PROGRAMME AND ABSTRACTS

1st International Conference on

Microbial Food & Feed Ingredients

New trends in academia, industry and regulatory affairs

2 - 4 May 2018
Copenhagen · Denmark

www.miffi.org

The Danish Microbiological Society DMS

MiFFI 2018 International Conference on Microbial Food and Feed Ingredients

THEMATIC ISSUE ON
Microbial Food and Feed Ingredients

Editors: ***Egon Bech Hansen, Gisèle LaPointe, Dennis Sandris Nielsen***

This thematic issue will be devoted to research on microbial food and feed ingredients. The science presented in the issue will be fundamental science in the applied field of food and feed ingredients.

The ultimate target of the research described will be to increase the quality of food and feed and the core scientific field applied will be microbiology. Microorganisms can be the ingredient, the production host, or the target. Microorganisms can be targeted for stimulation or elimination. The range of microorganisms will be wide, covering bacteria, yeasts, and moulds; pathogens as well as beneficial microorganisms; microbial metabolites with application in food and feed as preservatives, flavours, texturants, etc.; and also, methods to study and modify the microbiotas of humans and animals.

The issue is connected to the first International Conference on Microbial Food and Feed Ingredients, MiFFI, to be held from May 2nd - 4th, 2018 in Copenhagen, Denmark (www.MiFFI.org). Non-attendees are also welcome to submit.

All submitted papers will be subjected to our standard independent peer-review. Authors should specify '**MiFFI2018**' in the cover letter. Prospective authors for MiniReviews or Commentaries must contact the Editor in advance.

For instructions to authors please see the *FEMS Microbiology Letters* journal page:
academic.oup.com/femsle

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SUBMISSION DEADLINE: 1 AUGUST 2018

Join the 1st International Conference on Microbial Food and Feed Ingredients in Copenhagen, 2-4 May 2018.
Register here: **www.MiFFI.org**

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General Information

Conference venue
The Maersk Tower
Blegdamsvej 3B
2200 Copenhagen N

Conference language
The conference will be held in English.

Name badges
All participants and exhibitors must wear their name badge in the conference area at all times.
The badge must be visible.

Lunch and coffee breaks
Lunch and coffee is available in the exhibition area.
See programme for exact time of breaks.

Poster session
The poster area is in the foyer next to the Holst and Nielsine Nielsen auditorium

Social Programme

Welcome reception
(included in the registration fee)

Date	2 May 2018
Time	18.30 - 21.00
Place	15th floor, The Maersk Tower
Address	Blegdamsvej 3B, 2200 Copenhagen N

The welcome reception will take place at the Maersk Tower at 18:30-21.00. Make sure to be there – it will be an evening full of networking and one of Copenhagen's best views. The reception is included in the registration fee.

Conference dinner
(not included in the registration fee)

Date	3 May 2018
Time	18.30 - 23.00
Place	Carlsberg Museum & Business Centre
Address	Valby Langgade 1, 2500 Copenhagen

The conference dinner will take place from 18.30 - 23.00 in the old brewer's glyptotheque at Visit Carlsberg, the historical epicentre of Carlsberg. A Nordic-styled dinner will be served and accompanied with a range of beverages from Carlsberg.

Speaker information
Please bring your presentation to the session room before your session starts. We recommend you that you upload your presentation at least 30 minutes before your session. A technician will be present to assist in the upload, if necessary. Please bring your presentation on a USB stick.

Unless otherwise agreed all presentations will be deleted after the conference in order to secure that no copyright issues will arise at the end of the conference.

WiFi
Free WiFi is provided throughout the venue by logging on "KU Guest" and creating your own account.

Mobile phones
All mobile phones must be on silent mode during the sessions.

Lost and found
Found items should be returned to the registration desk. If you lose something, please report to this desk for assistance.

Conference secretariat

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Tel: +45 70 20 03 05
www.cap-partner.eu

Conference website
www.miffi.org

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@MiFFIconference

facebook.com/MiFFIConference

Welcome

Dear participant,

It is a great pleasure to welcome you to the 1st International Conference on Microbial Food and Feed Ingredients (MiFFI) 2018 in Copenhagen.

During the two conference days, you will experience a diverse programme that includes keynote sessions, poster presentations and workshops. You will get a unique opportunity to meet experts in the field and be updated on research in microbial food and feed ingredients, as well as the regulatory framework surrounding the area.

Microbial food and feed ingredients have a growing role both in terms of research, education and industry.
The MiFFI conference offers high-level scientific presentations, networking activities and is an excellent opportunity to exchange knowledge and experiences with peers from academia and industry.

The main subjects at the conference are:

- Microbiomes
- Microbial Food Cultures
- Fermented Beverages
- Direct-Fed Microbials
- Yeast and yeast extracts
- Proteins and peptide ingredients produced by fermentation e.g. enzymes
- Metabolites produced by fermentation used as ingredients in food and feed e.g. vitamins, acids, alcohols, and flavours.
- Regulatory aspects

We also have the pleasure to announce that a joint thematic issue on Microbial Food & Feed Ingredients will be published with FEMS Journals (Federation of European Microbiological Societies). We therefore encourage you to submit your manuscripts (full papers and/or reviews), before the 1st of August 2018. The issue will be out during Spring 2019.
We look forward to receiving your papers!

Furthermore, we have arranged some exciting social events, so you will get a chance to network and mingle with colleagues and peers from your field.

We hope you will enjoy the conference and your stay in Copenhagen!

On behalf of the Scientific and Local Organising Committee,

Egon Bech Hansen
Chair of the Scientific Committee
Technical University of Denmark, National Food Institute

Lars Bogø Jensen
Chair of the Organizing committee
Representative to the Danish Microbiological Society
Technical University of Denmark, National Food Institute

Organisation

The Local Organizing Committee

Lars Bogø Jensen
Technical University of Denmark,
National Food Institute (Chair)

Fergal P. Rattray
Dept. of Food Science, University of Copenhagen

Peder Fode
Confederation of Danish Industry

Anders Permin
Saxocon

Anemette Hansen
Danish Agriculture and Food Council

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THE DANISH MICROBIOLOGICAL SOCIETY

DMS Annual Congress

12 NOVEMBER 2018

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Keynotes

Microbial adaptive responses to the environment and their biotechnological applications
Gemma Reguera, Michigan State University

The medieval plagues: ecology, transmission modalities and routes of the infections
Nils Christian Stenseth, University of Oslo

The history of Danish microbiology
Søren Molin, Technical University of Denmark
Jan Sørensen, University of Copenhagen

Programme Wednesday - Thursday

Wednesday 02 May	
17.00 - 20.00	REGISTRATION
18.30 - 21.00	**WELCOME RECEPTION AT THE MAERSK TOWER** (included in the registration fee) Address: 15th floor, Blegdamsvej 3, 2200 Copenhagen
18.30	Welcome by Lars Bogø Jensen and Egon Bech Hansen (chairs)
18.45	"Crafted By, a nano brewery with customized portfolio" Presentation by Jon Bonne Eriksen, head brewer at Carlsberg's microbrewery "Crafted By"
19.15	Beers, snacks & networking

