



2026 SYMPOSIUM GUIDE



Symposium Sessions
Tokyo, Japan • October 6, 2026



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On behalf of the ISAPP board, I'd like to welcome you to ISAPP's 2026 annual scientific meeting. This meeting is particularly notable because we are convening in Tokyo, Japan, home to a rich diversity of fermented foods and stellar industry and academic communities engaged in biotics and microbiome research. I am excited that we were able to join those communities in cooperation with the Japanese Association for Food Immunology, The Intestinal Microbiology Society, and the Japan Society for Lactic Acid Bacteria to organize a joint symposium. With this event, our goal is to foster greater collaboration between researchers to address emerging and

urgent questions across disciplines and topics on probiotics, prebiotics, synbiotics, postbiotics, and fermented foods. Since ISAPP's first annual meeting in Asia, held in 2018, the landscape of biotics and microbiome research has experienced significant change across clinical, fundamental, industrial, and societal domains. Because interest in biotics and microbiome research continues to grow, now more than ever there is a need for standardized approaches and greater integration in biotics research at a global scale, transcending geographies.

In the past year, ISAPP has continued to provide leadership and guidance on biotic research. This year, we published the first consensus definition of gut health, a publication that was highlighted on the cover of a major medical journal. Addressing a common translational issue, an ISAPP expert panel and subsequent editorial explored how and why probiotic guidelines developed by clinical organizations differ and provided practical guidance for navigating conflicting recommendations. These authoritative papers complement ISAPP resources for the general public, including newly developed infographics on the vaginal microbiome and gut health, as well as ongoing scientific highlights shared through our podcast and blogs. We also launched a new webinar series to bring scientific and clinical information directly to our community of industry scientists. We thank all who have collaborated with us over the past year for helping advance the ISAPP mission.

We are particularly grateful to Seppo Salminen, whose term on the board is ending, but who has left an indelible mark through his many contributions over the past 14 years. We welcome our newest board member, Professor Maria Carmen Collado, who brings critical expertise in early-life nutrition, the gut microbiome, and probiotics, and whom we know will continue the tradition of excellence in advancing the ISAPP mission. Importantly, all of this is possible because of our Executive Director, Marla Cunningham, who has been steadfast in sustaining and building ISAPP. Last but not least, I hope you will join me in celebrating the ISAPP community. As participants in the annual meeting, you are strengthening this community by bringing diverse perspectives and distinct disciplinary expertise to advance biotic science to new heights for future generations.

Maria Marco, ISAPP President

Welcome to the ISAPP 2026 scientific symposium and annual scientific meeting, in Tokyo Japan! Through this international event, we are excited to bring together expert scientists from across the globe to share knowledge, discuss and debate diverse perspectives, and build synergy and collaboration around shared interests and complementary expertise.



The first day features a symposium plenary program co-developed with the Japanese Society for Lactic Acid Bacteria, the Intestinal Microbiology Society, and the Japanese Association for Food Immunology. The symposium will gather leading academic, clinical and industry experts in biotic science from North and South America, Europe, Australasia, and across Asia, with plenty of networking opportunities. Along with plenary presentations from high impact local and international researchers, the program also features interactive sessions including a discussion panel on international collaboration in biotic research, and a networking and poster session showcasing studies from around the globe. We will welcome an extended ISAPP community during these sessions, with additional delegates joining us from near and far.

These symposium sessions will be followed by ISAPP's annual meeting program over two days, for invited experts, industry member scientists and selected early career researchers. The program includes plenary lectures, late breaking news, talks from 2026 ISAPP award recipients, and an afternoon of workshops on current and topical scientific issues. The program will conclude with a member program for student and industry members to explore ongoing implementation of science in the field.

On behalf of ISAPP, we thank you for joining with the ISAPP community and sharing your contributions. Whether you're joining us for the one day symposium sessions or the full three day program, we hope your experience will be enriched with research and insights that excite you, new connections and collaborators, and time to enjoy the rich community of scientists in our field, within the wonderful setting of Tokyo. We are looking forward to connecting with you.

Marla Cunningham, ISAPP Executive Director

ISAPP Symposium Sessions have been co-developed with the following local scientific societies to curate a program featuring local and international scientists to maximize international knowledge exchange and professional connections.



Japanese Society for Lactic Acid Bacteria

The Japanese Society for Lactic Acid Bacteria (JSLAB) is dedicated to advancing research on lactic acid bacteria, bifidobacteria, probiotics, fermented foods, and related fields. Through scientific meetings, educational activities, and collaboration among researchers and industry, JSLAB promotes the exchange of knowledge and supports the development of research on lactic acid bacteria and related biotics in Japan. As Japan has a long tradition of fermented foods and lactic acid bacteria research, we hope that hosting this symposium in Tokyo will inspire new scientific discussions and collaborations across disciplines and countries. We look forward to welcoming colleagues from around the world and to advancing the science of biotics together.

Website: [Japan Society for Lactic Acid Bacteria](http://www.jslab.jp)



The Intestinal Microbiology Society (IMS)

Intestinal Microbiology Society

The Intestinal Microbiology Society (IMS) is a Japanese scientific society dedicated to advancing research and education in intestinal microbiology, including the gut microbiota, host-microbe interactions, probiotics, prebiotics, and related areas of health and disease. Through scientific meetings and interdisciplinary collaboration among academic and industrial researchers, IMS promotes the exchange of knowledge and the development of innovative research in this rapidly evolving field. We are delighted to collaborate with ISAPP in this symposium, which provides a valuable opportunity to share the latest scientific advances, foster international connections, and strengthen collaboration among researchers dedicated to improving human health through microbiome science.

Website: [The Intestinal Microbiology Society](http://www.ims-japan.org)



Japanese Association for Food Immunology

The Japanese Association for Food Immunology (JAFI) was established to promote food immunology as a scientific discipline, to disseminate scientific evidence on the immunomodulatory functions of foods, and to contribute to the maintenance and improvement of human health through the advancement of related scientific research. Probiotics and prebiotics are certainly among the key topics in food immunology, and we are delighted to collaborate with ISAPP in co-organizing this symposium.

Website: [Japanese Association for Food Immunology](http://www.jafi.or.jp)



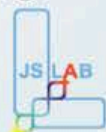
ISAPP 2026 ANNUAL MEETING

IN ATTENDANCE

Local researchers and clinicians



The Intestinal Microbiology Society (IMS)



日本乳酸菌学会

Japan Society for Lactic Acid Bacteria



Japanese Association for
Food Immunology

International representation



Delegates
arriving from
32 countries



31 Industry
member
companies (IAC)



63 academic
institutions
represented



Early career
researchers



ISAPP
Academic
Board

ISAPP award winners



Talks by recipients of 3
prestigious ISAPP awards

HIGHLIGHTS

Discover innovative biotics research from global leaders



- Plenary talks
- Symposium sessions

Connect with international researchers in biotic and microbiome science



- Networking breaks
- Welcome reception
- Social events

Gain insights for global research collaborations



- Expert panel - international research collaboration

Hear about the latest research from rising early career stars



- Featured Students and Fellows Association (SFA) talks
- Poster session

See how research is being commercialised and developed in industrial settings



- Featured ISAPP Industry Advisory Committee (IAC) talks
- Poster session

Workshop key scientific challenges with topic experts



- Discussion groups

All program events are held on the fourth floor (4F) of the Akasaka Intercity Conference Center (AICC), Akasaka, Tokyo, Japan unless otherwise noted below. Meeting language is English.

TUESDAY 6th OCTOBER: SYMPOSIUM SESSIONS

Sessions open to external registration. Registration is included in the annual meeting for invited experts, industry members and SFA delegates.

07:45 - 08:30 Registration
Foyer 4F

08:30 - 08:45 **Symposium welcome**
Plenary 4F AIR
Maria Marco, ISAPP President
Haruki Kitazawa, Japanese Society for Lactic Acid Bacteria, President
Hiroshi Ohno, Intestinal Microbiology Society, President
Satoshi Hachimura, Japanese Association for Food Immunology, President

Theme 1: Human and animal microbiomes in health and disease

In collaboration with Intestinal Microbiology Society of Japan  The Intestinal Microbiology Society (IMS)

08:45 - 09:15 **Gut microbial metabolites via GPCRs and host metabolic homeostasis**
Junki Miyamoto, Tokyo University of Agriculture and Technology, Japan

09:15 - 09:45 **Insights into the microbiota-gut-brain axis from the Kyotango Longevity Research.**
Yuji Naito, Kyoto Prefectural University of Medicine, Japan

09:45 - 10:15 **Applying machine learning to clinical biotic and microbiome research**
Hannah D. Holscher, University of Illinois Urbana-Champaign, USA

10:15 - 10:25 **Abstract: Diversity of *Lactobacillaceae*, *Bifidobacteriaceae*, and other taxa used by the probiotics industry in small intestinal samples**
Joseph Wang, Nimble Science, Canada

10:25 - 10:30 Students and Fellows Association: Introduction

10:30 - 11:00 *Morning break, posters displayed*
Foyer 4F

11:00 - 12:30 **Expert panel: International collaboration in biotic research**
Plenary 4F AIR
Maria Marco, University of California, Davis, USA (moderator)
Karsten Kristiansen, University of Copenhagen, Denmark
Sarah Lebeer, University of Antwerp, Belgium
Tahmeed Ahmed, International Centre for Diarrhoeal Disease Research, Bangladesh
Jan Knol, Danone Global Research and Innovation, Netherlands
Takuya Akiyama, Yakult Central Institute, Japan

12:30 - 13:30 *Lunch break, posters displayed*
Foyer 4F

2026 ANNUAL MEETING PROGRAM

Plenary 4F AIR

Theme 2: Ingested microbes: mechanisms and metabolites

In collaboration with Japanese Society for Lactic Acid Bacteria


日本乳酸菌学会
 Japan Society for Lactic Acid Bacteria

13:30 - 14:00

The phylogeny and ecology of lactobacilli determines their functionality in food fermentations

Michael Gänzle, University of Alberta, Canada

14:00 - 14:30

Molecular basis of *Bifidobacterium*-human symbiosis: Insights from microbial metabolism

Takane Katayama, Kyoto University, Japan

14:30 - 15:00

Smart bugs, smarter medicine: Tackling gut cancer

Susan L. Woods, University of Adelaide, Australia

15:00 - 15:10

Abstract: Translating pasteurized *Akkermansia massiliensis* LWHK0003 from preclinical models to clinical application as a next-generation postbiotic candidate for MASLD

Wei-Kai Wu, National Taiwan University, Taiwan

15:10 - 15:40

Afternoon break, posters displayed

Foyer 4F

Plenary 4F AIR

Theme 3: Biotics and immune interactions

In collaboration with Japanese Association for Food Immunology


Japanese Association for Food Immunology

15:40 - 16:10

Novel microbiome-related targets for colorectal cancer

Dirk Haller, Technical University Munich, Germany

16:10 - 16:40

Effects of dietary fiber on the intestinal immune system

Yukihiro Furusawa, Toyama Prefectural University, Japan

16:40 - 17:10

Maternal transfer of bifidobacteria and metabolic pathways in allergy development

Susanne Brix, Technical University of Denmark, Denmark

17:10 - 17:20

Abstract: Postbiotics derived from *Ligilactobacillus salivarius* improve mitochondrial homeostasis via TLR2-mediated signaling

Fu Namai, Tohoku University, Japan

17:30 - 19:30

Networking reception and poster session

Foyer 4F

Visit posters from researchers across the globe, and enjoy local catering and networking.

Symposium close

ISAPP is delighted to share this lineup of local and international scientists to speak on the latest advancements in biotic science. Familiarize yourself with our plenary speakers' biographies and abstracts below. The speakers are listed in order of program appearance.

Tuesday October 6 - 08:45-09:15

Gut microbial metabolites via GPCRs and host metabolic homeostasis Junki Miyamoto, Tokyo University of Agriculture and Technology, Japan

Excessive energy intake associated with the western diet habits has led to a continuous increase in the prevalence of metabolic disorders, including obesity and type 2 diabetes. In particular, the consumption of sucrose has markedly increased worldwide, and sucrose-induced metabolic dysfunction has become a major public health concern. Recent advances in gut microbiome research have revealed that gut microbiota play a pivotal role in maintaining host metabolic homeostasis. Among the diverse microbial metabolites, short-chain fatty acids (SCFAs) have attracted considerable attention because of their profound effects on immune and metabolic functions. SCFAs are primarily produced through microbial fermentation of indigestible polysaccharides, such as dietary fiber, in the intestine. Certain lactic acid bacteria are known to synthesize indigestible exopolysaccharides (EPS) from sucrose; however, their physiological significance in vivo remains unclear. Here, we focused on the EPS-producing bacterium *Leuconostoc mesenteroides* and investigated the impact of EPS on host energy metabolism and gut microbial SCFA production. We demonstrated that dietary EPS enhances SCFA production by the gut microbiota and improves glucose metabolism while suppressing obesity through SCFA receptor-mediated signaling pathways. Furthermore, EPS selectively promotes the growth of specific gut bacterial populations, resulting in elevated intestinal SCFA production and acting as a previously unrecognized prebiotic factor (Miyamoto et al., Gut Microbes. 2023). Moreover, we discovered that gut bacteria themselves can convert dietary sucrose into fiber-like EPS in the intestine. We found that EPS produced by *Streptococcus salivarius* from dietary sucrose serves as a substrate for other gut bacteria, particularly members of the genus *Bacteroides*, thereby enhancing SCFA production and contributing to the regulation of host energy metabolism. These findings revealed a novel microbiota-mediated metabolic network linking dietary carbohydrates, microbially derived EPS, SCFAs, and host receptor signaling to maintain energy homeostasis (Shimizu et al., Nature Commun. 2025). In this lecture, I will present our recent findings on the mechanisms by which gut microbiota-derived metabolites, particularly SCFAs, regulate host metabolic homeostasis through their receptors. I will also discuss emerging concepts regarding microbial carbohydrate metabolism and its implications for the prevention and treatment of metabolic diseases.



About this speaker:

Dr. Junki Miyamoto is an Associate Professor at Tokyo University of Agriculture and Technology, Japan. He received his PhD in Agriculture from Hiroshima University in 2017. His research focuses on diet-derived gut microbial metabolites and their roles in regulating host homeostasis. By employing interdisciplinary approaches, including microbiology, metabolomics, and nutritional science, he investigates host-microbiota interactions at molecular and physiological levels. His work aims to elucidate the mechanisms by which microbial metabolites mediate metabolic and immune functions, thereby linking dietary factors to host health. Through his research, he seeks to advance fundamental understanding of gut microbiota-host interactions and to contribute to the development of scientific bases for nutritional interventions and functional foods.

Tuesday October 6 - 09:15-09:45

Insights into the microbiota-gut-brain axis from the Kyotango Longevity Research.

Yuji Naito, Kyoto Prefectural University of Medicine, Japan

The Kyotango region, located in the northern part of Kyoto Prefecture, has an aging rate exceeding 38% among individuals aged 65 and older, making it the most rapidly aging region in the prefecture. However, this region also has one of the highest proportions of centenarians in Japan, with a prevalence approximately three times the national average, making it a notable area for healthy longevity. To investigate the factors contributing to this phenomenon, Kyoto Prefectural University of Medicine established the Department of Longevity and Regional Epidemiology and has been conducting health checkups and cohort studies in this region since 2017. In this cohort study, we analyzed data from 1200 individuals aged 65 years or older who had available gut microbiota profiles, frailty indices, and dietary nutrition survey data. Cognitive function was evaluated using the Mini-Mental State Examination (MMSE), with scores of ≤ 23 indicating suspected dementia, 24–27 indicating mild cognitive impairment (MCI), and 28–30 classified as normal cognition. Among the 578 individuals with MMSE data, 43 (7.4%) were suspected of having dementia, and 255 (44.1%) were classified as having MCI. Among the microbial features, Bifidobacterium showed a significant positive association with cognitive function. Age was inversely associated with Bifidobacterium, whereas Bifidobacterium was positively associated with MMSE. Mediation analysis demonstrated that Bifidobacterium partially mediated the association between age and MMSE (indirect effect: -0.0107 , 95% CI: -0.0196 to -0.0034 , $p = 0.004$). Furthermore, dietary analysis revealed that milk intake was positively associated with Bifidobacterium, suggesting a modifiable pathway linking diet, microbiota, and cognition. Our findings provide evidence that age-related changes in the gut microbiota, particularly reductions in Bifidobacterium, partially mediate cognitive decline in older adults. These results support the existence of a diet–microbiota–brain axis and highlight the gut microbiota as a potential target for interventions aimed at preserving cognitive function during aging.



About this speaker:

Yuji Naito, M.D., Ph.D., obtained his M.D. degree at the Kyoto Prefectural University of Medicine, Kyoto, Japan in 1983, and performed Ph.D. studies at the same university from 1983–1993. He is currently a Professor of Department of Human Immunology and Nutrition Science, Kyoto Prefectural University of Medicine. From 2017, his group has started the longevity cohort study in Kyotango, Japan, one of the world's most intensively studied regions for healthy aging. Research conducted in this population has revealed distinctive lifestyle patterns, dietary habits, and biological signatures that together support extended healthspan. At the center of these findings lies the microbiota-gut-brain axis, a critical integrative system connecting metabolism, immunity, inflammation, and cognitive health.

Tuesday October 6 - 09:45-10:15

Applying machine learning to clinical biotic and microbiome research

Hannah D. Holscher, University of Illinois Urbana-Champaign, USA

Clinical biotic and microbiome research increasingly generates complex, high-dimensional data from dietary assessments, clinical measures, multi-omics profiles, and the scientific literature itself. As datasets grow in scale and dimensionality, advanced analytical approaches are increasingly needed to extract meaningful patterns and advance research. Artificial intelligence (AI) technologies, particularly machine learning and deep learning, provide powerful tools for addressing these challenges. However, these tools are most useful when their selection and use are driven by biological questions and informed by the structure and scale of the available data.

This presentation will provide a practical overview of machine learning and AI approaches relevant to clinical biotic and microbiome research, including supervised learning, unsupervised learning, dimensionality reduction, deep learning, natural language processing, and large language models. Examples will illustrate how these approaches can be applied across the research pipeline. Case studies will include the use of random forest models to identify fecal bacterial, metabolomic, and metagenomic biomarkers of food intake; deep learning to predict personalized metabolite responses to dietary interventions; and natural language processing to extract diet-microbiome associations from the scientific literature. The final portion of the presentation will focus on practical considerations for selecting and evaluating machine learning approaches. AI is not an endpoint but rather a tool for extracting information from complex data. Thoughtful integration of these tools, grounded in methodological rigor and domain expertise, remains essential for analytical advances to contribute to scientific discovery and clinical translation.



About this speaker:

Dr. Hannah Holscher is an Associate Professor of Nutrition at the University of Illinois Urbana-Champaign, where she also serves as Associate Director of the Personalized Nutrition Initiative and holds affiliations with the Division of Nutritional Sciences, the Carl R. Woese Institute for Genomic Biology, and the National Center for Supercomputing Applications. Her research integrates clinical nutrition, microbiome science, and computational biology to explore how diet influences human health. She has authored more than 90 peerreviewed publications and leads multidisciplinary research projects supported by government agencies, foundations, commodity boards, and private industry. Dr. Holscher has held leadership roles within the American Society for Nutrition and serves on the editorial boards of The Journal of Nutrition and Nutrition Research.

Tuesday October 6 - 10:15-10:25

Diversity of *Lactobacillaceae*, *Bifidobacteriaceae*, and Other Taxa Used by the Probiotics Industry in Small Intestinal Samples

Joseph Wang, Nimble Science, Canada

We constructed a large-scale small intestinal metagenomic dataset comprising over 400 samples collected using a non-invasive sampling device (SIMBA capsules) across multiple observational clinical studies. All samples were paired with detailed clinical metadata, and most matched fecal samples were biobanked. The cohort represents a well-characterized Canadian population, providing a controlled demographic baseline that can support future clinical investigations and comparative studies.

Unlike conventional endoscopic sampling, which captures a single localized point, SIMBA capsule-derived samples demonstrated improved sample quality, as indicated by a higher proportion of nonhost sequencing reads against endoscopy (79.5% vs. 7.91%). The SIMBA capsule enables broader sampling through its larger inlet diameter and extended sampling time. This design likely contributes to increased sample volume and more representative microbial capture across the small intestine. In addition, a small-scale study involving three participants, each undergoing six repeated SIMBA sampling sessions over a two-week period, provided preliminary evidence supporting the reproducibility of small intestinal (SI) samples obtained using this approach appears higher than that observed in matched fecal samples from the same individuals.

Then we applied a high-resolution, non-redundant genome catalog to perform in-depth, subspecies-level profiling of whole-metagenome shotgun sequencing data derived from capsule samples. Building upon recent studies highlighting the extensive natural diversity of *Bifidobacterium* in fecal microbiomes, this work expands the scope to the small intestine, addressing a key gap in probiotic-related microbial ecology.

Our preliminary results reveal distinct patterns in the diversity and prevalence of probiotic-associated taxa. Well-known probiotic species such as *Limosilactobacillus reuteri* were detected with both high diversity and prevalence. Other taxa, including *Streptococcus thermophilus*, exhibited high prevalence but limited diversity. In contrast, species such as *Lactocaseibacillus paracasei*, *Lactocaseibacillus casei*, and *Limosilactobacillus fermentum* showed both low diversity and low prevalence, suggesting a more niche distribution within the small intestinal environment.

Collectively, this analysis provides a comprehensive view of probiotic-relevant taxa in the small intestine. These findings extend beyond microbial cataloging by informing the ecological fitness, persistence, and niche specificity of probiotic strains in their target site of action, thereby supporting more rational design, validation, and personalization of probiotic interventions. To date, a series of biotic interventions are underway (<https://clinicaltrials.gov/search?term=SIMBA%20Capsule>), including probiotics, antibiotics, prebiotics, FMT, dietary lipids, proteins, etc.



About this speaker:

Passionate and driven to share his technology with this emerging new field, co-founder and inventor Joseph Wang is leading Nimble Science to partner with leading industry partners from around the world. Dr. Wang has deep expertise in wearables and ingestibles with a track record of commercial success. He completed his Ph.D. in Biomedical Engineering at the University of Calgary and his Bachelor in Bioengineering at the National University of Singapore.

Tuesday October 6 - 13:30-14:00

The phylogeny and ecology of lactobacilli determines their functionality in food fermentations

Michael Gänzle, University of Alberta, Canada

Background. Members of the *Lactobacillaceae* with *Lactococcus lactis* and *Streptococcus thermophilus* are a major part of the microbial communities in most fermented foods. Alongside bifidobacteria, lactobacilli are also major representatives in probiotic preparations. The phylogeny of lactobacilli matches their ecology and metabolism (1), thus, the taxonomic identification is a suitable initial approach to identify suitable starter cultures and probiotics.

Starter cultures for traditional and novel animal foods. Some back-slopped dairy fermentations are dominated by domesticated lactic acid bacteria, i.e. *Lactobacillus delbrueckii* subsp. *bulgaricus*, a dairy-associated clade of *Lactococcus lactis* subsp. *lactis* and, likely, *Lactobacillus kefiranofaciens* subsp. *kefiranofaciens* (2). For domesticated organisms, phylogeny is the primary criterion for selection of starter cultures. Microbial communities in fermented meats and fishes are dominated by homofermentative lactobacilli, particularly psychrotrophic *Latilactobacillus* spp.; the same organisms are also suitable for novel fermented insect products (3).

Starter cultures for traditional and novel plant foods. Plant substrates generally provide maltose, sucrose and / or raffinose, i.e. the preferred carbon sources of food-fermenting heterofermentative lactobacilli. In most plant food fermentations, heterofermentative lactobacilli are thus the dominant, or among the dominant fermentation microorganisms (3,4). Heterofermentative lactobacilli fundamentally impact product quality by formation of CO₂ and acetate in their primary metabolism; in addition, the reduction of aldehydes resulting from lipid oxidation, and the reduction of oxidised glutathione strongly impact product quality (4). Glutathione reduces intra- and intermolecular disulfide bonds in proteins, which alters their technological functionality, allergenicity, and digestibility. An additional metabolic trait that is predominantly observed in heterofermentative lactobacilli is the conversion of lactate via 1,2-propanediol to propionic acid. The conversion aids in preservation of silage and sourdough bread, and facilitates refrigerated storage of sourdough starter cultures. The conversion of (poly-)phenolic compounds in plant fermentations is also strongly linked to lifestyle and phylogenetic position of lactobacilli.

Probiotic cultures. Lactobacilli with well-documented probiotic properties are predominantly or exclusively recruited from genera that are adapted to vertebrate hosts (*Lactobacillus*, *Ligilactobacillus*, and *Limosilactobacillus*) or nomadic organisms of the *Lactocaseibacillus casei* and *Lactiplantibacillus plantarum* groups (5). Here, phylogeny also provides a suitable initial screening tool before additional *in silico*, *in vitro* and *in vivo* assays identify suitable strains.

Conclusions. The strong link of phylogeny to ecology and metabolic traits enables the use of the phylogenetic position of lactobacilli as a relevant screening tool to select lactobacilli for food and probiotic applications.

References. (1) Doi: 10.1128/AEM.02116-15 (2) Doi: 10.1016/j.foodres.2025.117562, Doi: 10.1016/j.cell.2024.08.008 (3) Doi: 10.1146/annurev-food-072023-034207 (4) Doi: 10.1016/j.fm.2025.104945 (5) Doi: 10.1038/nrgastro.2017.75.



About this speaker:

Prof. Michael Gänzle is trained as Food Engineer at the University of Hohenheim in Stuttgart, Germany, and obtained his doctoral and post-doctoral degrees in Food Microbiology at the University of Hohenheim and the Technical University of Munich, respectively. He is Professor and Canada Research Chair in Food Microbiology and Probiotics at the University of Alberta, Edmonton, Canada, and was elected Fellow of the Royal Society of Canada in 2023. From 2021 to 2024, he was recognized as Clarivate Highly Cited Researcher. His research links microbial phylogeny, physiology and ecology to food quality, food safety and host health. More information is provided on the institutional [homepage](#).

Read ISAPP's recent [blog article](#) providing context for this talk and an interview with Prof. Gänzle.

Tuesday October 6 - 14:00-14:30

Molecular basis of *Bifidobacterium*-human symbiosis: Insights from microbial metabolism

Takane Katayama, Kyoto University, Japan

Since the isolation of two host glycan-degrading enzymes, 1,2- α -L-fucosidase and endo- α -N-acetylgalactosaminidase, in bifidobacteria, both of which had been annotated as proteins of unknown function in several gut microbes at that time^{1,2}, we have sought to elucidate the molecular mechanisms underlying the symbiotic relationship between bifidobacteria and humans mediated by host glycans, such as human milk oligosaccharides (HMOs) and mucin O-glycans, from the perspectives of bacterial enzymology and physiology. In this talk, I will summarize our research, with particular focus on the following three topics.

HMOs: A unique feature of HMOs is their richness in type 1 chains (Gal β 1-3GlcNAc-O-R) over type 2 chains (Gal β 1-4GlcNAc-O-R), and as reflecting this feature, infant gut-associated *Bifidobacterium* species were found to possess type-1 chain-specific enzymes³⁻⁶. Subsequent analysis using samples collected from mother-infant pairs revealed the importance of HMO-degrading enzymes for bifidobacteria to dominate in the gut microbial communities^{7,8}. Despite their importance, species-level predominance in the infant gut is frequently disproportionate to the HMO assimilation ability of each *Bifidobacterium* species, and we found that priority effects could contribute to this discrepancy^{9,10}. Although type-1 HMO-specific enzymes are highly prevalent in infant gut-associated *Bifidobacterium* species, several other bacterial taxa, including pathobionts, were found to be capable of utilizing HMOs at least in part^{11,12}.

Mucin O-glycans: *Bifidobacterium bifidum* expresses host glycan degrading enzymes as cell surface-anchored proteins, enabling the species to hydrolyze almost all glycosidic linkages present in mucin O-glycans extracellularly. Through the analysis of a unique enzyme, sulfoglycosidase, which releases 6-O-sulfated GlcNAc from mucin O-glycans, we have proposed two distinct strategies that mucinolytic gut microbes have evolved to assimilate mucin O-glycans¹³.

Indole salvage pathway: In collaboration with Danish groups, we identified aromatic lactate dehydrogenase (ALDH) in infant gut-associated bifidobacteria¹⁴. ALDH is involved in the synthesis of indole lactic acid (ILA), an agonist of the aryl hydrocarbon receptor (AhR) that plays pivotal roles in maintaining intestinal barrier function and suppressing excessive inflammatory responses. We have found that bifidobacteria upregulate the tryptophan biosynthetic pathway upon co-cultivation with intestinal epithelial cells, thereby producing higher levels of ILA than under mono-cultivation conditions¹⁵. We have also revealed that several human gut-associated *Bifidobacterium* species are able to salvage exogenous indole, an uremic toxin precursor produced by other gut microbes possessing tryptophan indole-lyase, and convert it into ILA via tryptophan¹⁶. This might represent a novel facet of probiotic functions of bifidobacteria.

