increased levels of glutathione (GSH) and nitric oxide (NO) in SHR+LAB serum suggested that the bacterium exerted vascular protection through antioxidative functions and

relatively high NO level that induced vasodilation. Collectively, Lb. casei strain C1 is a promising alternative for hypertension improvement.

Yaqoob, P. Ageing alters the impact of nutrition on immune function. Proc.Nutr.Soc. 11-8-2016; 1-5; PM: 27821203

ABSTRACT: Immunosenescence during ageing is a major challenge which weakens the ability of older individuals to respond to infection or vaccination. There has been much interest in dietary strategies to improve immunity in older people, but there is an assumption that modulation of the immune response in older people will be based on the same principles as for younger adults. Recent evidence suggests that ageing fundamentally alters the impact of nutrition on immune function. As a result, interpretation of data from studies investigating the impact of diet on immune function is highly dependent on subject age. Study design is critically important when investigating the efficacy of dietary components, and most studies involving older people include rigorous inclusion/exclusion criteria based on medical history, laboratory tests, general health status and often nutritional status. However, immunological status is rarely accounted for, but can vary significantly, even amongst healthy older people. There are several clear examples of age-related changes in immune cell composition, phenotype and/or function, which can directly alter the outcome of an intervention. This review uses two case studies to illustrate how the effects of n-3 PUFA and probiotics differ markedly in young v. older subjects. Evidence from both suggests that baseline differences in immunosenescence influence the outcome of an intervention, highlighting the need for detailed immunological characterisation of subjects prior to interventions. Finally, future work elucidating alterations in metabolic regulation within cells of the immune system as a result of ageing may be important in understanding the impact of diet on immune function in older people.

Yeo, S., Lee, S., Park, H., Shin, H., Holzapfel, W., and Huh, C. S. Development of putative probiotics as feed additives: validation in a porcine-specific gastrointestinal tract model. Appl.Microbiol.Biotechnol. 2016; 100(23):10043-10054; PM: 27633101

ABSTRACT: Enforced restrictions on the use of antibiotics as growth promoters (AGPs) in animal production have prompted investigations into alternative feed additives in recent decades. Probiotics are currently the main feed additive used in livestock. However, the selection of probiotic candidates relies on human-based methods and little is known about the verification criteria for host-specific selection. We investigated the probiotic potential of Lactobacillus salivarius strains isolated from fed pig feces for their use as porcine feed additives. Two methods were developed that simulated the pig gastrointestinal (GI) tract and the intestinal epithelium, and these were compared with human-based in vitro methods and used for selecting porcine probiotics. Lactobacillus salivarius strain LS6 was identified as a promising probiotic strain for potential use as a porcine feed additive. This strain prevented disruption of the epithelial integrity of pig small intestine (PSI) cells by inhibiting the adherence of enterotoxigenic Escherichia coli K88. It also showed high survival rates in the in vitro pig GI tract model and good adhesion to PSI cells. We propose that host target-specific screening and validation methods are important tools in the development of effective probiotic feed additives, and this approach may support future-oriented agriculture.

Yousaf, S., Hussain, A., Rehman, S., Aslam, M. S., and Abbas, Z. Hypoglycemic and hypolipidemic effects of Lactobacillus fermentum, fruit extracts of Syzygium cumini and Momordica charantia on diabetes induced mice. Pak.J.Pharm.Sci. 2016; 29(5):1535-1540; PM: 27731809

ABSTRACT: A lot of treatment strategies available for diabetes but its complications are still a medical problem around the globe. It demands to find out some alternative therapeutic measures. In order to investigate the anti-diabetic potential of probiotics and natural extracts, this study was designed. Accordingly, a local source of yogurt probiotic strain Lactobacillus fermentum was isolated and characterized that showed its probiotic properties. Besides this, natural extracts of plants fruits like java plum (Syzygium cumini) and bitter gourd (M. charantia) were made. Lactobacillus fermentum and the extracts were administered individually as well as in combination to diabetes induced mice. Different parameters like body weight, blood glucose level and lipid profile including total cholesterol, HDL & LDL were analyzed before and after treatment. The results showed that Lactobacillus fermentum and natural extracts have hypoglycemic as well hypolipidemic activity against diabetic mice. This study can further investigated to screen potential compounds from these extracts to control the glucose and the lipid levels in diabetic patients.

