Conference schedule and participant abstracts

2022 Annual Meeting

June 14-17, 2022
Sitges, Spain

For more details, join us at:
http://www.isapp-sfa.com
http://www.facebook.com/isappsfa

@ISAPPSFA #ISAPP2022

About the SFA

The ISAPP Students and Fellows Association (SFA) was created in 2009 as an initiative to link trainees working in fields related to probiotics, prebiotics and health effects of commensal microbes. We operate as a trainee-led branch of our parent group, ISAPP.

Our goal is to create an interactive network of graduate students and postdoctoral fellows across the globe working on probiotics, prebiotics or related fields, and thus promote real-time interactions, intellectual and technical exchanges, and other networking opportunities for our members. We intend to act as a resource for ISAPP and the industry, providing a communication platform to facilitate scientific discussions, internships, and employment opportunities among qualified researchers in the field.

Annual Meetings

The SFA began as an initiative by Gregor Reid and the students and fellows at the 2009 ISAPP meeting in Irvine, California. The SFA annual meetings are held in conjunction with the ISAPP annual meetings. Our first meeting began in 2010 in Barcelona, Spain, followed by: 2011 in Berkeley, California; 2012 in Cork, Ireland; 2013 in New York, New York; 2014 in Aberdeen, Scotland; 2015 in Washington D.C.; 2016 in Turku, Finland; 2017 in Chicago, Illinois, US; 2018 in Sinapore; 2019 in Antwerp, Belgium; and this year, 2022 in Sitges, Spain. Location of the 2023 meeting to be determined.

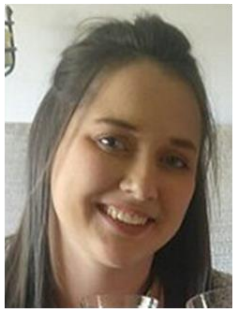
On behalf of the SFA committee, we welcome all SFA attendees to the 2022 annual SFA meeting, and we hope you enjoy your time in Spain!

2021-2022 SFA Executive Committee and Acknowledgements

SFA Executive Committee

2021-2022

President



Daragh Hill, PhD.
APC Microbiome, Ireland

Vice-President

Brendan Daisley, PhD.
University of Guelph, Canada

Secretary

Conall Strain, PhD.
APC Microbiome, Ireland & Teagasc Moorepark Food Research centre

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Local Organizer

Zaida Soler Luque
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Local Organizer

Zixuan Xie
Vall d'Hebrón Research Institute (VHIR), Spain

Acknowledgements

The SFA Executive Committee would like to thank Mary Ellen Sanders, Gregor Reid, and the ISAPP Board of Directors for integrating the SFA into the ISAPP annual meeting, and for their continued financial support of the SFA organization. We also thank all our meeting attendees who make the annual meeting so thought-provoking and fun.

Conference schedule

ISAPP meeting and general travel information:
https://isappscience.org/for-scientists/annual-meeting/2022-annual-meeting/

– Date of event –

Time
Location

LEGEND

SFA-only EVENTS
ISAPP EVENTS (SFA participating)
Talk title
Chair or presenter

Tuesday, June 14

15:00 - 18:00
Hotel Desitges

HOTEL CHECK-IN
Location: Carrer del Xarel·lo, nº2, 08812 Els Cards, Barcelona, Spain
Website: https://hoteldesitges.com

17:00 - 18:00

REGISTRATION
Pickup name badges in hotel lobby
Register in advance for discussion groups (see schedule for June 16)

18:00 - 20:00

MEET AND GREET
Casual gathering to meet the other SFA members attending the conference

Wednesday, June 15

8:30 - 9:30
Dolce Sitges Hotel

SFA REGISTRATION AND POSTER SETUP
Location: Av. Camí de Miralpeix, 12, 08870 Sitges, Barcelona, Spain
Website: https://www.dolcesitges.com

09:30 - 10:30

SFA WELCOME AND INTERACTIVE INTRODUCTION
This session is for SFA members only. The Executive Committee will present a 10 minute introduction. Subsequently, each SFA attendee will give a 90 second lightning presentation introducing themselves. Format will be 16:9 aspect ratio with 2 powerpoint slides – 1 on personal background and 1 on scientific research.
Chairs: Daragh Hill (SFA President, University College Cork, Ireland) and Brendan Daisley (SFA Vice President, University of Guelph, Canada
Daragh Hill, SFA president, University College Cork, Ireland

10:30 - 11:00

BREAK AND POSTER VIEWING

11:00 - 12:30

INDUSTRY FORUM
Precision probiotics and prebiotics

- Jens Walter (University of College Cork, Ireland)
- Niv Zmora (Weizmann Institute of Science, Rehovot, Israel)

Chairs: Marla Cunningham and Kelly Swanson, University of Illinois, Urbana - Champagne, USA

12:30 - 13:30 LUNCH

13:30 - 13:45 WELCOME TO ISAPP 2022

- Dan Merenstein, ISAPP President
- Marla Cunningham, Sr. IAC Representative
- Brendan Daisley, SFA Vice President

13:45 – 15:30 INTERACTIVE DISCUSSION

The Then, Now, and Future of the "Biotics" Family

- Eamonn Quigley, Weill Cornell School of Medicine, Houston, TX, USA
- Kristin Verbeke, KU Leuven, Belgium
- Clara Belzer, Wageningen University, The Netherlands

Chair: Dan Tancredi, University of California-Davis, USA

15:30 - 16:30 DEBATE SESSION

All probiotic effects must be considered strain-specific

- *Pro position: Hania Szajewska, The Medical University of Warsaw, Poland*
- *Con position: Sarah Lebeer, University of Antwerp, Belgium*
- *Panel: Dan Merenstein, Maria Marco, University of California – Davis, USA and Arthur Ouwehand, IFF, Kantvik, Finland, Gabriel Vinderola*

Chair: Colin Hill, University of College Cork, Ireland

16:30 - 17:00 BREAK AND POSTER VIEWING

17:00 - 18:30 FEATURED TALKS

Q&A following each talk

- *Personal predictions for the future of prebiotics (science only, don't ask me about economy)*
  Glenn Gibson, University of Reading, UK
- *Live dietary microbes and health.* An updates of an ISAPP project
  Maria Marco
- *Antiviral potential of topically applied lactobacilli in the respiratory tract: from mechanisms to application*
  Irina Spacova, University of Antwerp, Belgium
  *2021 Glenn Gibson Early Career Research Prize Winner
- *Human milk oligosaccharide-utilizing bifidobacteria produce immunomodulatory aromatic lactic acids in the infant gut*
  Martin Laursen, National Food Institute, Technical University of Denmark, Kgs. Lynbgy
  *2022 Glenn Gibson Early Career Researcher Prize Winner
- IAC talk: Individual and group-based difference in gut microbiota responses to in vitro fiber interventions: Can mixtures of prebiotics contribute to harmonized beneficial effects?
  Frank Schuren, TNO, The Netherlands

Chair: Kelly Swanson

18:30 - 19:30 LATE BREAKING NEWS
Chair: Gregor Reid, Lawson Research Institute, London, Ontario, Canada

19:30 - 21:30 WELCOME RECEPTION WITH POSTER VIEWING
SFA members will present posters for judging. Drinks and food to be served.
Please remove your poster at the end of the day.