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About this speaker:

Takane Katayama received his Ph.D. in Applied Microbiology from Kyoto University in 1999 and was appointed Professor at Kyoto University in 2015. He became interested in gut microbial research following the isolation of two novel glycosidases from bifidobacteria in 2002, and since then, his work has focused on the functional analysis of bifidobacterial genes and enzymes involved in the metabolism of host-derived carbohydrates, including human milk oligosaccharides and mucin O-glycans, as well as aromatic amino acids. His research has contributed to elucidating the molecular mechanisms underlying *Bifidobacterium*-human symbiosis.

Tuesday October 6 - 14:30-15:00

Smart bugs, smarter medicine: Tackling gut cancer

Susan L. Woods, University of Adelaide, Australia

Convincing preclinical studies and our clinical trial data demonstrate that certain bacteria, including the probiotic *E. coli* Nissle 1917 selectively colonise tumours, rather than normal tissue. Understanding these niche specific bacterial behaviours, together with advances in bacterial engineering, have underscored the development of living cell diagnostics and therapeutics. This includes engineered bacteria that respond to gut inflammation, intestinal bleeding, pathogens and hypoxic tumors.

Our team and others have engineered tumour homing bacteria to selectively release combinations of pro-apoptotic, cytotoxic and immune-activating payloads in tumours, with validated reductions in off-target toxicity through tumour directed delivery and anti-tumour and survival benefits in mouse models of cancer. We have also developed engineered probiotics to produce screening and therapeutic molecules, resulting in non-invasive detection of precursor lesions for bowel cancer. We have expanded on these studies to optimise probiotic chassis for the tumour microenvironment, understanding selection pressures for tumour-colonising strains and used alternate bacterial chassis species, to develop living biosensors engineered to detect specific cancer associated DNA sequences.



About this speaker:

Dr. Susan Woods established her Gut Cancer research group at Adelaide University within the South Australian Health and Medical Research Institute in 2019. She trained both in Australia and in San Francisco, USA, with Nobel Laureate J. Michael Bishop, and is the Gastroenterology Society of Australia (GESA) Bushell Research Fellow. Her group is supported by Australian and US (NIH) Government funding, GESA and Hospital Research Foundation funding. Research in Susan's group aims to improve early detection methods for gut cancers, so that they may be identified at a point when most can be cured. This involves the application of synthetic biology approaches to engineer bacteria to act as disease detectors in the gut. The team is also developing engineered bacteria to deliver treatments directly to tumours, to reduce unwanted side-effects of treatment. Her group is assessing the clinical utility of patient tissue samples cultured as organoids, to guide choice of personalised treatments for advanced disease.

Tuesday October 6 - 15:00-15:10

Translating pasteurized *Akkermansia massiliensis* LWHK0003 from preclinical models to clinical application as a next-generation postbiotic candidate for MASLD

Wei-Kai Wu, National Taiwan University, Taiwan

Metabolic dysfunction-associated steatotic liver disease (MASLD) affects approximately a quarter of the global population and can progress to steatohepatitis (MASH), cirrhosis, and hepatocellular carcinoma, yet effective pharmacotherapies remain scarce. Next-generation postbiotics (NGPs), such as pasteurized *Akkermansia muciniphila*, offer a promising alternative. *Akkermansia massiliensis*, the second most prevalent *Akkermansia* species in the human gut, remains largely uncharacterized. We isolated domestic *Akkermansia* spp. from healthy donors; among them, *A. massiliensis* LWHK0003 most strongly stimulated glucagon-like peptide-1 (GLP-1) secretion from enteroendocrine L-cells, and oral gavage raised circulating GLP-1 in germ-free mice. In a GAN diet-induced MASH model, both live and pasteurized LWHK0003 reduced the NAFLD activity score, mainly by attenuating lobular inflammation, alongside lower hepatic TNF- α , IL-1 β , and IL-18; the pasteurized form (pAma) markedly elevated plasma GLP-1. We then conducted a randomized controlled Phase I/II trial of low-, medium-, and high-dose pAma in patients with MASLD. After three months, pooled analysis showed significant reductions in FibroScan controlled attenuation parameter and liver stiffness, indicating improved steatosis and fibrosis, with improved insulin resistance (lower HOMA-IR) in the low- and medium-dose groups versus placebo. No adverse changes in renal function or inflammatory markers were observed, supporting a favorable safety profile. Pasteurized *A. massiliensis* LWHK0003 represents a promising NGP candidate for MASLD, advancing from mechanism to first-in-human evidence.



About this speaker:

Wei-Kai Wu, MD, PhD, is an Assistant Professor at the Graduate Institute of Microbiology, College of Medicine, National Taiwan University, and an attending physician at National Taiwan University Hospital. He is a physician-scientist specializing in diet-microbiome interactions, precision nutrition, and next-generation probiotics. His research focuses on translating gut microbiome discoveries into microbiome-based strategies for the prevention and treatment of noncommunicable diseases, particularly cardiometabolic disorders. His group integrates human microbiome research, culturomics, metabolomics, and gnotobiotic animal models to identify and functionally validate next-generation microbial candidates. His current translational work includes the development of *Akkermansia* strains, including LWHK0003, from preclinical efficacy studies toward clinical application as next-generation postbiotic candidates. He is also a co-founder of Leeuwenhoek Laboratories, a biotechnology company dedicated to translating microbiome science into precision health and microbiome-based therapeutic solutions.

Tuesday October 6 - 15:40-16:10

Novel microbiome-related targets for colorectal cancer

Dirk Haller, Technical University Munich, Germany

The prevalence of colorectal cancer (CRC) is constantly rising, with diet-microbe-host interactions being significant contributors to the pathogenesis of sporadic CRC. The regulation of endoplasmic reticulum (ER) stress responses in the intestinal epithelium is an important mechanism in tumor development and progression, and the activating transcription factor 6 (ATF6) is an emerging oncogene (Li et al, 2026). We recently demonstrated that chronic Atf6 activity changes the epithelial lipid metabolism to shape tumor-promoting bacterial communities in mouse models and colorectal cancer patients (Coleman et al., 2025). In addition, we show that refined, westernized diets profoundly increase tumor susceptibility by increasing the abundance of *Desulfovibrio* spp. and H₂S production, with strain-level functional variation. Metabolomic analysis revealed that diet is the most significant factor driving tumor-relevant metabolite changes, with amino acids and fatty acids as the primary differentiating factors. In conclusion, these studies indicate that diet shapes tumor-promoting microbiome niches in the colon.

Implications: Funding: This project is funded by Collaborative Research Center CRC 1371 (Microbiome Signatures) of the German Research Foundation (DFG; project number 395357507)

References:

Li X., Lebeaupin C., ..., Haller D.*, Kaufman R.J.*, Heikenwälder M.*. Activated ATF6 α is a hepatic tumour driver restricting immunosurveillance. *Nature* Mar;651(8106):796-807. * shared corresponding authorship
Coleman O., ..., and Haller D. (2025) ATF6 activation alters colonic lipid metabolism causing tumor-associated microbial adaptation. *Nature Metabolism* 2025 Sep;7(9):1830-1850.



About this speaker:

Prof. Dirk Haller pioneered the field of nutrition and microbiome research, and over the past two decades, he has developed highly innovative, truly interdisciplinary research. He holds the Chair of Nutrition and Immunology at the Technical University of Munich. In more than 250 publications, he developed novel concepts to elucidate the role of the microbiome in the pathogenesis of chronic inflammation and cancer. The question of how commensal microbes interact with the host at the gut-epithelial interface has become the primary research paradigm. Newly developed gnotobiotic animal models are the backbone of his translational research program. Receiving the Main Award of the German Society of Medical Microbiology, the distinguished Research Award of the United European Gastroenterology Association, and the recognition as a Highly Cited Researcher highlight his global scientific reputation across disciplines.

Tuesday October 6 - 16:10-16:40

Effects of dietary fiber on the intestinal immune system

Yukihiro Furusawa, Toyama Prefectural University, Japan

Dietary fibers are major modulators of gut microbiota composition and function, and their immunological effects are increasingly recognized as an important component of host health. However, dietary fibers are structurally diverse, and their impacts on intestinal immunity cannot be explained simply by total fiber intake. Our studies have focused on how individual dietary fibers regulate intestinal immune homeostasis, particularly through the induction of regulatory T cells (Tregs) and the modulation of inflammatory responses.

Short-chain fatty acids, especially butyrate, produced through microbial fermentation of dietary fibers, promote the differentiation of colonic Tregs, at least in part through inhibition of histone deacetylases. This pathway provides a mechanistic link between microbial metabolism and epigenetic regulation of intestinal immune tolerance. Based on this concept, we have been screening dietary fibers for their ability to induce colonic Treg. Our findings suggest that the immunological activity of dietary fibers differs substantially depending on their physicochemical properties and fermentability, and that only certain fibers efficiently promote Treg accumulation.

In parallel, we have also investigated dietary fibers that influence T helper 17 (Th17) cell responses. Intestinal Th17 cells are often associated with inflammation, but they also contribute to epithelial barrier protection and host defense. Thus, fiber-dependent regulation of Th17 immunity may have context-dependent beneficial effects.

In this presentation, I will summarize our experimental evidence linking dietary fibers to intestinal immune regulation, including the identification of Treg-inducing fibers and the modulation of Th17 responses. A better understanding of the fiber-microbiota-immune axis could contribute to the development of mechanism-based dietary or prebiotic strategies for maintaining gut health and preventing immune-mediated diseases.



About this speaker:

Yukihiro Furusawa, PhD, is a Professor in the Department of Pharmaceutical Engineering at Toyama Prefectural University, Japan. He earned his BS in Pharmaceutical Sciences from the University of Toyama (2006), completed an MS program in Pharmaceutical Sciences (2008), and received his PhD in Medical Science from the University of Toyama (2012). After his doctorate, Dr. Furusawa served as a Postdoctoral Researcher at RIKEN RCAI (Research Center for Allergy and Immunology) and as a Project Assistant Professor at The University of Tokyo, focusing on mucosal immunology. He subsequently joined the Faculty of Pharmacy at Keio University as an Assistant Professor (2014). Since 2015, he has been at Toyama Prefectural University, where he advanced to Associate Professor in 2020 and moved to Pharmaceutical Engineering in 2021.

Tuesday October 6 - 16:40-17:10

Maternal transfer of bifidobacteria and metabolic pathways in allergy development
Susanne Brix, Technical University of Denmark, Denmark

Maternal microbial transfer is a major determinant of infant gut microbiota development and may influence lifelong immune health. Through longitudinal analyses of microbiota, metabolites, and clinical outcomes from birth through childhood, we identified maternally transmitted aromatic lactate-producing bifidobacteria as key mediators linking early-life exposures to immune tolerance. Vaginal delivery, older siblings, and exclusive breastfeeding promoted colonization with these bifidobacteria and enhanced production of aromatic lactic acids. Elevated aromatic lactic acid levels, particularly 4-hydroxyphenyllactate, were associated with reduced allergen-specific IgE responses and lower risk of allergic disease, while simultaneously limiting expansion of opportunistic antimicrobial resistance gene-rich bacteria. These findings reveal a bifidobacteria-driven metabolic pathway through which maternal microbial transmission shapes both microbiota maturation and allergy development during a critical window of immune programming.



About this speaker:

Susanne Brix is Professor of Immune–Microbiota Interactions at the Department of Biotechnology and Biomedicine, Technical University of Denmark (DTU). She earned her MSc in Biotechnology from DTU and completed her PhD in Nutritional Immunology in 2005, during which she was a visiting researcher at the Nestlé Research Centre in Lausanne, Switzerland.

She heads the Section for Medical Biotechnology and has led the Disease Systems Immunology research group since 2014. Her research focuses on understanding host–microbiota interactions and immune regulation in health and disease.

Prof. Brix is Academic Leader of the Technical University Hospital (TUH), a partnership between DTU and 16 hospitals where she is responsible for strengthening clinical–academic integration and advancing cross-disciplinary research infrastructures that link innovation, patient needs, and health technologies.

She is an elected member of the Danish Academy of Natural Sciences; chair of the Research Council for Technology and Production Sciences, The Independent Research Fund Denmark; and serves on several national and international steering committees, including the Million Microbiomes of Humans Project.

Tuesday October 6 - 17:10-17:20

Postbiotics derived from *Ligilactobacillus salivarius* improve mitochondrial homeostasis via TLR2-mediated signaling

Fu Namai, Tohoku University, Japan

Objectives: Postbiotics are defined as “a preparation of inanimate microorganisms and/or their components that confers a health benefit on the host.” Owing to their greater stability and storability compared with live probiotics, postbiotics have attracted increasing attention for their potential applications in food and health-related fields. Mitochondrial function is known to decline with aging, and mitochondrial dysfunction is closely associated with various age-related diseases. In particular, the maintenance of mitochondrial homeostasis in immune cells is important for regulating inflammatory responses and tissue homeostasis. In this study, we aimed to investigate whether postbiotics contribute to the maintenance of mitochondrial homeostasis. To this end, mitochondrial dysfunction was induced in intestinal macrophages, and the effects of postbiotics prepared from several strains of *Ligilactobacillus salivarius* on mitochondrial dysfunction were evaluated.

Methods: An intestinal macrophage cell line, IPIM, was used to assess mitochondrial function. To induce mitochondrial dysfunction and mitochondrial reactive oxygen species (mtROS) production, cells were treated with antimycin A (AMA) at concentrations ranging from 5 to 500 μ M. mtROS production was measured using mtROS580, and the expression of redox-related genes was analyzed by RT-qPCR. Postbiotics were prepared from four strains of *L. salivarius* by culturing each strain followed by heat treatment. IPIM cells were pretreated with each postbiotic preparation for 24 h and subsequently exposed to AMA to induce mtROS production. The ability of each postbiotic preparation to activate innate immune receptor signaling was evaluated using a reporter assay in HEK293 cells. Finally, the effects of the postbiotics on mitochondrial function were examined using a flux analyzer.

Results: Treatment of IPIM cells with 500 μ M AMA induced mtROS accumulation and impaired SOD2 induction, suggesting disruption of mitochondrial homeostasis. Although the postbiotics were prepared from strains belonging to the same bacterial species, only postbiotics derived from specific *L. salivarius* strains significantly suppressed AMA-induced mtROS accumulation and enhanced SOD2 expression. Based on previous findings showing that SOD2 expression can be induced through innate immune receptor signaling, we examined the reactivity of the prepared postbiotics toward Toll-like receptor 2 (TLR2). Interestingly, the postbiotics that suppressed mtROS production showed higher reactivity toward TLR2. Consistently, the inhibitory effect of the postbiotics on mtROS production was abolished by the addition of a TLR2 inhibitor. Furthermore, postbiotic pretreatment enhanced mitochondrial respiratory function. These findings suggest that specific *L. salivarius*-derived postbiotics improve mitochondrial homeostasis by activating TLR2-mediated signaling.

Acknowledgements: AMED, BRAIN



About this speaker:

Fu Namai is an Assistant Professor at Shinshu University and a Project Assistant Professor at Tohoku University, Japan. He earned his Ph.D. from Shinshu University and was a JSPS Research Fellow in 2021. After working as a researcher at Nagasaki University and Shinshu University, he joined Tohoku University in 2023. In 2025, he joined the Faculty of Agriculture at Shinshu University and established his independent laboratory. His research focuses on host-microbe interactions, particularly how lactic acid bacteria and the gut microbiota regulate mitochondrial metabolism, barrier function, and immune homeostasis in intestinal epithelial cells.

ISAPP's interactive expert panel facilitates the sharing of best practice and learnings from real world international research collaborations.

Tuesday October 6 - 11:00-12:30

Expert Panel: International collaboration in biotic research

Cross-border scientific collaborations are of strong benefit to advance the field and can provide scientific, professional, commercial and community benefits. However, many barriers and challenges exist - ranging from compliance matters to cultural differences to logistical hurdles. In this interactive discussion, panelists will share insights from real-world research collaborations, (including multicentre trials, study replication, data sharing and aggregation efforts) and how they've overcome both practical and scientific challenges to execute effective research across geographies.

The expert panel includes the following moderator and speakers:



Maria Marco
University of California, Davis, USA
(moderator, speaker) See bio [here](#).



Karsten Kristiansen
University of Copenhagen, Denmark
(speaker) See bio [here](#).



Sarah Lebeer
University of Antwerp, Belgium
(speaker) See bio [here](#).



Tahmeed Ahmed
icddr,b: International Centre for
Diarrhoeal Disease Research,
Bangladesh (speaker) See bio [here](#).



Jan Knol, Danone Global Research and Innovation, Netherlands (speaker)

Professor Dr. Jan Knol is Senior Science Director of GIM, the Gut, Immunology & Microbiology Platform at Danone Global R&I Centre Utrecht (Netherlands), Paris and Singapore. He is responsible for the gut, immunology and microbiology related research programs focusing on nutrition and health. The platform brings together unique expertises in the area of immunology, microbiology and human (gut) physiology to substantiate nutritional concepts and to develop innovative products for the Danone Specialised Nutrition Division.



Takuya Akiyama, Yakult Central Institute, Japan (speaker)

Takuya Akiyama, PhD, is a Senior Researcher at the Yakult Central Institute with over a decade of experience in international research collaboration in nutrition and health. He holds a PhD in Bioscience and degrees in Agriculture and Life Sciences from the University of Tokyo. In 2012, he joined the Yakult Honsha European Research Center for Microbiology in Belgium, where he spent five years. He has since led and contributed to collaborative research and clinical studies with academic and governmental partners across Europe, the Asia-Pacific region, and the Americas, working with diverse populations and healthcare settings. His research expertise includes clinical study design, biostatistics, and advanced data analytics for nutrition and health research. He is currently involved in planning Yakult's new R&D hub in the Americas and building networks across disciplines, organizations, and sectors to explore new ways of conducting research that creates value for society.



Maria Marco, University of California, Davis, ISAPP President

Dr. Maria Marco, PhD, is a Professor in the Department of Food Science and Technology at the University of California, Davis. She earned her bachelor's degree in microbiology at The Pennsylvania State University and her PhD in microbiology at the University of California, Berkeley. As a postdoc at NIZO food research in The Netherlands, she developed a love for lactic acid bacteria and the importance of these microorganisms in our foods and the digestive tract. Her postdoctoral studies led to the discovery that probiotics are metabolically active in the intestine and responsive to dietary intake. Dr. Marco started her [lactic acid bacteria and gut health laboratory](#) at UC Davis in 2008 and has built an internationally-recognized, NIH, USDA, and NSF funded research program on probiotics, fermented foods, and dietary modulation of the gut microbiome. Dr. Marco also consults with and has received funding from international foundations and companies to investigate how certain microbes in foods or supplements may benefit health. She is active with science communication activities such as the [EATLAC project](#) and is the instructor for two food microbiology courses. Dr. Marco received a UC Davis Graduate Program Advising and Mentoring Award in 2024. She received the American Society for Microbiology Distinguished Lecturer award in 2012 and is currently a fellow in the American Academy of Microbiology. Dr. Marco attended her first ISAPP meeting as a postdoc and participated as an invited expert before joining the ISAPP Board of Directors in 2019.



Sarah Lebeer, University of Antwerp, ISAPP Vice President

Prof. Sarah Lebeer, PhD, is a research professor at the Department of Bioscience Engineering of the University of Antwerp, Belgium. She has studied bioscience engineering, with a specialisation in cell and gene technology/food & health, and obtained her Master at KU Leuven (Belgium). In 2008, she obtained a PhD degree with a topic on the mode of action of gastro-intestinal probiotics in inflammatory bowel diseases and a scholarship in the team of Prof. Jos Vanderleyden (KU Leuven). After a postdoc on the interaction between lactobacilli, viruses, and mucosal immunology, in November 2011, she was offered a tenure-track position at the University of Antwerp. Since then, she is leading the Laboratory for Applied Microbiology and Biotechnology. In 2020, she was awarded an ERC Starting Grant that enables her to gain more in-depth knowledge of the evolutionary history and ecology of [lactobacilli](#). This rationale was also an important driving force to revise the Lactobacillus genus taxonomy with a large international consortium. Within the ERC project, Sarah has also launched the [Isala citizen-science project](#) to gain new insights into the role of vaginal lactobacilli for women's health. Since 2018, Sarah is an academic board member of the International Scientific Association on Probiotics and Prebiotics. Communicating about beneficial microbes and probiotics for experts and laymen is an important inspiration for her daily work.



Daniel Merenstein, Georgetown University, ISAPP Past President

Dr. Daniel Merenstein, MD, is a Professor with tenure of Family Medicine at Georgetown University, where he also directs Family Medicine research. Dr. Merenstein has a secondary appointment in the undergraduate Department of Human Science, in the School of Health. Dr. Merenstein teaches two undergraduate classes, a research capstone and a seminar class on evaluating evidence based medical decisions. He has been funded by PCORI, NIH, USDA, foundations and industry. The primary goal of Dr. Merenstein's research is to provide answers to common clinical questions that lack evidence and improve patient care. Dr. Merenstein is a clinical trialist who has recruited over 2,000 participants for 10 probiotic trials since 2006. He is an expert on probiotics, on antibiotic stewardship in outpatient settings, and also conducts HIV research in a large women's cohort. He sees patients in clinic one day a week. Dan was nominated into the National Academy of Medicine (NAM) in 2024.



Anisha Wijeyesekera, University of Reading, ISAPP Secretary

Dr. Anisha Wijeyesekera, PhD, is an Associate Professor in Human Microbiome and Metabolome Studies, and Director of Postgraduate Research Studies in the Department of Food and Nutritional Sciences at the University of Reading, UK. She obtained her BSc (Hons) in Biomedical Sciences from the University of Durham (UK), MSc in Bioinformatics from the University of Exeter (UK), and PhD in Biochemistry from Imperial College London (UK). Her research interfaces analytical chemistry with microbiology and nutrition and applies 'omics technologies coupled with bio/chemoinformatics data analysis, for functional assessment of the human microbiota and to gain better insight into host-gut microbiota interactions. Dr. Wijeyesekera's research utilises in vitro laboratory model systems for studying the gut microbiota, as well as running human studies to capture information relating to microbial composition and function in vivo. She has wide ranging experience in the application of these analytical approaches in a number of research projects spanning the life course. These include pregnancy studies, infant early life, identifying signatures associated with disease outcome and in response to nutritional intervention in children, young and older adults, mining metabolite patterns associated with diet, microbiome and ageing that contribute to health status in large-scale human epidemiology studies, as well as stratified medicine clinical research projects. The ultimate aim of her research is to identify potential targets for therapeutic modulation through dietary intervention (in particular, prebiotics, probiotics and fermented foods), and capturing the impact of such interventions on gut and overall human health. Dr. Wijeyesekera's research portfolio includes projects funded by research councils, charities/societies, and the food industry. Dr. Wijeyesekera joined the ISAPP Board of Directors in 2021. She is also a member of the Academic Board for the Royal Society of Chemistry's Community of Analytical Measurement Sciences, and the London Metabolomics Network Committee.



Kelly Swanson, University of Illinois, ISAPP Treasurer

Prof. Kelly Swanson, PhD, is Director of the Division of Nutritional Sciences and the Kraft Heinz Company Endowed Professor in Human Nutrition at the University of Illinois Urbana-Champaign. He is also an Excellence in Nutrition Fellow of the American Society for Nutrition (ASN). His laboratory studies the effects of nutritional intervention on health outcomes, identifying how nutrients impact host physiology and gut microbiota, with primary emphasis on gastrointestinal health and obesity in dogs, cats, humans, and rodent models. Much of his lab's work has focused on dietary fibers, prebiotics, probiotics, synbiotics, and postbiotics. Over the past 2 decades, he has established an internationally recognized research program, highlighted by over \$28 million in research support, 185 invited lectures at scientific and professional meetings, 300 peer-reviewed journal articles, and 20 research and teaching awards. He has trained over 55 graduate students and post-doctoral fellows, hosted 18 international visiting scholars, and mentored over 40 undergraduate research projects. In addition to research, Kelly teaches a couple classes each year to undergraduate and graduate students and has been named to the university's 'List of Teachers Ranked as Excellent by Their Students' over 35 times. He also serves on advisory boards for many companies in the human and pet food industries as well as non-profit organizations, including the Institute for the Advancement of Food and Nutrition Sciences (IAFNS).



Karen Scott, Rowett Institute, University of Aberdeen

Prof. Karen Scott, PhD, is a Professor of Research in the Gut Microbiology Group at the Rowett Institute, University of Aberdeen. She leads a research team investigating the (molecular) mechanisms by which key members of the gut microbiota interact with the diet and host, at different life-stages. The fermentation products of gut bacteria contribute to gut health and are differentially expressed on different substrates, including prebiotics. In vitro bacterial growth studies utilising the Rowett's large culture collection of gut anaerobes illustrate niche-specific processes and bacterial interactions, identifying strains with potential for development as probiotic/live biotherapeutic products. Utilisation of pure culture, co-culture, and mixed culture fermentor incubations complemented by investigations into bacteria-host cell interactions, are essential to understand what happens in the complex gut ecosystem. This work encompasses a One Health approach building our understanding of gene flow between bacteria in diverse environments including soils, and animal and human guts, and identifying novel solutions for pathogen control and improving biodiversity. Karen has numerous highly cited publications and has attracted multiple research council and commercially funded grants.



Seppo Salminen, University of Turku

Prof Seppo Salminen, PhD, is a Professor at the Faculty of Medicine and the Nutrition and Food Research Center, University of Turku, Finland. He has been visiting professor at RMIT University, Melbourne, Australia, and BOKU University, Vienna Austria. His main research interests are probiotics, prebiotics, postbiotics and intestinal microbiota modulation as well as functional foods and health and regulatory issues in novel foods and health claims. He has served as an expert member of European Food safety authority (EFSA) in the NDA panel. He has been active in the International Life Sciences Institute Europe, the International Dairy Federation, and the International Scientific Association for Probiotics and Prebiotics (past President and current Board Member). He received his MS at Washington State University (USA) in 1978, MSc from the University of Helsinki in 1979, and PhD from the University of Surrey (United Kingdom) in 1982. He has around 500 journal articles and several textbooks and book chapters and he has received several international awards including the IDF-Institute Pasteur Metchnikoff Price, Swiss Price on Modern Nutrition and the Grand Prix du Yoplait.



Hania Szajewska, The Medical University of Warsaw, Department of Paediatrics

Prof. Hania Szajewska, MD, PhD, is Professor and Chair of the Department of Paediatrics at the Medical University of Warsaw. Among her various functions, she served as the Editor-in-Chief of the Journal of Pediatric Gastroenterology and Nutrition; a member of the Council, and then as the General Secretary of the European Society for Paediatric Gastroenterology, Hepatology and Nutrition (ESPGHAN); the Secretary of the ESPGHAN Committee on Nutrition. Most recently, she joined the Board of Directors of the International Scientific Association for Probiotics and Prebiotics (ISAPP). Prof. Szajewska has broad interests in paediatric nutrition but her research focuses on the effects of early nutritional interventions on later outcome (especially food allergy); and the gut microbiota modifications such as with various biotics (probiotics, prebiotics, synbiotics, postbiotics). She is or has been actively involved in several European Union-funded research projects. She is an enthusiastic advocate for the practice of evidence-based medicine. She has co-authored over 400 PubMed-indexed publications, which have been cited more than 24,000 times, with an H-index of 83.



Daniel Tancredi, University of California, Davis

Prof. Daniel J. Tancredi, PhD, is Professor in Residence of Pediatrics in the University of California, Davis School of Medicine. He has over 25 years of experience and over 300 peer-reviewed publications as a statistician collaborating on a variety of health-related research. A frequent collaborator on probiotic and prebiotic research, he has attended all but one ISAPP annual meeting since 2009 as an invited expert. In 2020, he joined the ISAPP Board of Directors. Colin Hill and Daniel co-host the ISAPP Podcast Series "Science, Microbes, and Health". On research teams, he develops and helps implement effective study designs and statistical analysis plans, especially in settings with clusters of longitudinal or otherwise correlated measurements, including cluster-randomized trials, surveys that use complex probability sampling techniques, and epidemiological research. He teaches statistics and critical appraisal of evidence to resident physicians; graduate students in biostatistics, epidemiology, and nursing; and professional scientists. Dan grew up in the American Midwest, in Kansas City, Missouri, and holds a bachelor's degree in behavioral science from the University of Chicago and masters and doctoral degrees in mathematics from the University of Illinois at Chicago. He lives in the small Northern California city of Davis, with his wife Laurel Beckett (UC Davis Distinguished Professor Emerita), their Samoyed dogs Simka and Milka, and near their two grandkids.



Gabriel Vinderola, Dairy Products Institute, Faculty of Chemical Engineering (UNL),

Dr. Gabriel Vinderola, PhD, graduated from the Faculty of Chemical Engineering at the National University of Litoral (Santa Fe, Argentina) in 1997 and obtained his Ph.D. in Chemistry in 2002 at the same University. He is presently Principal Researcher at the Dairy Products Institute (CONICET-UNL) and Adjoint Professor at the Biotechnology and Food Technology Department of the Faculty of Chemical Engineering (National University of Litoral). He participated in the development of the first commercial cheese carrying probiotic bacteria from Latin America, released in the market in 1999. In 2011, he was awarded the prize in Food Technology for young scientists, by the National Academy of Natural, Physic and Exact Sciences from Argentina. His interests are technological and microbiological aspects of lactic acid bacteria, bifidobacteria, fermented foods, and probiotics. He has joined several research groups in Brazil, Canada, Spain, Italy, France, Germany, and Finland. He has co-edited the fifth (2019) and the sixth (2024) editions of the book *Lactic Acid Bacteria: Microbiological and Functional Aspects*. He is engaged in communication of science to the general audience.