Yucel, Falco C., Sotres, J., Rascon, A., Risbo, J., and Cardenas, M. Design of a potentially prebiotic and responsive encapsulation material for probiotic bacteria based on chitosan and sulfated beta-glucan. J.Colloid Interface Sci. 2-1-2017; 487:97-106; PM: 27756004

ABSTRACT: ***HYPOTHESIS***: Chitosan and sulfated oat beta-glucan are materials suitable to create a prebiotic coating for targeted delivery to gastrointestinal system, using the layer by layer technology. ***EXPERIMENT***: Quartz crystal microbalance with dissipation (QCM-D), spectroscopic ellipsometry (SE) and atomic force microscopy (AFM) were used to assess the multilayer formation capacity and characterize the resulting coatings in terms of morphology and material properties such as structure and rigidity. The coating of colloidal materials was proven, specifically on L. acidophilus bacteria as measured by changes in the bacterial suspension zeta potential. Viability of coated cells was shown using plate counting method. The coatings on solid surfaces were examined after exposure to mimics of gastrointestinal fluids and a commercially available beta-glucanase. ***FINDINGS***: Successful build-up of multilayers was confirmed with QCM-D and SE. Zeta potential values proved the coating of cells. There was 2 log CFU/mL decrease after coating cells with four alternating layers of chitosan and sulfated beta-glucan when compared to viability of uncoated cells. The coatings were partially degraded after exposure to simulated intestinal fluid and restructured as a result of beta-glucanase treatment, mimicking enzymes present in the microflora of the human gut, but seemed to resist acidic gastric conditions. Therefore, coatings of chitosan and sulfated beta-glucan can potentially be exploited as carriers for probiotics and delicate nutraceuticals.

Yunes, R. A., Poluektova, E. U., Dyachkova, M. S., Klimina, K. M., Kovtun, A. S., Averina, O. V., Orlova, V. S., and Danilenko, V. N. GABA production and structure of gadB/gadC genes in Lactobacillus and Bifidobacterium strains from human microbiota. Anaerobe. 10-26-2016; 42:197-204; PM: 27794467

ABSTRACT: Gamma-amino butyric acid (GABA) is an active biogenic substance synthesized in plants, fungi, vertebrate animals and bacteria. Lactic acid bacteria are considered the main producers of GABA among bacteria. GABA-producing lactobacilli are isolated from food products such as cheese, yogurt, sourdough, etc. and are the source of bioactive properties assigned to those foods. The ability of human-derived lactobacilli and bifidobacteria to synthesize GABA remains poorly characterized. In this paper, we screened our collection of 135 human-derived Lactobacillus and Bifidobacterium strains for their ability to produce GABA from its precursor monosodium glutamate. Fifty eight strains were able to produce GABA. The most efficient GABA-producers were Bifidobacterium strains (up to 6 g/L). Time profiles of cell growth and GABA production as well as the influence of pyridoxal phosphate on GABA production were studied for L. plantarum 90sk, L. brevis 15f, B. adolescentis 150 and B. angulatum GT102. DNA of these strains was sequenced; the gadB and gadC genes were identified. The presence of these genes was analyzed in 14 metagenomes of healthy individuals. The genes were found in the following genera of bacteria: Bacteroidetes (Bacteroides, Parabacteroides, Alistipes, Odoribacter, Prevotella), Proteobacterium (Esherichia), Firmicutes (Enterococcus), Actinobacteria (Bifidobacterium). These data indicate that gad genes as well as the ability to produce GABA are widely distributed among lactobacilli and bifidobacteria (mainly in L. plantarum, L. brevis, B. adolescentis, B. angulatum, B. dentium) and other gut-derived bacterial species. Perhaps, GABA is involved in the interaction of gut microbiota with the macroorganism and the ability to synthesize GABA may be an important feature in the selection of bacterial strains - psychobiotics.