Thursday, June 16

8:30 - 9:45 INTRODUCTION AND ORAL PRESENTATIONS
Dolce Sitges Hotel
Day 2 introduction and selected oral presentations from SFA attendees

- Integrative multi-omics analyses reveal the therapeutic impact of probiotics via modulating microbial-based signatures in chronic kidney disease
  Hsiao-Wen Huang, National Taiwan University, Taipei, Taiwan
- Screening the prebiotic effects of human milk oligosaccharides on 330 bacterial strains derived from the infant gut microbiota
  Simone Renwick, University of Guelph, Canada
- Investigation of the gut microbiota composition and activity in acute myeloid leukemic patients: first results of the MicroAML study
  Sarah Pötgens, Université catholique de Louvain, Brussels, Belgium
- Gamma-aminobutyric acid (GABA) production by probiotics
  Andrea Monteagudo-Mera, University of Reading, UK

9:45 - 10:15 IMPACT OF THE COIVD-19 PANDEMIC IN SCIENTIFIC RESEARCH
Interactive discussion session on how COVID has impacted research worldwide with focus on strategies to overcome current workplace limitations.

10:15 - 10:30 BREAK

10:30 - 11:30 HARNESSING THE POWER OF SOCIAL MEDIA

- Social Media: How to Share Your Research Effectively
  Kristina Campbell, Consulting Communications Director, ISAPP

11:30 - 12:15 NICHE STUDY AREA BREAKOUT GROUPS

- Group 1: Probiotics and concept of using beneficial microbes for health
- Group 2: Prebiotics and concept of modulating fiber intake for health
- Group 3: Human microbiota and related bioinformatic analysis

12:15 - 12:30 BREAKOUT GROUP SUMMARIES
One member of each breakout group will give a 3-5 minute presentation on their niche study area discussion to share with the rest of the group.

12:30 - 13:00 NETWORKING ACTIVITIES
Reserved time for networking, idea generation, and collaborative planning.

13:00 - 14:00 LUNCH

14:00 - 15:30 PLENARY SESSION: MICROBIOTA-MEDIATED MECHANISMS DRIVING HEALTH

BENEFITS

- *Unravelling health promoting microbiota-mediated mechanisms using metabolic profiling*
  Anisha Wijeyesekera, University of Reading, UK
- *Do prebiotics promote health through microbiota-mediated mechanisms?*
  Michiel Kleerebezem, Wageningen University & Research, The Netherlands
- *Probiotics and chronic constipation: mechanisms of action and effectiveness*
  Eirini Dimidi, King's College London, UK
  *2022 Glenn Gibson Early Career Researcher Prize Winner

Chair: Karen Scott

15:30 - 16:00 **BREAK**

16:00 - 16:45 **TODD KLAENHAMMER MEMORIAL LECTURE**

- *Bacteria and bacteriophage - are they fighting or are they dancing?*
  Colin Hill, University College Cork, Ireland

Chair: Mary Ellen Sanders

16:45 - 17:30 **MINI-PLENARY: POSTBIOTICS**

- *A decade of research on Akkermansia muciniphila: what do we know now?*
  Clara Belzer
- *Regulatory perspectives on the first EFSA-approved novel food*
  Seppo Salminen, University of Turku, Finland

Chair: Seppo Salminen

19:00 Buses loading for Hotel Estela event

19:15 Buses depart

19:30 - 22:30 **DINNER AT THE HOTEL ESTELA NEAR THE SEA IN SITGES**
Join meeting participants and guests for a gala Catalonian dinner with Spanish wines and beers.

Friday, June 17

7:30 - 9:00 **REGISTRATION DESK**
Dolce Sitges Hotel

8:30 - 10:00 **PLENARY SESSION: CLINICAL DEVELOPMENTS**

- *Pain in the NEC: Choosing the Right "Biotic" for Preterm Neonates*
  Geoff Preidis, Baylor College of Medicine and Texas Children's Hospital, Houston, USA
- *Microbiota and gas related complaints*
  Francisco Guarner, University Hospital Vall d'Hebron, Barcelona, Spain
- *IAC talk: Prebiotic Galacto-oligosaccharides Impact Stool Frequency and Fecal Microbiota in Self-reported Constipated Adults: A Randomized Clinical Trial*

Marieke Schoemaker, Friesland Campina, The Netherlands

Chair: Eamonn Quigley

10:00 - 10:30 **BREAK**

10:30 - 12:30 **SUMMARY REPORTS FROM DISCUSSION & SFA EXECUTIVE TEAM**
Discussion Group Chairs and SFA President
Chair: Gabriel Vinderola

List of participants

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11	David Hourigan	University College Cork	Ireland	20
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Participant abstracts

Development of anaerobic fermentation techniques for measuring the impact of prebiotic supplementation on human gut microbiota in clinical studies

Alexander W. Thorman (University of Cincinnati, Cincinnati, OH, USA), David S. Newburg (University of Cincinnati, Cincinnati, OH, USA), Andre Groeneveld (FrieslandCampina, Amersfoort, The Netherlands), Arjen Nauta (FrieslandCampina, Amersfoort, The Netherlands), Ardythe L. Morrow (University of Cincinnati, Cincinnati, OH, USA).

Introduction: The ability of prebiotic dietary carbohydrates to influence the composition and metabolism of the gut microbiota is central to defining their impact in diverse individuals and populations. Unfortunately, most methods currently used to assess microbial metabolic products in the context of clinical trials are indirect and limited. This study aimed to develop an ex vivo anaerobic fermentation method to overcome this gap and thereby increase the information from prebiotic supplementation trials.

Methods: Stool samples from six volunteers were analyzed to detect differences in microbiome and metabolome based on comparison of different methods: sample collection vial (standard vial and two different BD anaerobic collection vials); storage (-80℃ or refrigerated); duration of fermentation (0, 4, 8, 16, 32, 48hr); and carbon source for growth (2'-fucosyllactose, glucose or galacto-oligosaccharides). All samples were grown anaerobically, measuring optical density (OD) and pH throughout culture.

Results: Neither the collection vial, nor cryopreservation influenced composition. Fermentation reached its maximum growth and minimum pH by 16hr, which was used as the end point for metagenomics and metabolomics, while 8hr identified intermediate metabolites. Metagenomic analysis distinguished samples at 16hr by subject and then by prebiotic. However, from metabolomic analysis, subjects were the primary driver of differences at 0hr, whereas the prebiotic became the dominant influence by 16hr.

Discussion: We identified a feasible and valid approach for prebiotic fermentation analysis of individual samples in large clinical studies: Collection of stool microbiota using standard vials; cryopreservation prior to testing; and collecting fermentation read-out at 8 and 16hr. Our findings that microbial community composition is primarily affected by the host, whereas microbial metabolism is most affected by the prebiotic at 16hr, supports the value of fermentation analysis to test prebiotic impact.