Thursday 03 May	Room. Niels K. Jerne
08.30	REGISTRATION & COFFEE
09.30 - 09.45	**Welcome - Opening ceremony** **Chairs:** **Egon Bech Hansen** (Chair), Technical University of Denmark, National Food Institute **Lars Bogø Jensen** (Chair), Technical University of Denmark, National Food Institute
09.45 - 10.30	**Keynote presentation** Brave new world of food functionality, *- What can we learn from the past -* **Wim Saris,** Maastricht University, The Netherlands
10.30 - 11.00	COFFEE BREAK & EXHIBITION

	Room. Auditorium Niels K. Jerne	Room: Auditorium Nielsine Nielsen	Room: Auditorium Holst
11.00 - 12.05	**Microbiome human** Chair. Giséle LaPointe, University of Guelph, Canada	**Direct fed microbials** Chair. Karoline Sidelmann Brinch, Novozymes, Denmark	**Innovation workshop:** **The future ingredients** **– opportunities and challenges** Chair. Anders Permin, Saxocon, Denmark
11.00	**Introduction**	**Introduction**	Panelists: Adam Hillestrøm, DACOFI, Denmark Jan Boeg Hansen, Tapperiet, Denmark Jens Legarth, Fermentationexperts, Denmark
11.05	**Understanding the impact of food ingredients through in vitro modelling of the fecal microbiome** Gisèle LaPointe, University of Guelph, Canada	**In situ delivery of beneficial compounds by Bacilli** Mette Dines Cantor, Chr. Hansen, Denmark	
11.25	**Mining the microbiome for novel anti-infectives for food and feed** Colin Hill, University College Cork, Ireland	**Formulation of Bacillus probiotics is key to product performance** Karoline Sidelmann Brinch, Novozymes, Denmark	
11.45	**Immune suppression after stroke. Evidence of bacterial translocation** Dragana Stanley, Central Queensland University, Australia	**Fermented canola meal product. Prebiotics, Probiotics and Bioactives** Søren Kjærulff, FermBiotics, Denmark	
11.55	**Fecal virome transplantation targeting type-2-diabetes and obesity in mice** Torben Sølbeck Rasmussen, University of Copenhagen, Denmark	**Microbial solutions for sustainable health and production of ruminants** Giuseppe Copani, Chr. Hansen, Denmark	
12.05 - 13.00	LUNCH & EXHIBITION	LUNCH & EXHIBITION	LUNCH & EXHIBITION
12.15 - 13.00			**INDUSTRY SYMPOSIA** **Industrial fermentation solutions by Lallemand. Danish roots with a global visionIndustrial fermentation solutions by Lallemand . Danish roots with a global vision** David Guerrand, Biotech Business Director, Lallemand Bio-Ingredients Rune Engell-Hansen, Plant Manager, De Danske Gærfabrikker
13.00 - 14.00	POSTER SESSION	POSTER SESSION	POSTER SESSION

Thursday 03 May	Room. Auditorium Niels K. Jerne	Room: Auditorium Nielsine Nielsen	Room: Auditorium Holst
14.00 - 15.05	**Microbial Food Cultures** Chair: Egon Bech Hansen, DTU Food - National Food Institute, Denmark	**Microbiome animals** Chair. Alexander Sulakvelidze, Intralytix, Inc., USA	**Workshop. From innovative student to entrepreneurial employee** Chair: Lars Bogø Jensen, Technical University of Denmark - National Food Institute, Denmark Panelists. Lars Bogø Jensen, Technical University of Denmark "Using real life scenarios in modern university teaching" Nanna Viereck, University of Copenhagen, Denmark "Industrial collaboration into courses and student projects enabling innovative & entrepreneurial alumni" Dorthe Lynnerup, University of Copenhagen, Denmark "SCIENCE Innovation Hub – how to support students from idea to business" Marie Louise M. Pollmann-Larsen, Technical University of Denmark "Connecting students and industry in hacks, spinout courses, food labs and open innovation challenge competitions" Peder Fode, Confederation of Danish Industry, Denmark "The importance of a close collaboration between knowledge institutions and industry" Harry Barraza, Arla Foods, Denmark "Collaborating with startups to scale innovation"
14.00	**Introduction**	**Introduction**	
14.05	**Product-yield selection in water-in-oil emulsions** Rinke van Tatenhove-Pel, Vrije Universiteit Amsterdam, The Netherlands	**Bacteriophages for Healthier Foods. Safety by Nature** Alexander Sulakvelidze, Intralytix, Inc., USA	
14.25	**Identification of efficient vitamin-secreting lactic acid bacteria through the droplet-based high-throughput screening** Jun Chen, DTU Food - National Food Institute, Denmark	**Ultra-high-resolution exploration of the microbiome** Henrik Bjørn Nielsen, Clinical-Microbiomics, Denmark	
14.45	**Microbial polysaccharides for texture improvement** Vera Kuzina Poulsen, Chr. Hansen, Denmark	**Confirmation of the presence of Enterococcus faecium M74 in the gut of 1-day-old and 7-day-old chickens using PFGE-typing after in ovo application** Line Skjoet-Rasmussen, Chr. Hansen, Denmark	
14.55	**Lactobacillus helveticus. spice up its life!** Ineke Van Boeijen, CSK food enrichment, The Netherlands	**New probiotic Bacillus strains to improve gut health in piglets** Bea Nielsen, Chr. Hansen, Denmak	
15.05	**Viability of microencapsulated Akkermansia muciniphila and Lactobacillus plantarum during freeze-drying, storage and in vitro upper gastrointestinal tract passage** Martin Marcial-Coba, University of Copenhagen, Denmark		
15.15 - 15.30	COFFEE BREAK & EXHIBITION	COFFEE BREAK & EXHIBITION (15.05 - 15.30)	COFFEE BREAK & EXHIBITION
15.30 - 16.30	**Keynote presentation** **CRISPR-Cas systems. from humble beginnings to today's headlines** Sylvain Moineau, Laval University, Canada		
16.30 - 17.15	**Networking Reception with fermented beverages** Exhibition area		
18.30	**Conference Dinner at Carlsberg Museum** (not included in the registration fee.) Carlsberg Museum & Business Centre, Valby Langgade 1, 2500 Copenhagen		