Zanjani, S. Y., Eskandari, M. R., Kamali, K., and Mohseni, M. The effect of probiotic bacteria (Lactobacillus acidophilus and Bifidobacterium lactis) on the accumulation of lead in rat brains. Environ.Sci.Pollut.Res.Int. 10-29-2016; PM: 27796979

ABSTRACT: Lead is a toxic metal present in different concentrations in a wide variety of food products. Exposure to lead, even to low levels, causes acute and chronic toxicities. Lead can cross the blood-brain barrier and accumulate in the nervous system. Probiotics are live microorganisms that, when used in adequate amounts, confer a health benefit on the host. Although a recent study demonstrated that the studied bacteria have a protective effect against acute lead toxicity, no research has been found that shows the long-term impact of these bacteria in vivo. The current study surveyed the protective effects of two species of probiotics, Lactobacillus acidophilus LA-5 and Bifidobacterium lactis BB-12, that are most widely used in many functional foods against oral lead exposure (4 weeks) in rat brains. The results revealed that, at the end of the second week of chronic exposure to lead and probiotic bacteria, the lowest level of lead belonged to the Lactobacillus group. At the end of the fourth week, the lowest amount of lead was related to the group receiving both types of probiotics. With the physiological benefits of probiotic consumption, the bacterial solution in this study did not show high efficacy in reducing brain lead concentrations.

Zhang, H. L., Li, W. S., Xu, D. N., Zheng, W. W., Liu, Y., Chen, J., Qiu, Z. B., Dorfman, R. G., Zhang, J., and Liu, J. Mucosa-reparing and microbiota-balancing therapeutic effect of Bacillus subtilis alleviates dextrate sulfate sodium-induced ulcerative colitis in mice. Exp.Ther.Med. 2016; 12(4):2554-2562; PM: 27698758

ABSTRACT: Gut microbiota composition of patients with ulcerative colitis (UC) is markedly altered compared with healthy individuals. There is mounting evidence that probiotic therapy alleviates disease severity in animal models and patients with inflammatory bowel disease (IBD). Bacillus subtilisis, as a probiotic, has also demonstrated a protective effect in IBD. However, the therapeutic mechanism of its action has yet to be elucidated. In the present study, a dextrose sulfate sodium (DSS)-induced UC mouse model was used to investigate the role of B. subtilis in the restoration of gut flora and determine its effective dose. Mucosal damage was assessed by performing alcian blue staining, cytokine levels were analyzed by ELISA and microbiota composition was investigated using 454 pyrosequencing to target hypervariable regions V3-V4 of the bacterial 16S ribosomal RNA gene. The results demonstrated that a higher dose B. subtilisis administration ameliorated DSS-induced dysbiosis and gut inflammation by balancing beneficial and harmful bacteria and associated anti- and pro-inflammatory agents, thereby aiding intestinal mucosa recovery from DSS-induced injuries. These findings indicate that choosing the correct dose of B. subtilis is important for effective UC therapy. The present study also helped to elucidate the mechanisms of B. subtilis action and provided preclinical data for B. subtilis use in UC therapy.

Zhang, M. and Yang, X. J. Effects of a high fat diet on intestinal microbiota and gastrointestinal diseases. World J.Gastroenterol. 10-28-2016; 22(40):8905-8909; PM: 27833381

ABSTRACT: Along with the rapid development of society, lifestyles and diets have gradually changed. Due to overwhelming material abundance, high fat, high sugar and high protein diets are common. Numerous studies have determined that diet and its impact on gut microbiota are closely related to obesity and metabolic diseases. Different dietary components affect gut microbiota, thus impacting gastrointestinal disease occurrence and development. A large number of related studies are progressing rapidly. Gut microbiota may be an important intermediate link, causing gastrointestinal diseases under the influence of changes in diet and genetic predisposition. To promote healthy gut microbiota and to prevent and cure gastrointestinal diseases, diets should be improved and supplemented with probiotics.

Zhang, R., Dong, X., Zhou, M., Tu, Y., Zhang, N., Deng, K., and Diao, Q. Oral administration of Lactobacillus plantarum and Bacillus subtilis on rumen fermentation and the bacterial community in calves. Anim Sci.J. 9-14-2016; PM: 27628956

ABSTRACT: The objective of this study was to assess the effect of dietary probiotics on rumen fermentation and the bacterial community in dairy calves. Twelve Holstein calves were randomly allocated to three treatments: a basal diet, the basal diet supplemented with Lactobacillus plantarum GF103 (LB) or basal diet supplemented with a mixture of Lactobacillus plantarum GF103 and Bacillus subtilis B27 (LBS). A milk replacer was fed to calves from 8 days of age. A starter and alfalfa hay was offered ad libitum from 21 and 28 days of age, respectively, and the orts were weighted daily. The ruminal fluid was sampled at 56 and 83 days of age to determine the rumen fermentation characteristics. The bacterial community was analyzed by denaturing gradient gel electrophoresis (DGGE) and the number of certain bacteria was quantified by real-time polymerase chain reaction. The ratio of total dry matter intake to average body wieght was higher in the control ($P < 0.05$). The DGGE fingerprint of the 16S ribosomal RNA gene was affected by the blended probiotics at 83 days of age. The number of Ruminococcus albus was

lower in the LB and LBS treatment ($P < 0.05$). Oral administration of the probiotics affected the rumen bacterial community and the numbers of cellulolytic bacteria decreased.