Gamma-aminobutyric acid (GABA) production by probiotics

Andrea Monteagudo-Mera (University of Reading); Carolyn McNabb (University of Reading), Valentina Fanti (University of Reading), Andreas Karatzas (University of Reading), Anisha Wijeyesekera (University of Reading), Glenn Gibson (University of Reading), Bhismadev Chakrabarti (University of Reading).

Introduction: Gamma-aminobutyric acid (GABA) plays a central role in inhibitory neurotransmission in the human brain. Abnormal GABA levels have been linked to certain psychopathological conditions. The potential to alter GABA levels through diet opens up possibilities for novel interventions. To this end, we assessed the ability of 7 commercial probiotic strains to produce GABA.

Methods: Seven commercial probiotic strains were screened for GABA production. Probiotics were inoculated in 10 mL MRS tubes supplemented with monosodium glutamate (1% w/v) in combination or not with the prebiotic inulin Synergy-1 (1% w/v). The two strains with the highest production of GABA were assessed for 48 h in pH-controlled anaerobic batch cultures inoculated with faecal samples. LC-MS was used for the quantification of GABA and microbiota composition was determined using 16S rRNA gene sequencing.

Results: *Lactobacillus brevis* LB01 and *Lactobacillus plantarum* 299v were the most efficient probiotic strains tested for GABA production in pure cultures. High GABA levels (62.9 mM ± 14.3) were obtained by the probiotic strain *L. brevis* when added in the fermentation vessels at pH 5.4-5.6. This result was significantly higher compared to GABA levels obtained with *L. plantarum* (4.8 mM ± 6.8) and negative control (2.9mM ± 3.1). The prebiotic did not stimulate GABA production under the conditions tested.

Discussion: The ability of different commercial probiotics to produce GABA in-vitro was evaluated. *L. brevis* LB01 was found to be the most efficient probiotic strain tested in a faecal microbiota environment. High GABA levels obtained make this probiotic formulation a candidate for a potential interventions aimed at increasing GABA levels. Ongoing work in our lab will test this possibility.

Physicochemical and microbiological evaluation of a probiotic carrot bio-yogurt stored under refrigeration conditions

Angel David Camargo-Herrera (Facultad de Ciencias Agrarias, Maestría en Ciencia y Tecnología de Alimentos, Universidad Nacional de Colombia, Bogotá, Colombia), Camila Bernal-Castro (Instituto de Biotecnología (IBUN), Universidad Nacional de Colombia, Bogotá, Colombia), Carolina Gutiérrez-Cortes (Universidad Nacional Abierta y A Distancia (UNAD), Escuela de Ciencias Agrícolas, Pecuarias y del Medio Ambiente (ECAPMA)), Consuelo Díaz-Moreno (Instituto de Ciencia y Tecnología de Alimentos (ICTA), Universidad Nacional de Colombia, Bogotá, Colombia).

Introduction: Carrot (Daucus carota) and mango (*Manguifera indica*) are considered a substrate for the growth of lactic acid bacteria, due to the content of carotenoids, antioxidants and fibre. Phytochemicals and bioactive molecules are attractive in designing alternative fermented dairy beverages such as bio- yogurt. The objective of this work was to evaluate microbiological (cell viability) and physicochemical variables (pH, titratable acidity and soluble solids) of a bio-yogurt during 21 days of storage (4°C).

Methods: Bio-yogurt formulations based on oak honeydew, FOS, carrot and mango pulp. The probiotics was activated for 48 hours and inoculated with 1%v/v, the probiotic was VEGE092 from HOWARU Danisco®. As a control, the starter culture YOFLEX CHR Hansen®. Growth and acidification kinetics were evaluated by measurements during 0,1,7,14 and 21days. Viability was determined by depth plate counting on MRS agar. The measurement of pH and titratable acidity was carried out by the potentiometric method (AOAC 937.05)

Results: The addition of carrot, mango, oak honeydew and FOS, influenced growth and acidification kinetics due to nutritional compounds available for probiotics. Regarding pH, during 21 days storage days stabilized at values of 4 and 4.1 for VEGE092 and the control respectively. In cellular viability there was a significant difference during storage, VEGE092 culture showed a greater adaptation reaching 10,3 Log CFU/mL, while the starter culture showed a lower survival 8,3 Log CFU/mL by day 21 of storage.

Discussion: The results suggest an adaptation strains to carrot and mango pulp, they assimilate phytochemicals and bioactive compounds to be able to survive during storage. Microbial growth and adaptation have a strong correlation with the decrease in pH due to the enzymatic complex that assimilate carbon sources. The prebiotic potential of oak honeydew is remarkable due to the composition of carbon molecules. The symbiotic bioyogurt represent an opportunity to generate value in dairy fermented products

In silico study of efficiently producing fructooligosaccharides using a novel strain of *Fusarium* sp. HKF- 74

Atul Rajkumar Chavan (Environmental Biotechnology and Genomics Division, CSIR-National Environmental Engineering Research Institute, Nehru Marg, Nagpur 440020, India) 2. Anshuman Arun Khardenavis (Environmental Biotechnology and Genomics Division, CSIR-National Environmental Engineering Research Institute, Nehru Marg, Nagpur 440020, India).

Introduction: Agricultural wastes are a renewable and huge biomass resource that can be effectively digested by a mix of carbohydrate-active enzymes released by fungal extracellular enzymes. Indentifying enzymes with innovative features to improve conversion processes in the manufacturing of lignocellulosic-based products is critical.

Methods: Potential isolates were screened for extracellular transfructosylation activity. For qualitative analysis HPLC has been performed. The whole genome sequencing was performed using NGS platform and genome annotation using GeneMark ES and Augustus. For CAZyme analysis, CAZyme annotation using dbCAN is in progress, all data in dbCAN will be generated based on the family classification from CAZy database.

Results: HKF-74 resulted in highest hydrolytic activity at the end of 96h which was found to be 17.52 U/mL and highest transfructosylation activity of 19.88 U/mL after 96h of incubation. Indeed, the qualitative analysis using HPLC proved that the strain HKF-74 is capable of producing different enzymatic activities. The whole genome sequencing was performed using NGS platform and genome annotation using GeneMark ES and Augustus. For CAZyme analysis, CAZyme annotation using dbCAN is in progress.

Discussion: Optimum bioprocess parameters for the enhanced production of prebiotic oligosaccharides from carbohydrate rich substrates such as pure sugars and agricultural wastes. In addition to the prebiotic produced which are of use in health supplement production, the enzymes can directly find potential application in prebiotic producing industries

Extending probiotic science beyond human health: Design and application of a novel spray- based formula for sustainable disease management in California honey bees

Brendan Daisley (University of Guelph, Guelph, Canada), Andrew Pitek (Western University, London, Canada), Christina Torres (University of California, Davis, Davis, USA), Robin Lowery (University of California, Davis, Davis, USA), Bethany Adair (Western University, London, Canada), Kait F Al (Western University, London, Canada), Bernardo Niño (University of California, Davis, Davis, USA), Jeremy Burton (Western University, London, Canada), Emma Allen-Vercoe (University of Guelph, Guelph, Canada), Graham Thompson (Western University, London, Canada), Elina Niño (University of California, Davis, Davis, USA), Gregor Reid (Western University, London, Canada).