Friday 04 May			
	Room. Auditorium Niels K. Jerne		
09.00 - 10.00	**Regulatory affairs. Challenges and opportunities** Chair. Svend Laulund, Chr. Hansen, Denmark		
09.00 - 09.10	**Introduction**		
09.10 - 09.30	**Effect of the Nagoya protocol on future access to industrial lactic acid bacteria cultures** Eric Johansen, Chr. Hansen, Denmark		
09.30 - 09.45	**Challenges facing the Microbial food culture industry** Paul Tenning, European Food and Feed Cultures Association (EFFCA)		
09.45 - 10.00	**Developing science and today's regulations** Lisa Jensen, Association of Manufacturers and Formulators of Enzyme Products (AMFEP)		
	COFFEE BREAK & EXHIBITION	**COFFEE BREAK & EXHIBITION**	**COFFEE BREAK & EXHIBITION**
	Room. Auditorium Niels K. Jerne	Room: Auditorium Nielsine Nielsen	Room: Auditorium Holst
10.30-11.35	**Fermented beverages** Chair. Jochen Förster, Carlsberg Research Laboratory, Denmark	**Metabolites and proteins/peptide (enzymes)** Chair: Vibe Glitsø, Nozozymes, Denmark	**Regulatory affairs: Challenges and opportunities cont.** Chair: Svend Laulund, Chr. Hansen, Denmark
10.30	Introduction	Introduction	Introduction
10.35	**A systems approach to generating superior industrial yeasts** Kevin Verstrepen, University of Leuven, Belgium	**Hydrolysis of microbial turnover products in the gut. a case study in broiler chickens** Mikkel Klausen, Novozymes, Denmark	**Safety Demonstration of Microbial Food Cultures. The IDF EFFCA Initiative** François Bourdichon, Food Safety and Hygiene Consultant, France
10.55	**The use of Pichia kluyveri yeast for the production of low alcohol and non-alcoholic beer** Sofie Saerens, Chr. Hansen, Denmark	**Refining proteins from green crops using lactic acid fermentation and obtaining high quality feed products for animals** Mette Lübeck, Aalborg University, Denmark	**Role of the Qualified Presumption of Safety concept in the EFSA risk evaluations** Lieve Herman, Flanders Research Institute for Agriculture, Fisheries and Food, Belgium
11.15	Panel discussion	**Lactic Acid Bacteria for Efficient Delivery of Vitamin K2** Yue Liu, Wageningen University, The Netherlands	**Bacteriophages for the wine industry. a way to a healthier wine** Ifigeneia Kyrkou, Aarhus University, Denmark
11.25		**Developing Lactic Acid Bacteria for the conversion of brown macroalgae to green chemicals and fuels** Elleke Bosma, The Novo Nordisk Foundation Center for Biosustainability, Technical University of Denmark	**The challenge of lactic acid bacteria to metabolize phenolic compounds in elderberry juice** Annalisa Ricci, University of Parma, Italy
11.35 - 13.00	**LUNCH & EXHIBITION**	**LUNCH & EXHIBITION**	**LUNCH & EXHIBITION**
13.00 - 14.00	**POSTER SESSION**	**POSTER SESSION**	**POSTER SESSION**
	Room. Auditorium Niels K. Jerne	Room: Auditorium Nielsine Nielsen	Room: Auditorium Holst
13.00 - 14.05	**Food and feed enzymes** Chair. Hans van den Brink Chr. Hansen, Denmark	**Food and feed waste - waste upgrade** Chair. Nicoletta Ravasio, CNR ISTM, Italy	
13.00	Introduction	Introduction	
13.05	**Development of novel chymosins for cheese production** Christian Jaeckel, Chr. Hansen, Denmark	**Upgrading of whey and other food supply chain waste** Nicoletta Ravasio, CNR ISTM, Italy	
13.25	**Enzymes for Fruit Processing** Wolfgang Aehle, BRAIN, Germany	**Value added products from dairy waste** Peter Ruhdal Jensen, Technical University of Denmark, Denmark	
13.45	**Cassava non-starch polysaccharide composition & degradation by carbohydrases** Larissa Staack, Novozymes, Denmark	**Phage Therapy Against Soft Rot Enterobacteriaceae in Potatoes Under Simulated Storage Conditions** Amaru M. Djurhuus, Aarhus University, Denmark	
13.55	**Camel Chymosin for Camel Cheese Making Properties** Yonas Hailu, Haramaya University, Ethiopia	**Production of nutrient-rich yeast from wood-derived sugars** Ricardo Ekmay, Arbiom Inc., USA	
14.05 - 15.00	**Nordic Food Lab –fermenting for deliciousness** Michael Bom Frøst, Nordic Food Lab, Denmark		
15.00 - 15.15	**CLOSING REMARKS AND POSTER PRIZES**		

Industry Symposium

Thursday 3 May, 12.15 – 13.00
Auditorium: Holst

Industrial fermentation solutions by Lallemand: Danish roots with a global vision

Speakers

David Guerrand
Biotech Business Director, Lallemand Bio-Ingredients – Introduction to biotech and fermentation, our offer - 15'

Rune Engell-Hansen
Plant Manager - De Danske Gaerfabrikker plant presentation – 15'

Description
Lallemand is a privately owned Canadian company, a leader in the development, production and marketing of yeast, bacteria and specialty ingredients. Lallemand Bio-Ingredients produces inactive yeast and yeast derivatives, such as yeast extracts, for the savory, health and fermentation markets. The DDGF factory, based in Grenaa in Denmark is the most important industrial site for producing Lallemand yeast extracts.

The industrial fermentation market players, including both the producers of biomass and the producers of various metabolites (enzymes, pharmaceutical molecules...) are looking for new solutions to optimize their production yield and end-products quality. Lallemand Bio-Ingredients will present recent developments in yeast ingredients addressing the needs of the industrial fermentation industry, highlighting the central role of the DDGF Danish facility from pilot scale development to production.

It is possible to bring your lunch into the auditorium.

LALLEMAND

LALLEMAND BIO-INGREDIENTS

Keynote speakers' abstracts

[K1] BRAVE NEW WORLD OF FOOD FUNCTIONALITY -WHAT CAN WE LEARN FROM THE PAST-?

Wim H.M. Saris[1]

[1] *Maastricht University, The Netherlands*

After the initial successful discoveries of the role of vitamins and minerals in the first part of the nineteenth century, the nutritional sciences has devoted most of the time in the second part of that century on the role of dietary fats and carbohydrates in relation to chronic diseases like CHD, diabetes and obesity. In contrast to the reductionist approach in vitamins/minerals research, randomized controlled trials on macronutrients such as fats are impossible due to the energetic interrelationship. High fat intake leads automatically to low carbohydrates intake in iso-energetic diets. Therefore scientific evidence to change dietary recommendations based on observed food functionality is difficult and risky. In the seventies and eighties the recommendation to reduce fat intake in particular saturated fat initiated a boost in low/fat free food products and diets high in unsaturated fats. However the availability of these alternatives did not lead to lower obesity rates, On the contrary. Reason why more research attention was given to carbohydrates in particular sugar. The scientific arguments are except for the sweetened beverages turned out not very convincing again. These changing opinions showed clearly that it is difficult to transfer trial outcomes into diets recommendations. The introduction of the Health Claim regulations in Europe has reinforced the reductionist approach of testing new ingredients on functionality. Next to this one nutrient – one physiological effect/biomarker pharmacological approach is the problem of multi-target and relative small effects of nutrients compare to drugs, leaving much room for discussion about the functionality.