Kristin Verbeke, KU Leuven

Prof. Kristin Verbeke, PharmD, PhD, is Professor at the Department of Chronic Diseases and Metabolism of KU Leuven, Belgium, where she conducts research at the interface of gastroenterology, nutrition, and the gut microbiome. Trained as a pharmacist, she has extensive expertise in the application of stable isotope technologies and mass spectrometry to investigate gastrointestinal function, host-microbiome interactions, and microbial metabolism in humans. Her research focuses on understanding how diet, prebiotics, probiotics, and other nutritional interventions influence microbial metabolism and contribute to gastrointestinal and metabolic health.

Her group has developed and applied innovative analytical approaches to study intestinal transit, permeability, nutrient metabolism, microbial fermentation, and metabolite production in both healthy individuals and patient populations. Through interdisciplinary collaborations, her research has provided important insights into the role of microbial metabolism in gastrointestinal and metabolic physiology and gut-brain communication, as well as the role of nutrition in modulating microbiome function. She has published more than 250 peer-reviewed papers and has successfully led numerous nationally and internationally funded research projects. Prof. Verbeke currently serves as Vice-Chair of the Leuven Food Science and Nutrition Centre and Co-Chair of the ILSI Europe Prebiotics Task Force.



Geoffrey Preidis, Baylor College of Medicine and Texas Children's Hospital

Dr. Geoffrey A. Preidis, M.D., Ph.D., Associate Professor of Pediatrics at Baylor College of Medicine in Houston, Texas, is a board-certified Pediatric Gastroenterologist with basic and translational research expertise in nutrition-sensitive mechanisms that regulate intestinal and liver physiology, growth, and development. Based at Texas Children's Hospital, which contains the largest neonatal intensive care unit in the United States, Dr. Preidis explores how the intestinal microbiome develops in extreme prematurity and how probiotics might be used to reduce the risk of necrotizing enterocolitis, sepsis, feeding intolerance, growth failure, and death in this vulnerable population. His biomedical research career began during undergraduate studies at Harvard University. He developed an interest in Neonatal Gastroenterology while in the Medical Scientist Training Program at Baylor College of Medicine. He completed internship and residency training in Pediatrics, as well as fellowship training in Pediatric Gastroenterology, Hepatology & Nutrition, at Baylor College of Medicine.



Hannah Holscher, University of Illinois Urbana-Champaign

Dr. Hannah Holscher is an Associate Professor of Nutrition at the University of Illinois Urbana-Champaign, where she also serves as Associate Director of the Personalized Nutrition Initiative and holds affiliations with the Division of Nutritional Sciences, the Carl R. Woese Institute for Genomic Biology, and the National Center for Supercomputing Applications. Her research integrates clinical nutrition, microbiome science, and computational biology to explore how diet influences human health. She has authored more than 90 peer-reviewed publications and leads multidisciplinary research projects supported by government agencies, foundations, commodity boards, and private industry. Dr. Holscher has held leadership roles within the American Society for Nutrition and serves on the editorial boards of The Journal of Nutrition and Nutrition Research.



Jens Walter, University College Cork, APC Microbiome Ireland

Jens Walter serves as the Professor of Ecology, Food, and the Microbiome at University College Cork and the APC Microbiome Ireland. His expertise lies at the interface of evolutionary ecology of the gut microbiome and human nutrition. His research focuses on the evolutionary and ecological processes that have shaped host-microbiome symbiosis and the translation of basic microbiome science into therapeutic and nutritional strategies. Dr. Walter and his collaborators have pioneered the application of ecological theory to elucidate ecological and nutritional factors that shape gut microbiomes and have achieved targeted modulations of microbiomes via dietary strategies and live microbes. Prof. Walter has published >140 peer-reviewed publications (google scholar H-index 69, >23,000 citations) and is a 'highly cited researcher' according to the Web of Science group.



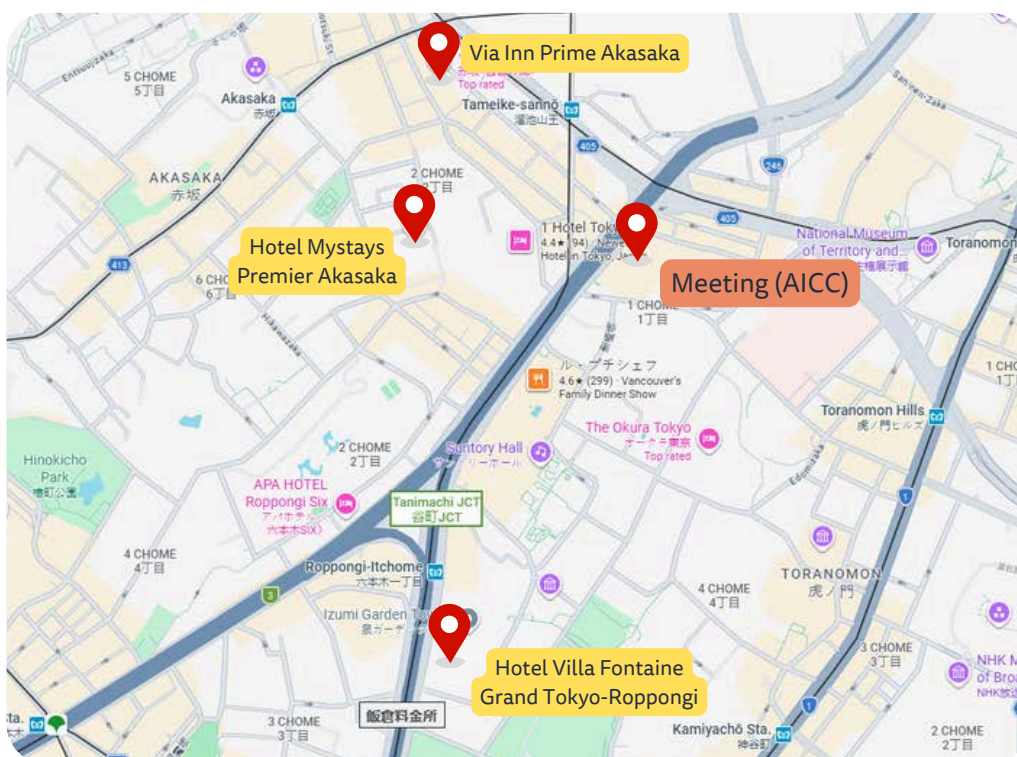
Maria Carmen Collado, Institute of Agrochemistry and Food Technology (IATA-CSIC), Spain

M.Carmen Collado is a Full Research Professor at IATA-CSIC, Spain where she leads the MAINbiotic Lab. Her research focuses on diet-microbiota interactions across the life course, with particular emphasis on maternal-infant and women's health. She investigates how human milk and dietary bioactives, and biotics shape the human microbiome and contribute to health. Her team combines cohorts and clinical studies, multi-omics, mechanistic studies, and translational research to investigate microbiome-targeted interventions, including biotics and dietary strategies, for health promotion and disease prevention. Beyond research, she is actively engaged in science communication, citizen science, and outreach initiatives to bring microbiome science closer to families and society. Prof. Collado has authored more than 300 peer-reviewed publications and has coordinated numerous national and international research projects. She serves on international expert panels, scientific committees, and consensus initiatives, advancing the translation of microbiome science into evidence-based nutrition and health applications.



Marla Cunningham, ISAPP Executive Director

Marla Cunningham is the Executive Director of ISAPP. Marla has a background as an industry scientist within the ISAPP community and brings over 20 years of experience in the probiotic, prebiotic, and natural products industry, working across innovation, clinical research, product development, regulatory compliance, and education. She trained as a healthcare practitioner in complementary medicine and nutrition and has written and presented extensively for clinical audiences on the implementation of biotic-related science. Marla has broad scientific literacy across the fields of biotic substances, microbiome, gut health, nutrition, and human physiology, and brings a passion for harnessing science to deliver beneficial changes in health for our community and our planet.



The meeting venue (AICC) is noted in orange and hotels in yellow.

Use the links below for walking directions on Google Maps from your hotel to the meeting venue.

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ISAPP is pleased to offer a dynamic poster session, showcasing international research on probiotics, prebiotics, synbiotics, postbiotics and fermented foods as well as the microbiome. Posters were chosen from abstracts submitted by members of the ISAPP Students and Fellows Association, symposium participants and industry members. Read the complete list of poster abstracts [here](#), and be sure to visit them during the evening poster session and networking event.

Theme 1: Fermentation and novel food ingredients

Poster 1 IAC

Enhancing probiotic bacteria growth via balanced RNA and DNA precursors in an innovative fermentation nutrient

Introduction: Probiotic bacteria have gained popularity over the past two decades due to growing scientific data pointing to their positive benefits on human health. *Lactobacillus reuteri*, *Lactobacillus rhamnosus*, *Bifidobacterium*, *Bacillus coagulans*, *Lactobacillus casei*, the *Lactobacillus acidophilus* group, *Escherichia coli*, and various *enterococci*, are some of the more prevalent probiotic bacteria [1]. Probiotic lactobacilli are nutritionally fastidious organisms, meaning their growth activity and viability are commonly influenced by growth factors such as medium formulations, pH, temperature, and others [2-4]. Understanding their nutritional needs is essential for maximizing production and viability of probiotic biomass. Lallemand Bio-Ingredients in collaboration with AS TFTAk have investigated the specific needs of probiotic lactobacilli especially for nucleotides, nucleosides, and nucleobases.

Methods: Three model bacterial strains, *Lactobacillus rhamnosus*, *Pediococcus acidilactici*, and *Lactobacillus acidophilus*, were cultured in 2L bioreactors with an industrial complete media supplemented with three distinct nitrogen sources: yeast peptone rich in nucleotides, yeast peptone containing nucleosides and nucleobases, and standard yeast extract. The consumption of nucleotides, nucleosides, and nucleobases was quantified using ultra-performance liquid chromatography-tandem mass spectrometry (UPLC-MS/MS). Design of experiments and microbioreactors Biolector XT system were utilized to identify the optimal synergy between a yeast peptone rich in nucleotides and a yeast peptone containing nucleosides and nucleobases.

Results & Conclusions: Most bacteria are able to produce nucleotides de novo, while others, including some lactic acid bacteria require addition of purines and/or pyrimidines to growth medium [5]. These auxotrophic bacteria utilize salvage pathways for conversion of the required nucleobases or nucleosides to nucleotides. Based on this observation, Lallemand bio-ingredients and AS TFTAk studied the consumption of DNA and RNA precursors, highlighting the absence or variable capability to metabolize nucleotides for *P. acidilactici*, *L. rhamnosus*, *L. acidophilus*. A balanced combination of RNA and DNA precursors significantly enhanced the growth of *Lactobacillus rhamnosus*. The yeast extract containing nucleosides and nucleobases reduced the lag phase duration by 50%, and boosted the total viable cell count by a factor of 6. This finding highlights a promising opportunity to synergistically combine various yeast-based nutrients, through Design of Experiments, to optimize the effects of RNA and DNA precursors.

Lallemand Bio-Ingredient has developed a rational design for next-generation fermentation nutrients, enabling probiotic producers to create optimal culture media. This approach should be extended to more challenging species to produce as *Bifidobacterium* and microbiome bacteria.

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Poster 2 SFA

Functional and Bioaccessible Profiling of Probiotic and Postbiotic Metabolites Derived from Fermented Tuna By-Products

Valorisation of marine by-products into functional "biotics" supports a circular economy while providing a sustainable source of bioactive compounds. While fermentation improves the nutritional profile of tuna waste, the impact of subsequent processing, specifically the transition from live probiotics to heat-inactivated postbiotics, on the biochemical composition and bioaccessibility remains underexplored. This study incorporates a comprehensive functional characterisation to evaluate the changes in digestates of raw, fermented, and postbiotic tuna products. Raw, probiotic (fermented), and postbiotic (heat-treated) tuna samples were subjected to the INFOGEST 2.0 static in vitro digestion protocol. Soluble digestates (<3 kDa) were isolated to represent the bioaccessible fraction. Protein quality was quantified using the Digestible Indispensable Amino Acid Score (DIAAS), and a detailed screen of organic acids, amino acids, and fatty acids was performed. Antioxidant activity and cytocompatibility in C2C12 myoblasts in Solid-Phase Extracted eluates were additionally assessed. Analysis identified lactic acid as the dominant organic acid across all fermented stages. Notably, the postbiotic variant exhibited the highest relative protein quality, with DIAAS scores ranging from 44.40% to 74.31%, suggesting that thermal processing may optimise the release or stability of essential amino acids during digestion. Cellular assays revealed a fraction-dependent safety profile; while the less hydrophobic SPE fractions (E1) were well-tolerated by C2C12 cells at concentrations ≤0.05% (w/v), the more hydrophobic E2 fractions exhibited dose-dependent cytotoxicity. These results highlight how processing modulates the concentration of soluble, low-molecular-weight metabolites. This research demonstrates that the transition from a probiotic to a postbiotic state significantly alters the bioaccessible profile of fermented tuna waste. By integrating standardised digestion with safety screening, this study provides a foundation for the development of stable, marine-derived postbiotics with nutritional and functional relevance.

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Poster 3 Symposium

Enzymatic transformation of whey permeate into a prebiotic ingredient: FTIR-multivariate approach

Galactooligosaccharides (GOS) are produced via transgalactosylation reactions catalyzed by β -galactosidases using lactose as substrate. The initial lactose concentration and enzyme source determine yield and product composition. Higher lactose levels increase the likelihood that lactose, rather than water, acts as the acceptor molecule, thus favoring transgalactosylation over hydrolysis. The enzyme source further influences catalytic efficiency and its preference for transgalactosylation relative to hydrolysis. This work addresses key challenges in the cost-effective and sustainable production of GOS through the synthesis from whey permeate, a lactose-rich dairy by-product. To this end, the performance of β -galactosidases from *Bacillus circulans*, *Kluyveromyces lactis*, and *Aspergillus oryzae* were evaluated, revealing distinct catalytic behaviors in terms of lactose conversion, GOS yield, and enzyme productivity. A straightforward purification strategy combining an additional enzymatic treatment with ethanol precipitation was further implemented to increase the content of GOS, thus to increase the prebiotic effect of the product. The synthesis and purification processes were followed through HPLC and FTIR spectroscopy. Principal Component Analysis (PCA) allowed the grouping of spectra according to sample composition, explaining more than 91% of the total variance in the first two components (PC1, PC2). Then, a Partial Least Square (PLS) regression model, calibrated with HPLC chromatograms and validated with independent spectra, enabled accurate prediction of sample composition ($R^2 > 0.95$, Error $< 10\%$). The integration of optimized enzymatic synthesis, purification, and FTIR-multivariate monitoring provided a scalable approach for producing GOS-rich syrups, simultaneously valorizing whey permeate into high-value food and nutraceutical applications.

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Poster 4 Symposium

In vitro fermentation of brewer's spent yeast β -glucans: microbiota modulation and short-chain fatty acids production

Brewer's spent yeast, a major by-product of the beer industry, is a rich source of cell wall β -glucans with potential prebiotic properties. However, their fermentation dynamics and effects on human gut microbiota remain poorly understood. This study evaluated the *in vitro* fermentability of purified yeast β -glucans and their impact on intestinal microbiota metabolism and composition. β -glucans were extracted from brewer's spent yeasts using an alkaline-acidic method, structurally characterized, and subjected to *in vitro* batch fermentation using a pooled fecal inoculum from four healthy adults (35–55 years old, omnivorous diet, no

antibiotic use within the previous 6 months). Fermentations were conducted in a carbohydrate-free basal medium supplemented with 2 g/L yeast β -glucans, while inulin (prebiotic control), cellobiose, glucose, and a no-added-sugar basal medium served as controls. After 48 h of anaerobic incubation at 37°C, short-chain fatty acids (SCFA) were quantified by gas chromatography, and microbial composition was analyzed by 16S rRNA gene amplicon sequencing (NGS). β -glucans fermentation induced rapid acidification (from an initial pH of 7.5 to 5.7 after 24 h) and significant SCFA production, confirming its fermentability by the gut microbiota. Although cellobiose and inulin produced the highest butyrate concentrations (11.18 ± 1.02 and 11.32 ± 0.11 mM, respectively), β -glucans generated significantly more butyrate (9.07 ± 2.31 mM) than the no-added-sugar control (4.82 ± 0.11 mM), together with substantial acetate and propionate production. NGS analysis revealed a reduction in alpha diversity across all substrates relative to the initial inoculum, reflecting the selective pressure of the *in vitro* environment. β -glucans fermentation increased the relative abundance of phylum Firmicutes (56% to 73%) while decreasing Bacteroidota and promoted the enrichment of families *Selenomonadaceae* (mainly *Megamonas*), *Veillonellaceae* (*Megasphaera*), and *Coriobacteriaceae* (*Collinsella*). In contrast, genera associated with simple carbohydrate utilization, such as *Escherichia-Shigella*, were less enriched than in glucose control fermentation. Overall, yeast β -glucans were effectively fermented by human gut microbiota, producing beneficial SCFAs, particularly butyrate, while selectively modulating microbial composition. These findings position brewer's spent yeast-derived β -glucans as promising functional ingredients and support the valorization of brewery by-products as sustainable resources for human nutrition.

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Poster 5 Symposium

Influence of Native Milk Cream on Probiotic Adhesion to Intestinal Epithelial Cells

Background: The milk fat globule membrane (MFGM) is a biologically active membrane that surrounds native milk fat globules and contains phospholipids, glycoproteins, and membrane-associated proteins that may influence probiotic functionality. However, the relationship between probiotic attachment to native milk cream and subsequent adhesion to intestinal epithelial cells remains poorly understood. This study investigated the interactions between probiotics and native milk cream and evaluated how different milk cream fractions influence probiotic adhesion to intestinal epithelial cells.

Methods: Four probiotic strains (*Lactocaseibacillus rhamnosus* LGG, *Lactocaseibacillus rhamnosus* HN001, *Bifidobacterium infantis* (NLS), and *Bifidobacterium breve*) were initially incubated with native milk cream,

and flow cytometry was used to quantify the percentage of bacterial cells associated with milk fat globules. In the cell culture assay, probiotics were cocultured with different dairy fractions, including native milk cream, butter oil, buttermilk, combined treatment of butteroil and buttermilk, beta serum, milk phospholipids, and Dulbecco's Modified Eagle Medium (DMEM) to investigate their adhesion to Caco-2 intestinal epithelial cells. Ongoing work is investigating the contribution of bacterial surface layer proteins in mediating probiotic attachment to native milk cream.

Results: Flow cytometry demonstrated strain-dependent differences in probiotic adhesion to native milk cream. *L. rhamnosus* LGG and *B. breve* exhibited greater adhesion to native milk cream than *L. rhamnosus* HN001 and *B. infantis*. In Caco-2 adhesion assays, probiotics that had been exposed to native milk cream showed greater adhesion to intestinal epithelial cells than those exposed to butter oil, buttermilk, beta serum, milk phospholipids, or DMEM control. The strains exhibiting higher adhesion to native milk cream, particularly LGG and *B. breve*, also demonstrated greater adhesion to Caco-2 cells, suggesting that interactions with intact native milk fat globules may enhance probiotic adhesion to the intestinal epithelium.

Conclusions: Native milk cream enhanced probiotic adhesion to intestinal epithelial cells compared with individual milk cream fractions. These findings suggest that the intact milk fat globule structure plays an important role in promoting probiotic host interactions. Ongoing investigations into bacterial surface layer proteins aim to elucidate the mechanisms underlying probiotic attachment to native milk cream and support the development of dairy-based probiotic delivery systems with enhanced functionality.

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Poster 6 Symposium

Isolation of High Putrescine-Producing Bacteria from Cheese and Their Potential Application to Polyamine-Enriched Fermented Ingredients

Polyamines, represented by putrescine, spermidine, and spermine, are aliphatic compounds with two or more amino groups and are involved in gene transcription, translation, and cell differentiation. Their intake is expected to exert health-promoting effects, such as lifespan extension and anti-inflammatory effects (Science 359:eaan2788, 2018). Polyamines are supplied through host-cell biosynthesis, gut bacterial production, and dietary intake (Med. Sci. 9:8. (2021)). Host polyamine biosynthesis decreases with age, whereas polyamine-rich foods increase blood polyamine levels (Annu. Rev. Nutr. 40:135–159. (2020)).

In a human randomized controlled trial conducted to show memory improvement by oral polyamine intake, memory performance in the group receiving 1.2 mg/day polyamines slightly increased compared with placebo, although the statistical significance was weak (Wirth M. et al. Cortex 109:181–188. (2018)). This dose accounted for only 3.3–7.5% of usual daily polyamine intake in the United States, Turkey, Japan, and Sweden. In contrast, the polyamine concentration in high-polyamine diets showing lifespan extension in animal studies was fourfold higher than that in low-polyamine diets (Exp. Gerontol. 44:727–732. (2009)). One reason for the low dose used in human trials is the limited availability of food materials containing high

concentrations of polyamines. To solve this problem, we searched foods for microorganisms that highly produce polyamines.

Fermented foods contain high concentrations of polyamines. Therefore, bacteria were isolated from seven types of cheese, and putrescine concentrations in culture supernatants were quantified by the PuO-POD-4AA-TOPS method (Anal. Biochem. 593:113607. (2020)) and HPLC. *Levilactobacillus brevis* FB215 and *Hafnia paralvei* FB315 were isolated as bacteria producing high concentrations of putrescine from Blue Stilton cheese and Langres cheese, respectively. When cultured in MRS medium, *L. brevis* FB215 and *H. paralvei* FB315 accumulated 176 μ M and 221 μ M putrescine in the culture supernatants, respectively. Next, the strains were cultured in MRS medium supplemented with 1 mM ornithine or agmatine, precursors of putrescine, and putrescine concentrations in the culture supernatants were measured. Compared with no precursor addition, putrescine concentrations with ornithine or agmatine were 4.5-fold and 4.4-fold in *L. brevis* FB215, and 7.3-fold and 2.0-fold in *H. paralvei* FB315, respectively.

When *L. brevis* FB215 was cultured in Sakekasu extract containing ornithine and agmatine, approximately 1 mM putrescine accumulated in the culture supernatant. Therefore, novel foods with high putrescine content could be developed by fermenting food substrates containing polyamine precursors in combination with high polyamine-producing bacteria derived from fermented foods.

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Poster 7 SFA

Plant cell cultures: novel sustainable food ingredients for promoting gut health

Diet is a central determinant of human health, acting through complex interactions between nutrients, the gut microbiota, and host physiological barriers. Understanding these interactions is essential for developing sustainable dietary strategies that support gut health. Plant cell culture (PCC) technology represents a promising approach for producing nutritionally balanced and environmentally sustainable food ingredients. This study evaluates the safety, nutritional quality, and microbiota-mediated effects of plant cell cultures (PCCs) derived from scurvy grass (*Cochlearia danica*) and rowan (*Sorbus aucuparia*), and their potential impact on gut barrier integrity. Oral safety, toxicity, and protein quality of PCCs were evaluated in in vivo models, with protein quality assessed using the Digestible Indispensable Amino Acid Score (DIAAS). Macro- and micronutrient composition was also characterised. PCC samples were processed using the INFOGEST digestion method followed by an in vitro colonic fermentation model, after which microbiota composition and metabolic profiles were analysed using 16S rRNA gene sequencing and UHPLC-based non-targeted metabolomics. Both PCCs displayed high protein and fibre content and contained relevant micronutrients, including vitamins C and D. Acute and subacute toxicity assessments revealed no signs of toxicity. DIAAS confirmed protein quality comparable to conventional plant-based proteins. After 24 hours of colonic fermentation in both PCCs no changes in the overall alpha-diversity were observed but distinct metabolomic shifts were observed. These metabolic alterations were primarily driven by metabolites such as cytidine, sebatic acid, and cymatherelactone. The most enriched pathways included lysine degradation, arginine and proline metabolism, and the urea cycle. To investigate the impact of PCCs on host physiological barriers, an dynamic in vitro gut barrier model is being developed. Together, these findings

support PCCs as safe, nutritionally balanced, and sustainable dietary ingredients that modulate microbial metabolism, providing a rationale for future studies on diet-microbiota-host barriers interaction.

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Poster 8 SFA

Soy-derived indigestible peptides serve as substrates for putrescine production by gut microbes

Polyamines produced by human gut microbes accumulate in the intestinal lumen at millimolar levels and are absorbed by the host, where they exert health-promoting effects. Arginine, a conditionally essential amino acid, is a precursor of polyamines. During gastrointestinal digestion, dietary proteins are degraded into peptides and free amino acids; however, a fraction of peptides remains indigestible and escapes host digestion and absorption. These dietary indigestible peptides reach the large intestine, where gut microbes reside. We hypothesized that dietary indigestible peptides serve as a source of arginine for gut microbiota and thereby support microbial polyamine production.

10 human fecal samples were cultured in media supplemented with soy-derived indigestible peptides, which are insoluble peptides generated by pepsin and pancreatin digestion of soy protein. Supplementation with soy-derived indigestible peptides increased putrescine levels in 30% of cultures and agmatine levels in 70%.

To identify bacteria responsible for agmatine-to-putrescine conversion, 51 predominant gut bacterial species from European and Japanese microbiota were cultured with or without 1 mM agmatine. Although genomic analysis predicted this capacity in 15 species, only *Enterococcus faecalis* produced putrescine from agmatine in vitro. When *E. faecalis* was inoculated together with fecal microbiota into the same medium, putrescine production increased in the presence of soy-derived indigestible peptides, whereas *E. faecalis* alone did not increase putrescine levels. *Levilactobacillus brevis* FB215, isolated in our laboratory from blue cheese and capable of converting agmatine to putrescine (Ami Y, Kurihara S, et al. Int J Mol Sci. 2023;24(11):9668.), also increased putrescine concentrations when co-inoculated with fecal microbiota.

Fecal samples showing increased putrescine production were plated on minimal media containing soy-derived indigestible peptides as the sole nitrogen source, yielding strains of *Bifidobacterium breve*, *Escherichia coli*, and *Bifidobacterium ruminantium*. Among these, *E. coli* OK26 accumulated up to 124 µM putrescine in vitro. Colonization of germ-free mice with *E. coli* OK26 resulted in fecal putrescine concentrations of 600 µM in vivo.

These results demonstrate that certain gut bacteria produce polyamines from soy-derived indigestible peptides as substrates in the large intestine.

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Poster 9 SFA**Dietary microorganisms and fermented foods in Swiss adults: consumption, metabolomic profiling, and associations with cardiometabolic health**

Background: Dietary microorganisms and fermented foods (FFs) are hypothesised to contribute to human health through modulation of gut microbiota composition and function, although evidence of their impact on health at the level of the overall diet is still emerging. Untargeted metabolomics approaches may help uncover diet-microbiota-host interactions.

Objectives: We aimed to classify dietary intake in Swiss adults by live microorganism levels and FF descriptors, examine associations with cardiometabolic health, and explore metabolomic profiles linked to these dietary patterns.

Methods: We analysed cross-sectional data from 2086 adults (18-75 years) in the Swiss National Nutrition Survey menuCH 2014-2015 and 440 adults (≥ 18 years) from the Swiss Kidney Stone Cohort (SKSC). Foods were categorised by live microorganism level (Low, $<10^4$ CFU/g; Medium, 10^4 - 10^7 CFU/g; High, $>10^7$ CFU/g) and FF descriptors, including core microbiota. A novel microbial richness index was calculated from unique microbial species in FFs. Adjusted linear regression models assessed associations of Medium or High (MedHi) food intake, FF intake, and microbial richness with cardiometabolic parameters including weight, waist circumference, BMI, blood pressure, glycated haemoglobin, glucose, triglycerides, and total, HDL, and LDL cholesterol in SKSC. Untargeted LC-MS metabolomics were performed on SKSC 24-h urine samples with biological pathway analysis (mummichog).

Results: Mean MedHi food intake accounted for 7-8% of total intake (228-269 g/day), primarily from fruits, vegetables, and fermented dairy; mean FF intake represented 20-21% (667-717 g/day), mainly from coffee, bread, alcoholic beverages, and fermented dairy. We identified 186 unique microorganisms across six taxonomic levels in FFs. Higher MedHi food intake was associated with lower diastolic blood pressure ($\beta = -0.020$, CI: -0.037, -0.003; $p = 0.02$). Increased microbial richness was associated with lower systolic blood pressure ($\beta = -0.0021$, CI: -0.0039, -0.0003; $p = 0.02$) and glycated haemoglobin ($\beta = -1.22 \times 10^{-5}$, CI: -2.14×10^{-5} , -3.11×10^{-6} ; $p = 0.009$). No other significant associations were observed. Metabolomics analyses linked MedHi food intake to tryptophan metabolism, FF intake to galactose metabolism, and microbial richness to tryptophan, tyrosine, and galactose pathways.