Abstract: Managed honey bee (*Apis mellifera*) populations play a crucial role in supporting adequate pollination of food crops but are facing unsustainable colony loss as the result of rampant disease spread within agricultural environments. Antibiotics have failed to resolve the issue so far, whereas mounting evidence from *in vitro* experiments suggest that select lactobacilli strains (some of which are symbionts in honey bees) can inhibit a broad range of important pathogens via multifaceted mechanisms. Importantly, there has been very little validation at the field-level potentially due to the fact that delivery methods for applying viable lactobacilli to the hive are lacking. Here, we compare how different delivery systems (standard pollen patty and a novel spray-based formula) influence the efficacy of a three-strain lactobacilli consortium (LX3) in reducing overall bacterial and fungal disease burden within a pathogen-dense region of California post-almond harvest. Hives were supplemented for 4-weeks,
followed by a 20-week monitoring period. Results reveal long-lasting beneficial effects of LX3 in delivery-dependent and -independent manners. The most striking finding was that spray-based LX3 supplementation led to >100-fold reduction in *Ascosphaera apis* (deadly fungal agent of Chalkbrood disease), whereas patty-based LX3 showed unique nutritional benefits. In addition, spray-based LX3 was also highly active against many well-known opportunistic plant pathogens in the hive suggesting this method may be especially useful for reducing honey bee vectoring of plant diseases. The collective
scope of this work is expansive and broadly relevant to microbial disease management in terrestrial ecosystems.

In silico prediction and in vitro assessment of microbial substrate utilisation among recently identified health-associated gut taxa

Cathy Lordan (Teagasc, Fermoy, Co. Cork, Ireland and University College Cork, Cork, Ireland), Aaron M Walsh (Teagasc, Fermoy, Co. Cork, Ireland), Dinesh Thapa (Teagasc, Fermoy, Co. Cork, Ireland), R. Paul Ross (APC Microbiome Ireland, Cork, Ireland), Paul D. Cotter (Teagasc, Fermoy, Co. Cork, Ireland and APC Microbiome Ireland, Cork, Ireland).

Introduction: Targeting more recently identified beneficial bacteria, including strict anaerobes, such as *Akkermansia muciniphila*, *Faecalibacterium prausnitzii* and *Eubacterium rectale*, through supplementation with different substrates, can enhance their growth and/or activity. Identification of substrates through in silico approaches can aid in elucidating which substrates may be best appropriate. However, validating these in silico-based tools is pertinent to developing robust genome-based predictions.

Methods: Seven strains from *A. muciniphila*, *E. rectale*, *F. prausnitzii*, *R. inulinivorans* and *R. bromii* were sequenced. The phenotypic microbial trait analyser, Traitar, and metabolic modelling tool, CarveMe, were used to predict substrate utilisation. Five simple sugars identified in both tools were chosen to be evaluated in vitro through growth experiments. Additionally, CAZymes were detected in the genomes using dbCAN2 to further elucidate carbohydrate degradation capacities on a genomic-level.

Results: Mean accuracy of the predictions acquired from Traitar (71.4%) and CarveMe (62.9%) were determined, although strain variability was observed. *R. bromii* 6883 had the capacity to consume all carbohydrates tested, whereas this was not the case for *R. bromii* X-30. CAZymes reflected strain-level differences between two *R. bromii* strains. A. muciniphila YL44 had the highest number of detected CAZymes, while *R. bromii* 6883 had the lowest. CAZyme number did not reflect the number of substrates consumed.

Discussion: Overall, both tools were fairly accurate in their predictions based on growth curves performed, demonstrating experimental validation is important in evaluating the accuracy of predictive in silico tools, while also highlighting the potential for identifying substrates that could enhance the growth and/or activity of microbes. Strain-level sensitivity of these tools shows potential with computational-based predictions for microbes well-studied and those less-studied.

Bacteriocin structural gene shuffling reveals multiple diverse structural gene homologues in the *S. bovis/S. equinus* complex pangenome.

Daragh Hill[1], Catherine Stanton[1], Paul Ross[2,3]

[1]APC Microbiome Ireland, Cork, Ireland [2]Teagasc Moorepark, Cork, Ireland [3]University College Cork, Cork, Ireland / APC Microbiome Ireland, Cork, Ireland).

Introduction: Using a combination of genome analysis and laboratory experiments, we discovered shuffling of bacteriocin structural genes in *Streptococcus infantarius* and *Streptococcus gallolyticus. Streptococcus gallolyticus* LL009 produces gallocin D, which is distinct from gallocin A produced by other *S. gallolyticus* strains. *S. gallolyticus* gene clusters share a high degree of gene synteny while the structural genes are highly variable. This prompted further investigation into all sequenced strains in the *Streptococcus bovis/Streptococcus equinus* complex (SBSEC) to determine the incidence and organisation of bacteriocin genes within this taxonomic group.

Methods: Whole genome sequencing led to the *in silico* identification of gallocin D. Gallocin D and infantaricin A peptides were synthesized and studied for activity. All sequences of the SBSEC were downloaded and analysed for bacteriocin potential using Bagel4. Bacteriocin operons were manually annotated for predicted peptides and aligned using ClustalO, entire operons were compared using MAUVE.

Results: Gallocin D is a narrow spectrum two component bacteriocin with mature peptides of 3343Da and 3019Da. These peptides were synthesized and display activity against VRE strain EC300 with a MIC value of 1.56 µM (Hill et al. 2020). Screening the SBSEC pangenome identified sixty-seven areas of interest (AOIs). These contained 166 predicted structural peptides, comprised of both class I and class II bacteriocins. The AOIs were aligned and grouped into six clusters based on gene synteny. Individual AOIs contained between two and fifteen structural genes.

Discussion: The operons containing multiple bacteriocin structural genes display remarkable diversity in their predicted mature peptide and signal sequences, however in some cases different bacteriocins shared almost identical leader sequences. It is tempting to speculate that the possession of such a variety of bacteriocin structural genes within this bacterial group offers competitive advantages to the producers in different microbial niche.

Honey varietals impact survivability of Bifidobacterium animalis ssp lactis in commercial yogurt through simulated in vitro digestion

David Alvarado (University of Illinois, Urbana, IL), Luis Alberto Ibarra-Sánchez (University of Illinois, Urbana, IL), Annemarie Mysonhimer (University of Illinois, Urbana, IL), Michael J. Miller (University of Illinois, Urbana, IL), and Hannah D. Holscher (University of Illinois, Urbana, IL).

Introduction: While numerous studies evaluated the probiotic properties of *Bifidobacterium animalis* ssp. lactis (*B. animalis*), how this bacterium interacts with honey in yogurt through the digestive process is limited. The study objective was to evaluate the effects of different honey varietals and concentrations on *B. animalis* survivability in yogurt through in vitro digestion. We hypothesize that adding the varietals to a yogurt matrix would improve probiotic survivability through in vitro complete digestion.