Zhang, S. F., Tang, Z. S., Tong, L., Tao, X. X., Suo, Q. F., and Xu, X. M. Effects of clostridium butyricum and bifidobacterium on BTLA expression on CD4+ T cells and lymphocyte differentiation in late preterm infants. Microb.Pathog. 2016; 100:112-118; PM: 27622346

ABSTRACT: ***BACKGROUND & AIMS***: Probiotics is recognized to promote growth performance and immune function via balancing the intestinal microflora. Live clostridium butyricum and bifidobacterium combined powder (LCBBCP) has been widely to treat intestinal dysbacteriosis in newborns in China. This study was undertaken to investigate the effects of the combined probiotics on the expression of B and T lymphocyte attenuator (BTLA) on CD4+ T cells and the differentiation of lymphocyte subsets in late preterm infants. ***METHODS***: Eighty eligible late preterm infants were equally randomized into LCBBCP therapy group (oral LCBBCP dissolved in formula milk before intake) and control group (treated with simple formula milk for preterm infants) by random digit table. Flow cytometry was used to determine the expression level of BTLA on CD4+ T cells and the percentage of individual subpopulation of lymphocytes in peripheral-blood mononuclear cells (PBMCs) obtained from the late preterm infants in both groups. ***RESULTS***: BTLA protein expression on CD4+ T cells showed no significant change in LCBBCP therapy group before and after intervention, yet was rapidly and significantly down-regulated in the controls. The percentage of increased CD4+ T cells, decreased CD8+ T cells and increased ratio of CD4+/CD8+ T cell proportion were seen in both groups after treatment, yet the increasing or decreasing extent in LCBBCP therapy group was more obvious than in control group. The proportion of NK cells and B lymphocytes remained no significant difference between the two groups before and after therapy. ***CONCLUSIONS***: LCBBCP appears capable of facilitating the activation, proliferation and differentiation of T lymphocytes, which is beneficial to improving immunity in late preterm infants. The continuous high expression of BTLA on CD4+ T cells in LCBBCP therapy group may be involved in the inhibiting of excessive activation of T lymphocytes. Our findings may lay a basis for further clinical evaluation of the efficacies and wider clinical recommendation of probiotics containing live clostridium butyricum and bifidobacterium for late preterm infants.

Zhang, W., Zhu, Y. H., Zhou, D., Wu, Q., Song, D., Dicksved, J., and Wang, J. F. Oral administration of a select mixture of Bacillus probiotics affects the gut microbiota and goblet cell function in newly weaned MUC4 resistant pigs following Escherichia coli challenge. Appl.Environ.Microbiol. 11-23-2016; PM: 27881419

ABSTRACT: Structural disruption of the gut microbiota and impaired goblet cell function are collateral etiologic factors in enteric diseases. Low, moderate, or high doses of a Bacillus licheniformis-B. subtilis mixture (BLS-mix) were orally administered to MUC4 resistant piglets for 1 week before the F4-expressing ETEC strain (F4+ ETEC) challenge. The luminal contents were collected from the mucosa of colon on day 8 after F4+ ETEC challenge. BLS-mix attenuated E. coli-induced expansion of Bacteroides uniformis, Eubacterium eligens, Acetanaerobacterium and Sporobacter populations. Clostridium and Turicibacter populations increased following F4+ ETEC challenge in pigs pretreated with low-dose BLS-mix. Lactobacillus gasseri and L. salivarius populations increased after administering BLS-mix during E. coli infection. The beneficial effects of BLS-mix were due in