Conclusions: Our comprehensive dietary classification enabled the first characterisation of dietary microbial richness among Swiss adults, suggesting potential cardiometabolic health benefits. Metabolomics analyses provided the first insight into metabolomic signatures of dietary microorganisms and fermented foods."

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Poster 74 SFA**High-Throughput fermentation and metabolome fingerprinting for ML-guided design of sustainable dairy-plant hybrid foods**

Background: Developing sustainable fermented foods with desirable flavour and health-promoting properties requires rapid identification of effective microbial strain-substrate combinations. High-throughput fermentation combined with metabolome fingerprinting can capture strain-specific biotransformation patterns across diverse food matrices. Integrating these datasets with machine learning could enable prediction of fermentations that reduce off-flavours while generating desirable aroma and potentially beneficial metabolites.

Objectives: The study was aimed to establish a high-throughput pipeline for evaluating the fermentation and aroma-transforming potential of microorganisms in oat-dairy and fava-dairy hybrid substrates.

Methods: Five dairy, plant-based and hybrid formulations were fermented using lactic acid bacterial strains. Fermentation performance was assessed through acidification kinetics and endpoint pH. Strain, substrate and interaction effects were statistically evaluated, while profiles were compared using principal component analysis and hierarchical clustering. The fermented products were further characterized by untargeted metabolomic fingerprinting.

Results: Acidification was significantly influenced by strain, substrate and their interaction, demonstrating strain-matrix dependency. Plant-based and hybrid formulations generally exhibited greater acidification than the dairy control, although responses varied among strains. Metabolome fingerprinting revealed distinct strain and inoculation dependent volatilome profiles. Fermentation commonly decreased aldehydes while increasing corresponding alcohols, indicating carbonyl-reduction activity relevant to off-flavour mitigation.

Conclusion: Combining high-throughput fermentation with metabolomic fingerprinting provides a systematic framework for identifying effective microbial cultures and their flavour-transforming activities. The resulting multidimensional dataset establishes a foundation for machine-learning models that can guide culture selection and accelerate the development of sustainable fermented foods with improved flavour, functionality and potential health benefits.

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Theme 2: Genomic technologies and engineering

Poster 10 IAC**A genomic framework for probiotic strain characterization, functional potential, and safety assessment**

Whole-genome sequencing has become an important tool for probiotic strain characterization because it enables complex genomic data to inform biologically meaningful decisions. We developed a genomic analysis pipeline to systematically evaluate properties of potential benefit to hosts, strain uniqueness, and safety-related features in candidate probiotic strains.

The pipeline uses comparative genomics to characterize and evaluate taxonomic identity, relatedness to closely associated strains, and strain-specific genomic content. Functional annotation is then performed using multiple complementary databases to improve confidence in trait prediction. Genomes are screened for genes and pathways associated with carbohydrate metabolism, stress tolerance, vitamin biosynthesis, short-chain fatty acid production, antimicrobial potential, and other traits relevant to probiotic performance. These features provide insight into strain adaptation to the gut, interaction with the host, and the overall functional benefits.

In parallel, the pipeline evaluates safety-relevant genomic features, including antimicrobial resistance genes, virulence factors, mobile genetic elements, and genes associated with biogenic amine production. We use comparative genomics to distinguish core, accessory, and strain-specific features, allowing each candidate strain to be evaluated in the context of closely related organisms.

This pipeline, combined with AI tools, provides a practical framework for integrating functional annotation, comparative genomics, and safety screening into candidate probiotic strain evaluation. By organizing genomic information into interpretable categories, the approach supports strain selection, risk assessment, and research and development decisions. Overall, this work demonstrates how genomics can be used not only to describe probiotics but also to predict and prioritize candidate strains with desirable functional potential and appropriate safety profiles.

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Poster 11 Symposium

Analysis of acid tolerance mechanisms in a novel *Lactiplantibacillus plantarum* strain JN18 via optimized genetic engineering platforms

Lactic acid bacteria (LAB) are utilized as probiotics and industrial hosts for polylactic acid production. However, LAB encounter severe environmental stresses during both industrial processing and gastrointestinal transit. Among these stresses, acid stress is a primary bottleneck for LAB survival; therefore, elucidating the acid tolerance mechanisms is crucial to expanding their applications. In this study, we characterized a novel acid-tolerant LAB strain, *Lactiplantibacillus plantarum* JN18, isolated from a traditional fermented rice bran bed (nukadoko), which exhibits high viability for over three hours at pH 2.0. To reveal the genetic determinants of acid tolerance in JN18, we performed functional analysis by developing transformation and genome editing platforms.

Through UV mutagenesis, we generated two mutants exhibiting reduced acid tolerance. Comparative genomic analysis between the wild-type and mutants identified ten acid-tolerance candidate genes (*cg1-cg10*). To verify the function of these candidates, we first optimized an electroporation-based transformation method using a pHSP02-based *gfp* expression vector. The obtained transformants exhibited stable plasmid maintenance and GFP fluorescence. Subsequently, we constructed expression vectors by replacing the *gfp* with each candidate gene to obtain overexpression strains. The acid tolerance assay

revealed that the individual overexpression of *cg4* and *cg5* significantly elevated cell viability at pH 2.0, demonstrating their direct roles in conferring acid tolerance.

For loss-of-function analysis of candidate genes, we optimized base editing strategy using Target-AID, which couples dCas9 with cytidine deaminase PmCDA1 to induce targeted cytosine-to-thymine (C-to-T) conversions. Developing this optimized Target-AID platform, we efficiently introduced nonsense or missense mutations into multiple loci, including the candidate genes. Phenotypic analysis revealed that the *cg10* mutant exhibited a drastic reduction in viability at pH 2.0. Taken together, we successfully established transformation and base-editing platform for *L. plantarum* JN18 and identified *cg4*, *cg5*, and *cg10* as critical genetic determinants modulating acid tolerance. These tools and findings provide promising approaches for engineering robust LAB strain for various applications.

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Poster 12 SFA

CRISPR/Cas engineering of the human gut cornerstone crAssphage and their Bacteroidota hosts

CrAss-like phage are ubiquitous in the human gut virome and sometimes can account for up to 99% of viral metagenomic reads. These viruses, despite making up such a large proportion of the virome, remain poorly characterised, with only a handful of cultured representatives. crAssphage dysbiosis is correlated with a number of disease states in humans, with functional characterisation being an important step in discerning between co-occurrence or causal relationships. Dynamic relationships between the phage-host pairs and potentially unique replication lifestyles are permissive of sustained persistence of both phage and host without classical integration mechanisms.

We herein develop modular CRISPR-Cas systems applicable across a broad range of Bacteroidota members for the purpose of genetic editing in both phage and host. This system consists of two plasmids: (i) an integrative pNBU2-derived vector carrying the cas assembly under the control of either IPTG or aTC inducible promoters, and (ii) a replicating plasmid carrying the constitutively expressed guide RNA and Homology Directed Repair template sequences. This segregation of CRISPR components confers potential benefits allowing iterative editing in the same host population, and reducing the burden on the plasmid replication machinery theoretically improving cloning efficiencies. Replicating plasmids are also designed using Golden Gate cloning restriction sites allowing single-tube, single-step introduction of new CRISPR targets and edits. This system's design is suitable for a broad range of applications and ease of use in the hopes of allowing other research groups to apply the same system in their own work.

Preliminary results demonstrate >2-log decreases in viable CFU when targeting non-essential host chromosomal genes in the combined presence of gRNA and cas inducer, demonstrating highly efficient CRISPR-Cas activity. Future work will focus on characterising critical aspects of the phage-host dynamic and phage life cycle including host receptor recognition, cargo protein functional characterisation and early gene transcription. Understanding of abundant phage and bacterial gut species provides an avenue for modulating complex gut communities through inter-species ecological interactions. The tools developed here permit highly specific microbiome modulation, with edited phage providing a strain level high specificity platform for phage therapy, VLP peptide tagging, and genetic material delivery.

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Poster 13 Symposium

Development of genetically modified lactic acid bacteria producing monosialotetrahexosylganglioside-binding protein

Objective: Monosialotetrahexosylganglioside (GM1) is a prominent cell-membrane ganglioside that mediates critical immunomodulatory signaling pathways. Proteins that specifically bind GM1, termed monosialotetrahexosylganglioside-binding proteins (MGBPs), can modulate the function of antigen-presenting cells and reduce the production of pro-inflammatory cytokines. These properties suggest that MGBPs have significant potential as anti-inflammatory biomolecules. Lactic acid bacteria (LAB) are established hosts for recombinant protein production and delivery of bioactive molecules. The expression of MGBP in LAB therefore offers a potential platform for both production and therapeutic delivery of this functional protein. This study aimed to construct genetically modified LAB (gmLAB) expressing MGBP and to evaluate the anti-inflammatory activity of the produced MGBP.

Methods: The gene encoding an MGBP with high specificity for GM1 was inserted into a nisin-inducible gene expression vector and introduced into *Lactococcus lactis* strain NZ9000 via electroporation, generating the gmLAB strain NZ-MGBP. MGBP expression was induced by nisin and confirmed by western blot analysis. MGBP production was determined across multiple culture times to identify the optimal induction period. The ganglioside-binding ability of the produced MGBP was evaluated by a GM1-ELISA assay. The anti-inflammatory activity of MGBP was evaluated in RAW 264.7 cells pretreated with MGBP for 24 h and subsequently stimulated with 10 ng/mL lipopolysaccharide (LPS). At 5 h after LPS stimulation, culture supernatants were collected and tumor necrosis factor- α (TNF- α) levels were quantified by ELISA.

Results: Western blot analysis confirmed that NZ-MGBP expressed MGBP in a nisin-dependent manner, demonstrating successful gmLAB strain construction. Intracellular MGBP production peaked at 6 h after induction, but MGBP secretion into the culture supernatant did not vary significantly across induction times. In the GM1-ELISA, when GM1 was immobilized on a solid phase and NZ-MGBP cell lysate was added, the absorbance at 450 nm was significantly higher than in the control group. Furthermore, NZ-MGBP-derived MGBP significantly suppressed LPS-induced TNF- α production in RAW264.7 cells. These results demonstrate that MGBP produced by NZ-MGBP retains both ganglioside-binding and anti-inflammatory activities. Future studies will evaluate the efficacy of NZ-MGBP in intestinal inflammation models and explore its potential as a mucosal delivery vehicle for anti-inflammatory therapy.

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Poster 14 IAC**Genome-based Quality Assessment for Probiotic Manufacturing and Strain Purity Verification**

Maintaining strain purity from seed stock to finished product is essential throughout probiotic manufacturing, where unexpected microorganisms may affect product quality, safety, consistency, and strain integrity. Traditional industrial approaches to monitor quality, including culture-based testing and targeted microbial assays, remain foundational for probiotic production. However, these methods may have limitations when distinguishing between closely related organisms, resolving strain-level identity, detecting non-viable microbial cells, or identifying contaminants that are difficult to culture or classify. These limitations are especially relevant when investigating strain misclassification or unexpected microbial signals that may create challenges for product characterization and quality assurance.

We developed a whole-genome sequencing-based quality assessment pipeline to complement existing commercial testing standards and to provide a more comprehensive evaluation of probiotic strain purity. The approach is designed for production-related samples, including seed, fermentation, and finished product materials, and integrates genome-level data to support interpretation of strain identity, purity, and potential contamination. Rather than one single analytical output, the pipeline combines multiple lines of genomic evidence, including alignment to the expected strain genome, taxonomic profiling to identify organisms represented in the sequencing data, investigation of unmapped reads from foreign DNA or potential contaminants, and assessment of unexpected genetic differences. Together, these complementary analyses provide a more rigorous basis for identifying potential genus, species, or strain-level contamination.

The pipeline is also designed to distinguish biologically meaningful contamination signals from technical artifacts. Low complexity reads, sequencing noise, and database misclassification are considered during interpretation to avoid misinterpretation of low-level signals or misclassification of closely related taxa. Detection thresholds are estimated based on total sequencing depth and the number of supporting reads, providing a quantitative framework for determining whether a potential contaminant signal is statistically and biologically meaningful.

This work highlights the potential application of whole-genome sequencing as an added layer of scientific rigor in probiotic manufacturing QA/QC. By complementing established industrial standards with genomic analysis, this pipeline supports improved strain verification, transparent and reproducible contamination assessment, root-cause analysis, product consistency and confidence in probiotic strain identity across production stages.

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Poster 15 Symposium**Genomic characterization of *Lacticaseibacillus rhamnosus* GG and its derivative strains to define the strain boundaries of LGG**

Lacticaseibacillus rhamnosus GG (LGG) is one of the most widely commercialized and extensively studied probiotic strains. Despite its widespread global use, the genomic definition and boundaries that distinguish LGG from closely related *L. rhamnosus* strains remain unclear. In this study, we analyzed genomes of 606 strains of *L. rhamnosus* obtained from a public database to define strain LGG and to establish a genomic framework for identification and protection of commercial probiotic strains.

A core-gene phylogenetic analysis revealed several distinct clades within *L. rhamnosus*. All strains annotated as LGG (n=5) clustered together in a single clade with very short branch lengths, which contained a total of 53 strains. We therefore designated this group as the LGG-phylo-clade. Next, pairwise average nucleotide identity (ANI) values were determined among the 606 strains and used for hierarchical clustering. All strains annotated as LGG again formed a single clade together with several closely related strains, which was designated as the LGG-ANI-clade. Notably, strains comprising the LGG-ANI-clade completely matched those of the LGG-phylo-clade. Strains within this clade showed markedly high ANI values (>99.8%), which were much higher than the values obtained between LGG-clade strains and non-LGG strains. Based on these findings, we concluded that the strains within the LGG-clade represent LGG and its derivatives. The origins of strains in this clade included human gastrointestinal tracts, breast milk, human blood, dairy products, supplements, and unknown sources.

Comparative genomic analysis between the LGG-clade strains and non-LGG strains identified several LGG-specific genes, some of which were located in the galactose-rich exopolysaccharide biosynthesis cluster. These genomic features provide potential markers for distinguishing LGG and its derivatives from other *L. rhamnosus* strains and will help to establish a robust genomic basis for strain identification and commercial strain protection.

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Poster 16 Symposium**ProFiT-MAGs: An automated pipeline for designing strain-specific qPCR primers to detect and quantify target microbes in complex communities**

Tracking a defined microorganism within a complex community requires an assay that detects and quantifies that target very specifically. Quantitative PCR is the most accessible tool for this purpose, but designing primers specific to the target in a complex microbial community can be challenging due to primer specificity and cross-reactivity with other microbial members. To address this, we developed ProFiT-MAGs, an automated bioinformatics pipeline that designs and validates specific primers directly from metagenomic data.

The pipeline takes whole-genome sequences or metagenome-assembled genomes (MAGs) as the target, either a single genome (strain-level) or a defined group (group-level), together with an exclusion set of off-target genomes. Our in-house tool ProFiT (PRObe FInding Tool) extracts short sequences that are unique to the target and Primer3 designs qPCR-grade primer pairs within these regions. Each candidate primer pair is then screened by virtual PCR and retained only if it amplifies the target exactly once and none of the off-target genomes.

We evaluated our pipeline both for tracking an isolate in synthetic microbial communities and for tracking uncultured MAGs from a natural gut microbial community. In both cases, the designed primers showed no cross-reactivity against a large microbial isolate collection. Generating primers that detect and quantify uncultured organisms in their source samples pave the way to their targeted isolation. Furthermore, our method could be used to track candidate probiotics, monitor microbial engraftment, or quantify other microorganisms of interest in complex microbial communities.

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Poster 17 Symposium

In Silico Characterization of Immunomodulatory Potential in *Bifidobacterium animalis* subsp. *lactis* BR2-5 from Indonesian Infant Feces

Background: *Bifidobacterium* sp. is a key commensal species with established roles in host-microbiome immune modulation. *Bifidobacterium animalis* subsp. *lactis* is a widely commercialized species, but strain-level variation within the species can influence immunomodulatory efficacy. While phenotypic screening remains the gold standard for probiotic validation, putative insight into immunomodulatory capacity offers a scalable approach for prioritizing candidate strains. We performed in silico characterization of *B. animalis* subsp. *lactis* BR2-5 whole genome to predict its capacity for host immune modulation and metabolic contributions to gut homeostasis.

Methods: *B. animalis* subsp. *lactis* BR2-5 was isolated from the feces of a healthy Indonesian infant (Dosan et al., Biodiversitas, 2024, 25:337-343) and subjected to whole-genome sequencing using Oxford Nanopore Technology. In silico characterization utilized a modular pipeline including: (1) genome annotation using Bakta (v1.12) for structural and functional characterization, (2) genome-scale metabolic model (GEM) reconstruction using CarveMe (v1.6.5) with model quality assessment via Memote (v0.17.0), (3) carbohydrate utilization prediction using GlycoBif (v0.1.1), a machine-learning pipeline trained on human-derived *Bifidobacterium* subsystems, and (4) systematic gene mining for immunomodulatory markers, including exopolysaccharide (EPS) biosynthesis, CpG motif (GTCGTT), and surface adhesins.

Results: *B. animalis* subsp. *lactis* BR2-5 harbors a complete EPS biosynthetic cluster (*epsF*, *epsH*, *wzy*, *cpsA*) for host immune modulation, alongside peptidoglycan remodeling enzymes (*dacA*, *ddl*) that reduce pro-inflammatory signaling. Oleate hydratase (*ole*) enables conjugated linoleic acid (CLA) production with established anti-inflammatory effects. Colonization is mediated by sortase-dependent fimbriae and moonlighting adhesins (*eno*, *tuf*, *gap*, *pgi*) for competitive exclusion. Moreover, the absence of sialidase confirms the strain does not degrade host mucin (non-mucinolytic). GEM result confirmed the strain's capacity to produce short-chain fatty acids (SCFAs) and essential B-vitamins (folate and riboflavin), key metabolites for inflammation regulation and Treg differentiation. GlycoBif predicted utilization of

immunologically relevant prebiotics (scFOS and IMO). In addition, genomic characterization identified 1,283 (683 forward and 600 reverse) copies of the human immunostimulatory CpG motif for TLR9 activation.

Discussion: These findings position *B. animalis* subsp. *lactis* BR2-5 as a promising candidate for host immune modulation. The genomic identification of EPS biosynthetic clusters, CpG motif, and specialized adhesins underscores the basis for mucosal and epithelial interaction. Furthermore, BR2-5's capacity for SCFAs and vitamin biosynthesis suggests a broader role in driving systemic immune homeostasis. Leveraging the resolution of long-read sequencing, this work provides a holistic functional reconstruction of BR2-5, supporting its further development as a commensal and personalized microbiome-based immune intervention.

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Theme 3: Gut-brain axis

Poster 18 SFA

Beneficial modulation of the microbiota-gut-brain axis in epilepsy using a GABA-producing microbial approach

Introduction: γ -Aminobutyric acid (GABA) is a key inhibitory neurotransmitter that also functions as a conserved microbial signal, facilitating communication between gut bacteria and host systems. Emerging evidence suggests that GABA-producing probiotics, or psychobiotics, can modulate the gut-brain axis and influence neurophysiological processes, including seizure susceptibility. This study aimed to identify and characterize potent GABA-producing gut commensals, evaluate their neuroprotective potential in epilepsy, and elucidate their mechanisms of action through functional genomics, metabolite profiling, and gut-brain axis assessments. By linking microbial GABA production to host neurophysiology, this work provides a rationale for developing targeted psychobiotic interventions to mitigate neuroinflammation and seizure pathology.

Methods: Forty-five bacterial strains (20 *Lactobacillus* and 25 *Bifidobacterium*) were screened qualitatively and quantitatively for GABA production ability. Whole-genome sequencing of potential strain (Illumina NovaSeq 6000) was assembled in CLC Genomics Workbench 21 and annotated via RAST, followed by intraspecific comparative genomics. Comprehensive safety was evaluated using *in silico*, *in vitro*, and *in vivo* approaches, including acute, subacute, and sub-chronic oral administration following OECD guidelines. Chronic epilepsy was induced with PTZ (35 mg/kg, *i.p.*), and LAB6 administered alone at different doses or co-supplemented with 10% modified kodo millet bran (mKMB), was evaluated as a gut-brain modulatory intervention. Outcomes included neurotransmitters, cytokines, corticosterone, BDNF, SCFAs, gut microbiota (16S rRNA), colon histology, Golgi-Cox staining, and qRT-PCR to investigate underlying mechanistic pathways.

Results: Screening of 45 indigenous strains identified LAB6 (*Levilactobacillus brevis*) and LAB19 (*Limosilactobacillus fermentum*) as potent GABA producers, with LAB6 showing the strongest anti-inflammatory and anti-epileptic activity. *In silico* analysis confirmed LAB6 harbored a complete

glutamate decarboxylase system (gadA, gadB, gadC) and probiotic traits, including acid/bile tolerance and adhesion proteins. Safety assessments demonstrated LAB6 was non-pathogenic with no adverse hematological or histological effects. In chronic PTZ-induced kindling, medium-dose LAB6 restored gut barrier integrity, behavioral deficits, and peripheral GABA. Colonization studies showed rapid GI adhesion and distal colon/cecum translocation. Co-supplementation with 10% modified kodo millet bran further reduced seizures, preserved neuronal spines, restored hippocampal GABA, attenuated inflammation, and mitigated dysbiosis by enriching *Turicibacter* and *Lactobacillus*.

Discussion: Our study demonstrates that *Levilactobacillus brevis* LAB6 is a potent GABA-producing psychobiotic with robust anti-inflammatory and neuroprotective effects. LAB6, especially when combined with modified kodo millet bran, modulates the gut-brain axis by restoring hippocampal GABA, reducing neuroinflammation, and improving gut microbiota composition. These findings highlight its strain-specific potential as a therapeutic intervention for epilepsy and related gut-brain axis aberrations.

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Poster 19 IAC

Elucidation of the Mechanism of Action of the Psychobiotic Strain *Lactiplantibacillus plantarum* HEAL9.

Short Introduction to the Study: Previous research on the probiotic strain *L. plantarum* HEAL9 has demonstrated improved clinical outcomes in healthy individuals in terms of cognition, memory function, overall wellbeing and sleep. To describe the molecular mechanisms underlying these clinical findings, an early-stage Alzheimer's disease (AD) mouse model was used.

Methods: An early-stage AD mouse model consisting of senescence-accelerated mouse prone 8 (SAMP8) mice and control SAM-Resistant 1 (SAMR1) mice with four arms; SAMP8 vs SAMR1 +/- HEAL9 (1B CFU/mouse/day) was used. Treatment with probiotic or placebo was done for two months in two separate phases.

Phase 1: Cognitive impairment (assessed using Morris Water Maze (MWM) Test), colonic motility, astrogliosis, microgliosis, brain and colonic amyloid- β 1-42 levels, and inflammasome components activation were assessed.

Phase 2: Levels of butyrate, butyrate transporter expression and tryptophan metabolism were measured from ileum, colon and brain tissue, as well as in stool and plasma samples.

Results:

Phase 1: Two months administration of HEAL9 to SAMP8 mice significantly attenuated cognitive impairment as seen by increased number of target entries in MWM test, and counteracted colonic dysmotility. In addition, HEAL9 decreased astrogliosis and microgliosis, as well as A β 1-42 accumulation and inflammasome signaling activation in the colon and brain.

Phase 2: Two months intake of HEAL9 led to significantly elevated levels of fecal butyrate, compared to placebo SAMP8 mice. No differences in butyrate levels were seen in the SAMR1 mice. Expression of butyrate transporters significantly increased in the distal colon of HEAL9 SAMP8 mice compared to placebo. In terms of tryptophan metabolism, intake of HEAL9 led to decreased expression of IDO-1 (Indoleamine 2,3-dioxygenase), responsible for the conversion of tryptophan into kynurenine, as well as increased

expression of TPH-1 (Tryptophan hydroxylase 1), responsible for the conversion of tryptophan into serotonin, in the distal colonic tissue of SAMP8 mice.

Conclusions HEAL9 intake alleviated cognitive decline and normalized colonic motility which may be due in part to its ability to increase butyrate production and transporters important for butyrate absorption. Moreover, the favored conversion of tryptophan to serotonin rather than neurotoxic kynurenine may also explain the findings of decreased plaque and inflammation as well as the clinically documented improvements in cognitive functions, mood, and sleep quality seen with HEAL9 intake. This study provides valuable insights into the psychobiotic properties of HEAL9 as evidenced in several clinical trials.

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2. Unpublished study / Manuscript in preparation (2026)

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Poster 20 Symposium

GABA-producing *Levilactobacillus brevis* strains modulate neuroactive metabolite production in an *in vitro* human faecal gut microbiota model

Gamma-aminobutyric acid (GABA) is a neuroactive microbial metabolite involved in gut-brain axis signalling. One promising direction in next-generation probiotic development is to modulate the gut microbiota (GM) to enhance the production of bioactive metabolites, including GABA. Therefore, live psychobiotic bacteria with probiotic potential and confirmed GABA-producing capacity may represent an alternative to direct GABA intake by supporting local metabolite formation in the gut. This study aimed to evaluate whether two selected *Levilactobacillus brevis* strains, KKP 3500 and KKP 3516, can enhance GABA production in the presence of GM and modulate microbial composition and metabolite production, with particular focus on short-chain fatty acids (SCFA), in an *in vitro* human faecal GM fermentation model.

The standardised pooled faecal inoculum from 6 healthy human donors was used to study strain-GM metabolic interactions. GM was inoculated into deoxygenated Schaedler medium (pH 6.0) supplemented with 0.3% monosodium glutamate (MSG) as a GABA precursor. Fermentations were conducted anaerobically at 37°C for 72 h in three variants: GM, GM with *L. brevis* KKP 3500, and GM with *L. brevis* KKP 3516. After 0, 24, 48 and 72 h of faecal fermentation, bacterial growth was assessed by plate counting and OD₆₀₀ measurements, pH and GABA concentration were determined, and samples were collected for GM taxonomic profiling and metabolomic analysis.

Both *L. brevis* strains maintained high viability in the presence of GM, indicating adaptation to the GM environment. GABA was markedly enhanced by both strains compared with the GM control. In the control, GABA increased from 51.0 ± 7.5 mg/L at baseline to 733.2 ± 31.7 mg/L after 48 h. *L. brevis* KKP 3500 reached 1433.3 ± 46.7 mg/L after 24 h and 1421.8 ± 61.6 mg/L after 48 h, whereas *L. brevis* KKP 3516 achieved the highest GABA concentration after 48 h. A slight decrease in GABA concentration after 72 h was observed in all variants. Fermentation was accompanied by alkalization from approximately pH 6.1 to 6.9-7.0, consistent with glutamate decarboxylation-associated metabolism.

These findings demonstrate enhanced GABA production by *L. brevis* KKP 3500 and *L. brevis* KKP 3516 in a GM model, supporting their potential as psychobiotic strains for targeted modulation of neuroactive microbial metabolites. Ongoing taxonomic and metabolomic analyses will further elucidate their impact on GM composition and metabolic activity (SCFAs), with GABA biosynthesis remaining the primary functional outcome.

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Poster 21 IAC

Modulation of the Gut-Brain Axis by Prebiotic Inulin-Type Fructans

Introduction: Evidence from preclinical studies suggests that prebiotics, such as chicory-derived inulin-type fructans, influence mental health and behavior via the gut-brain axis. Recently, randomized controlled trials in humans have begun to support this concept, linking improvements in mood and perceived stress to selective modulation of the gut microbiota. This presentation synthesizes mechanistic and translational evidence from human intervention studies, including two randomized controlled trials, and highlights key insights and remaining challenges in understanding gut-brain interactions.

Methods: A narrative synthesis based on two randomized, placebo-controlled trials investigating the effects of inulin-type fructans on stress-related outcomes in different populations was conducted. The SYN-ReduSTRESS study evaluated healthy men exposed to an acute stress paradigm, while the EFFICAD trial included adults with mild-to-moderate anxiety. Outcomes included psychological measures (e.g., anxiety, mood), physiological stress markers (e.g., salivary cortisol, cortisol awakening response), and gut microbiota composition. Findings are interpreted in the context of the broader literature on prebiotics and the gut-brain axis.

Results: Prebiotic supplementation reduced stress-related markers and improved mood in both trials. In the SYN-ReduSTRESS study, oligofructose-enriched inulin reduced subjective anxiety and salivary cortisol during the Trier Social Stress Test (TSST) paradigm, particularly in participants with high baseline cortisol levels. The EFFICAD study demonstrated significant improvements in mood scores (BDI-II, STAI, PANAS) and cortisol awakening response in response to oligofructose supplementation. Microbiota analysis showed increases in *Bifidobacterium* and *Faecalibacterium prausnitzii*, which positively correlated with mood improvements.

Discussion: Human intervention studies provide converging evidence that inulin-type fructans can modulate stress responsiveness and mood via the gut-brain axis. Due to variability in study populations and outcome measures, direct comparability across trials is limited. However, these randomized controlled trials build a strong foundation for further research in this field. To advance the field, future studies should align endpoints with hypothesized mechanisms, incorporate functional biomarkers, and adopt stratified designs to capture context-dependent effects. Addressing these points will help elucidate underlying mechanisms and may add to the role of inulin-type fructans as nutritional approach to support stress-related conditions.