Methods: Yogurt, honey or control treated, underwent in vitro simulated oral, gastric, and intestinal digestion. Probiotic cells were enumerated on MRS medium and anaerobic incubation, followed by an overlay of selective MRS medium and anaerobic incubation. Probiotics were quantified at pre-digestion and after oral, gastric, intestinal digestion. Phase 1 included 4 honey varietals at 20% w/w per 170g of yogurt; phase 2 tested 7 levels of clover honey (20, 14, 10, 9, 8, 6, and 4% w/w) per 170g of yogurt.

Results: Similar probiotic counts were observed between all treatments after oral and gastric digestion (<1 Log CFU/g probiotic reduction after gastric phase). Higher *B. animalis* survivability was observed in yogurt with clover honey after exposure to simulated intestinal fluids (~3.5 Log CFU/g reduction) compared to all control treatments (~5.5 Log CFU/g reduction). The 20%, 14%, and 10% w/w clover honey similarly supported *B. animalis* survivability after exposure to simulated intestinal fluids.

Discussion: There was comparable *B. animalis* survivability in yogurt with alfalfa, buckwheat, and orange varietals relative to controls in all phases of in vitro digestion. In addition, the effective dose was demonstrated to be 1 to 2 tablespoons (10 to 20% w/w) of clover honey per serving (170g) of yogurt for increased probiotic survivability during in vitro digestion. In summary, the culinary combination of yogurt with clover honey supports *B. animalis* survivability during in vitro digestion.

The bacteriocinogenic potential of the Hungate1000 culture collection of rumen isolated micro-organisms

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Introduction: The Hungate1000 collection is a catalogue of rumen isolates. Bacteriocins are secondary metabolites with antibacterial activity and roles in niche clearing, colonisation resistance and spatial segregation. Production is considered a probiotic trait. Using in silico methods gives insights into the mechanisms and ecology of bacteriocin production. This study aimed to identify putative novel bacteriocin biosynthetic gene clusters (BGCs) from the rumen microbiome.

Methods: BGCs were predicted using Antismash and functionally annotated using InterPro. BGCs were classified on core biosynthetic machinery within the predicted operon, and distribution was overlayed on a phylogenetic tree. Propeptides were predicted using homology to existing peptides, key motifs within short open reading frames (sORF), a Neuripp score, and the genomic context of the sORF. Peptides were aligned using ClustalO, and regions of synteny were analysed using progressiveMauve and Easyfig.

Results: A total of 1072 BGCs were predicted across 349 genomes (85.1%); of these, 525 are putative RiPP/bacteriocin BGCs. The most abundant BGC determined were putative ranthipeptide BGCs, mainly among Clostridia. Lanthipeptides were predicted across multiple genera, with class II lanthipeptides the most diverse and large subset predicted. The majority of the BGCs identified belonged to Firmicutes, with the remainder among Proteobacteria, Actinobacteriota, Spirochaetota and Bacteroidota.

Discussion: In silico screening identified putative novel bacteriocin BGCs, including natural nisin variants. We hypothesise that screening the rumen microbiome for antimicrobial peptides may help find in situ solutions to reducing ruminant methane production whilst increasing animal feed efficiency. The abundance and diversity BGCs highlight the potential of the rumen as a reservoir for novel antimicrobial peptides for potential use in the agricultural, food and medical settings.

The Microbiome Modulating Effects of Superheated Steam (SHS) Treatment of Fibers

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Introduction: While a high fiber diet is associated with a healthy gut microbiome, daily fiber intake is below than the recommended level with associated negative impact on gut health. SHS treatment is a method used to inactivate oxidative enzymes and increase bioactive compounds in bran fibers. In this study, wheat and oat bran fibers were subjected to SHS treatment to investigate the potential prebiotic effect of SHS treated fibers compared to untreated controls.

Methods: Wheat and oat were processed using SHS treatment prior to in vitro digestion, followed by exposure to MicroMatrixTM in vitro fermentation. 16s rRNA sequencing, and short chain fatty acid analysis with GC-FID system were used to assess changes in gut microbiota and metabolomics, respectively. The DADA2 version 1.16 was used in the analysis of amplicon sequencing data. Alpha, beta diversity and taxonomic composition analysis were performed to explain microbiome data.

Results: SHS treatment of wheat and oat bran fibers resulted in significant differences in beta diversity and in several taxa, compared to untreated controls. PCoA revealed significant separation between SHS- treated oat and non-treated (NT) oat, and between SHS-treated wheat and NT wheat. Ruminococcus and Lactobacillus were higher in SHS-treated oat than NT oat, while Escherichia_Shigella and Alistipes were lower in SHS-treated oat group than NT oat group. Butyrate was higher in SHS treated oat than NT oat.

Discussion: This study showed that SHS treatment of oat and wheat bran have positively modulated gut microbiome. Lactobacillus was higher in SHS-treated oat than NT oat while the abundance of Escherichia_Shigella and Alistipes were found to be significantly lower in SHS-treated oat group than NT oat. Our results indicated that the microbiome modulating effect of oat bran was more effective than wheat bran. In conclusion, SHS treatment of bran fibers results in improvement of their prebiotic potential.

Probiotic bifidobacteria mitigate the deleterious effects of para-cresol in a *Drosophila melanogaster* toxicity model

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Introduction: Renal impairment associated with chronic kidney disease (CKD) causes the build-up of uremic toxins that are deleterious to the patient's health status. Current standard therapies that manage toxin accumulation in CKD offer an incomplete therapeutic effect against toxins like p-cresol and p-cresyl sulfate. As these toxins continuously build-up, they contribute to the production of reaction oxygen species which can accelerate CKD progression.

Methods: Using in vitro culture techniques, strains of lactobacilli and bifidobacteria from a 24 strain synbiotic were investigated for their ability to reduce p-cresol. Toxin clearance was measured using HPLC. To assess if p-cresol clearance was maintained in vivo a *Drosophila melanogastor* model was used.

Results: Four bifidobacterial strains tested internalize p-cresol. Toxin clearance was maintained when the multi-strain product was cultured under the same conditions. Oral supplementation of toxin-clearing bifidobacteria improved longevity ($P<0.0001$) of p-cresol exposed flies compared to controls by more than twenty days. We also showed p-cresol increased reactive oxygen species in the Malpighian tubules (i.e. fly kidney) of exposed flies and two of the four bifidobacteria reduced this oxidative stress.

Discussion: Using a *Drosophila* model, this work highlights why dosing with certain probiotic strains may be clinically useful in CKD. Cost effective oral supplementation of toxin clearing probiotics may be a useful adjunct therapy in CKD and should be evaluated in humans. In addition to reducing p¬-cresol produced by the gut microbiota, the synbiotic or its components might normalize the dysbiotic gut microbiota observed in CKD patients.

Compositional quality and quantity of mass marketed probiotic products and their Regulatory Insights in developing countries

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Introduction: The global retails are full of various probiotic supplements with a range of health assertion. The continuous scientific investigation and sales causes a substantial growth in the probiotic field. Reports on commercial product failures has driven the attention towards consuming live microorganisms solely based on label claims is not worth the risk. The study evaluates the commercialized products for their core microbial components in terms of quantity and quality.