part to expansion of certain Clostridium, Lactobacillus, and Turicibacter populations, with a corresponding increase in the number of goblet cells in the ileum via upregulated Atoh1 expression, in turn increasing MUC2 production, thus preserving the mucus barrier and enhancing host defenses against enteropathogenic bacteria. However, excessive BLS-mix consumption may increase the risk for enteritis, partly through disruption of colonic microbial ecology, characterized by expansion of Proteobacteria and impaired goblet cell function in the ileum. Our findings suggest that oral administration of BLS-mix reprograms the gut microbiota and enhances goblet cell function to ameliorate enteritis. IMPORTANCE: The present study is important for improving our understanding the protective role of probitics against the infection of Escherichia coli in piglets. Structural disruption of the gut microbiota and impaired goblet cell function are collateral etiologic factors in enteric diseases. In this study, low, moderate, or high doses of a Bacillus licheniformis-B. subtilis mixture (BLS-mix) were orally administered to MUC4 resistant piglets for 1 week before the F4-expressing ETEC strain (F4+ ETEC) challenge. Our findings suggest that oral administration of BLS-mix reprograms the gut microbiota and enhances goblet cell function to ameliorate enteritis.

Zhang, Z., Liu, Z., Tao, X., and Wei, H. Characterization and sulfated modification of an exopolysaccharide from Lactobacillus plantarum ZDY2013 and its biological activities. Carbohydr.Polym. 11-20-2016; 153:25-33; PM: 27561468

ABSTRACT: An exopolysaccharide (EPS) from probiotics Lactobacillus plantarum ZDY2013 was purified to illustrate its molecular weight, monosaccharide composition and biological activities. The yield of EPS (429.4+/-30.3mg/L) was obtained with a purity of 96.06%. The EPS was characterized to have only one symmetrical sharp peak by high-performance size-exclusion chromatography and its molecular weight was 5.17x10(4)Da. The GC analysis revealed that EPS only consisted of xylose and galactose, and the galactose possessed as high as 98.3% (w/w) of the total monosaccharides. By sulfonation, a sulfated EPS was successfully synthesized with the degree of substitution (DS) of 0.29, which was confirmed using FT-IR spectroscopy. Both EPS and sulfated EPS showed radical scavenging activities, and the antioxidant activities increased after sulfonation. In addition, sulfated EPS was more effective in counteracting the cytotoxicity induced by B. cereus enterotoxins on Caco-2 cells when compared with EPS. In summary, sulfonation is a feasible strategy for improving the biological activities of EPS from L. plantarum ZDY203.

Zhao, S., Liu, W., Wang, J., Shi, J., Sun, Y., Wang, W., Ning, G., Liu, R., and Hong, J. Akkermansia muciniphila improves metabolic profiles by reducing inflammation in chow diet-fed mice. J.Mol.Endocrinol. 2017; 58(1):1-14; PM: 27821438

ABSTRACT: Abnormal shifts in the composition of gut microbiota contribute to the pathogenesis of metabolic diseases, including obesity and type 2 diabetes (T2DM). The crosstalk between gut microbes and the host affects the inflammatory status and glucose tolerance of the individuals, but the underlying mechanisms have not been elucidated completely. In this study, we treated the lean chow diet-fed mice with Akkermansia muciniphila, which is thought to be inversely correlated with inflammation status and body weight in rodents and humans, and we found that A. muciniphila supplementation by daily gavage for five weeks significantly alleviated body weight gain and reduced fat mass. Glucose tolerance and insulin sensitivity were also improved by A. muciniphila supplementation compared with the vehicle. Furthermore, A. muciniphila supplementation reduced gene expression related to fatty acid synthesis and transport

in liver and muscle; meanwhile, endoplasmic reticulum (ER) stress in liver and muscle was also alleviated by A. muciniphila. More importantly, A. muciniphila supplementation reduced chronic low-grade inflammation, as reflected by decreased plasma levels of lipopolysaccharide (LPS)-binding protein (LBP) and leptin, as well as inactivated LPS/LBP downstream signaling (e.g. decreased phospho-JNK and increased IKBA expression) in liver and muscle. Moreover, metabolomics profiling in plasma also revealed an increase in anti-inflammatory factors such as alpha-tocopherol, beta-sitosterol and a decrease of representative amino acids. In summary, our study demonstrated that A. muciniphila supplementation relieved metabolic inflammation, providing underlying mechanisms for the interaction of A. muciniphila and host health, pointing to possibilities for metabolic benefits using specific probiotics supplementation in metabolic healthy individuals.