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Poster 22 SFA**The (neuro)epigenetic potential of butyrate in emotional learning and memory**

Background: The microbiota–gut–brain axis has emerged as a key pathway through which diet and gut bacteria influence brain function and behavior. Animal studies and initial human evidence suggest that microbial metabolites, particularly short-chain fatty acids (SCFAs), modulate stress- and anxiety-related processes. Butyrate, an SCFA produced by bacterial fermentation of dietary fiber and an endogenous histone deacetylase inhibitor (HDACi), enhances fear extinction and memory persistence in rodents. However, translational evidence in humans remains scarce.

Methods: We investigated the role of butyrate in threat–safety learning and fear extinction using a Pavlovian fear conditioning paradigm across two complementary studies. First, associative data were analyzed from 217 healthy male participants to examine relationships between endogenous SCFA levels and threat–safety discrimination, indexed by skin conductance responses (SCRs). Second, causal effects were tested in a six-arm, randomized, triple-blind, placebo-controlled trial (n = 180; healthy males and females). Sodium butyrate (NaBu) or placebo was administered either before or after extinction learning, and extinction learning duration was manipulated (short vs. long). Fear learning, extinction, and one-week extinction memory retrieval were assessed using physiological (SCRs) and subjective expectancy ratings.

Results: Higher endogenous butyrate levels, but not acetate or propionate, predicted superior discrimination between threat and safety cues in the associative study. In the randomized trial, NaBu administration significantly enhanced long-term extinction memory retrieval following long extinction learning duration, as reflected in reduced threat expectancy ratings at one-week follow-up. These effects were observed when NaBu was administered either before or after extinction training. No significant effects emerged under short extinction learning duration, and NaBu did not significantly alter differential SCRs during extinction or return-of-fear tests.

Discussion: These findings provide the first human evidence that butyrate facilitates emotional learning and memory, and particularly enhances the consolidation and retention of strongly encoded extinction learning. The results highlight epigenetic mechanisms as a potential pathway linking gut microbiota–derived metabolites to fear regulation. This work underscores the translational relevance of diet and the microbiota–gut–brain axis for modulating fear-related processes and supports the potential use of butyrate as an adjunct to exposure-based treatments for anxiety and stress-related disorders.

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Poster 23 SFA**The Role of *Saccharomyces boulardii* CNCM I-745 Gut-Brain-Microbiome Axis in Social Memory**

The gut brain axis constitutes a bidirectional neuroimmune endocrine communication network linking the gastrointestinal tract and the central nervous system, critically influencing social behavior, emotional regulation, and cognition. Emerging evidence suggests that gut dysbiosis alters social memory through inflammatory and neuropeptidergic pathways; however, the underlying molecular mechanisms remain incompletely understood.

This study investigated the role of microbiota-mediated signaling in modulating social cognition in male Swiss albino mice. Fifty-six-week-old mice (20–25 g) were randomly assigned to five groups: control, antibiotic cocktail (Ampicillin, Metronidazole, Neomycin, and Vancomycin), antibiotic plus probiotic (Econorm, 90 mg/kg), probiotic only, and social isolation. Behavioural paradigms, including the Three Chamber Social Interaction Test, Tube Dominance Test, Elevated Plus Maze, Marble Burying Test, and Open Field Test, were employed to assess social memory, dominance behaviour, anxiety-like responses, and locomotor activity. Fecal samples were analyzed using RT-qPCR for microbial profiling. Peripheral inflammatory markers (IL-6, IL-1 β , TNF- α) and neuropeptides (oxytocin, vasopressin) were quantified biochemically. Oxytocin receptor (OXTR) expression was evaluated, and histopathological analyses examined hypothalamic architecture (including neuronal density and the structural integrity of the paraventricular and supraoptic nuclei), as well as intestinal permeability and mucosal integrity.

Antibiotic-induced dysbiosis significantly impaired social recognition memory and elevated pro-inflammatory cytokines, accompanied by alterations in oxytocin signaling. Probiotic administration mitigated these behavioral and molecular abnormalities, suggesting restoration of microbiota-dependent neuroimmune homeostasis.

These findings demonstrate that gut microbiota critically regulate social memory through inflammatory modulation and oxytocinergic signaling pathways. The study provides mechanistic insight into dysbiosis-driven alterations in social cognition and highlights microbiota-targeted interventions as potential therapeutic strategies.

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Poster 24 SFA**Uncovering GABA-producing Psychobiotics Candidates in Individuals Undergoing Recovery from Methamphetamine Use Disorder**

Background: The rising global prevalence of methamphetamine addiction imposes significant burdens on healthcare and social welfare systems. Methamphetamine's strong addictive properties lead to escalating use, resulting in severe short- and long-term physical and mental health damage. Currently, there is no definitive treatment beyond abstinence. An increasing body of research indicates that probiotics may influence addiction-related pathways, yet definitive psychobiotic candidates with reliable efficacy remain lacking. Despite this gap, psychobiotics hold considerable potential as a supportive strategy to alleviate mental health effects and enhance the recovery process.

Objective: This study aims to identify potential psychobiotics candidates derived from individuals undergoing methamphetamine recovery and elucidate their potential to support mental health improvement and recovery outcomes.

Methods: Stool samples were collected from individuals diagnosed with methamphetamine use disorder at a rehabilitation centre and a healthy control. Microbial DNA was analysed using high throughput 16S rRNA sequencing targeting the V3 region. Bioinformatic and statistical analyses were performed to compare gut microbiota composition between groups. Functional prediction was conducted to identify microbial biomarkers and infer metabolic pathways associated with GABA production.

Results: A total of 32 male individuals recovering from methamphetamine use disorder (mean age 27 ± 4.22 years; range 18–35) and 31 healthy male controls (mean age 19 ± 7.73 years; range 14–36) with no prior history of illicit substance use were recruited. Significant differences in gut microbial diversity were observed between individuals in recovery and healthy controls, particularly in α -diversity (Chao1 index and Observed ASVs, $p < 0.05$) and β -diversity (Bray–Curtis, $p = 0.001$). LEfSe analysis indicated that Clostridiaceae was more prevalent among individuals in recovery, whereas *Bifidobacterium adolescentis* was predominant in healthy controls. Functional prediction using PICRUST2 highlighted pathways related to GABAergic metabolism in individuals undergoing withdrawal. In addition, MaAsLin2 analysis identified potential microbial biomarkers associated with recovery profiles, including GABA-producing *Lactococcus* sp. and *Weissella* sp.

Conclusion: This study highlights the potential of next-generation psychobiotics, particularly GABA-producing genera, as novel adjunctive therapies to support recovery from methamphetamine addiction. Targeting gut microbiota may offer a promising strategy to enhance mental health and facilitate the recovery process.

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Theme 4: Immunity & inflammation

Poster 25 Symposium

Effect of CO₂ on the physiological state of *Akkermansia muciniphila*

The human gut is home to over 100 trillion bacteria, where various environmental factors, such as nutrients and gases, exist and influence them. In addition, fermentation gases in gut, including CO₂, fluctuate depending on the host's diet. However, it remains poorly understood how CO₂, as an environmental factor, affects the physiological states of these bacteria, whereas CO₂ is often utilized for gut anaerobes. *Akkermansia muciniphila* (*A. muciniphila*) is widely studied as one of the beneficial human gut microbes. But the effects of its growth conditions on the physiological state of *A. muciniphila*, and how these resulting physiological changes affect the host, are not fully understood. In this study, we focused on CO₂ as an environmental factor and aimed to clarify how CO₂ affects the physiological state of *A. muciniphila* and how these resulting changes affect host cells.

In the culture experiment, the results showed that the presence of CO₂ (CO₂ (+)) stimulated the growth of *A. muciniphila* and that condition-dependent morphological changes were observed. Metabolomic analysis using stable ¹³C (¹³CO₂) flux analysis demonstrated that CO₂ was incorporated into various metabolites in the

TCA cycle and other pathways. Furthermore, the ligand activities of *A. muciniphila* for Toll-like receptors (TLRs) also changed depending on culture conditions.

These results suggested that *A. muciniphila* utilizes CO₂ to produce various metabolites, and the changes in its physiological state may affect the host immune response.

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Poster 26 IAC

A postbiotic derived from *Saccharomyces cerevisiae* rapidly modulates inflammatory cytokines and immune surveillance markers in healthy adults: a randomized double-blind placebo-controlled cross-over trial

Introduction: Postbiotic dietary ingredients have exhibited bioactive properties that are capable of modulating immune pathways. However, the short-term dynamics of systemic immune responses following acute intake remain incompletely characterized.

Methods: In a randomized, double-blind, placebo-controlled crossover study, 30 healthy adults received a single oral dose of 500 mg of the yeast-derived EpiCor® postbiotic or placebo on separate clinic visits with a one-week washout period. Blood samples were collected at baseline, 1-, 2-, and 3-hours post-consumption. Circulating immune cell populations were assessed by flow cytometry, including evaluation of the activation-associated markers CD25 and CD69, and serum samples were analyzed for a broad panel of cytokines, chemokines, and growth factors over the 3-hour time course.

Results: Consumption of EpiCor postbiotic was associated with early, time-dependent changes in selected immune-related biomarkers compared to placebo, including selective comparative decreases in pro-inflammatory cytokine levels and transient redistribution of CD25⁺ immune cell subsets, consistent with altered immune surveillance dynamics. Within the first hour, a subset of cytokines, including interferon-gamma (IFN-γ) and interleukin-1 receptor agonist (IL-1RA), showed significantly reduced levels ($p \leq 0.05$) compared to changes after consuming placebo, accompanied by a transient decrease in CD25⁺ immune cells in circulation. At 2 hours, IL-4, IL-7, and IL-8 were significantly reduced ($p \leq 0.05$) compared to placebo alongside trends toward decreased IL-1β, IL-5, tumor necrosis factor-alpha (TNF-α), and, at 1 hour, a trend toward reduced granulocyte colony-stimulating factor (G-CSF) (all $p \leq 0.1$). These effects occurred within 1–3 hours of ingestion and reflected targeted modulation of specific inflammatory markers rather than uniform downregulation of immune activity.

Discussion: Collectively, these findings suggest that consuming EpiCor postbiotic was associated with a rapid anti-inflammatory response and altered immune cell trafficking consistent with augmented immune surveillance. The results suggest that consumption of EpiCor may act as an acute nutritional modulator of immune signaling, potentially engaging gut-associated immune and neuroimmune pathways to influence systemic inflammatory status.

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Poster 27 Symposium

Anti-osteoporotic, anti-inflammatory and in vivo safety evaluation of native *Lactobacillus* probiotics for osteoporosis management

Background and aims: Osteoporosis is a rising health burden in India and globally with current therapies limited by cost, side effects, lack of awareness and diagnosis. Probiotics particularly *Lactobacillus* have shown to modulate gut microbiota, enhance nutrient absorption, immunomodulation and production of short chain fatty acids (SCFAs). This study investigated the potential of indigenous lactobacilli strains for anti-osteoporotic and anti-inflammatory activity and their synergistic application with calcium and vitamin D.

Methods: A robust screening of indigenous lactobacilli isolates from fermented Indian foods, infant feces and breast milk for probiotic attributes (acid and bile tolerance, adhesion to binding proteins and Caco2 cells, antimicrobial activity, non-hemolytic and sensitive to tested antibiotics) was done. Anti-osteoporotic, anti-inflammatory activity and safety assessment were evaluated for potential isolates. Whole genome sequencing was done to understand genomic characteristics.

Results: Four isolates of *Limosilactobacillus fermentum* and *Lactiplantibacillus plantarum* out of a total 424 isolates were selected based on probiotic attributes. The cell filtrate of lactobacilli isolates was non-toxic to osteoblast cells MC3T3 and induced osteoblast differentiation at 10^4 - 10^9 CFU/ml. Alizarin red staining indicated mineralization in MC3T3 cells upon induction with lactobacilli cell suspensions. In vitro fecal fermentation increased the production of SCFA. The isolates showed downregulation of TNF- α , IL-6 and upregulation of IL-10 upon pre- and post-inflammatory challenge. The isolates were non-toxic to female rats in a single dose toxicity study and are being evaluated for anti-osteoporotic effect in ovariectomized rat model. Whole genome sequencing revealed presence of probiotic marker genes. The strains were finally incorporated in a freeze-dried fermented mix fortified with calcium and vitamin D.

Conclusion: Results indicate that indigenous lactobacilli could be potential anti-osteoporotic probiotics for the management of osteoporosis and strain-specific adaptability to the Indian population.

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Poster 28 Symposium

Adjunctive Probiotic/Prebiotic Interventions in Inflammatory Bowel Disease: A Retrospective Practice-Based Case Series

Background: Inflammatory bowel disease (IBD), including Crohn's disease and ulcerative colitis, is primarily managed by gastroenterologists using evidence-based medical therapy. Some patients/clients also seek wellness coaching regarding diet, probiotics, prebiotics, and other supplements as adjuncts to physician-directed care. Practice-based descriptions of these experiences may identify questions for future controlled research.

This retrospective practice-based case series reviewed practitioner-recorded coaching notes from normal wellness-coaching practice.

Methods: Adults with self-reported clinician-diagnosed Crohn's disease or ulcerative colitis who remained under the care of their own gastroenterologists and treating physicians were included when practitioner-recorded notes documented probiotic and/or prebiotic use and follow-up information. The author's role was wellness coaching and did not include gastroenterology care, diagnosis of IBD activity, prescribing medication, or directing changes to prescription IBD therapy. Notes documented participant-reported bowel habits, gastrointestinal symptoms, self-rated energy, supplement use, dietary guidance, medication changes reported by participants, antibiotic use, and health events mentioned during routine contacts. Probiotic/prebiotic guidance was generally part of normal coaching practice and was not delivered as a research protocol or standardized clinical trial regimen.

Results: Clinical observations included practitioner-documented participant reports of improved bowel habits, including reduced diarrhea or constipation, fewer bowel movements, and more formed stools; reduced or resolved abdominal pain; and improvement in selected other gastrointestinal symptoms, including bloating, reflux, indigestion, and appetite. Participants/clients also reported increases in self-rated energy, weight gain or regain in some cases, and improved mood in some cases. Outcomes varied across individuals, interventions, concurrent dietary and supplement use, medication changes, antibiotic exposure, and follow-up duration.

Conclusion: This retrospective practice-based case series describes practitioner-documented participant reports of improved bowel habits, abdominal symptoms, selected gastrointestinal symptoms, self-rated energy, weight, and mood during adjunctive probiotic/prebiotic use in a wellness-coaching setting alongside physician-directed IBD care. These observations may help identify practical questions regarding tolerability, timing, symptom monitoring, and the role of microbiome-targeted strategies as adjuncts to standard care. However, the uncontrolled design, practitioner-recorded nonstandardized notes, variable follow-up, self-reported information, concurrent dietary and supplement interventions, and changes in physician-directed treatment preclude conclusions regarding efficacy, disease control, remission, safety, or causality. Controlled, strain-specific research with standardized clinical outcomes is needed.

Conflict of interest: Inna Melamed is founder and owner of Real Health Solutions LLC, doing business as Digestive Reset and the Melamed Institute for Functional Medicine. She receives commissions through Fullscript and has commission-based relationships with Designs for Health, Moss Nutrition, and ProLon. Products used by individual clients varied, and not all probiotic/prebiotic products discussed were sourced from these companies. This presentation was independently funded; no manufacturer or external sponsor funded the analysis, poster preparation, travel, or symposium attendance.

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Poster 29 Symposium

Dietary fructooligosaccharides promote intestinal and systemic antibody production by modulating cellular responses in cecal patches.

Aims: Fructooligosaccharides (FOS) are representative prebiotics expecting several health benefits through improving gut microbiota. Dietary supplementation with FOS has been shown to promote IgA production in mucosal tissues such as saliva and the intestine in mice and rats. However, the cellular mechanisms underlying this phenomenon are still not understood, and its effects on the other antibody isotypes remain unclear. We aimed to examine the effect of dietary supplementation with FOS on intestinal and systemic

antibody production as well as the cellular responses of gut-associated lymphoid tissues as inductive sites.

Methods: 6 weeks-old BALB/c female mice were divided into Ctrl group and FOS group. Those mice were administered 0% or 7.5% FOS containing AIN-76 based purified diet for 2 weeks, respectively. The concentration of antibodies in serum and intestinal tissues was measured by ELISA. Flowcytometric analysis was performed to characterize B cells and follicular helper T (Tfh) cells within Peyer's patches and cecal patches. To assess the contribution of CD4⁺ T cells and innate immune signaling to the effect of dietary FOS on the antibody responses, mice depleted of CD4⁺ cells by treatment with a neutralizing antibody, as well as MyD88-deficient mice lacking the adaptor molecule for toll-like receptor signaling, were also used and analyzed.

Results: FOS group increased the amount of IgA and IgG2b in serum and intestinal tissues. FOS group increased pre-germinal center (GL7⁺CD95⁺) B cells and Tfh (CD4⁺CXCR5⁺PD-1⁺) cells in cecal patches. FOS containing diet did not increase the amount of IgA and IgG2b in serum and intestinal tissues from MyD88-deficient mice and the BALB/c mice depleted CD4⁺ cells. These results indicate that dietary FOS promotes intestinal and systemic antibody production by modifying cellular responses in cecal patches through innate and acquired immune signaling.

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Poster 30 Symposium

Identification of Th17-Inducing Dietary Fibers and Investigation of Their Underlying Mechanisms

Background: The emergence of infectious diseases, including COVID-19, has heightened interest in strategies for maintaining and enhancing immune function. Furthermore, extensive antibiotic use has raised concerns regarding antimicrobial resistance and gut microbiota dysbiosis. Given that approximately 70% of immune cells reside in the gastrointestinal tract, modulation of the intestinal microenvironment has emerged as a promising strategy for immune regulation. Among dietary components, dietary fibers are known to modulate the intestinal microenvironment by altering the composition of the gut microbiota. In addition, their fermentation by gut microbiota results in the production of metabolites that modulate host immune responses. In the present study, we investigated the effects of different dietary fibers on the intestinal microenvironment and immune cell populations to elucidate their immunomodulatory properties.

Methods: C57BL/6J mice were fed diets containing different dietary fibers, followed by analyses of fecal short-chain fatty acids (SCFAs), gut microbial composition, and colonic immune cell populations. Mice in the control group received a diet containing 5% cellulose (CE), a poorly fermentable dietary fiber. The other groups were fed diets containing 2.5% corn arabinoxylan (CX), wheat bran arabinoxylan (WX), or partially hydrolyzed guar gum (PHGG), with the remaining 2.5% consisting of CE to maintain an equivalent total dietary fiber content. To investigate the mechanisms underlying dietary fiber-induced immune modulation, dietary fibers or fecal suspensions were added to cultures of dendritic cells, naïve CD4⁺ T cells, or their co-culture, and their effects on T-cell differentiation were evaluated.

Results and Discussion: Dietary supplementation with WX or PHGG significantly increased fecal butyrate concentrations and the frequency of colonic regulatory T cells (Tregs). On the other hands, CX and PHGG supplementation significantly increased the frequency of colonic Th17 cells. Although butyrate has been well documented to promote Treg differentiation, the observed expansion of Th17 cells could not be explained by increased butyrate production, suggesting the involvement of additional mechanisms.

To further investigate the mechanism underlying Th17 cell induction by dietary fibers, we performed in vitro Th17 differentiation assays. Direct exposure to dietary fibers did not induce Th17 differentiation, whereas fecal suspensions obtained from CX- or PHGG-fed mice significantly promoted Th17 differentiation compared with those obtained from CE-fed mice. These findings indicate that certain dietary fibers indirectly promote the induction of colonic Th17 cells, possibly through alterations in the gut microbiota and/or microbiota-derived metabolites.

We are currently performing microbiome and metabolomic analyses to identify the factors responsible for Th17 induction associated with dietary fibers. These analyses may provide mechanistic insights into dietary fiber-mediated immune regulation.

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Poster 31 SFA

Live and heat-treated *Lactiplantibacillus plantarum* induce distinct metabolic and immune responses in intestinal epithelial cells

The health-promoting effects of lactic acid bacteria are traditionally attributed to their structural components and metabolic activities. However, whether bacterial viability fundamentally determines the quality and direction of host responses remains incompletely understood. Although both live and non-viable bacterial preparations are widely used, direct mechanistic comparisons under physiologically relevant host-microbe interaction settings are limited.

Bacterial-epithelial co-culture studies have historically been limited by technical challenges, primarily bacterial overgrowth and mismatched oxygen requirements. Advances in microfluidic systems have enabled precise control of microbial-epithelial co-cultures under physiologically relevant conditions, preserving epithelial integrity while preventing bacterial over proliferation. we employed a microfluidic co-culture platform (Shimadzu, Kyoto, Japan) to systematically investigate the differential regulation of swine intestinal epithelial cells (SIECs) by live and heat-treated *L. plantarum* JCM 1149T. Our aim was to elucidate the bacterial molecules and host signaling pathways that mediate viability-dependent metabolic and immunological responses by directly comparing live and heat-treated preparations of the same bacterial strain, thereby addressing key gaps in the mechanistic understanding of probiotic and postbiotic functionality. Transcriptomics and metabolomics revealed fundamentally distinct epithelial responses dependent on bacterial viability. Live *L. plantarum* induced a hypoxia-associated glycolytic shift and PPAR γ -related transcriptional responses independently of HIF1 α , accompanied by the production of lipid mediators including 12,13-diHOME and 9,10-diHOME. In contrast, heat-treated *L. plantarum* induced NF κ B-associated pro-inflammatory gene expression in a TLR4-dependent manner, likely in response to alterations in bacterial surface components.

These findings demonstrate that variations in treatment, even within the same strain, can fundamentally alter the qualitative nature of host responses. Systematic comparison of live and non-viable bacteria under physiologically controlled conditions enabled a mechanistic distinction between probiotic and postbiotic strategies. These insights provide a foundation for rationally selecting microbial preparations according to specific metabolic or immunological outcomes.

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Poster 32 Symposium

Postbiotics derived from *Ligilactobacillus salivarius* improve mitochondrial homeostasis via TLR2-mediated signaling

Objectives: Postbiotics are defined as “a preparation of inanimate microorganisms and/or their components that confers a health benefit on the host.” Owing to their greater stability and storability compared with live probiotics, postbiotics have attracted increasing attention for their potential applications in food and health-related fields. Mitochondrial function is known to decline with aging, and mitochondrial dysfunction is closely associated with various age-related diseases. In particular, the maintenance of mitochondrial homeostasis in immune cells is important for regulating inflammatory responses and tissue homeostasis. In this study, we aimed to investigate whether postbiotics contribute to the maintenance of mitochondrial homeostasis. To this end, mitochondrial dysfunction was induced in intestinal macrophages, and the effects of postbiotics prepared from several strains of *Ligilactobacillus salivarius* on mitochondrial dysfunction were evaluated.

Methods: An intestinal macrophage cell line, IPIM, was used to assess mitochondrial function. To induce mitochondrial dysfunction and mitochondrial reactive oxygen species (mtROS) production, cells were treated with antimycin A (AMA) at concentrations ranging from 5 to 500 μ M. mtROS production was measured using mtROS580, and the expression of redox-related genes was analyzed by RT-qPCR. Postbiotics were prepared from four strains of *L. salivarius* by culturing each strain followed by heat treatment. IPIM cells were pretreated with each postbiotic preparation for 24 h and subsequently exposed to AMA to induce mtROS production. The ability of each postbiotic preparation to activate innate immune receptor signaling was evaluated using a reporter assay in HEK293 cells. Finally, the effects of the postbiotics on mitochondrial function were examined using a flux analyzer.

Results: Treatment of IPIM cells with 500 μ M AMA induced mtROS accumulation and impaired SOD2 induction, suggesting disruption of mitochondrial homeostasis. Although the postbiotics were prepared from strains belonging to the same bacterial species, only postbiotics derived from specific *L. salivarius* strains significantly suppressed AMA-induced mtROS accumulation and enhanced SOD2 expression. Based

on previous findings showing that SOD2 expression can be induced through innate immune receptor signaling, we examined the reactivity of the prepared postbiotics toward Toll-like receptor 2 (TLR2). Interestingly, the postbiotics that suppressed mtROS production showed higher reactivity toward TLR2. Consistently, the inhibitory effect of the postbiotics on mtROS production was abolished by the addition of a TLR2 inhibitor. Furthermore, postbiotic pretreatment enhanced mitochondrial respiratory function. These findings suggest that specific *L. salivarius*-derived postbiotics improve mitochondrial homeostasis by activating TLR2-mediated signaling.

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Poster 33 IAC

Protective Effects of Probiotic Lactobacilli Attenuate Blue Light-Induced Retinal Degeneration in a Murine Model

Background: Blue light (400–500 nm), a high-energy visible light capable of penetrating ocular tissues to the retina, induces photooxidative stress and has been implicated in retinal degeneration, including age-related macular degeneration, as well as digital eye strain associated with prolonged digital device use. This study investigated whether oral administration of selected lactic acid bacteria (LAB) strains protects against chronic blue light-induced retinal damage in mice and explored the potential involvement of the gut–retina axis.

Methods: Male ICR mice received oral LAB supplementation for 17 weeks, including one week of pretreatment followed by 16 weeks of concurrent supplementation during blue light exposure. To mimic chronic digital device exposure, mice were exposed to low-intensity blue light for 6 hours daily over 16 weeks. Visual function, retinal morphology, photoreceptor integrity, oxidative stress, inflammation, apoptosis, gut microbiota composition, and short-chain fatty acid (SCFA) production were evaluated.

Results: Chronic blue light exposure significantly impaired visual acuity and retinal electrophysiological responses and induced retinal structural damage, including thinning of the outer nuclear layer (ONL) and photoreceptor inner/outer segment (IS/OS) layers, accompanied by reduced ONL nuclei counts. LAB supplementation significantly attenuated these changes, preserving visual function and electroretinogram responses while restoring ONL thickness, IS/OS thickness, and ONL nuclei counts to levels comparable with non-exposed controls. LAB treatment also preserved rhodopsin and S-opsin expression, indicating

improved maintenance of photoreceptor integrity under blue light stress. Furthermore, LAB reduced retinal gliosis, as evidenced by decreased glial fibrillary acidic protein (GFAP), and attenuated oxidative stress and inflammatory responses, including reduced 8-hydroxy-2'-deoxyguanosine (8-OHdG), malondialdehyde (MDA), and tumor necrosis factor- α (TNF- α), together with suppression of NF- κ B activation and caspase-3 expression. Gut microbiota and SCFA analyses demonstrated that LAB supplementation altered microbial composition and metabolite production, suggesting that modulation of the gut-retina axis may contribute to the observed retinal protection.

Conclusions: Oral supplementation with selected LAB strains mitigated chronic blue light-induced retinal dysfunction and structural degeneration in mice while reducing oxidative stress, inflammation, and apoptosis. These findings support microbiota-mediated modulation of the gut-retina axis as a potential mechanism underlying probiotic-mediated retinal protection and warrant further investigation of probiotics as a strategy to promote retinal health during chronic blue light exposure.

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Poster 34 SFA

Restoring Immune Balance through Ancestral Microbes: Functional Insights into host-microbe interactions of VANISH and BloSSUM Taxa

The incidence of non-communicable inflammatory diseases, including multiple sclerosis (MS), allergies, and inflammatory bowel disease (IBD), has risen drastically across industrialized societies over the past century. A prevailing hypothesis attributes this increase to lifestyle transitions from traditional, non-industrialized communities such as the Hadza in Tanzania and the Yanomami in the Amazon to Westernized diets and environments characterized by low fibre intake, high consumption of processed and sugary foods, and widespread antibiotic exposure. These lifestyle changes reshape the gut microbiome by depleting ancestral VANISH (Volatile and/or Negatively Influenced by Societies of Humans) taxa while enriching BloSSUM (Bloom/Selected Societies of Urbanization/Modernization) taxa. These alterations may disrupt host-microbe homeostasis, drive immune dysregulation, and promote chronic inflammation. Although there is growing interest in the VANISH taxa and their metabolites as next-generation biotic candidates, their immunological roles remain poorly understood.

To investigate how the depletion of VANISH taxa and the expansion of BloSSUM taxa influence immune pathways, we examined cell-free bacterial supernatants (CFBS) from cultured VANISH and BloSSUM species. CFBS were applied to reporter cell lines (HT-29 (AhR) and THP-1 Dual (NF- κ B/IRF)) and primary healthy donor PBMCs at 1, 5, and 10% v/v for 24 hours. Innate pathway signalling was assessed through luminescent reporters, and cytokine secretion (TNF- α , IL-1 β , IL-6, IFN- γ , and IL-10) was quantified using ELISA. BloSSUM species induced consistent proinflammatory cytokine secretion, including IL-1 β , IL-6, and TNF- α , accompanied by compensatory IL-10 responses and maintained PBMC viability across doses. VANISH species exhibited greater variability, with robust responses limited to selected strains. Notably, *Treponema succinifaciens*, a rarely studied VANISH taxon, showed measurable immunomodulatory activity, highlighting its potential immune relevance despite its near absence in modern microbiomes. Reporter assays revealed distinct patterns: BloSSUM strains primarily activated NF- κ B, while IRF signalling was broadly induced, and

AhR activation was limited to specific taxa, suggesting metabolite-driven selective interactions.