What can we learn from these past experiences, now metagenomics analyses are providing valuable knowledge of the diversity and functionality of the gut microbiota? Is it possible to define a "healthy microbiota" based on a reductionist approach or to prove the functionality of specific gut microbes in relation to specific chronic disease endpoint like CVD, T2D and obesity? Most probably we have to adopt new more holistic and integrative approaches to show the functionality of the food matrix on the gut microbiome in relation to our health.

[K2] CRISPR-CAS SYSTEMS: FROM HUMBLE BEGINNINGS TO TODAY'S HEADLINES

Sylvain Moineau[1]

[1] *Université Laval, Canada*

Bacteriophages are now recognized at the most abundant and diverse biological entities on the planet. Their bacterial hosts have a plethora of defense mechanisms to combat phages, including CRISPR-Cas systems. Exploiting this system has also resulted in the development of the much-publicized CRISPR-Cas9 technology for precise genome manipulation of various organisms. This seminar will recall the roles played by phages in the discovery and understanding of CRISPR-Cas systems as well as the means use by phages to bypass this system. Finally, the use of the CRISPR-Cas9 technology for viral genome editing to better understand phage-host interactions will be presented.

Workshops

Innovation workshop: The future ingredients – opportunities and challenges

Chair Anders Permin, Saxocon, Denmark
Day Thursday 3 May 2018
Time 11.00 – 12.05
Auditorium Holst

In a world where the human population is growing rapidly and where the pressure on natural resources increases day-by-day, there is a need to rethink our supply and production of food and feed. Can we utilize the resources better? Can we find new resources? Can we produce our food and feed in a smarter way? Can we improve the durability of food and feed? Or can we improve our health by intake of special food with specific features? Can we change our eating habits or introduce novel foods? There are many answers to these questions. However, to begin at the beginning we need to rethink how we use the natural resources and which food and feed stuffs we produce from these resources. This process has already started which is among other technologies reflected in the increasing use of ingredients in the food and feed production. Our workshop will focus on the future needs of ingredients in the food industry and in which directions the innovation is going or should go.

Panelists

- Adam Hillestrøm, DACOFI, Denmark
- Jan Boegh Hansen, Tapperiet, Denmark
- Jens Legarth, Fermentationexperts, Denmark

Workshop: From innovative student to entrepreneurial employee

Chair Lars Bogø Jensen, DTU Food - National Food Institute, Denmark
Day Thursday 3 May 2018
Time 14.00 – 15.05
Auditorium: Holst

By integrating entrepreneurship in today teaching at universities as an intra curriculum activity and possibilities for students to participate in extra curium activities at students' hub the gap between being part of the education system and their first job as graduates is drastically reduced. At the same time students will be able to look at the real life challenges seen in companies and challenge existing limitation hereby create new innovative solution. Challenges can be introduced to students as part of the learning objective in course at universities or competitions like OIX (Open Innovation), Hackathons. In this session, speakers from universities and companies will focus on the importance of this cooperation's between companies and universities and examples on students' solutions will be resented.

Panelists:

- Nanna Viereck, University of Copenhagen, Denmark
- Dorthe Lynnerup, University of Copenhagen, Denmark
- Marie Louise Møllebæk Pollmann-Larsen, Technical University of Denmark , Denmark
- Peder Fode, Confederation of Danish Industries, Denmark
- Harry Barraza, Arla Foods, Denmark

Session and panel debate

Regulatory affairs: Challenges and opportunities

Chair Svend Laulund, Chr. Hansen, Denmark
Day Friday 4 May 2018
Session time 9.00 – 10.00 in Auditorium Niels K. Jerne
Panel debate 10.30 – 11.35 in Auditorium Holst

Today, we experience that the speed of science is a great deal faster than the speed of regulation. Science has the advantage that it is generally recognized in the same way worldwide. Regulation, on the contrary, is an area with a slower evolution. It is also implemented differently at regional level and in many cases down to individual national level; differing from country to country. This is a challenge for international food and feed ingredients producers. At the same time, being dependent on microbes adds to the challenge due to the Nagoya protocol that regulates access and benefit-sharing of genetic resources.

Two European associations with manufacturers of ingredients products (Association of Manufacturers & Formulators of Enzymes products, AMFEP, and European Food Cultures Association, EFFCA) where the use of microbes is the core, will explain their challenges. One is struggling with relatively new regulatory demands, and the other struggles due to the opposite: no specific regulation!

Since 2002, the International Dairy Federation (IDF) and EFFCA have published and managed an inventory of microorganisms with a historical safe use in food. Five years later, in 2007, the European Food Safety Authorities (EFSA) published their first Qualified Presumption of Safety (QPS) list with microbial species safety assessed (by scientists) for use in production of feed and food additives. Despite updates twice a year, the QPS list lacks several microbes which have been used for producing safe fermented food for years. This is due to differences in EU regulation. This creates uncertainty and confusion for food producers and authorities as QPS is seen as "the positive list" by many. We will hear about the 2 systems, their differences, their differences from other systems like the US GRAS and discuss whether there is a possibility of finding a way to eliminate the uncertainties.

Speakers

- Eric Johansen, Chr. Hansen, Denmark
- Paul Tenning, European Food and Feed Cultures Association (EFFCA)
- Lisa Jensen, Association of Manufacturers and Formulators of Enzyme Products (AMFEP)
- Francois Bourdichon, Food Safety and Hygiene Consultant, France
- Lieve Herman, Flanders Research Institute for Agriculture, Fisheries and Food, Belgium

The session will turn into a panel discussion.

Oral Abstracts

[O1] UNDERSTANDING THE IMPACT OF FOOD INGREDIENTS THROUGH IN VITRO MODELLING OF THE FECAL MICROBIOME

Gisele LaPointe[1]
[1]*University of Guelph, Canada*

In vitro modelling is most appropriate for testing interactions from ingredients that may have a negative impact on the health of human subjects. Specific examples are experimentation with foodborne pathogens and potentially toxic substances. We used a metabolomics approach to assess the impact of tartrazine on the metabolism of the gut microbial community, standardized in a chemostat over 36 days. Metabolite profiling by nuclear magnetic resonance (NMR) showed that short chain fatty acids (SCFA: acetate, butyrate and propionate) decreased after 12 hours of incubation in the sample containing tartrazine compared to the control. This indicates that tartrazine and its potential degradation products impact microbial metabolism.

[O2] MINING THE MICROBIOME FOR NOVEL ANTI-INFECTIVES FOR FOOD AND FEED

Colin Hill[1]
[1]*APC Microbiome institute, University College Cork*

The human microbiome is a vast repository of potential interventions which could be deployed in human and animal food, feed and medicine. These include bacterial cultures, bacterial metabolites or end-products, and bacteriophages. Evidence for the efficacy of these interventions in food and in animal models will be presented.