Zhao, Y., Duan, C., Gao, L., Yu, X., Niu, C., and Li, S. Genome shuffling of Lactobacillus plantarum C88 improves adhesion. Biosci.Biotechnol.Biochem. 2017; 81(1):184-193; PM: 27691905

ABSTRACT: Genome shuffling is an important method for rapid improvement in microbial strains for desired phenotypes. In this study, ultraviolet irradiation and nitrosoguanidine were used as mutagens to enhance the adhesion of the wild-type Lactobacillus plantarum C88. Four strains with better property were screened after mutagenesis to develop a library of parent strains for three rounds of genome shuffling. Fusants F3-1, F3-2, F3-3, and F3-4 were screened as the improved strains. The in vivo and in vitro tests results indicated that the population after three rounds of genome shuffling exhibited improved adhesive property. Random Amplified Polymorphic DNA results showed significant differences between the parent strain and recombinant strains at DNA level. These results suggest that the adhesive property of L. plantarum C88 can be significantly improved by genome shuffling. Improvement in the adhesive property of bacterial cells by genome shuffling enhances the colonization of probiotic strains which further benefits to exist probiotic function.

Zheng, H., Gao, M., Ren, Y., Lou, R., Xie, H., Yu, W., Liu, X., and Ma, X. An improved pH-responsive carrier based on EDTA-Ca-alginate for oral delivery of Lactobacillus rhamnosus ATCC 53103. Carbohydr.Polym. 1-2-2017; 155:329-335; PM: 27702519

ABSTRACT: A pH-responsive carrier based on an ethylenediaminetetraacetic-calcium-alginate (EDTA-Ca-Alg) system was developed by controlling the release of Ca2+. The system remained in the solution state at neutral pH since EDTA completely chelated the Ca2+. In contrast, a hydrogel immediately formed when the pH was below 4.0, which triggered the in situ release of Ca2+ from the EDTA-Ca compound and led to alginate-Ca binding. Taking advantage of the pH sensitivity, we prepared hydrogel microspheres with uniform size to entrap Lactobacillus rhamnosus ATCC 53103 through emulsification. In an acidic environment, the hydrogel structure remained compact with negligible pores to protect L. rhamnosus ATCC 53103. However, in a neutral intestinal environment, the hydrogel structure gradually disassembled because of the Ca2+ release from the hydrogel, which caused cell release. Therefore, a pH-responsive carrier was developed for the protection and the controlled release of cells in gastrointestinal tract, thus providing potential for oral delivery of probiotics.

Zoghi, A., Khosravi-Darani, K., Sohrabvandi, S., Attar, H., and Alavi, S. A. Effect of Probiotics on Patulin Removal from Synbiotic Apple Juice. J.Sci.Food Agric. 10-27-2016; PM: 27785791

ABSTRACT: ***BACKGROUND***: Studies have reported the occurrence of mycotoxin patulin in apple products. The aim of this study was to produce synbiotic apple juice and investigate the detoxification of patulin by Lactobacillus acidophilus and Lactobacillus plantarum as probiotic strains. Impact of seven process variables on efficiency of toxin removal was investigated using Plackett-Burman design and presence of the surface-layer proteins as binding site of probiotics to patulin was confirmed during 6 weeks cold storage. ***RESULTS***: Results showed that the removal of patulin by probiotic bacteria from apple juice depends significantly ($P<0.05$) on the fructooligosaccharide content (as a prebiotic), concentration of patulin and adding ascorbic acid. Sodium dodecyl sulphate polyacrylamide gel electrophoresis of cell surface proteins of probiotic strains revealed that surface layer proteins have an important role in patulin removal of apple juice. In the best condition, 91.23% of initial patulin concentration was removed from juice during 6 weeks refrigerated storage. No significant different was observed in organoleptic properties of the synbiotic apple juice and raw sample. ***CONCLUSION***: In the best condition reported in this study, contaminated synbiotic apple juice by patulin will be safe for consumers after 1st day of probiotic inoculation.

Interested in contributing to the Biotherapy Journal Review?

We are looking for professionals in the biotherapy field to submit journal reviews to our quarterly publication. If you have a journal article you would like to review, or are interested in becoming a regular reviewer for the ***Biotherapy Journal Review***, please contact Ronald Sherman at RSherman@uci.edu.



BioTherapeutics, Education & Research Foundation

36 Urey Court, Irvine, CA 92617

www.BTERFoundation.org

Phone: 949-246-1156 / Fax: 949-679-3001