These findings demonstrate that bacterial secretomes fine-tune immune signalling in a strain- and dose-dependent manner. The combined depletion of VANISH taxa and enrichment of BloSSUM species in industrialized microbiomes may therefore promote heightened immune activation, supporting microbiota restoration strategies using live biotherapeutics and biotic interventions to re-establish immune balance.

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Theme 5: Metabolic health and liver disease

Poster 35 IAC

A Next-Generation Butyrate-Producing Commensal for Metabolic Health: Translational Evidence for *Anaerobutyricum soehngenii*

The modulation of gut microbial functionality is increasingly recognized as a promising strategy to support metabolic health. In particular, next-generation commensal strains with defined metabolic outputs offer opportunities beyond traditional probiotics. *Anaerobutyricum soehngenii* is a strictly anaerobic, butyrate-producing bacterium with the capacity to influence host energy metabolism through microbial metabolite production and host-microbe interactions.

The work on AS integrates findings from preclinical studies, over dose-escalation studies^{1,2} to human RCTs investigating the safety, tolerability, and metabolic effects of *A. soehngenii*^{3–7}. Across these studies, administration of the strain—either orally or via targeted intestinal delivery—was consistently associated with improvements in markers of metabolic regulation. These included improvements in postprandial hormone excursions, decreased glucose variability, and suggested improved insulin sensitivity in individuals with impaired metabolic profiles.

Mechanistically, these effects are supported by the strain's capacity to enhance butyrate availability and transform bile acids, as demonstrated in preclinical models, and by observed changes in intestinal gene expression linked to nutrient sensing, barrier function, and metabolic signaling pathways^{1,2}. Notably, modulation of these pathways occurred without adverse safety signals, and dose-dependent responses suggest a direct functional contribution of the strain to host metabolic processes. In addition to effects on glycemic variability replicated across trials, exploratory analyses indicated favorable shifts in cardiometabolic parameters, including blood pressure regulation. While these findings remain preliminary, they point toward a broader role of microbial-derived metabolites in systemic metabolic homeostasis. Given the well-established link between sustained metabolic health and healthy aging, such effects may also be relevant in the context of longevity-oriented interventions.

Collectively, the evidence positions *A. soehngenii* as a well-characterized next-generation microbial candidate with translational potential to support metabolic health. Its targeted functional profile, centered

on butyrate production and host pathway modulation, underscores its relevance for future strategies aimed at maintaining metabolic resilience and promoting long-term health.

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Poster 36 Symposium

***Roseburia hominis*, Identified via a Human Microbiota-Associated Mouse Model, Protects Against MASLD**

Metabolic dysfunction-associated steatotic liver disease (MASLD) is closely linked to gut microbiota dysbiosis, which contributes to hepatic injury, metabolic dysfunction, and disease progression. To identify microbiota-derived factors protective against MASLD, human microbiota-associated (HMA) mice were established by transplanting fecal microbiota from MASLD patients of varying disease severity into germ-free mice. Among donor-derived groups, mice colonized with microbiota from donor D (HMA_D) exhibited the strongest protection against metabolic dysfunction-associated steatohepatitis (MASH). Comparative profiling identified *Roseburia hominis* (Rh) and *Bacteroides stercoris* (Bs) as bacterial species enriched in HMA_D mice, implicating them in the observed protection.

To test their functional significance, Rh and Bs were evaluated in the Gubra-Amylin NASH (GAN) diet mouse model. Using the nonalcoholic fatty liver disease activity score (NAS), a combined Rh and Bs bacterial suspension reduced the proportion of mice meeting MASH criteria by 33%, exceeding the effect of either strain alone. Although individual Rh or Bs administration showed limited efficacy against GAN diet-induced

hepatic injury, both significantly reduced liver weight gain, plasma total cholesterol, and plasma non-HDL cholesterol. Notably, Rh outperformed Bs on metabolic endpoints: Rh significantly lowered fasting blood glucose ($p = 0.0061$) and improved insulin resistance ($p = 0.0428$), accompanied by increased expression of key insulin-signalling molecules IRS1 and PI3K.

Thus, while the combination of Rh and Bs conferred the greatest overall protection against MASLD progression, Rh alone produced pronounced metabolic benefits, including reduced body weight gain, improved glucose homeostasis, and enhanced insulin sensitivity. Together, these findings position Rh as a promising candidate for development as a next-generation probiotic (NGP) for MASLD.

Building on these findings, future studies will assess the preventive efficacy of distinct Rh preparations—live bacteria, pasteurized bacteria, proteinase K-digested pasteurized bacteria, and cell-free supernatant—in a high-fat-diet-induced MASLD model. These studies aim to elucidate how Rh improves insulin sensitivity, hepatic lipid accumulation, and inflammation, and to identify the most effective Rh-based strategy for therapeutic development.

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Poster 37 Symposium

Soy-derived resistant protein attenuates high-fat diet-induced obesity

Dietary composition critically influences host metabolism, and excessive intake of energy-dense diets, particularly high-fat diets (HFD), increases the risk of obesity and related metabolic disorders. Obesity is associated with gut microbiota dysbiosis, characterized by reduced microbial diversity and altered bacterial composition. Although dietary proteins can modulate gut microbiota, the metabolic effects of protein (e.g., soy-derived resistant protein) remain unclear. In this study, we investigated the impact of soy-derived resistant protein on HFD-induced obesity in mice. We found that the body weight of soy-derived resistant protein-fed mice was significantly lower than that of HFD-fed mice during growth. In addition, the fat mass of white adipose tissue (WAT) was significantly reduced in soy-derived resistant protein-fed mice compared to HFD-fed mice at 12 weeks of age. Moreover, we found that levels of the plasma GLP-1 (glucagon-like peptide-1) were significantly higher in soy-derived resistant protein-fed mice than in HFD-fed mice. These metabolic improvements were accompanied by attenuation of hepatic lipid accumulation and marked alterations in gut microbial composition. Furthermore, bile acid profiles differed between HFD-fed and soy-derived resistant protein-fed mice, suggesting a link between soy-derived resistant protein, bile acid metabolism, and host energy regulation. Notably, these beneficial metabolic effects of soy-derived resistant protein were abolished in germ-free mice, indicating that the gut microbiota is essential for mediating the effects of soy-derived resistant protein. Collectively, our findings demonstrate that soy-derived resistant protein contributes to the regulation of host energy homeostasis under obese conditions. These results suggest that targeted dietary protein interventions may represent a promising strategy for preventing metabolic disorders by modulating the gut microbiota.

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Poster 38 SFA

Strain-Specific Modulation of Host Lipid Metabolism by *Lactobacillus paracasei* DTA81 Through Gut Microbiota Restructuring and Immunoregulation

Diet-induced hypercholesterolemia is closely associated with gut microbiota dysbiosis, metabolic inflammation, and impaired host lipid homeostasis. We investigated the mechanistic cholesterol-lowering potential of the probiotic strain *Lactobacillus paracasei* DTA81 using an integrated in vitro, in vivo, and genomic approach. In vitro analyses demonstrated strong gastrointestinal stress tolerance and cholesterol assimilation capacity. In a BALB/c mouse model receiving a high-fat diet, six weeks of oral DTA81 supplementation significantly reduced total serum cholesterol and fasting blood glucose compared with untreated controls. Colon tissue cytokine profiling revealed a significant reduction in interleukin-6, indicating attenuation of diet-induced intestinal inflammation. High-throughput 16S rRNA gene sequencing showed that DTA81 administration reshaped gut microbial community structure, characterized by increased relative abundance of Bacteroidetes and enrichment of the butyrate-associated genus *Coprococcus*, taxa commonly linked to improved metabolic outcomes and short-chain fatty acid production. These microbial shifts suggest restoration of microbial functionality potentially contributing to host lipid regulation. Whole-genome sequencing confirmed the presence of genes associated with cholesterol assimilation and bile salt hydrolase activity, while comprehensive safety assessment demonstrated the absence of transmissible antibiotic resistance genes, plasmids, and intact prophage regions. Collectively, these findings indicate that *L. paracasei* DTA81 modulates host metabolic parameters through coordinated effects on gut microbial ecology and inflammatory signaling, supporting its development as a safe, mechanistically grounded, non-pharmacological strategy for metabolic health improvement under high-fat dietary conditions.

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Poster 39 SFA

Targeted delivery of short-chain fatty acids affects circulating concentrations, GLP-1 and PYY release and self-reported appetite

Short-chain fatty acids (SCFA), predominantly acetate, propionate and butyrate, are microbial metabolites associated with various metabolic health benefits. In the human colon, SCFA are produced through fiber fermentation and absorbed by colonocytes, which use them as energy source. In contrast, fermented foods contain SCFA that are absorbed in the small intestine. Unlike colonocytes, small intestinal enterocytes do not utilize SCFA as energy source. Therefore, we hypothesized that a greater proportion would reach the systemic circulation following small intestinal (SI) versus colonic (Co) administration. Additionally, SCFA stimulate enteroendocrine L-cells to release glucagon-like peptide 1 (GLP-1) and peptide YY (PYY), which regulate appetite and glucose homeostasis. Because L-cells are more abundant in the colon, we hypothesized that colonic SCFA administration would result in greater gut hormone secretion.

Both hypotheses were tested in a randomized, crossover study involving 28 healthy individuals. On separate visits, a single dose of SCFA (230 mmol, Ac:Pr:Bu 6:2:2) was administered into either the small intestine or colon using targeted-delivery capsules. A placebo (Pl) visit enabled correction for meal-induced GLP-1 and PYY secretion. Postprandial blood samples were collected up to 8h to quantify GLP-1, PYY, circulating SCFA, glucose and c-peptide. Appetite scores were assessed using VAS.

SI SCFA administration increased GLP-1 versus Co administration and placebo (iAUC mean [95% CI] SI: 69.99 [56.15–83.83] vs. Co: 53.99 [39.50–68.49] vs. Pl: 39.73 [26.09–53.38] pM.h, $\text{padj} < 0.05$), whereas Co administration increased PYY versus SI administration and placebo (SI: 183.67 [124.63–242.71] vs. Co: 295.13 [197.28–392.98] vs. Pl: 87.13 [38.50–135.76] pg.ml-1.h, $\text{padj} < 0.0001$). Circulating SCFA concentrations were higher following SI versus Co delivery (iAUC (mean [95% CI]) acetate: 379 [258–501] vs. 19.1 [–65–103] $\mu\text{M.h}$, $\text{padj} < 0.0001$; propionate: 24.6 [19.7–29.6] vs. 6.62 [3.66–9.58] $\mu\text{M.h}$, $\text{padj} < 0.0001$; butyrate: 9.92 [6.92–12.9] vs. 2.71 [1.35–4.06] $\mu\text{M.h}$, $\text{padj} < 0.0001$). SI delivery reduced hunger and prospective food consumption, and increased fullness and satiation ($\text{padj} < 0.01$) versus placebo. Colonic delivery also increased fullness and satiation and reduced prospective consumption ($\text{padj} < 0.05$). Glucose homeostasis remained unaffected.

Thus, the site of SCFA administration impacts gut hormone release and systemic SCFA exposure. These findings highlight the importance of administration site when evaluating the metabolic effects of SCFA.

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Poster 40 Symposium

The Potential of Proteinase K-digested Pasteurized *Parabacteroides merdae* as a Next-Generation Postbiotic Against MASLD

Metabolic dysfunction-associated steatotic liver disease (MASLD) is a major health burden that affects approximately one-quarter of the global population. MASLD can progress to metabolic dysfunction-associated steatohepatitis (MASH), increasing the risk of fibrosis, cirrhosis, and hepatocellular carcinoma. The gut microbiota is a critical driver of MASLD pathogenesis, and next-generation probiotics (NGPs) have recently emerged as promising therapeutic candidates for metabolic disorders. Among them, *Parabacteroides merdae* (PM) has been reported to exert cardioprotective effects and ameliorate hepatic steatosis. Across three complementary mouse studies, we demonstrated that both live and pasteurized PM (pPM) alleviate MASLD, with the NTUH_O15 strain showing the strongest improvement in insulin resistance. To determine whether the active compounds are proteinaceous, we compared pPM with proteinase

K-digested pPM (PK-pPM). Both improved glucose–insulin homeostasis and reduced hepatic steatosis, with transcriptomic analysis indicating enhanced retinoic acid signaling as a potential mechanism. The greater efficacy of PK-pPM suggests that these functional components may be non-proteinaceous bioactive metabolites. Additionally, pPM from the type strain reduced hepatic lobular inflammation by decreasing M0 macrophages while increasing M2 macrophages. To further validate the therapeutic potential of PK-pPM, we examined its efficacy in an independent mouse model of MASLD. PK-pPM reproducibly improved metabolic dysfunction, significantly lowering serum triglycerides, cholesterol, non-HDL cholesterol, and OGTT AUC. Importantly, these beneficial effects were recapitulated in a HepG2 steatosis model, where PK-pPM markedly reduced intracellular lipid accumulation. Ongoing histological and transcriptomic analyses are expected to provide mechanistic insights into these protective effects. Collectively, these findings support the potential of PK-pPM as a next-generation postbiotic for the prevention and treatment of MASLD.

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Poster 41 Symposium

Translating pasteurized *Akkermansia massiliensis* LWHK0003 from preclinical models to clinical application as a next-generation postbiotic candidate for MASLD

Metabolic dysfunction-associated steatotic liver disease (MASLD) affects approximately a quarter of the global population and can progress to steatohepatitis (MASH), cirrhosis, and hepatocellular carcinoma, yet effective pharmacotherapies remain scarce. Next-generation postbiotics (NGPs), such as pasteurized *Akkermansia muciniphila*, offer a promising alternative. *Akkermansia massiliensis*, the second most prevalent *Akkermansia* species in the human gut, remains largely uncharacterized. We isolated domestic *Akkermansia* spp. from healthy donors; among them, *A. massiliensis* LWHK0003 most strongly stimulated glucagon-like peptide-1 (GLP-1) secretion from enteroendocrine L-cells, and oral gavage raised circulating GLP-1 in germ-free mice. In a GAN diet-induced MASH model, both live and pasteurized LWHK0003 reduced the NAFLD activity score, mainly by attenuating lobular inflammation, alongside lower hepatic TNF- α , IL-1 β , and IL-18; the pasteurized form (pAma) markedly elevated plasma GLP-1. We then conducted a randomized controlled Phase I/II trial of low-, medium-, and high-dose pAma in patients with MASLD. After three months, pooled analysis showed significant reductions in FibroScan controlled attenuation parameter and liver stiffness, indicating improved steatosis and fibrosis, with improved insulin resistance (lower HOMA-IR) in the low- and medium-dose groups versus placebo. No adverse changes in renal function or inflammatory markers were observed, supporting a favorable safety profile. Pasteurized *A. massiliensis* LWHK0003 represents a promising NGP candidate for MASLD, advancing from mechanism to first-in-human evidence.

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Poster 42 IAC

Modulation of Microbiome-Derived Uremic Toxins and Short-Chain Fatty Acids in Healthy Senior Cats

Chronic kidney disease (CKD) is estimated to affect 30-40% of cats >10 years old and is considered a primary cause of mortality in cats overall. With aging, microbiome-derived uremic toxins (UT) increase and are associated with declining kidney function, but this process remains poorly understood in otherwise healthy senior cats. The objective was to characterize baseline microbiome-metabolite relationships in healthy senior cats to better understand the gut-renal axis in this population. Forty-eight healthy senior cats (≥ 7 yr) were fed a standard maintenance diet for 30 d prior to sampling. Fecal microbiome composition was assessed via shotgun metagenomics (MetaPhlAn SGBs, MAGs, CAZymes, glycans, HUMAnN pathways). Serum, urine, and fecal metabolites were quantified by metabolomics. Microbiome-metabolite associations were assessed by Spearman correlations (FDR-corrected). Effects on CLR-transformed abundances were tested using linear models that were adjusted for sex, location, age, and body condition score. At baseline, fecal microbiota and serum metabolite profiles were significantly concordant (Procrustes correlation = 0.53, $P < 0.001$). Coprostanol was strongly correlated with Firmicutes SGB4304 ($\rho = 0.81$, $q < 0.01$), valerate with *Megasphaera elsdenii* ($\rho = 0.66$, $q = 0.003$), and indole-derived metabolites (indoleacetate, indoleacetylglutamine) with *Mogibacterium kristiansenii* ($\rho = 0.62-0.73$, $q < 0.02$). Integration of additional metagenomic layers (MAGs, CAZymes, glycans, pathways) is underway to elucidate the microbial mechanisms driving these metabolic shifts. By characterizing microbiome-metabolite networks tied to uremic toxin production in healthy aging cats, these results provide an observational foundation for future studies of the gut-kidney axis.

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Theme 6: Microbial discovery and ecology

Poster 43 Symposium

Sporulation characteristics of human gut microbes, a focus on butyrate-producing bacteria

Objective: Butyrate-producing gut bacteria are regarded as promising candidates for next-generation probiotics because of their beneficial impacts on host health. However, most gut bacteria are strict anaerobes and rapidly lose viability under atmospheric conditions, which limits their practical application as probiotics and live biotherapeutic products (LBPs). The present study evaluated the sporulation properties of human gut bacteria, with a particular focus on butyrate producers, and studied the impacts of sporulation on their survival under atmospheric conditions.

Materials and Methods: Potential spore-forming bacteria were isolated from ethanol-treated fecal samples obtained from six healthy individuals. A total of 77 strains, including the fecal isolates and strains obtained from culture collections, were examined for sporulation using phase-contrast microscopy and ethanol treatment. Conservation of sporulation signature genes was further characterized by genome-based *in silico* analysis. Oxygen tolerance of spore-forming strains was also evaluated.

Results: A total of 100 strains belonging to 25 species were isolated from ethanol-treated fecal samples, including potentially beneficial species *Faecalibacterium duncaniae* and disease-associated species *Mediterraneibacter gnavus* and *Enterocloster bolteae*. The number of spores in feces was estimated to be approximately 10^5 SCFU/g, suggesting that only a very small fraction of vegetative cells form spores in the human gut. Among the 77 gut bacterial strains examined, sporulation was observed in 22 strains. *Anaerostipes* spp. and *Blautia* spp. showed relatively high spore-formation frequencies, reaching up to 2%. Genome-based analysis revealed that several strains that did not show sporulation under the tested conditions possessed sporulation-related gene sets comparable to those of known spore-formers. Spore-forming *Anaerostipes* and *Blautia* strains survived under atmospheric conditions for more than 15 weeks, whereas non-spore-forming *Anaerostipes* and *Blautia* strains rapidly lost viability within a few hours. Notably, *Anaerostipes caccae* survived for more than three years when spores were formed.

Conclusions: The present study demonstrated that endospore formation markedly enhances oxygen tolerance in human gut anaerobes. These findings suggest that spores may serve as an effective delivery form for beneficial, oxygen-sensitive gut microbes as probiotics and LBPs.

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Poster 44 Symposium

Novel *in vitro* gut model with anaerobic coculture capability to enhance probiotic product development

Development of probiotics and live biotherapeutic products (LBPs) is constrained by a shortage of *in vitro* models that faithfully recreate host-microbe interactions at the gut epithelial interface. A central obstacle is the incompatibility between oxygen-requiring epithelial cells and the obligate anaerobes that dominate the gut microbiota, resulting in a need for expensive custom fabricated cell culture units or use of animal models.

To address this limitation, we have developed a novel proprietary model using Transwell™ cell culture workflows. This included validating oxygen states, suitability for anaerobic exposure cell growth, and optimising growth media to support both epithelial cells and anaerobic microbes. Caco-2 cells (an industrial

standard human intestinal cell line) were grown for three weeks under standard aerobic conditions before being integrated with our system, providing a dedicated compartment of oxygenated media to support epithelial cells from below. This is complemented by an anaerobic compartment which allows cultures of anaerobic microbes directly above epithelial cells, mimicking the anatomical compartmentalisation of the gut lumen and barrier. Four microbiome-relevant anaerobic bacteria were tested in the system and barrier responses of epithelial cells during coculture were monitored via changes in permeability. Barrier responses were further evaluated in inflamed and non-inflamed states, with and without bacterial coculture, and host responses profiled by qPCR.

Caco-2 gut epithelial monolayers maintain strong barrier function before and during anaerobic exposure in our model, with no significant loss of barrier function observed. All four anaerobic microbes tested remained viable during experiments, demonstrating that bacterial growth is supported over 24 hours of coculture. Microbial coculture modulated barrier function during inflammatory stimulation experiments, including *Bifidobacterium bifidum* coculture improving epithelial barrier response to inflammatory challenge.

The *in vitro* gut model presented enables physiologically relevant, direct live coculture of obligate anaerobes and gut epithelium in a scalable high-throughput manner without reliance on animal models. It offers a reproducible, accessible research tool for screening and derisking probiotic and LBP candidates during development, while offering traditional cell culture data outputs to support mechanistic and regulatory data generation for probiotic products.

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Poster 45 IAC

Genomic and Safety Assessment of an Understudied *Bifidobacterium* Species with Probiotic Potential

Bifidobacterium pseudocatenulatum is a common member of the healthy human gut microbiota, yet it remains relatively understudied, and its probiotic potential and safety profile is poorly characterized compared to commercial bifidobacterial species. We previously described properties of *B. pseudocatenulatum* iVE-2 (formerly ED02) that had been isolated from a healthy human. The objective of this study was to evaluate the probiotic potential of this novel strain using genome mining and clinical safety assessment. Following complete genome sequencing of *B. pseudocatenulatum* iVE-2, we identified genes involved in prebiotic and complex carbohydrate utilization, stress tolerance, B vitamin biosynthesis, and host adaptation. The genome was also screened for genes of safety concern including antimicrobial resistance determinants, virulence factors, and biogenic amine production pathways, and none were present. To further establish safety, *B. pseudocatenulatum* iVE-2 was evaluated in a randomized, double-blind, placebo-controlled clinical trial involving healthy adults (n = 30). Participants consumed iVE-2 (2×10^{10} CFU) or placebo daily for four weeks and were monitored for adverse events and gastrointestinal tolerance throughout the trial. No adverse events due to product consumption compared to the placebo, serious adverse events, or adverse event withdrawals were reported during the intervention, and no safety concerns were identified. Collectively, these findings demonstrate that *B. pseudocatenulatum* iVE-2 possesses genetic characteristics consistent with probiotic functionality and exhibits a favorable safety profile in humans. This work contributes to the growing body of evidence supporting the development of understudied bifidobacterial species as next-generation probiotics and highlights the value of integrating genomic and clinical approaches for evaluating potential probiotic strains.

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Poster 46 SFA

Lactocaseibacillus rhamnosus GG metabolic modeling for analysis of nutrients, secretions, and probiotic community interactions

Lactocaseibacillus rhamnosus GG (LGG) is an important gut commensal bacterial strain that has been extensively studied in both industrial and health applications. Despite its long history of study, a high-quality genome-scale metabolic network model (GEM) for LGG has yet to be reconstructed. Furthermore, defined media have not been established for well-controlled in vitro study of LGG. Here we present the first curated GEM for LGG using a new approach for reconstruction and validation that leverages automatically-generated draft models, applied study literature, and high-throughput defined media experiments. In addition, our results include a series of chemically defined media representing various nutritional richness, extensive single-component nutrient dropout growth data, insights from in silico and in vitro experiments into secretion products lactate and host-beneficial indole-3-carboxaldehyde, a minimal medium and in silico characterization of LGG's metabolic requirements. Using this toolbox, we designed genetic and nutrient interventions for increases in therapeutic microbial secretions from LGG growing in a three-species consortium. We leveraged community modeling approaches to assess cross-feeding interactions leading to butyrate secretion. This work presents a mechanistic metabolic toolbox for the important probiotic bacterium LGG, identifies targeted interventions to improve LGG's probiotic potential, and establishes a framework for systems-level interdisciplinary mechanistic study and improvement of other probiotic strains.

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Poster 47 Symposium

Antagonistic properties of bacteriocin-producing strain *Streptococcus salivarius* K12

Introduction: Antimicrobial resistance is a global problem. A new term has emerged within the global medical community: "antibiotic-sparing technologies." *Streptococcus salivarius* K12 (SsK12) is a probiotic that naturally inhabits the oropharyngeal area. Research suggests that SsK12 reduces the risk of ENT infections and decreases the need for antibiotics.

Aim: To evaluate the antagonistic activity of SsK12 against ENT and oral cavity infections pathogens (*S. pneumoniae*, *S. pyogenes*, *S. aureus*), gram-negative bacteria (*E. coli*, *P. aeruginosa*) and *C. albicans*.

Materials and methods: The probiotic strain SsK12 was isolated from dietary supplement, containing at least 1×10^9 CFU per tablet. The tablet was dissolved in the enrichment broth. The resulting suspension was seeded on 5% blood agar and incubated at 35°C in 4-6% CO₂ for 48 hours. The raised culture was identified

as *Streptococcus salivarius* with MALDI-TOF mass spectrometry method. The evaluation of SsK12 antagonistic activity was carried out using a perpendicular streak technique. The daily SsK12 culture was inoculated as heavy streaks with a loop at one side of Petri dish with the Muller-Hinton agar (MHA) and incubated for 24 hours at 35° C in anaerobic conditions. It was supposed that bacteriocins would diffuse over the whole area of the agar media. On the next day *S. pneumoniae*, *S. pyogenes*, *S. aureus*, *E. coli*, *P. aeruginosa* and *C. albicans* clinical isolates were streaked at the clear side of MHA Petri dish. MHA Petri dish inoculated with SsK12 (one part) and with the respective clinical isolates (another part) streaked perpendicularly on the same day was used as the control.

Results: There was no growth of *S. pyogenes* on the Petri dish with SsK12 daily culture; the growth of a few colonies of *S. pneumoniae* was noted. The growth of *S. aureus*, *E. coli*, *P. aeruginosa* and *C. albicans* was noted along the inoculated streak. On the control Petri dish with simultaneous inoculating of the SsK12 strain and the test cultures, the growth of all the testes isolates was noted.

Conclusions:

1. SsK12 possesses antagonistic activity against *S. pyogenes* and *S. pneumoniae*.
2. There was no antagonistic activity of SsK12 against *S. aureus*, *E. coli*, *P. aeruginosa* and *C. albicans*.
3. SsK12 antagonistic properties make it possible to use it as antibiotic-sparing technology.

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Poster 48 SFA

A case study of gastrointestinal microbiome biogeography in the endangered François' lutung (*Trachypithecus francoisi*): Implications for probiotics and dietary management

The François' lutung (*Trachypithecus francoisi*) is an endangered folivorous primate with a highly specialized foregut-fermenting digestive system, yet data on microbial communities across its gastrointestinal (GI) tract remain limited. Here, we present a case study of a hand-reared juvenile François' lutung in which we characterized the microbiome, luminal pH, and histology across multiple GI sites and compared these findings with fecal and GI microbiomes from three adult conspecifics that were maternally reared in captivity. Distinct microbial communities were associated with different GI compartments, emphasizing the importance of microbiome biogeography beyond fecal samples alone. Compared with adults, the juvenile exhibited an altered microbiome enriched in lactic acid-producing and potentially inflammatory taxa, consistent with a microbial community adapted to readily fermentable substrates. The distal gut of the juvenile also exhibited a lower pH compared to the adults. These shifts may have been influenced by the juvenile's modified diet, which included probiotic supplements and foods that were easily digestible to support its weight gain. Additionally, the individual suffered from acute gastric dilation, raising the possibility that altered foregut fermentation contributed to disease progression. Our findings suggest that diet may disproportionately affect dietary specialists with immature microbiomes, highlighting its potential influence on vulnerable individuals. Furthermore, they highlight the value of characterizing microbial communities throughout the GI tract to better understand digestive function in folivores. The results also suggest that probiotics developed for humans or omnivorous species should be applied cautiously in animals with specialized digestive ecologies, as disruption of host-adapted microbial communities may have unintended consequences. Ultimately, understanding species-specific microbiome composition will help inform evidence-based husbandry, veterinary care, and conservation of endangered folivorous primates.

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Poster 49 Symposium

Diversity of *Lactobacillaceae*, *Bifidobacteriaceae*, and Other Taxa Used by the Probiotics Industry in Small Intestinal Samples

We constructed a large-scale small intestinal metagenomic dataset comprising over 400 samples collected using a non-invasive sampling device (SIMBA capsules) across multiple observational clinical studies. All samples were paired with detailed clinical metadata, and most matched fecal samples were biobanked. The cohort represents a well-characterized Canadian population, providing a controlled demographic baseline that can support future clinical investigations and comparative studies.