[O3] IMMUNE SUPPRESSION AFTER STROKE: EVIDENCE OF BACTERIAL TRANSLOCATION

Dragana Stanley[1], **Dena Lyras**[2], **Monica D. Prakash**[3], **Kulmira Nurgali**[3], **Rob J. Moore**[4] **and Connie H. Y. Wong**[5]
[1]*School of Medical and Applied Sciences, Central Queensland University, Australia*
[2]*Department of Microbiology, School of Biomedical Sciences, Monash University, Australia*
[3]*College of Health and Biomedicine, Victoria University , Australia*
[4]*School of Applied Sciences, RMIT University, Australia*
[5]*Centre for Inflammatory Diseases, Department of Medicine, School of Clinical Sciences, Monash University, Australia*

Stroke is a leading cause of morbidity and mortality worldwide. Despite its recognised debilitating neurological deficits, the major cause of death after stroke is infection. Bacterial pneumonia is the most frequent complication and microbiologic data of patients with post-stroke pneumonia is showing a pattern of mostly early onset nosocomial infections. Regardless of this, there is often no causative organism detected, or the yield of culture is usually very low in infected patients. Antibiotic administration after stroke does not prevent the infection. Here we present the role of gut microbiota in post stroke complications due to bacterial infection. We demonstrate that stroke injury has far-reaching effects, including the capacity to induce profound cellular and barrier functional changes in the gut. We show that the bacteria found in the post-stroke peripheral tissue originates from the translocated host gut mucosa associated microbiota.

[O4] FECAL VIROME TRANSPLANTATION TARGETING TYPE-2-DIABETES AND OBESITY IN MICE

Torben Sølbeck Rasmussen[1], **Caroline M. Junker Mentzel**[2], **Witold Kot**[3], **Lars Hestbjerg Hansen**[3], **Finn Kvist Vogensen**[1], **Axel Kornerup Hansen**[2], **Dennis Sandris Nielsen**[1]
[1] *University of Copenhagen; Department of Food Science; Section of Microbiology and Fermentation*
[2] *University of Copenhagen; Dept. of Veterinary and Animal Sciences; Section of Experimental Animal Models*
[3] *Aarhus University; Dept. of Environmental Science; Section of Environmental Microbiology & Biotechnology*

Development of obesity and type-2-diabetes is associated with gut microbiota (GM) alterations and in mice experiments the disease phenotype can be transferred via GM to a germ-free host. Bacteriophages (phages) are viruses attacking bacteria in a specific manner. Alterations in the gut viral community is associated with changes in the prokaryotic GM component preceding flares of inflammatory bowel disease. Further, transfer of cell-free (but phage containing) fecal extracts seemingly cure *Clostridium difficile* associated diarrhoea with same efficacy as fecal transplants. As a proof-of-concept we here demonstrate the efficacy of fecal virome transplantation (FVT) for shifting the phenotype of obese mice into closer resemblance of lean mice. The FVT cocktail consisted of viromes extracted from cecum content from 18 mice fed a low-fat diet (LFD) for 14 weeks. The virome titer was $2*10^{10}$ Virus-Like-Particles/mL. Subsequently 40 male C57BL/6NTac mice (5 weeks old) were divided into 5 groups: LFD (as control), High-Fat Diet (HFD), HFD + Ampicillin (A), HFD+A+FVT and HFD+FVT. After 6 weeks on their respective diets, the HFD+FVT and HDF+A+FVT mice were treated with FVT twice with one-week interval by oral gavage. The remaining LF, HFD+A, and HFD mice received no treatment. For the HFD+A and HFD+A+FVT, the ampicillin was added once to the drinking water one day before the FVT treatment. HFD+FVT and HFD+A mice showed a significant decrease in weight gain compared to HFD group (adj. p-value = 0.0033 and 0.0007, respectively). Surprisingly, as the only group the HFD+FVT showed a comparable glucose tolerance as determined by OGTT to the lean LFD group (adj. p-value = 0.9648). Thus, the HFD+FVT group exhibited decreased weight gain and maintained an efficient glucose tolerance. We hypothesise that bacteriophages within the FVT altered the abundance of species which, directly or indirectly, are involved in metabolic syndrome development. In conclusion, transfer of gut viral communities from a lean phenotype to an obese phenotype reduce weight gain and improve blood glucose parameters.

[05] IN SITU DELIVERY OF BENEFICIAL COMPOUNDS BY BACILLI

Mette Dines Cantor[1], Karin Bjerre[1], Rute Neves[1], Patrick Derkx[1], Gunnar Oeregaard[1]
[1] *Chr. Hansen A/S*

Bacillus spores are used as probiotics for both animals and humans and have been used in the animal feed industry for at least 25 years. Their use in the feed industry is related to their probiotic effect including improved intestinal health and growth performance of pigs and poultry, thereby reducing or replacing the use of antibiotics. Bacilli are also used extensively in the biotechnology industry to produce a vast range of enzymes, which are used for both food and feed applications. By combining the knowledge of beneficial properties and the ability to produce and secrete the compounds important for the positive effect, it is possible to improve strains in a natural way that will produce a desired component *in situ*, in this case the gut environment.

Obtaining strains with the right phenotype can be done by two routes: screening a large pool of wild type isolates or performing natural strain improvement, which encompasses e.g. classical mutagenesis, selection or adaptive evolution. As an example of classical mutagenesis, *in vitro* proof of concept has been obtained for a *Bacillus subtilis* overproducing tryptophan, a limiting amino acid in pig feed (Bjerre et al., 2016). Also production of fiber degrading enzymes *in situ* would be beneficial for the feed utilization of many animals as the amount of non-starch polysaccharides in feed is increasing.

Examples of strategies for making probiotic Bacilli produce beneficial compounds *in situ*, which species would be relevant and which compounds to aim for will be discussed.

Bjerre, K., Cantor, M. D., Nørgaard, J. V., Poulsen, H. D., Blaabjerg, K., Canibe, N., Jensen, B. B., Stuer-Lauridsen, B., Nielsen, B. and Derkx, P. M. F. (2016). Development of *Bacillus subtilis* mutants to produce tryptophan in pigs. Biotechnol. Lett. DOI 10.1007/s10529-016-2245-6.

[06] FORMULATION OF BACILLUS PROBIOTICS IS KEY TO PRODUCT PERFORMANCE

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[3] *Adisseo*

Purpose: Probiotic-based products have received more attention in poultry productions since the focus on reduction of antibiotics has increased in recent years. Several Bacillus-based products are available but often users find a lack of consistent effects. Much focus has been on selection of the right strain but this factor is only one of several which needs to be considered to develop a consistent probiotic product.

In this talk the screening of Bacillus strain is briefly described and followed by in depth studies of how formulation of the probiotic strain is key to product performance.