Methods: We adopted a poly-phasic approach of classical culturing and molecular identification techniques e.g; 16srRNA, multi-locus sequence typing and ribotyping. A full array of in-vitro probiotic characterization assays were performed. The effect of gastro-intestinal phases on cell viability in the absence or presence of non-antibiotic drugs was tested using in-vitro static cost INFOGEST model. The strains were tested for all safety aspects as recommended in WHO/FAO guidelines.

Results: The data created declare the poor labeled species and dose harmonization. Only a third of the tested products were found to comply with their label claims. Strain sequence types showed different genome sequences of *L. acidophilus* LA-5, *L. rhamnosus* LGG, *B. animalis* BB12, and *C. butyricum*. Pheno- and genotypic resistance to more than three antibiotic classes, low microbial viability in GIT, weak antagonistic activity, and poor functional attributes compromise their usefulness.

Discussion: The unregulated and rampant use of these commercial probiotic strains carries the risk of spreading antibiotic resistance to more well-known intestinal infectious bacteria and the current regulatory systems do not consider this discrepancy. Current regulatory procedures fail to address these industry-related inconsistencies. Probiotics despite of having internationally endorsed definition and increasing consumers interest are not given a discrete trade category.

Milk fermented by *Lacticaseibacillus casei* improve barrier function and alter sterol metabolism in intestinal epithelial cells

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Introduction: How fermented dairy products can modify immunity and metabolism to benefit human health is largely unknown. In this study, we investigated the capacity of milk and milk fermented by *Lacticaseibacillus casei* BL23 (BL23-milk) and ATCC334 (A334-milk) to improve transcellular barrier integrity of intestinal epithelial cells (IEC).

Methods: Cell-free preparations of UHT milk fermented by *L. casei* BL23 or ATCC334 were applied onto differentiated Caco-2 cell monolayers in transwell inserts. IFNg was then applied and transepithelial electrical resistance (TER) across cell the monolayers was quantified 24 h later. To determine IEC responses to milk and *L. casei*, transcriptome analysis was performed using RNA-seq. An average of 2 million reads were obtained per sample. Follow up studies were performed using EGFR inhibitor, AG1478.

Results: BL23-milk and A334-milk, but not milk alone, significantly increased TER in an IFNg-dependent manner. IFNg application increased expression of genes in known IFNg modulatory pathways. Several of these genes were downregulated in IECs exposed to milk, BL23-milk, or A334-milk ($p < 0.05$). Only BL23-milk reduced expression of CLDN2 ($p = 0.02$). Only BL23- and A334-milk conferred increased expression of genes required for sterol metabolism ($p < 0.05$). AG1478 prevented *L. casei*-induced increase in TER.

Discussion: Secreted compounds resulting from *L. casei* growth in milk improved barrier function of Caco-2 cells exposed to IFNg. These results were not strain specific and were dependent on EGFR activation. Transcriptomic analysis strongly indicates a role for sterol metabolism in *L. casei*-dependent regulation of intestinal cell responses. These results support ongoing efforts to understand how the dairy matrix influences the capacity of probiotics to promote gastrointestinal health.

Integrative multi-omics analyses reveal the therapeutic impact of probiotics via modulating microbial-based signatures in chronic kidney disease

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Introduction: Growing clinical evidence supports that gut dysbiosis is a major contributor to adverse chronic kidney disease (CKD) progression, resulting in generation of gut-derived uremic toxins and aggravating kidney failure. Therefore, microbiota-based therapeutic interventions could be considered potent approaches to reduce uremic toxins and alleviate CKD progression. In the present study, we integrated multi-omics analyses to investigate the mechanisms of therapeutic impact of probiotics in feline CKD.

Methods: An open-label single-arm clinical study was conducted with cats with CKD stage 2-3 for 8 weeks. Each cat collected stool, urine, and blood specimens at three time points, i.e. before, after probiotic administration for 4 and 8 weeks (one capsule per day). Each capsule contained approximately 5 billion live freeze-dried bacteria of *Lactiplantibacillus plantarum* and *Lacticaseibacillus paracasei* (Lm). Strains were selected due to their functionality against CKD progression in our previous study.

Results: After 8 weeks of Lm administration, creatinine and blood urea nitrogen were decreased in 57% and 64% of CKD cats, respectively. Lm also decreased the level of serum gut-derived uremic toxins. Alpha diversity indices were increased significantly by Lm, indicating the transition to a more diverse gut environment. Moreover, Lm changed the level of specific bacteria and metabolites in cats, and these biomarkers showed a significant correlation with adverse factors associated with kidney function.

Discussion: We demonstrated Lm downregulated gut-derived uremic toxins and improved intestinal diversity, leading to alleviated CKD progression via modulation of microbial composition and metabolite production. Bacteria and metabolites showing correlation with kidney function indicators could be utilized as novel CKD prognostic/diagnostic biomarkers. Our findings suggest these biomarkers have the potential to be developed as next-generation/precision probiotics and medicine for alleviating CKD progression.

Comparison of the effect of fidaxomicin, thuricin CD, vancomycin and nisin on the human gut microbiota, both in vitro and ex vivo.

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Introduction: Vancomycin and metronidazole are commonly used treatments for *Clostridium difficile* infection (CDI). However, these antibiotics have been associated with high levels of relapse in patients. A new treatment for CDI, fidaxomicin, is described as a narrow spectrum antibiotic that is minimally active on the commensal bacteria of the gut microbiome. The aim of this study was to investigate the effect of fidaxomicin on the human gut microbiome and to compare it to thuricin CD, vancomycin and nisin.

Methods: The spectrum of activity of each antimicrobial was tested against 50 bacterial strains, including a variety of antibiotic resistant and gut strains, by well diffusion assay. MIC's were performed for each antimicrobial against a select number of those strains. The micro-Matrix™ mini fermentation system was used to simulate the environment of the colon. A pooled faecal slurry of 6 donors with 100µM of each antimicrobial and a no treatment control was assessed in the mini-fermentation system.

Results: Fidaxomicin, vancomycin and nisin were active against most Gram-positive bacteria tested, although fidaxomcin and vancomycin produced larger zones of inhibition. In contrast, activity of thuricin CD was specific to *C. difficile* and some Bacillus strains. Thuricin CD exhibited low MIC's for *C. difficile* and B. firmus. Whereas, fidaxomcin, vancomycin and nisin demonstrated lower MIC's for other strains tested when compared to thuricin CD. These results were mirrored in the micro-Matrix™ system.

Discussion: We conclude that the spectrum of activity of fidaxomicin is comparable to that of the broad spectrum antibiotic vancomycin, and is therefore broad spectrum. While active against *C. difficile*, fidaxomicin does show activity against gut commensal bacteria such as Bifidobacterium, Ruminococcus and some Lactobacillus strains, unlike thuricin CD which is very narrow spectrum.

Diversifying the offer of regional plant-based functional foods: development of a probiotic fermented drink of beets and strawberries

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Introduction: The consumption of fermented products has risen due to their proven beneficial effects, in particular on the intestinal microbiota. They are mainly home-made, which could imply risks for public health and lack of standardization. The state of Santa Fe in Argentina is one of the main producers of strawberries and other vegetables such as beets in the region. The aim of this work was to add value to regional produce by developing a fermented drink as a potential plant-based functional food.