Unlike conventional endoscopic sampling, which captures a single localized point, SIMBA capsule-derived samples demonstrated improved sample quality, as indicated by a higher proportion of nonhost sequencing reads against endoscopy (79.5% vs. 7.91%). The SIMBA capsule enables broader sampling through its larger inlet diameter and extended sampling time. This design likely contributes to increased sample volume and more representative microbial capture across the small intestine. In addition, a small-scale study involving three participants, each undergoing six repeated SIMBA sampling sessions over a two-week period, provided preliminary evidence supporting the reproducibility of small intestinal (SI) samples obtained using this approach appears higher than that observed in matched fecal samples from the same individuals.

Then we applied a high-resolution, non-redundant genome catalog to perform in-depth, subspecies-level profiling of whole-metagenome shotgun sequencing data derived from capsule samples. Building upon recent studies highlighting the extensive natural diversity of *Bifidobacterium* in fecal microbiomes, this work expands the scope to the small intestine, addressing a key gap in probiotic-related microbial ecology.

Our preliminary results reveal distinct patterns in the diversity and prevalence of probiotic-associated taxa. Well-known probiotic species such as *Limosilactobacillus reuteri* were detected with both high diversity and prevalence. Other taxa, including *Streptococcus thermophilus*, exhibited high prevalence but limited diversity. In contrast, species such as *Lactocaseibacillus paracasei*, *Lactocaseibacillus casei*, and *Limosilactobacillus fermentum* showed both low diversity and low prevalence, suggesting a more niche distribution within the small intestinal environment.

Collectively, this analysis provides a comprehensive view of probiotic-relevant taxa in the small intestine. These findings extend beyond microbial cataloging by informing the ecological fitness, persistence, and niche specificity of probiotic strains in their target site of action, thereby supporting more rational design, validation, and personalization of probiotic interventions. To date, a series of biotic interventions are underway (<https://clinicaltrials.gov/search?term=SIMBA%20Capsule>), including probiotics, antibiotics,

prebiotics, FMT, dietary lipids, proteins, etc.

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Poster 50 SFA

Functional and Safety Characterization of Microbes from Traditional Nepali Fermented Foods: Insights from the KiKhaIK Project

Background: Traditional Nepali fermented foods are central to household diets and sustainable food systems and may represent reservoirs of functionally relevant microbes. The Himalayan Fermented Food Microbiome Project (KiKhaIK) was initiated to systematically characterize cultured isolates through integrated genomic and phenotypic analyses, with emphasis on functional potential and safety.

Methods: We cultured 363 isolates from 35 fermented foods spanning vegetable-, cereal-, and dairy-based substrates. Of these, 231 yielded genomic data and 209 high-quality genomes (>99% completeness, <5% contamination). Re-analysis of failed assemblies recovered additional high-quality MAGs, resulting in 210 genomes. Dereplication at 99.99% ANI identified 96 nonredundant strains representing 22 species across eight genera, including *Lactococcus*, *Lactiplantibacillus*, *Limosilactobacillus*, *Weissella*, *Pediococcus*, *Enterococcus*, and *Bacillus/Priestia*. Genomes were screened for GABA and B-vitamin biosynthesis, bacteriocins, exopolysaccharide and biosurfactant pathways, stress tolerance genes, antibiotic resistance genes (ARGs; CARD/RGI), and mobile genetic elements (mobileOG-db). Phenotypic assays evaluated acid, bile, and salt tolerance, bile salt hydrolase (BSH) activity, and antimicrobial activity against *Staphylococcus aureus* (including MRSA) and *Escherichia coli*. Genotype–phenotype associations were assessed.

Results: Acid and bile tolerance were broadly conserved and significantly associated with oxidative stress (*katA*, *sodA*) and bile salt hydrolase (*bsh/cbh*) genes. Complete GABA pathways were concentrated in *Limosilactobacillus fermentum* and *Lactiplantibacillus brevis*. *Bacillus* spp. exhibited greater metabolic versatility, including enrichment of complete vitamin B1 (thiamine) and B2 (riboflavin) biosynthesis pathways, whereas B9 and B12 were rare across taxa. Lactic acid production positively correlated with anti-MRSA ($p=0.51$, $p<0.001$) and anti-*E. coli* activity ($p=0.44$, $p<0.001$). Pln and Lcn bacteriocins were positively associated with antimicrobial activity. Across 96 genomes, 755 ARG hits were detected; resistance profiles were largely species-specific (PERMANOVA $R^2\approx 0.90$, $p<0.05$). LAB harbored few ARGs with fair genotype–phenotype concordance, whereas *Bacillus* and Gram-negative isolates carried more ARGs and mobile elements, indicating higher mobilization potential.

Conclusion: Traditional Nepali fermented foods harbor multifunctional microbes with conserved gut survival traits and largely favorable safety profiles, particularly among LAB. KiKhaIK establishes a curated genomic and biobank resource (>1,500 samples) to advance mechanistic microbiome research and probiotic development locally and globally.

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Poster 51 SFA

Initiating a Longitudinal Healthy Donor Biobank of Comprehensive Regional Microbiota to Advance Personalized Microbiota Transplantation Therapies

Fecal microbiota transplantation, or microbiota transplant therapy (MTT), has demonstrated substantial efficacy in recurrent *Clostridioides difficile* infection (rCDI), with over 90% clinical success rates. These outcomes have garnered interest in expanding MTT to conditions beyond rCDI such as inflammatory bowel disease (IBD). Reported efficacy in IBD appears to depend strongly on donor effects, described as “super-donors”, suggesting that therapeutic benefit hinges on specific microbial features that enable durable engraftment and immunomodulatory function in recipients. Donor microbial diversity, community stability, and functional capacity to produce short chain fatty acid (SCFA) have been implicated as contributors to MTT response. However, determinants of donor superiority, recipient compatibility, and sustained community establishment remain poorly defined, limiting the ability to select donors rationally and to design consistent, scalable interventions. To address this, we are building a program to longitudinally evaluate healthy donor microbiota using metagenomic sequencing and metabolomic approaches to optimize donor selection. The program will screen >100 healthy individuals, collecting detailed clinical, dietary, and lifestyle metadata alongside biospecimens. Donor enrollment will involve meeting extensive inclusion criteria for clinical safety, including rigorous medical and laboratory screening as well as defined dietary challenges to identify health associated metabolite production. Individuals meeting study criteria and demonstrating high microbial diversity and favorable functional profiles will be enrolled in a one-year longitudinal cohort. Donors will undergo routine bookend clinical reassessment to confirm continued eligibility, while providing weekly stool donations and small-intestinal capsule samples paired with dietary and health data. Integrated metagenomic and metabolomic profiling will enable enterotype profiling, and longitudinal evaluation of microbial stability and functional output associated with “super-donor” status and sustained engraftment potential. This initiative will generate a deeply characterized longitudinal dataset capturing healthy donor phenotypes of both large and small intestinal microbial communities. We then plan to carry out MTT clinical trials in IBD to quantify the therapeutic potential of donors. Further, we will measure strain engraftment events that correlate with clinical success and isolate response-associated strains to generate Live Biotherapeutic Products and next-generation probiotic strains. These data will provide the critical infrastructure to advance personalized microbiota therapeutics in IBD and related diseases.

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Poster 52 SFA

The Effect of Probiotics on the Growth and Toxigenic Expression of Emetic *Bacillus cereus*

Introduction: Cereulide, a potent cyclic dodecadepsipeptide which is produced by emetic *Bacillus cereus*, poses a critical challenge to food safety due to its extreme resistance to heat, low pH and proteolysis. This study investigated the antagonistic potential of probiotic strains- *Bifidobacterium bifidum* *Lactobacillus casei*, *Lactobacillus plantarum*, and in consortium-against the growth and toxigenic activity of *B. cereus*.

Methodology: The research employed a multi-phasic approach. *B. cereus* was grown in monoculture and co-culture with probiotics in broth. Growth dynamics were monitored over 72 hours using the Standard Plate count Technique on selective *Bacillus* agar. Cereulide levels were quantified via LC-MS at 24 and 48 hours. Molecular mechanisms were analyzed using RT-qPCR to measure the transcriptional response of the *cesA* and *cesB* genes, utilizing the $2^{-\Delta\Delta Ct}$ method for fold-change calculation.

Results: Data revealed a significant impact on growth rates; while monoculture *B. cereus* peaked at 7.55 ± 0.61 log CFU/mL, co-cultivation with *L. casei* significantly suppressed proliferation ($p=0.0035$), demonstrating effective niche exclusion. LC-MS analysis showed that while control cereulide reached 72.89 µg/kg, co-culture with *L. casei* and the probiotic consortium reduced toxin levels to 55.62 µg/kg at 48h ($p=0.00036$). but in presence of *B. bifidum* the growth of *B. cereus* and cereulide expression was increased.

Molecular Insights: Gene expression analysis confirmed a profound silencing of the cereulide synthetase operon. *cesB* expression was drastically reduced to just 0.19% of the control at 24h (fold change = 0.00194) and remained suppressed at 0.97% at 48h. *cesA* expression followed a similar downward trend.

Conclusion: These findings conclude that specific probiotics, including in consortium, does not merely inhibit growth but actively downregulate the genetic machinery of toxin production, offering a promising "clean-label" strategy for enhancing food safety.

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Poster 75 SFA

Phagebiotics as a Potential Member of the Biotics Family

The "biotics family" currently includes prebiotics, probiotics, synbiotics and postbiotics. These can be generally defined as a group of interventions involving microbes that confer a health benefit on the host. Bacteriophages (phages) are viruses that specifically infect bacteria, are the most abundant biological entities on Earth and are integral components of the human microbiome. Here, we propose that phages could be considered a new member of the biotics family. Phage-based treatments offer highly targeted therapies that can have benefits ranging from infection control to impacts on the gut-brain axis and neuromodulation. The term "phagebiotics" has appeared inconsistently in academic and non-academic sources, highlighting the absence of a shared definition. We suggest that the term "phagebiotics" would be reserved for 'preparations of characterised viable bacteriophage that confer a health benefit on the host'. This new term could harmonise communication between academia, industry, regulators and consumers.

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Poster 76 SFA**Microbiome of captive and endangered animals as a platform for antimicrobial discovery**

The global antimicrobial resistance crisis highlights the need for novel alternatives to antibiotics. Bacteriocins are ribosomally synthesised antimicrobial peptides and proteins that mediate microbial competition and are increasingly being explored as potential antimicrobial therapeutics. Here, we investigated the faecal microbiomes of 40 captive zoo animals, the majority representing vulnerable or endangered species, using complementary culture-based and in silico approaches to assess their potential as a reservoir of bacteriocins. We isolated 92 distinct bacterial strains with antimicrobial activity from 34 animals, with activity most frequently observed against the foodborne pathogens *Clostridium perfringens* and *Listeria monocytogenes*. Using genome-resolved metagenomics, we identified 569 novel species-level genome bins (SGBs), highlighting endangered animals as reservoirs of previously undescribed microbial diversity that may also be lost as host biodiversity declines. Across 1,085 metagenome-assembled genomes (MAGs), we predicted 1,787 biosynthetic gene clusters (BGCs), the majority of which were novel including a novel *Roseburia* species recovered from a François' langur monkey encoded a novel nisin variant. Our findings highlight understudied captive and endangered animal microbiomes as a valuable reservoir of novel bacterial and biosynthetic diversity for bacteriocin discovery, while demonstrating the importance of combining culture-based and in silico approaches, as culturing alone misses a substantial proportion of bacteriocin BGCs.

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Theme 7: Prebiotics & microbiota modulation

Poster 53 SFA**Structural Determinants of Microbial Glycans Modulate Gut Microbial Ecology and Metabolic Output In vitro**

Microbial glycans are desirable polymers for use in foods, due to their rheological and physicochemical properties, novel functionality and compatibility with human digestion and environmental processes. These glycans are considered as promising candidates for prebiotics through fermentation by specific populations of gut microbiota. Here, we hypothesized that microbial glycans with different glycosidic linkages and varying degree of polymerization would be fermented differently by gut microbiota of the same individuals, leading to alterations in gut microbiota community structure and function. To test this hypothesis, we isolated microbes from different environments capable of producing glycans (fructans & glucans) and structurally characterized them. Glycans were produced using sucrose as a substrate in different media and purified through ethanol precipitations of the supernatant. These glycans were then treated via in vitro digestion with salivary α -amylase, pepsin and pancreatin, simulating passage through the upper gastrointestinal tract. We then performed an in vitro fermentation assay in which glycans from 10 different microbes were inoculated with fecal microbiota of healthy donors. Samples were collected at different time points (0, 4, 8, 12 & 24) and gas production was also measured at each time point. From these samples, 16S rRNA gene sequencing and SCFAs analysis were performed. SCFA analysis revealed that different microbial glycans structures led to differential metabolite production by otherwise-identical gut microbiota. Microbial community analysis by 16S rRNA gene sequencing revealed that these metabolic outcomes were

accompanied by the development of divergent microbial community structures. All fructan molecules provided higher amount of butyrate but less amount of acetate and propionate than slow-fermenting glucans. All the glycans also showed a beneficial effect on overall microbial community structures that highly depends on donor's gut microbiota compositions. These data suggest that variation in microbial glycan structure exerts significant impacts on metabolic function through alteration in microbial community structure. These findings contribute to the growing evidence base on structure-function relationships of prebiotics and provide an understanding that how specific glycan structure, linkage patterns and degree of polymerization differentially shape microbial ecology and metabolite production. This study advances us towards design of next-generation synbiotics and microbiome-targeted nutritional strategies for diverse populations.

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Poster 54 SFA

7-KLCA: A Promising Bile Acid in a New Wave of Prebiotic Therapeutics

Background: The ageing process is linked to alterations in the gut microbiota, reductions in beneficial microbial metabolites, and dysregulated metabolic and immune signalling. Bile acids have emerged as potential modulators of host-microbe interactions. This study investigates the effects of 7-ketolithocholic acid (7-KLCA), a secondary bile acid, on gut microbiota composition, microbial metabolism, and host gene expression, to evaluate its potential as a bileotic/prebiotic therapeutic for promoting healthy ageing.

Methods

A validated three-stage continuous in vitro fermentation model, simulating the human colon, was inoculated with faecal samples from healthy adult donors. 7-KLCA was administered daily during the second steady state. Microbial composition was assessed using FISH-flow cytometry and 16S rRNA gene sequencing. Metabolite analysis included short-chain fatty acids (SCFAs) measured by gas chromatography and bile acid profiling by UPLC-MRM-MS. Cell culture studies, using intestinal and hepatic cell lines, were used to evaluate the effect of 7-KLCA on gene expression to gain insights into more detailed cellular and molecular mechanisms, and to assess any potential off-target effects of this bile acid.

Findings: Treatment with 7-KLCA led to increased abundance of key butyrate-producing bacteria, including *Faecalibacterium prausnitzii*, *Eubacterium rectale*, and *Roseburia* spp. This was accompanied by increased SCFA production. Bile acid profiling revealed microbial transformation of 7-KLCA to ursodeoxycholic acid (UDCA), with region-specific changes in microbial composition and metabolite profiles across the colon model. Gene expression analysis indicated that 7-KLCA modulated pathways associated with metabolic and immune regulation, consistent with a healthy ageing phenotype.

Interpretation: 7-KLCA may exert prebiotic effects by reshaping the gut microbiota, enhancing beneficial microbial metabolites, and modulating host gene expression. These findings support its potential as a novel nutraceutical agent to promote healthy ageing and maintain metabolic homeostasis. A placebo-controlled parallel human intervention study is currently investigating these effects and potential benefits in a healthy adult population.

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Poster 55 SFA

Human milk oligosaccharides reshape the gut microbiome and metabolome in irritable bowel syndrome: a randomised, double-blind, placebo-controlled crossover trial

Background: Human milk oligosaccharides (HMOs) are prebiotic glycans with established roles in shaping the infant gut microbiota and supporting host health. Their potential as dietary interventions for adults with irritable bowel syndrome (IBS) remains underexplored.

Objective: To evaluate the effects of a 3'-sialyllactose/lacto-N-tetraose (3'SL/LNT) blend on IBS symptoms as the primary outcome, and on secondary outcomes including gut microbiota composition and metabolic activities, host metabolism, sleep quality, anxiety, and depression in adults with Rome IV-diagnosed IBS.

Methods: The GUT-HEAL-IBS study was a 16-week randomised, double-blind, placebo-controlled, two-by-two crossover trial (NCT06281600). Forty-two adults received 5 g/day of HMO mix (A) and placebo (maltodextrin, B) in randomised order. Primary outcome was change in IBS Symptom Severity Score (IBS-SSS). Secondary outcomes included anxiety, depression, sleep, gut microbial composition (16S rRNA sequencing, FISH), and urine and faecal metabolomic profiles (1H-NMR spectroscopy, GC-MS).

Results: Treatment B (placebo) significantly reduced IBS-SSS ($P = 0.00055$), while Treatment A (HMO) did not significantly alter symptom severity ($P = 0.543$). Adverse events were mild and transient. Despite the absence of symptomatic benefit beyond placebo, HMOs induced reproducible biological changes, including increases in *Bifidobacterium* and *Faecalibacterium*, and metabolomics shifts away from proteolytic/aromatic amino-acid fermentation. No significant microbial or metabolic changes were associated with placebo.

Conclusions: A 3'SL/LNT HMO blend was safe, well-tolerated, and biologically active, modulating gut microbiota and metabolism in IBS adults. However, symptom improvements were not greater than placebo, highlighting the challenge of high placebo responsiveness in IBS trials. Larger and longer studies are warranted to establish whether microbiome modulation by HMOs can translate into consistent clinical benefit.

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Poster 56 IAC

A lifestyle intervention with galacto-oligosaccharide supplementation selectively increases gut *Bifidobacterium* in health-compromised older adults

Background: Age-related health complaints are often associated with declines in skeletal muscle health, immune function, and metabolic regulation. Lifestyle factors, particularly physical activity and nutrition, influence these processes. Emerging evidence also suggests a role for the gut microbiome in age-related dysfunction, but its responsiveness to multidomain lifestyle interventions in health-compromised older adults remains incompletely understood. The VOILA study is a multidimensional intervention shown to improve muscle health and physical functioning (1). This abstract evaluates its effects on the gut microbiome.

Method: This study evaluated the impact of a 12-week lifestyle intervention (VOILA) on gut microbiota in health-compromised older adults (>60 years, n=94), including individuals with impaired mobility, compromised metabolic health, or recovering from total knee replacement. The intervention included supervised high-intensity resistance training three times per week, nutritional counselling, and morning and evening supplementation. The morning supplement was taken with breakfast and provided 25 g whey protein (Nutri Whey™ Isolate), 5.5 g Vivinal® GOS, 800 IU vitamin D, and 250 mg calcium. The evening supplement was consumed between dinner and bedtime and provided 20 g whey protein and 116 mg calcium (2). Gut microbiota composition from fecal samples collected at baseline and post-intervention was assessed using shotgun metagenomic sequencing, and taxonomically profiled with MetaPhlAn4. Differential abundance was assessed using MaAsLin3.

Results: After excluding participants who were using antibiotics at the time of fecal sample collection, 90 participants were included in the microbiota analysis. At baseline, no differences in overall microbial community composition (beta-diversity) were observed (PERMANOVA, multiple distance metrics). The intervention did not significantly affect alpha diversity (Shannon index, observed species, Berger-Parker dominance, Faith's phylogenetic diversity). However, a significant increase in relative abundance of *Bifidobacterium* was observed ($p<0.001$), primarily driven by *Bifidobacterium longum* ($p<0.001$) and *Bifidobacterium adolescentis* ($p<0.01$). No other genera showed significant changes after multiple testing correction.

Conclusion: A 12-week lifestyle intervention combining resistance training, nutritional counseling, and prebiotic GOS-containing supplementation selectively increased beneficial *Bifidobacterium* species in health-compromised older adults. These microbiome changes occurred alongside improvements in physiological and functional outcomes assessed within the VOILA study.

Clinical trial registration: clinicaltrials.gov, identifier NCT05762965.

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Poster 57 IAC

***Bifidobacterium animalis* subsp. *lactis* BPL1® laxative effects in loperamide-induced constipated sprague dawley rats**

Introduction: Constipation is a prevalent gastrointestinal state affecting around 12% of global population. Given its multifactorial nature, a comprehensive investigation into the underlying mechanisms is essential to identify more effective and safer interventions. Probiotics have emerged as a promising option given their non-invasive nature. This research not only evaluated the physiological effect of probiotic *Bifidobacterium animalis* subsp. *lactis* CECT 8145 (BPL1®) in a loperamide-induced constipation murine

model, a metagenomic analysis was also conducted to understand its ecological impact on the gut microbiota.

Material & Methods: This research was divided in two groups, control and loperamide-induced constipation. Both groups received a 3-day intervention with placebo or probiotic BPL1® with two doses ($1.5 \cdot 10^8$ CFU (low) and $3 \cdot 10^9$ CFU (high)). The study assessed several parameters to determine the probiotic's effect, including: gene expression and microbiota composition.

Results: While loperamide significantly decreased stool number, weight and humidity, BPL1® supplementation effectively restored these parameters, being more pronounced at a high dose. Microbiome analysis showed that BPL1® at a low dose reduced the abundance of *Muribaculaceae* and *Muribaculum gordoncarteri*, associated with constipation. In addition, *Muribaculaceae* abundance was negatively correlated with stool humidity. Functional microbiome profiling indicated that BPL1® suppressed pathways related to mucin degradation, vancomycin resistance and isoleucine biosynthesis while promoting L-lactate and pyridoxal-P (vitamin B6) biosynthesis, which may support gut motility and barrier integrity.

Conclusion: The study findings suggest that the BPL1® exhibits potential as a functional probiotic for relieving constipation through improving stool excretion and consistency, inducing taxonomic changes and beneficial functional modulation of the intestinal microbiome. These findings justify further investigation into the mechanisms of BPL1® as a probiotic for constipation management.

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Poster 58 IAC

Classifying ingredients as prebiotics: a case study for the resistant dextrins

Following 2017 scientific definition from ISAPP where it was stated that a prebiotic is a substrate that selectively utilized by host microorganisms conferring a health benefit, Hutkins et al. recently published recommendations gathering the scientific perspectives that should be examined before considering an ingredient or a compound as a prebiotic. In the present poster, we would like to develop the case study of resistant dextrins (RD). The first criteria of the definition implies a substrate that is identified and isolated from a diet – considering RD as a supplement in many placebo-controlled clinical trials, it is easy to set apart its own effects on gut microbiota a health benefits. The second criteria involves the identification and characterization of the compound before and after food processing when it is incorporated into a food matrix: RD structure and chain-length were not altered neither by low pH nor by high temperature that are commonly used in some food processes such as fruit compotes. The resistance to gastrointestinal conditions (low pH and human enzymes) did not impact neither the fiber content of RD which is not the case for all soluble fibers. The selective use of RD by gut microbiota was demonstrated several times through ex vivo experiments and confirmed in clinical trials with a main impact on Parabacteroides genus

and more especially on *P. distasonis* and *P. merdae*. Some RD were able to demonstrate a modulation of the gut microbiota composition and function simultaneously with a long-term impact on blood glucose homeostasis and satiety. Through repeated clinical investigations on different populations and confirmation using *in vitro* and *ex vivo* tools, some mechanisms of action linking gut microbiota modulation, metabolites production and their impact of blood glucose regulation were highlighted for RD. The complementary *in vitro* tools such as *ex vivo* colonic fermentations and metabolomics were useful to identify cross feeding pathways within gut bacteria explaining the trophic networks involved in RD degradation. In addition, after many years of presence on the market, RD are considered as safe and well tolerated, even in sensitive populations such as IBS. At least, the demonstration of the prebiotic properties of RD also involves dose and duration that are compatible with everyone's diet as it can fit in many serving sizes thanks to its high solubility and low viscosity.

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Poster 59 Symposium

Difructose Anhydride III Selectively Enriches *Blautia* and Complements Raffinose and Fructo-oligosaccharides to Promote Concurrent Enrichment of *Blautia* and *Bifidobacterium* in an *In Vitro* Human Fecal Fermentation Model

Background and Objective: Difructose anhydride III (DFA III) is a non-digestible oligosaccharide composed of two fructose molecules linked by two anhydride bonds. While raffinose (RAF) and fructo-oligosaccharides (FOS) are well known to selectively stimulate *Bifidobacterium* growth in clinical trials, DFA III has been reported to be utilized primarily by *Blautia* species. Recent studies suggest that *Blautia* plays a beneficial role in host metabolic health. This study investigated the fermentation characteristics of DFA III alone and in combination with RAF and FOS using an *in vitro* human fecal fermentation model.

Methods: Fecal samples from 10 healthy adult males living in Japan were individually cultured anaerobically for 48 h using cellulose (CEL), RAF, FOS, DFA III, RAF+FOS (1:1), RAF+DFA III (1:1), FOS+DFA III (1:1), or RAF+FOS+DFA III (1:1:1) as carbon sources. The culture medium contained 2% (v/v) fecal inoculum, 3% (v/v) carbon source, and 0.8% (v/v) nitrogen source. Anaerobic incubation was performed at 37 °C for 48 h under a mixed nitrogen-carbon dioxide atmosphere, with the pH maintained above a lower limit of 6.5. Microbial composition was analyzed at 24 and 48 h using 16S rRNA gene amplicon sequencing, and metabolites, including short-chain fatty acids (SCFAs), were measured at 0, 6, 12, 24, and 48 h by HPLC.

Results: Among single-substrate treatments, RAF and FOS significantly increased the relative abundance of *Bifidobacterium* compared with CEL at 24 and 48 h. In contrast, DFA III significantly increased *Blautia* abundance compared with CEL, RAF, and FOS. Lactate production, a characteristic metabolite of *Bifidobacterium*, was observed in RAF and FOS but not in DFA III. Total SCFAs concentrations at 48 h did not differ significantly among RAF, FOS, and DFA III treatments; however, total SCFAs production at 24 h was higher in RAF and FOS than in DFA III, suggesting slower fermentation of DFA III by gut microbiota. In mixed-substrate treatments, RAF+DFA III, FOS+DFA III, and RAF+FOS+DFA III significantly increased *Bifidobacterium* abundance compared with DFA III alone. Notably, RAF+FOS+DFA III significantly increased the abundances of both *Bifidobacterium* and *Blautia* compared with CEL. Although oligosaccharide

mixtures did not improve α -diversity or SCFAs production, the results suggest that *Blautia* and *Bifidobacterium* can coexist without mutual inhibition under combined oligosaccharide supplementation *in vitro*.

Conclusion: Combined supplementation with DFA III, RAF, and FOS simultaneously promoted *Blautia* and *Bifidobacterium*, suggesting a potential strategy for selectively enriching multiple beneficial bacterial taxa in the human gut.

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Poster 60 Symposium

Distinct effects of different dietary fibers and sugars on gut microbiomes of young and older adults in an in-vitro simulated gut model

Gut microbiome has high inter-individual variability, but how this variance differently regulates dietary carbohydrates metabolism remains unclear. Herein, we evaluated age-specific responses of the human gut microbiome to simple sugars and dietary fibers using a simulated in-vitro fecal fermentation model. Fecal samples were obtained from healthy young adults (N=12; 26.42 $\pm$ 3.98 years) and older adults (N=5; 64.20 $\pm$ 9.65 years). A basal control medium (Ct) was supplemented (5% w/v) with 13 carbohydrates, including monosaccharides (glucose, fructose), disaccharides (lactose, sucrose), oligosaccharides (fructooligosaccharides [FOS], galactooligosaccharides [GOS], human milk oligosaccharides [HMO]), soluble fibers/starch (inulin, potato starch), and resistant starches extracted from black-eyed peas (RS1), pinto beans (RS2), lentils (RS3), and chickpeas (RS4). Fresh fecal suspensions (40 μ m filtered), prepared within 30 minutes of defecation under anaerobic conditions, were inoculated (2% v/v) into treatment and control anaerobic media and incubated at 37°C for 24 hours. Metagenomic DNA was extracted and sequenced for 16S rRNA gene on an Illumina MiSeq platform. At baseline, younger subjects exhibited higher abundance of *Bifidobacterium* and *Prevotella*, whereas older subjects had higher *Bacteroides*. Compared to control group, α -diversity (Chao1 and Shannon indices) significantly decreased across treatments in both age groups, with statistically stronger effect in young cohort ($p < 0.001$). Beta-diversity analysis (Bray-Curtis PCoA) showed distinct clusters between control vs. carbohydrate-treated samples in young subjects ($p < 0.001$), but not in older subjects (likely due to smaller sample size). Across cohorts, most carbohydrates increased the abundance of Actinobacteriota/*Bifidobacterium* and Bacteroidota/*Prevotella*, while reducing Firmicutes particularly an unclassified taxa from Lachnospiraceae family, compared to control. LEfSe analysis identified discriminatory taxa enriched by specific substrates: *Bifidobacterium* (all treatments); *Faecalibacterium* (FOS, HMO, inulin, potato starch, RS1–RS4); *f_Lactobacillaceae;g_* (mono-, di-, and oligosaccharides); *Leuconostoc* (RS1, RS3); *Exiguobacterium* (RS2, RS4); and *Paenibacillus* and *Stenotrophomonas* (RS3). These findings demonstrate distinct host microbial responses to simple and complex carbohydrates and highlight age-dependent differences in microbiome modulation. This work supports the identification of targeted prebiotic strategies for precision microbiome interventions across the lifespan.