Description: A probiotic strain screened and selected for performance in broilers was assessed in a range of assays related to formulation. Simple and optimized formulations were compared and analyzed with respect to demixing, flowability and particle size distribution (PSD) to assess the impact on homogeneity in feed.

Results: The optimized formulation had several beneficial characteristics when compared to simple formulations: The PSD was 233 um (Dv50) where other products were at either <100 or >500. In assessments of simple and optimized formulations of products, significant differences were found in a demixing study: In comparisons of bacterial counts (CFU) between bottom, middle and upper layer of a feed formulation the difference between upper and lower layer was a significant 142% in the simple formulation, while the difference in the optimized formulation was insignificant at 7%. In recovery trials from nine in vivo studies the average in-feed recovery was above 80% and the CV below 20% proving good in-feed homogeneity.

Conclusion: A careful development of the correct formulation is an often overlooked key feature to ensure product performance. Our studies have shown that an optimized formulation with a correct PSD will result in less demixing, better flowability and thus higher in-feed homogeneity - as well as more hassle-free usage. Especially in the starter feed it's crucial that the in-feed counts of Bacillus spores are equal in each feed pellet as the chicks only ingest tiny amounts. Therefore, product performance is closely linked not only to strain selection but also to formulation features of the product.

[07] FERMENTED CANOLA FEED AND FOODPRODUCTS: PREBIOTICS, PROBIOTICS AND BIOACTIVES

Søren Kjærulff[1]
[1] *Fermentationexperts As ; Fermbiotics Aps; Copenhagen Bio Science Park*

Fermentationexperts and Fermbiotics have developed several fermented plant and seaweed products for pigs, poultry and human health. The products consist of prebiotics, probiotics and bioactives from the fermented plant and seaweed material. We have demonstrated very good *in vitro* anti-microbial and anti-inflammatory activities of the fermented material. A new pig trial shows that fermented feed called EP100i containing fermented canola meal can replace zinc in pig's feed. The study shows that pigs fed with EP100i achieve the same or better growth rate than pigs fed with zinc oxide. The pigs were divided into a positive and negative zinc oxide group, and another seven groups that received different quantities of EP100i in the feed. Results show that the piglets fed with EP100i without addition of zinc, actually achieved similar or better growth rates compared with pigs receiving zinc in their feed. An additional pig trial showed a high (58%) reduction of LDL-cholesterol in pigs given 4 % EP100i feed compared with normal feed.

[08] MICROBIAL SOLUTIONS FOR SUSTAINABLE HEALTH AND PRODUCTION OF RUMINANTS

Giuseppe Copani[1], Zhigang Zhu[1], Bea Nielsen[1], Nina Milora[1], Hugo Alonso Ramirez-Ramirez[2]
[1] *Chr. Hansen*
[2] *Iowa State University*

Application of probiotics has the potential to improve health and performance in ruminants. The objective of this study was to investigate the administration of *Pediococcus acidilactici* (PED) or *Bacillus subtilis* (BAC) as direct-fed microbials (DFMs) in dairy cow ration on milking performance, ruminal pH and immune status. Forty-eight multiparous Holstein dairy cows (121 ± 22 DIM) were blocked by milk yield in a randomized complete block design. All treatments consisted of a basal TMR with top-dressed supplements: Control (CON) with no probiotics; PED fed at 1×10^{10} CFU/d; BAC fed at 1×10^{10} CFU/d and; a combination of *Enterococcus faecium* at 1×10^{10} CFU/d and yeast (PRO). Cows were housed in a free stall barn with individual feeding gates, milked twice a day and fed on treatment diet twice a day for 105 d; daily feed intake and milk yield data were recorded and weekly averaged. Blood samples collected bi-weekly were analyzed for complete blood count. Ruminal pH of eight rumen fistulated cows, two cows per treatment, were monitored every two hours during a 24-h period on d 105. Dry matter intake was improved ($P = 0.03$) by 6% in cows consuming PED (24.9 vs 23.6 ± 0.55 kg/d) with milk yield being similar ($P \geq 0.78$) across the treatments (37.4 ± 1.36 kg/d). There were no differences in milk composition ($P \geq 0.79$) between the four treatments, with similar milk fat (3.63 ± 0.16%) and milk protein (3.05 ± 0.06%) percentage. There were no differences in mean daily ruminal pH (5.69 ± 0.05) between the treatments ($P = 0.29$). Digestibility of organic matter (68.9 ± 1.48) was similar across the groups. However, supplementing PED resulted in greater ($P < 0.01$) white blood cell count 9.97 $\times 10^3$ cells/μL compared with 9.52, 9.36 and 8.74 ± 0.20 $\times 10^3$ cells/μL for CON, PRO and BAC, respectively. This was likely due to greater neutrophil count ($P < 0.01$) in PED group. Overall, the present study demonstrates that supplementation of *PED* or *BAC* as DFMs support rumen function with no negative effects on pH while having an immune-modulatory effect.

[09] PRODUCT-YIELD SELECTION IN WATER-IN-OIL EMULSIONS

Rinke van Tatenhove-Pel[1], Iris van Swam[1], Cristian Picioreanu[2], Bas Teusink[1], Herwig Bachmann[1]
[1] *Vu Amsterdam*
[2] *TU Delft*

Many fermentation industries aim for the production of extracellular products, such as flavor compounds. These processes can be improved by using strains with a higher product-yield. To have non-GMO strains, it would be ideal to directly select high product-yield mutants. A cultivation method suited for extracellular product-yield selection is compartmentalization of single cells in water-in-oil emulsions. Compartmentalization ensures that each cell has its own substrate pool and it allows for coupling of the phenotype (extracellular product concentration) to the corresponding producer cell. This coupling of extracellular product to producer is only possible when cells and products stay inside the microdroplet. However, quite some interesting extracellular products with hydrophobic properties, such as diacetyl, ethanol or acetaldehyde, leak into the oil-phase.

We here analyzed the effect of product leakage on high product-yield selections in water-in-oil emulsions. A *Lactococcus lactis* strain was used as producer and fluorescent indicator strains were used as read-out of the product-concentration in microdroplets. We created conditions which allow or prevent product leakage and we analyzed mixtures of droplets with indicator strains only and droplets with both producer and indicator strains. The fluorescence of microdroplets was analyzed using flow cytometry.

Under conditions of product leakage all droplets with indicator strains show a measurable signal response, indicating that under these conditions we cannot discriminate between droplets with and droplets without producer strains. Upon incubation under conditions without product leakage only droplets with both indicator and producer strains showed a response, which should allow FACS sorting of high product-yield mutants. The obtained results indicate therefore that coupling of the product to the corresponding producer is essential to product-yield selections in water-in-oil emulsions.