Methods: Juice from beets and strawberries (20% and 10% w/v respectively) was obtained in a food processor, distributed in bottles and pasteurized. Drinks were then inoculated (1% v/v) with washed cultures of the probiotic strain *L. plantarum* 299v (Lp299v) or with L. plantarum F1B-GW (isolate from strawberries, LpF1B) and incubated (37°C;16h). Microbiological counts, pH and soluble solids(SS), total phenols, antioxidant capacity, betalains, and color measurment. were determined pre and post-fermentation.

Results: Fermented drinks (pH 3.5/SS 4.6°Bx for both Lp) with 8.8 and 8.7 logCFU/ml for Lp299v and LpF1B, were obtained. Controls (C) maintained their pH and SS. Microbiological indicators were satisfactory. Fermented drinks showed greater intensity of color and red tone than C (Lp299v >LpF1B) and betalains were also increased by lactic fermentation (Lp299v>LpF1B). The content of total phenols did not change whereas a significant increase in the antioxidant capacity for fermented drinks was observed.

Discussion: Fermented drinks were safe for consumption and obtained in a standardized way, with levels of *L. plantarum* 299v or the autochthonous strain *L. plantarum* F1B-GW according to the recommended doses for probiotic foods. They also presented enhanced color and antioxidant capacity, and were enriched with compounds with bioactive potential as betalains. In conclusion, a plant-based fermented drink was obtained based on regional produce with potential functional properties to be further studied.

Influence of food matrix on prebiotic efficacy of inulin-type fructans

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Introduction: The impact of food matrices on the prebiotic efficacy of inulin-type fructans (ITF) is of growing interest amongst the scientific community as previous research suggests that the food can either hinder or enhance the bioavailability of bioactive molecules including polyphenols. Yet, while prior studies have utilised numerous food products for ITF supplementation, due to differences in experimental design, drawing decisive conclusions on the food matrix impact on ITF efficacy cannot be undertaken

Methods: Study design was a prospective parallel-group, randomised trial involving 96 healthy adults lasting 10 days. Volunteers were assigned 1 of 4 ITF-fortified food products: rice milk, chocolate, shortbread, or pure inulin which were consumed 2x per/day resulting in a total ITF intake of 10g/day. Stool & urine samples were collected at Day-0 & Day-10 along with food & daily bowel habit diaries. Changes in microbial communities were analysed via FISH-FLOW & 16S rRNA sequencing.

Results: Targeted analysis via FISH-FLOW and 16S rRNA sequencing revealed that the selective effect of ITF towards bifidobacteria is unaltered in differing food matrices ($P < 0.01$ and $P < 0.001$). Further individual bacterial groups varied among the food products and the analysis method applied i.e. increases in Bacteroides/Prevotella (shortbread), Roseburia spp. & F. prausnitzii (shortbread and rice milk) & decreases in Clostridium cluster IVXA + IVXB & Blautia (inulin, shortbread, & rice milk).

Discussion: Our study aimed to determine the effects that differing food matrices have on the prebiotic efficacy of ITF using a standardised protocol. We confirm that irrespective of the food application and matrix, prebiotic ITF are selectively utilized and lead to specific changes in the gut microbiota. Bifidobacterium was the only genus consistently impacted by inulin-type fructans.

Prebiotic attribute of chitin oligosaccharides derived from sea food waste

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Introduction: Biodegradation of sea food waste leads to the formation of various value-added products such as chitin oligosaccharides (COS). *Paenibacillus* sp. AD has been reported to produce chitin oligosaccharides after degradation of sea food waste. Conditions were optimized for the degradation of sea food waste in solid state and concomitant production of COS. Prebiotic potential of COS was evaluated by in vitro fermentation of oligosaccharides with various intestinal microorganisms.

Methods: Chitin oligosaccharides were analysed in terms of degree of polymerization, molecular weight and degree of acetylation using various techniques. Furthermore, prebiotic potential of Chitin oligosaccharides of various degree of polymerization was evaluated by in vitro fermentation of chitin oligosaccharides with various intestinal microorganisms.

Results: COS was found to have beneficial effect on the growth of various *Lactobacillus* sp. and also limit the growth of various enteric pathogens, therefore having huge potential as prebiotic agents.

Discussion: Current research revolutionizes the production and availability of prebiotic chitin oligosaccharides as this is the study in which COS were produced directly from solid state fermentation of sea food waste. This is the economical and eco-friendly method in which chemical and enzymatic production of COS are not required. Moreover, this study shows prebiotic potential of N-acetylated glucosamine oligosaccharides by their in-vitro fermentation with various intestinal microorganisms.

Riboflavin-overproducing Limosilactobacillus reuteri for biofortification of fermented foods

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Introduction: Up to 50% of the population in developing countries displays riboflavin deficiency with negative impact on energy levels, reproductive function, lactation and pregnancy outcomes. This essential, water-soluble vitamin B2 is important for macronutrient and energy metabolism, and has antioxidant effects. Lactobacilli are promising candidates to simultaneously ferment and fortify fermented foods with vitamins, while additionally producing anti-pathogenic and immunomodulatory bioactive molecules.

Methods: Within the Isala citizen science project on women´s health, we isolated over 2000 bacterial strains. Here, 74 of these were genomically and functionally characterized for antimicrobial, epithelial barrier and immunostimulatory capacity. Riboflavin production was evaluated in laboratory media as well as in a milk or plant/derived matrices. Active transport riboflavin by intestinal Caco-2 cells, and passive transport in a gastrointestinal dialysis model was also evaluated, as well as survival.

Results: *Limosilactobacillus reuteri* AMBV339 showed a riboflavin production of 18.7 µg/mL, the highest described for non-genetically modified lactobacilli, with concomitant increase of riboflavin content in coconut and butter milk. AMBV339 also displayed anti-pathogenic, epithelial barrier-enhancing and immunostimulatory properties. The strain survived for 72h in a gastrointestinal dialysis model while releasing an active and passive stable riboflavin concentration with minimal gut microbiome modulation.

Discussion: We establish the properties of an efficient riboflavin-overproducing *L. reuteri* AMBV339 isolated from a healthy volunteer and its potential for fermented food fortification and implementation as a probiotic for the gut and potentially other body niches. Our results suggest that AMBV339 could provide food enrichment and health benefits in humans which could provide cost-effective solution, especially in disadvantaged populations.

Investigation of the Gut Microbiota Composition and Activity in Acute Myeloid Leukemic Patients: First Results of the MicroAML Study

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Introduction: The gut microbiota, a key regulator of host metabolism and immunity, is affected in preclinical models of leukaemia and cachexia. Reversing these changes in the gut microbiota can provide benefits to the host. To evaluate the translational value of pre-clinical studies, a multi-centric, prospective, observational study was initiated: MicroAML. The main objective of this study is to assess the composition and activity of the gut microbiota in patients diagnosed with acute myeloid leukaemia (AML).