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Poster 61 IAC**Impact of a synbiotic combination of resistant dextrin and *Lactcaseibacillus paracasei* strain LPCR on gut microbiota composition, metabolic activity and intestinal epithelial responses in a SHIME® model**

Synbiotics are a promising strategy to modulate the gut microbiota and support host health by enhancing the growth, activity, and interactions of beneficial microorganisms. This study evaluated the effects of a synbiotic combination of resistant dextrin and *Lactcaseibacillus paracasei* strain LPCR on gut microbiota composition, microbial metabolism, and intestinal epithelial responses using a dynamic SHIME® (Simulator of the Human Intestinal Microbial Ecosystem) model. A semi-continuous SHIME® system simulating the ascending colon was inoculated with fecal microbiota from six healthy donors and maintained under controlled anaerobic conditions, temperature, retention time, and pH to closely mimic the colonic environment. Four conditions were tested in parallel: control, resistant dextrin (prebiotic), *Lactcaseibacillus paracasei* strain LPCR (probiotic), and their synbiotic combination, allowing a direct assessment of synergistic effects compared to individual components. After stabilization, formulations were administered daily for two weeks. Samples were collected from luminal and mucosal compartments to capture microbial communities across different gut niches. Microbiota composition was analyzed via 16S rRNA gene sequencing, while metabolic activity was assessed through short-chain fatty acids (SCFAs) and bile acids profiling. To investigate host-relevant functional effects, SHIME® supernatants were applied to human intestinal organoids, enabling the assessment of epithelial responses, including barrier integrity, mucus production, antimicrobial peptides, and inflammatory pathways. After two weeks, compartment-specific shifts in alpha diversity emerged. In the lumen, diversity significantly increased with the probiotic but significantly decreased with the prebiotic, a drop more pronounced than in the control, whereas the synbiotic maintained stable diversity, suggesting a compensatory effect. In the mucin, diversity remained stable except for a significant increase induced by the probiotic. In terms of specific taxonomic shifts, the prebiotic enriched luminal *Alistipes* and *Parabacteroides* compared to the control, and mucosal *Bifidobacterium*. The synbiotic increased luminal *Anaerostipes* and mucosal *Enterococcus* compared to the prebiotic. Conversely, the probiotic only reduced the potential luminal pathobionts *Enterobacter* and *Sutterella*. Metabolically, both the prebiotic and synbiotic increased acetate from day one. The synbiotic accelerated the propionate rise, which peaked at day two compared to day four for the prebiotic. Butyrate remained unchanged. Bile acids and organoid analyses are ongoing. This study highlights the value of combining complementary in vitro models, including SHIME® and human intestinal organoids, with multi-dimensional analyses to optimize next-generation synbiotic strategies, enabling targeted modulation of gut microbiota, microbial metabolism, and epithelial health to improve host outcomes.

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Poster 62 IAC**Prebiotic Chicory Fructooligosaccharides for Bowel Regularity: A Systematic Review of Clinical Evidence**

Background: Dietary fiber plays a central role in supporting normal bowel function, yet habitual intakes remain below recommended levels worldwide, making the enrichment of foods with functional and isolated fiber sources a practical and effective approach. Chicory-derived oligofructose (synonym fructooligosaccharides; FOS) are well-established dietary fibers and prebiotics that selectively stimulate beneficial gut microbiota and have been associated with improvements in stool frequency and consistency.

Their effects are mediated through fermentation in the colon, resulting in the production of short-chain fatty acids (SCFAs), which influence intestinal motility, epithelial function, and fluid balance via microbiota–host signaling pathways. Recent human data show that fermentation of FOS occurs primarily in the colon, alongside rapid shifts in microbiota composition. The resulting SCFAs are largely absorbed and metabolized by the host, linking microbial activity directly to host metabolic processes.

Objective: The aim of this review is to evaluate the effects of chicory FOS on bowel habit in healthy and/or constipated adults aged 18 years and older.

Methods: PubMed and Scopus were searched from inception to March 2026, supplemented by reference screening and grey literature searches. Randomized controlled trials of at least one week in adults (≥ 18 years) evaluating chicory-derived FOS versus placebo or usual diet and reporting stool frequency or stool consistency were included. Study selection, data extraction, and risk of bias assessment were performed independently by two reviewers.

Results: Eight randomized, placebo-controlled trials ($n = 471$ healthy adults) published between 1996 and 2024 were included. Intervention durations ranged from 2 to 12 weeks and FOS doses from 2.5 to 30 g/day. Five of eight studies reported a statistically significant increase in stool frequency with FOS compared with control or baseline. Reported mean differences ranged from 0.3–1.2 bowel movements/day. Stool consistency was assessed in seven studies, with largely no significant effect; one study reported significantly softer stools with 15 g/day.

Conclusion: Chicory FOS improves bowel regularity in healthy adults, with consistent increases in stool frequency observed across a broad dose range, including low doses. Mechanistically, these effects are supported by rapid colonic fermentation, gut microbiota modulation, and efficient SCFA absorption, providing a direct link between prebiotic intake, microbial activity, and host physiology. Overall, the evidence highlights the role of chicory FOS as a functional ingredient to support bowel regularity, with effects that are consistent with its established prebiotic mode of action.

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Poster 63 SFA

Pure $\beta(1-4)$ Galacto-oligosaccharides Enable the Persistence of a Rationally Designed Consortium in the Intestinal Tract

A major limitation of probiotic therapies is limited persistence in the gut, which reduces long-term efficacy. Building on prior research, we hypothesized that a galacto-oligosaccharide (GOS) diet would ecologically prime the gut environment to support persistence of a probiotic-commensal consortium assembled using an ecology-driven bioreactor approach. We evaluated a 16-strain rationally designed consortium (RDC-L1), assembled in packed bioreactors, in a mouse model to assess how a GOS diet affects microbial persistence. Forty-eight female C57BL/6J SPF mice were fed a defined diet for two weeks, then switched to a GOS-supplemented diet, or continued the defined (control) diet. At that time, mice received RDC-L1, and persistence was monitored for one week (time point 1, TP1). They then received antibiotics cocktails, after

which RDC-L1 was reintroduced to assess recolonization (TP2). Finally, mice were challenged with *Clostridioides difficile* spores, received a third RDC-L1 dose, and were monitored for 7 days to assess pathogen clearance and RDC-L1 persistence (TP3).

16S rRNA sequencing data from stool samples collected at TP1 revealed increased persistence of RDC-L1 *Bifidobacterium*, *Lactobacillus*, and *Lactococcus* species under the GOS diet. To achieve strain-level resolution, we designed 90 strain-specific probes based on target sequences of RDC-L1 strains. This approach had a high specificity, with 75-90% of reads aligning to probe-targeted regions compared with 10-25% in unenriched samples.

Probe-enriched WGS sequencing revealed that the GOS diet increased the persistence of the RDC-L1 strains at TP1, including three *Bifidobacterium* strains. At TP2, 15 of the 16 RDC-L1 strains successfully recolonized both diet groups. At TP3, following the *C. difficile* challenge, *Parabacteroides johnsonii* dominated the gut community (~30% relative abundance) in RDC-L1-treated mice. GOS supplementation further increased the persistence of *B. longum* and *B. breve* after *C. difficile* challenge.

These findings indicate that GOS-mediated ecological priming reshaped the gut environment to support colonization and persistence of the bioreactor-assembled RDC-L1 consortium, particularly *Bifidobacterium* species. By reducing community diversity and opening niches, GOS enhanced strain persistence across baseline, post-antibiotic recolonization, and *C. difficile* challenge. This ecology-guided strategy, dietary niche conditioning followed by targeted consortium delivery, offers a rational framework for improving the durability of microbiome-based therapeutics.

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Poster 64 SFA

Short-term mixed-fiber supplementation associates with selective mucosal microbial shifts and alterations in mucosal metabolism

Background: Dietary fiber is a key modulator of colorectal health, yet ~95% of Americans do not meet the recommended adequate intake. Over-the-counter fiber supplements are recommended in routine anorectal care to improve stool form and reduce straining. Mixed-fiber formulations combine physicochemical properties that may shape the rectal mucosal environment through coordinated effects on microbiota composition, SCFA handling, and host transcriptional programs. We aimed to characterize multi-compartment changes associated with a pragmatic mixed-fiber add-on.

Methods: In a 28-day clinical intervention, participants added 15g/day mixed fibers (7.5g psyllium husk, 8g wheat dextrin) to their habitual diet. Rectal mucosal biopsies and serum were collected at baseline (PRE) and post-intervention (POST). Final analyses included participants whose POST biopsy occurred within 7 days of the final dose (n=37). The microbiota was profiled by 16S rRNA gene sequencing. Targeted and untargeted metabolomics were quantified in tissue and serum by GC-MS and LC-MS. Host mucosal RNA-seq was performed in a paired subset (n=10), with pathway analysis assessed using KEGG gene set enrichment analysis (GSEA).

Results: The fiber supplementation did not significantly alter overall microbiota composition, while PRE host and dietary factors explained substantial compositional variance. Although global structure was stable, 10/302 mucosal amplicon sequence variants (ASV) were significantly associated with the fiber supplementation, including increases in *Parabacteroides distasonis* and *Lachnospiraceae* NK4A136 group. In rectal tissue, propionate and isobutyrate decreased while butyrate and acetate were unchanged. In serum, acetate and propionate did not change, while butyrate showed a decrease. RNA-seq identified 177 differentially expressed genes. KEGG GSEA indicated POST enrichment of metabolic/mitochondrial pathways (carbon metabolism, oxidative phosphorylation, TCA cycle) and SCFA-related pathways, while PRE-enriched pathways were dominated by immune-related signaling.

Conclusion: Short-term mixed fiber supplementation was associated with selective mucosal taxonomic shifts and enrichment of rectal mucosal metabolic programs, alongside changes in circulating metabolites.

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Poster 65 Symposium

Structural Basis for the Selective Promotion of *Roseburia intestinalis* by Tamarind Seed Gum Oligosaccharides

Tamarind seed gum (TSG) is a plant-derived polysaccharide composed mainly of xyloglucan and is widely used in Japan as a food thickener and stabilizer. Butyrate produced by gut bacteria serves as a major energy source for colonic epithelial cells and exerts health-promoting effects, including anti-inflammatory effects mediated by regulatory T-cell differentiation. However, conventional prebiotics that increase intestinal butyrate concentrations often promote multiple gut bacterial species and increase several short-chain fatty acids simultaneously. Thus, few dietary materials selectively promote a specific butyrate-producing bacterium and preferentially increase butyrate production.

TSG comprises hepta-, octa-, and nonasaccharide units with a glucose tetrasaccharide backbone, three xylose side chains, and galactose in the latter two. The growth-promoting effects of a hepta- to nonasaccharide-rich oligosaccharide fraction from enzymatically degraded TSG (Sugar C) were first evaluated in pure cultures of 51 predominant human gut commensal bacterial species commonly found in Japanese and European individuals. Sugar C selectively promoted the butyrate-producing bacterium *Roseburia intestinalis*, whereas most other dominant gut bacterial species showed little or no growth response.

The effects of Sugar C were next examined in cocultures of *R. intestinalis* with human fecal microbiota from 10 donors. Sugar C increased the abundance of *R. intestinalis* and approximately doubled butyrate concentrations, while other major short-chain fatty acids, including acetate and propionate, remained largely unchanged.

Gnotobiotic mice were colonized with *R. intestinalis*, *Bacteroides caccae*, *Bacteroides thetaiotaomicron*, *Coprococcus comes*, *Collinsella aerofaciens*, and *Ruminococcus torques*. In mice fed a Sugar C-containing low-fiber diet, relative abundances of the other five species decreased or increased by at most 1.1-fold versus the control diet, whereas that of *R. intestinalis* increased 15.8-fold. Cecal butyrate increased 4.9-fold, whereas acetate and propionate remained unchanged. Sugar C preferentially increased intestinal butyrate by selectively promoting *R. intestinalis*.

To investigate the basis of this species selectivity, we further analyzed the growth-promoting effects of TSG-derived pentasaccharides (Sugar E) and trisaccharides (Sugar F) in pure cultures of the same 51 species. As glycan chain length decreased, a greater number of bacterial species exhibited growth promotion, suggesting reduced species selectivity. Their chain length and structural complexity may restrict utilization by gut bacteria.

Taken together, these findings suggest that the hepta- to nonasaccharide chain length and structural complexity of these oligosaccharides contribute to the species-selective growth-promoting effect on *R. intestinalis*. Sugar C represents a promising candidate next-generation prebiotic that preferentially increases *R. intestinalis* and its metabolite butyrate without substantially promoting the growth of non-target gut bacteria.

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Theme 8: Pregnancy, infancy & early life

Poster 66 SFA

Breastfeeding as a Natural Biotic Strategy to Limit Early-Life Antimicrobial Resistance

Antimicrobial resistance (AMR) represents a growing global health challenge, particularly during early life, a critical developmental window in which the gut microbiota, immune system, and resistome are established in parallel. Antibiotic resistance genes (ARGs) can be rapidly acquired and stabilised in the infant gut within weeks after birth, especially following caesarean delivery or antibiotic exposure. Identifying natural, microbiome-targeted strategies to limit early-life AMR acquisition is therefore a public health priority.

Here, we present breastfeeding as a powerful biotic-driven intervention capable of mitigating AMR through interconnected prebiotic and probiotic mechanisms. Using shotgun metagenomic sequencing of over 256 infant faecal samples from MAMI longitudinal birth cohort, we demonstrate that exclusive breastfeeding during the first month of life is consistently associated with reduced ARG abundance and diversity, even among high-risk infants born by caesarean section or exposed to antibiotics. These effects were strongly linked to the selective enrichment of *Bifidobacterium* species, as infants with high bifidobacterial colonisation exhibited a significantly lower resistome burden.

Bifidobacteria are key early-life probiotics whose dominance in the infant gut is largely driven by their

capacity to metabolise human milk oligosaccharides (HMOs), a structurally diverse group of prebiotics uniquely present in human milk. In this study, maternal secretor status—determined by genotyping—and milk HMO profiles—characterised by HPLC-FL—emerged as major determinants of infant resistome outcomes. Maternal secretor status was associated with a lower infant gut ARG load and appeared to partially offset the effects of caesarean delivery. Furthermore, targeted isolation of infant-derived bifidobacteria revealed that HMO-specialist species, including *Bifidobacterium bifidum* and *Bifidobacterium longum* subsp. *infantis*, displayed increased antibiotic susceptibility, supporting a mechanistic link between prebiotic-driven microbial selection and reduced ARG persistence.

Beyond microbial ecology, human milk bioactives—specially HMOs—also contribute to immune maturation and epithelial barrier development, reinforcing colonisation resistance. Ongoing work integrates microbiome, immune, and functional readouts to determine how milk-driven immune signals influence microbial competition, pathogen exclusion, and horizontal gene transfer. Together, these findings position breastfeeding as a naturally evolved biotic system and highlight the translational potential of HMO-informed prebiotic and bifidobacteria-based probiotic strategies to limit AMR acquisition in early life.

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Poster 67 IAC

Building a Healthier Start: Maternal Probiotic Supplementation Promotes Beneficial Microbial Transfer and Infant Microbiome Establishment

It is now well established that a healthy early-life microbiome is critical for optimal development and long-term health, and that multiple perinatal factors influence the establishment, evolution, and functionality of the infant gut microbiota. Recent studies conducted in industrialized countries report a marked decline in *Bifidobacterium*, a key component of the infant microbiome. This decline has been associated with altered metabolic and immune functions, as well as increased gastrointestinal discomfort. In this context, probiotic-driven modulation of the gut microbiome has shown promising potential to support infant health. More specifically, maternal supplementation during the perinatal period may represent an effective strategy to enhance neonatal protection by promoting beneficial microbial transfer and mitigating environmental influences such as antibiotic exposure and birth by C-section.

Two recent clinical trials, a pilot study including 52 mother–infant dyads and a large randomized controlled trial involving 180 dyads, investigated the impact of maternal probiotic supplementation on infant microbiome development using metagenomic, biomolecular, and advanced bioinformatics approaches. Both studies demonstrated that maternal probiotic supplementation enhances early microbial exposure in infants through vertical transmission during delivery and breastfeeding. Strain transfer and engraftment were strain-specific, suggesting differential functional contributions of the tested probiotics.

In infants, these interventions led to increased colonization by beneficial bacteria, particularly *Bifidobacterium*, and promoted a more interconnected and structured microbial ecosystem organized around key taxa.

Overall, findings support the safety and potential of probiotic supplementation during the last trimester of pregnancy and lactation as an effective strategy to promote optimal infant microbiome establishment.

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Poster 68 IAC

Galacto-oligosaccharides promote dose-dependent gut microbial-, metabolic-, and barrier benefits in children, *ex vivo*

Background: Galacto-oligosaccharides (GOS) are well-established prebiotic fibers; however, their dose-response relationships and functional effects on the gut microbiota of children remain incompletely defined.

Methods: The dose-dependent effects of GOS (1.25, 2.5 and 5 g/day equivalents) on the gut microbiome of children from two age groups (3–6 years and 6–12 years, n = 6 per group) was evaluated using the predictive *ex vivo* SIFR® technology platform. Specifically, the impact of GOS fermentation on microbial composition, metabolite production and intestinal epithelial barrier integrity was assessed.

Results: GOS induced robust, dose-dependent bifidogenic effects and significantly increased other beneficial microbial taxa, including butyrate-producing members of the Lachnospiraceae and Ruminococcaceae families, in both age groups. These changes in microbiota composition were accompanied by increased concentrations of short chain fatty acids (SCFA: acetate, propionate, butyrate), with significant effects already evident at the lowest dose. In addition, untargeted metabolomics revealed that GOS fermentation

enhanced production of a broad range of other microbial metabolites such as indoles, N-acetylated amino acids, *p-hydroxyphenyllactic* acid, agmatine and polyamines. Many of these metabolites were already significantly elevated at lower GOS doses. GOS fermentation also supported gut barrier integrity, a key-indicator of gut health, reflected by increased TEER (trans-epithelial electrical resistance) values under normal as well as LPS-induced inflammatory conditions. The improvements were evident from the lowest dose onward. Overall, the effects on microbiome composition, metabolite production and gut barrier integrity were highly comparable for the two different age groups.

Conclusions: *Ex vivo* fermentation of GOS positively altered child gut microbiome composition and microbial metabolic activity, resulting in improved barrier function. The observation of efficacy at low doses positions GOS as a promising nutritional option for supporting gut health in children aged 3–12 years.

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Poster 69 Symposium

Impact of Dietary Urbanization on the Gut Microbial Structure and Metabolic Activity of School-aged Children from Urban and Rural Areas of Leyte Island, Philippines

Urbanization has profoundly influenced lifestyles, including dietary preferences, particularly in developing countries undergoing rapid socioeconomic transitions. Increased urbanization facilitates access to energy-dense, high-fat, and processed foods, contributing to the rising prevalence of lifestyle-related diseases among both children and adults.

The Asian Microbiome Project (AMP) previously categorized the Asian gut microbiota (GM) into two distinct enterotypes: the *Prevotella*-driven enterotype (P-type), commonly associated with high-fibre, plant-based diets and prevalent in developing regions, and the *Bacteroides/Bifidobacterium*-driven enterotype (BB-type), which is more frequently observed in developed countries and Western populations. Previous research on the GM of rural and urban children from Leyte Island, Philippines, demonstrated that dietary shifts towards Westernized diets in urban children were associated with a transition from P-type to BB-type. However, the metabolic changes accompanying this microbial transition remain poorly understood.

In this study, stool samples from school-aged children in urban Ormoc and rural Baybay, Leyte Island (n = 40 per area) were subjected to 16S rRNA sequencing to characterise the GM composition associated with dietary differences. Bile acids (BAs) were quantified using liquid chromatography-tandem mass spectrometry (LC-MS/MS), while short-chain fatty acids (SCFAs) were quantified using proton nuclear magnetic resonance (¹H-NMR) spectroscopy. Dietary analysis revealed differences between the two populations, with urban children consuming more fat-rich and processed foods, whereas rural children maintained a diet richer in plant-based foods and dietary fibre.

Baybay children tended to harbour a higher abundance of taxa associated with high-fibre diets and SCFA production, including the genera *Prevotella* and *Lachnospira*. Redundancy analyses (RDAs) showed associations between urban- and rural-type GMs and their corresponding BA and SCFA profiles. Urban-type GM exhibited higher concentrations of secondary and tertiary BAs, whereas the rural-type GM BA profile was characterized by a higher level of conjugated BAs. Total SCFA concentration was higher in rural-type GM, although the difference did not reach statistical significance (p = 0.09). These findings suggest that urban and rural GMs are driven by differing dietary habits, resulting in distinct metabolic capacities for BA and SCFA metabolism. Overall, dietary urbanization influences both the structure and metabolic activity of the gut microbiota in Filipino children.

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Poster 70 SFA**In vitro fermentation of 2'-fucosyllactose and/or galacto-oligosaccharides by an infant gut microbial community: effects on microbial composition and metabolite profiles**

The early-life gut microbiota is an important contributor to infant development during this crucial stage of life. Several species of infant-associated *Bifidobacterium* have demonstrated an ability to directly metabolise human milk oligosaccharides (HMO) such as 2'-fucosyllactose (2'-FL) as well as complex carbohydrates such as galacto-oligosaccharides (GOS). This study aimed to evaluate in vitro the temporal compositional and functional changes in an early-life microbial community when supplemented with 2'-FL, GOS or in combination (GOS+2'-FL). Stool samples from 16 infants aged 1 to 8 weeks were collected to form a representative infant microbial community. Anaerobic in vitro fermentation experiments were performed with this faecal community using the various carbohydrate substrates and a carbohydrate-free control. Samples were obtained at 0, 6 and 24 hours. pH measurement, shotgun metagenomic sequencing, and untargeted metabolomics revealed substrate-mediated effects. At 24 hours the mean relative abundance of *Bifidobacterium* was highest in the presence of GOS alone (78.95%) compared to the other test substrates. Similarly, alpha diversity analysis revealed that all carbohydrate-supplemented groups had a lower diversity at 6 and 24 hours (i.e., illustrating the *Bifidobacterium*-dominated community), reflective of a more desirable infant microbiota profile, compared to the control. GOS alone induced the lowest alpha diversity at the end of fermentation. At species level *Bifidobacterium breve* remained the most abundant species in all carbohydrate groups highlighting its ability to persist in the early life microbiota. Carbohydrate supplementation induced the production of metabolites such as N-acetylated amino acids, p-hydroxyphenyllactic acid, and γ -aminobutyric acid, which may support infant gut health, including intestinal barrier integrity and immune function.

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Poster 71 Symposium**Species-specific Shifts in *Faecalibacterium* Associated with Improved Clinical Outcomes in Children with Cow Milk Allergy Following 1-Kestose Intervention**

Objective: The gut microbiome is reported to play a critical role in food allergen tolerance. A previous study reported that 1-kestose supplementation improved clinical outcomes in children with cow's milk allergy (CMA)¹. *Faecalibacterium* is one of the key genera associated with host health, exerting its effects through the production of butyrate and microbial anti-inflammatory molecule (MAM). However, its species-level properties and composition in diseased patients remains unclear. This study aims to explore species-level *Faecalibacterium* abundance in pediatric CMA patients, evaluate the impact of 1-kestose on this abundance, and assess its association with clinical allergy markers post-intervention.

Materials and Methods: In an open-label, parallel-group study (Ethics Committee of the National Hospital Organization Fukuoka National Hospital, Approval No. F30-17), children aged 4–15 with severe CMA underwent oral immunotherapy (OIT) daily, divided into groups supplemented with daily 1-kestose (n=23) or no supplement (control, n=7) over 6 months. Fecal samples and clinical data, including milk- and

casein-specific IgE and oral food challenge (OFC) thresholds, were collected at baseline and study completion. Species-level quantification was performed using *Faecalibacterium* specific *rpoA*-based qPCR. Statistical analysis included Wilcoxon signed-rank tests, Spearman correlations, and stepwise multiple linear regression.

Results and Discussions: After 6 months, the kestose group exhibited a significant increase in *F. longum* abundance, and the changes in *F. longum* abundance were inversely correlated with both milk- and casein-specific IgE levels, while *F. duncaniae* abundance correlated positively with milk protein threshold dose. Stepwise regression analysis identified *F. longum* as a significant negative predictor for both milk-specific ($R^2=0.23$, $p=0.041$) and casein-specific IgE ($R^2=0.257$, $p=0.038$). Furthermore, levels of specific *Faecalibacterium* species positively correlated with relative abundance of beneficial commensals, including *Alistipes* and *Roseburia*. These results demonstrate that species-level investigation provide deeper insights into the gut microbiota, essential for identifying probiotic and biomarker candidates to ensure host health.

References

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Structural and biochemical study of aromatic lactate dehydrogenase from infant gut-associated bifidobacteria

Breastfeeding-associated *Bifidobacterium* colonization of the infant gut is crucial for early-life immune development. These bacteria utilize a specific aromatic lactate dehydrogenase (ALDH) to convert aromatic pyruvates into aromatic lactic acids, such as indolelactic acid. These microbial metabolites regulate intestinal immune responses and maintain homeostasis by acting as agonists of the host aryl hydrocarbon receptor and hydroxycarboxylic acid receptor 3 (Laursen et al., *Nat. Microbiol.* 6, 1367-1382, 2021). However, the molecular basis and structural mechanisms for how ALDH recognizes and converts its substrates to generate these key immunomodulatory molecules remain unknown.

It has been known that ALDH is activated by inorganic phosphate. Through biochemical experiments aimed at identifying the physiological allosteric effector of ALDH, we discovered that fructose 6-phosphate, an intermediate of the bifidobacterial glycolysis (the bifid shunt), serves as a heterotropic activator.

To further investigate the molecular basis of these unique properties of ALDH, we determined the crystal structures of ALDH in its apo and cofactor-bound (NADH-bound) states. To facilitate crystallization, we used a mutant protein in which the surface cysteine residue was substituted with serine. In the homotetrameric structure of ALDH, a large, positively charged surface, which may account for its unique allosteric activation by phosphate, was present. We also determined a ternary complex with NADH and an analogue of a substrate, phenylpyruvic acid. The active site of ALDH has a large hydrophobic pocket, which may account for the substrate specificity for aromatic substrates.

Together, these biochemical and structural insights indicate that ALDH retains traces of precise molecular evolution from a canonical L-lactate dehydrogenase involved in central carbon metabolism to the

specialized enzyme in aromatic amino acid metabolism. Our findings provide a structural basis for understanding the early-life host-microbiome interactions mediated by breastfeeding-associated *Bifidobacterium*.

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Poster 73 SFA

Vaginal microbiome dysbiosis and inflammation in preterm birth

Preterm birth (PTB), defined as delivery before 37 weeks of gestation, affects ~10% of pregnancies worldwide and remains the leading cause of infant mortality. Emerging evidence implicates cervico-vaginal microbiome and vaginal infections, particularly by Group B *Streptococcus* (GBS) and *Ureaplasma*, as risk factors for PTB. Additionally, higher bacterial diversity microbiomes (fewer protective lactobacilli) are linked to immune activation (increased proinflammatory cytokines), leading to spontaneous PTB. Yet, it is unclear how specific vaginal microbial shifts trigger inflammatory pathways, limiting reliable early-pregnancy biomarkers and targeted preventive strategies against PTB.

In this study, the vaginal microbiome of women is followed from preconception, through pregnancy, and postpartum, focusing on the link of specific bacteria with host immune responses and pairing observational data with mechanistic assays to inform early prediction and intervention. Importantly, the anti-GBS activity of vaginal lactobacilli (single cultures and synthetic communities) is tested.

Three complementary clinical cohorts were integrated: (1) women undergoing fertility treatments, (2) women at high risk of PTB (with and without synbiotic intervention), and (3) women who delivered preterm (<30 weeks gestation, Infancy trial-NCT06342752). Gynecologist-collected vaginal swabs underwent DNA extraction and 16S rRNA V4 amplicon sequencing.

Preliminary results from an Infancy cohort subset (n = 44) show high-diversity microbiome profiles in 57% (25/44) of participants, whereas only 43% were *Lactobacillus*-dominated. *Ureaplasma* was positive in 57% (25/44) of the participants, from which 40% (10/25) were diagnosed with chorioamnionitis. To provide alternative probiotics, we leveraged the lab's extensive vaginal isolates biobank and isolates from the Infancy cohort to screen vaginal lactobacilli for anti-GBS activity. Results demonstrated a strong antipathogenic activity of *L. crispatus*, *L. jensenii*, *L. reuteri* strains against GBS, with synergistic inhibition observed by mixed cultures. The selected strain's anti-GBS activity will be mechanistically validated in a vagina-on-chip microfluidic model, which supports host-microbe-immune co-culture under physiologically relevant conditions, for instance, the inclusion of estrogen. Immunomodulatory responses will be profiled using the Olink Target 96 Inflammation panel, quantifying key proinflammatory biomarkers (e.g., IL-6, IL-8, TNF- α) linked to PTB. Collectively, our study combines unique datasets to contribute to the understanding of the vaginal microbiome dynamics across assisted reproduction, pregnancy, and postpartum.

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