[O10] IDENTIFICATION OF EFFICIENT VITAMIN-SECRETING LACTIC ACID BACTERIA THROUGH THE DROPLET-BASED HIGH-THROUGHPUT SCREENING

Jun Chen[1], Mike Vestergaard[1], Thomas Glasdam Jensen[2], Martin Dufva[2], Christian Solem[1], Peter Ruhdal Jensen[1]
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[2] *Technical University of Denmark, Department of Micro- and Nanotechnology*

The industry prefers to use classical approaches, e.g., random mutagenesis followed by screening, to improve microorganisms used in food production, as the use of recombinant DNA technologies is still not widely accepted. Although modern automated screening platforms are widely accessible, screening remains as a bottleneck in strain development, especially when a mild mutagenesis approach is applied to reduce the chance of accumulating unintended mutations, which may cause unwanted phenotypic changes. Here, we incorporate a droplet-based high-throughput screening method into the strain development process and readily capture *Lactococcus lactis* (*L. lactis*) variants with more efficient vitamin secretion from low-error-rate mutagenesis libraries. In the example, *L. lactis* mutants producing 400% more vitamin B2 were isolated by iterative rounds of random mutagenesis and droplet screening, where a total of 200,000 mutants were screened within 1 hour with a significantly reduced cost of experimental consumables. Genome resequencing of the mutants revealed that, during each round of mutagenesis, only four mutations were introduced. The study shows that useful mutants showing strong phenotypes but without extensive mutations can be identified with efficient screening technologies. It is, therefore, possible to avoid accumulating detrimental mutations while enriching beneficial ones through iterative mutagenesis screening. Due to the low mutation rates, the genetic determinants are also readily identified

[O11] MICROBIAL POLYSACCHARIDES FOR TEXTURE IMPROVEMENT

Vera Kuzina Poulsen[1], Gunnar Oeregaard[1]
[1] *Chr. Hansen A/S*

Polysaccharides produced by lactic acid bacteria can be used as viscosifying, stabilizing, emulsifying, sweetening, gelling, water-binding agents, in food as well as in non-food applications. Polysaccharide-producing lactic acid bacteria (LAB) are of interest for dairy products as they may contribute to better texture in fermented milk. For this reason, methods for screening and selection of polysaccharide-producing LAB have been of big interest by both academia and industry.

A rapid screening assay combining small scale 96-well microtiter plates and automated liquid handling was used to find *S. thermophilus*, *L. lactis*, *Lactobacillus* and *Leuconostoc* strains that give good texture in fermented milk. More than 200 *Leuconostoc* strains were additionally screened for ability to produce slimy colonies on agar media, with or without supplementation of sucrose or raffinose, which can be used to generate either glucan- or fructan-type homo-polysaccharides. Only a fraction of the slimy colonies were observed to give better texture in acidified milk.

We found that some *Leuconostoc* strains contain a hetero-polysaccharide gene cluster represented by the Wzy-dependent pathway, in addition to the genes responsible for homo-polysaccharide production. The Wzy pathway is responsible for the hetero-polysaccharide production in *S. thermophilus*, *L. lactis* and *Lactobacillus* spp, where a small fraction of strains show superior texturing properties. The genetic content of the strains can be used to predict milk texturing properties.

[O12] LACTOBACILLUS HELVETICUS: SPICE UP ITS LIFE!

Ineke Van Boeijen[1], Martijn Bekker[2], Anne Wiersma[1], Marjo Starrenburg[2], Tiffany Williams[1], Neleke Van Nieuwenhuijzen[1], Wilco Meijer[1]
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[2] *Nizo Food Research*

Traditionally, the dairy industry uses *Lactobacillus helveticus* as an adjunct culture for hard, semi-hard and Swiss type cheese. It functions as a tastemaker as this culture has a very high proteinase and peptidase activity. The culture requires these enzymes, as it is incapable of producing all amino acids itself. Hence, yeast extract is an essential growth component during fermentation.

In our research we have investigated the influence of both medium composition (ratio whey permeate-yeast extract; WP-YE) and pH during pH controlled batch fermentations for one *Lactobacillus helveticus* strain. To reduce the amount of experiments we have used an experimental design consisting of these two factors, which we have analysed with Minitab Statistical Software. For the settings used (pH range of 5,4 – 6,0 and ratio WP-YE of 5:1 – 2:1) we found significant correlations for the ratio WP-YE with growth rate, yield and biomass formation per lactate formed. In all cases a lower ratio WP:YE resulted in a higher growth rate, increased yield and higher biomass formation per lactate formed. This outcome shows that yeast extract is an essential component for growth. Optimized medium conditions can increase yield of fermentations.

[O13] VIABILITY OF MICROENCAPSULATED AKKERMANSIA MUCINIPHILA AND LACTOBACILLUS PLANTARUM DURING FREEZE-DRYING, STORAGE AND IN VITRO UPPER GASTROINTESTINAL TRACT PASSAGE

Martin Marcial-Coba[1], Tomasz Cieplak[1], Thiago Cahú[1], Andreas Blennow[2], Susanne Knøchel[1], Dennis Sandris Nielsen[1]
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[2] *University of Copenhagen; Faculty of Science; Department of Plant and Environmental Sciences*

Akkermansia muciniphila, an abundant member of the human gut microbiota (GM), has been suggested as a potential next-generation probiotic for the therapeutic treatment of GM dysbiosis-associated diseases and metabolic disorders. Nevertheless, its high sensitivity to oxygen limits the development of dosage protocols. Here, we describe microencapsulation, in a xanthan and gellan gum matrix, and a subsequent freeze drying protocol for *A. muciniphila* DSM22959 and *Lactobacillus plantarum* subsp. *plantarum* ATCC14917. Four different cryoprotectants were tested individually: sucrose 5%, trehalose 5% (ST), agave syrup 10% (AS), skim milk 10%, glucose 1%, yeast extract 0.5%, mannitol 2.5% (MGYM) and peptone 0.1%, sorbitol 1.2% (PS) in addition to MilliQ-water serving as control. Only cryoprotectants with high sugar or protein content significantly improved the survival of both strains during freeze-drying. Microencapsulated strains were stored anaerobically during 1 month at 4 °C or 25 °C. Storage temperature had a major impact on survival of *A. muciniphila,* as demonstrated by 2-3 higher log CFU/g reduction upon storage at 25 °C as compared to 4°C. The survival of microencapsulated *L. plantarum*, was relatively stable at both temperatures. During *in vitro* simulated gastric transit, the viability loss of microencapsulated bacteria was significantly lower than that of naked cells for both strains during both fasted (pH 2) and fed (pH 4) conditions. The Smallest Intestine (TSI), a newly developed *in vitro* model of the small intestine, was used to assess the survival of both strains during small intestine transit. A gradual release of cells from microcapsules during small intestine passage was observed as well as a remarkable tolerance of naked *A. muciniphila* cells against bile salts during both fasted and fed conditions. Our study provides data for selecting a proper cryoprotectant and controlling several physical parameters, in order to improve survival of *A. muciniphila* during lyophilisation, storage and upper GIT transit.