Methods: Patients newly diagnosed with AML were recruited (n=30). Biological samples and clinical data were collected before any therapeutic intervention. Patients' food habits and cachectic hallmarks (e.g. appetite, muscle strength, body composition) were also collected. Control subjects from the general population were matched (1:1) for age, sex and BMI. Blood, faeces and urine were analysed using 1H- NMR metabolomics. The gut microbiota composition was assessed using shotgun metagenomics.

Results: Compared to control subjects, AML patients at diagnosis do not show significant difference in body composition and quality of their diet. The gut microbiota of AML patients shows some specific changes. AML patients also display clinical and metabolic alterations (inflammation, hyperglycaemia, anorexia, muscle weakness). We are now investigating the contribution of the gut microbiota to those metabolic changes by integrating metabolomics and metagenomics data.

Discussion: Our first results show significant changes in the composition of the gut microbiota as well as early signs of metabolic alterations in AML patients at diagnosis. Using a multi -omics strategy, we are now investigating whether these signs are related to changes in gut microbiota composition and activity.
ClinicalTrials.gov Identifier: NCT03881826

Screening the prebiotic effects of human milk oligosaccharides on 330 bacterial strains derived from the infant gut microbiota

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Introduction: Human milk oligosaccharides (HMOs) perform several functions critical to infant development, including acting as prebiotic stimulants for gut microbes. Several species of Bifidobacterium have demonstrated an ability to directly metabolize HMO structures in vitro, but data on the capacity of other species is limited. The aim of this study was to evaluate the ability of infant gut- associated bacterial strains to metabolize HMOs in monocultures.

Methods: A total of 330 bacterial strains, spanning 160 species, were isolated from infant fecal cultures, using a variety of selective and non-selective media types under aerobic and anaerobic conditions. Strains were treated with 15 g/L pHMOs (or a no-treatment control) in biological triplicate under anaerobic conditions. Growth responses were monitored over 48 h, and the degradation of 19 HMO structures were evaluated by HPLC-glycoprofiling to determine microbial HMO structure preferences.

Results: A wide variety of taxa were capable of degrading HMOs in vitro. Some strains demonstrated the ability to metabolize numerous HMO structures while others displayed structure-based specificity. In most cases, strains of the same species exhibited similar HMO metabolism capabilities, but exceptions included strains of [*Ruminococcus*] *gnavus* and *Alistipes finegoldii*. HMOs also suppressed the growth of several strains of species from the *Sellimonas* and *Sutterella* genera.

Discussion: HMOs interacted with a wide range of species, including those reported to be beneficial to human health as well as potential pathogens. Previously, direct HMO metabolism has only been reported in 17 species not including Bifidobacterium members. This study has greatly increased this figure through the discovery of HMO use in 60 previously untested species. Overall, this study has considerably expanded our knowledge of HMO–gut microbiota interactions.

Effect of an *Aspergillus oryzae* derived postbiotic on heat stress in *Drosophila melanogaster* and dairy cows

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Introduction: *Drosophila melanogaster* (DM) has been increasingly applied in nutrition research in the past years. Due to its characteristics in its nutritional physiology and molecular targets, findings in DM could be relevant for livestock like dairy cows. Heat stress has been shown to impair cellular function and performance in animals. Thus, we tested the efficacy of a postbiotic derived from *Aspergillus oryzae* (AOP) to modulate thermal tolerance in ectothermic fruit flies and endothermic dairy cows.

Methods: Two DM strains were cultivated for 24h in a standard culture sucrose-yeast or same culture with 5% (v/v) AOP. Bodyweight and composition, metabolic rate, and heat stress (HS) tolerance (39°C,75 min) was measured. For 36 d, 48 lactating dairy cows were fed 0 (C), 3 (L), 6 (M) or 18 g/d (H) AOP in a TMR (41:59; F:C). Cows were in summer heat for 10 d with heat abetment. On d 11, heat abetment was removed. Intake, milk yield, body temperature, respiration rate and inflammation were evaluated.

Results: Feeding AOP improved survivability of HS flies compared to the control (58% vs. 25%, respectively). This response was associated with downregulation of genes associated with modulation of oxidative stress and immunity. In dairy cows, AOP reduced circulating acute phase proteins and expression of IL-6 expression in white blood cells. Additionally, AOP increased energy-corrected milk yield and its components, suggesting improved lactogenesis and metabolic efficiency.

Discussion: Collectively, our data demonstrate that a postbiotic from *Apergillus oryzae* significantly enhances thermal tolerance and performance both in Drosophila melanogaster as well as in lactating dairy cows. *Drosophila melanogaster* is as a versatile model organism which can be applied to get mechanistic insights by which postbiotics relevant to animal nutrition modulate heat related stress response.

Integrating Dietary Data into Microbiome Studies: A Step Forward for Nutri-Metaomics

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Introduction: Nowadays it is accepted that gut microbiota, plays a key role in human's health and that diet may influence its composition. Commonly, dietary intake is obtained from long dietary assessment methods, what could lead to misreporting and low collaboration. Furthermore, this relationship is typically evaluated using specific populations groups together with concrete or extreme diets. Thus, we decided to (1) design a new semi-quantitative and simplified FFQ (sFFQ); (2) undertake a relative validation analysis; (3) perform a reproducibility analysis; and (4) correlate dietary intake with microbiome data.

Methods: We conducted two consecutive studies (n = 84: a first pilot study (n = 40) to build a web-based, semi-quantitative simplified FFQ (sFFQ) based on three 24-h dietary recalls (24HRs); a second study (n = 44) served to validate the newly developed sFFQ using three 24HR as reference method. Information about microbiota was obtained from 2 fecal samples by each participant. Gut microbiome profiling (16S rRNA gene) was combined with the extracted dietary and lifestyle data.

Results: Relative validation analysis provided acceptable classification and agreement for 13 out of 24 (54%) food groups and 20 out of 29 nutrients (69%) based on intraclass correlation coefficient, cross-classification, Spearman's correlation, Wilcoxon test, and Bland–Altman. Microbiome analysis showed that higher diversity was positively associated with age, vaginal birth, and intake of fruit. In contrast, microbial diversity was negatively associated with BMI, processed meats, ready-to-eat meals, sodium, and saturated fat.

Discussion: Our results suggest that the new sFFQ could be applied for future dietary studies. On the one hand, through the sFFQ, we were able to associate high microbial diversity, which is considered a health-promoting factor, with the intake of fruits and low diversity with less "healthy options" such as total fat, saturated fatty acids, and sodium intake. However, still there is need to test the sFFQ in a larger cohort as well as to improve information from microbiota using –omics data.

FunOMIC: Pipeline with built-in Fungal Taxonomic and Functional Databases for Human Mycobiome Profiling

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While analysis of the bacterial microbiome has become routine, that of the fungal microbiome is still hampered by the lack of robust databases and bioinformatic pipelines. Here, we present FunOMIC, a pipeline with built-in taxonomic (1.6 million marker genes) and functional (3.4 million non-redundant fungal proteins) databases for the identification of fungi. Applied to more than 2,600 human metagenomic samples, the tool revealed fungal species associated with geography, body sites, and diseases. Correlation analysis provided new insights into inter-kingdom interactions. With this pipeline and two of the most comprehensive fungal databases, we foresee a fast-growing resource for mycobiome studies.