[O14] BACTERIOPHAGES FOR HEALTHIER FOODS: SAFETY BY NATURE

Alexander Sulakvelidze[1]
[1] *Intralytix, Inc.*

Lytic bacteriophages/phages are the oldest and most ubiquitous microorganisms on Earth. Because of their potent, highly specific antibacterial activity, phages can provide an all-natural, nontoxic, and effective means for significantly reducing or eliminating bacterial pathogens present in various foods. These natural phage products, when properly applied, reduce significantly the levels of their bacterial hosts contaminating various foods without altering their flavors, aromas, or appearances. The presentations will give the audience an overview of the bacteriophage technology and a current and novel perspective on the crucial technical, regulatory, and human safety issues of this emerging technology for improving food safety.

[O15] ULTRA-HIGH-RESOLUTION MICROBIOMICS

Henrik Bjørn Nielsen[1]
[1] *Clinical-Microbiomics A/S*

SNV level microbiomics provides an unprecedented high-resolution description of the microbiome that facilitate strain tracking e.g. of probiotic strains, explorations of pan-genomes in metagenomics data, and ultra-high resolution association. The latter, however, is challenged by high dimensionality. Here we explore the opportunities and limitations for associations with ultra-high-resolution microbiomics and suggest a solution for this.

[O16] CONFIRMATION OF THE PRESENCE OF ENTEROCOCCUS FAECIUM M74 IN THE GUT OF 1-DAY-OLD AND 7-DAY-OLD CHICKENS USING PFGE-TYPING AFTER IN OVO APPLICATION

Line Skjoet-Rasmussen[1], Tina Styrishave[1], Alfred Blanch[1], Jannie Schnabl[1], Elke Brockmann[1], Dorthe Sandvang[1]
[1] Chr. Hansen A/S

Enterococcus faecium M74 was fed to broiler embryos in ovo at day 18 of incubation. The purpose of the study was to determine the presence and concentration of M74 in the intestinal tract of newly-hatched and 7-day-old chickens.From 1-day-old (app. 12 hours post-hatch) and 7-day-old chickens, the following samples were retrieved: Yolk sac (YS), caecal tonsils (CT), and intestinal tract (the rest of the gut) (IT). Twenty samples from both 1-day-old and 7-day-old chickens were analysed. The samples were diluted 10-fold and plated on TSA blood agar and Enterococcus selective agar (bile aesculin azide agar). Counting of CFUs was done using the selective agar results. In general, the samples revealed a high number of uniform bacterial cultures. When visually inspecting the colonies, many looked homologous and had the morphology of *Enterococcus* colonies. One randomly-collected colony from a subset of the samples was cultivated to ensure purity in preparation for PFGE typing. The YS, CT and IT isolates from the 1-day-old and 7-day-old chickens and the M74 reference strain were typed by PFGE with the rare cutting restriction enzyme SmaI. Enterococci were found in high concentration in the samples: YS ($1.4x10^7$–$2.2x10^{10}$ CFU/g), CT ($7.5x10^4$–$3.1x10^9$ CFU/g) and IT ($1.0x10^4$–$1.8x10^9$ CFU/g). When compared to the initial inoculation dose ($2x10^5$ CFU/g), this indicates that M74 has been multiplying in the animals. Isolates with M74 PFGE pattern were found in samples from both 1-day-old and 7-day-old chickens. The prevalence of M74 was highest in 1-day-old (88%) as compared to 7-day-old chickens (67%). Looking at the pattern distribution in relation to sample site, the caecal tonsils and the intestinal tract showed an identical distribution of M74 intermixed with other found PFGE patterns, whereas all isolates from yolk sacs displayed the M74 fingerprint pattern.

After *in ovo* feeding of the probiotic strain *E. faecium* M74, it was found in high concentration in YS, CT and IT samples from both 1-day-old and 7-day-old chickens. This demonstrates that the embryos ingested M74 before hatching, that M74 is viable for intestinal colonization through *in ovo* feeding, and that the strain multiplies in the chicken gut post-hatching.

[O17] NEW PROBIOTIC BACILLUS STRAINS TO IMPROVE GUT HEALTH IN PIGLETS

Bea Nielsen[1], Mette Dines Cantor[1], Juergen Zentek[2]
[1] Chr. Hansen A/S
[2] Freie Universität Berlin

The scheduled phase-out of zinc oxide in EU and restrictions of the use of antibiotic growth promoters in the US requires management changes in the pig industry to counteract post-weaning diarrhea. Probiotics have shown to improve health and production in piglets. The objective of this work was to screen a wide range of new spore formers to select promising probiotic leads for *in vivo* trials. Analyses for selection included antibiotic resistance profiles, bile and acid tolerance, growth in different media, sporulation capability and antimicrobial activity against *Escherichia coli* F4 and F5, *Clostridium perfringens Type A* and *Type C* as well as *Salmonella typhimurium and Staphylococcus aureus*. Two strains were selected and identified as *Bacillus subtilis* subsp. *subtilis* (BS) and *Bacillus amyloliquefaciens* subsp. *amyloliquefaciens* (BA) by sequencing of 16S rDNA, *gyrB* and *rpoB* genes.

For the *in vivo* performance trial (Trial 1) 324 newly weaned piglets were fed standard diets based on corn-soybean-barley randomly allocated to control (CON) or to BA (DSM25840) or BS (DSM25841) respectively (18 pens per treatment group; 1.28×10^9 CFU/ kg feed).

The two probiotic candidates were also tested under experimental F4 *E. coli* challenge conditions in weaned piglets from 24 to 45 days of age fed corn-wheat-soybean meal based diets (Trial 2). A total of 64 piglets were allocated to either a non-infected group (Con), an infected control group (CI) or an infected group added one of the 2 probiotics, BA or BS in the diet (1.28×10^9 CFU kg/feed).

In Trial 1, at day 14 after weaning probiotic supplementation had numeric or significant effect on daily gain (218, 222 and 235 g/day) and feed conversion (1.21, 1.15 and 1.14 kg/kg) for CON, BA and BS respectively. In Trial 2, piglets of the CI group showed a lower faecal score (more firm) and dry matter content in faeces compared to infected piglets fed the probiotics and to non- infected piglets of the CO group ($P<0.001$).

In conclusion, combined *in vitro* and *in vivo* approaches are essential to select probiotic strains with health and performance promoting properties.

[O18] EFFECT OF THE NAGOYA PROTOCOL ON FUTURE ACCESS TO INDUSTRIAL LACTIC ACID BACTERIA CULTURES

Eric Johansen[1]
[1]Chr. Hansen A/S

Access to a diversity of microorganisms is fundamental to research in microbiology as well as to product development in industries, which depend on the activity of microbes to produce valuable products. In 1993, the United Nations Convention on Biological Diversity determined that states have sovereign rights over genetic resources found in their territories and that they can make their own rules for access and benefit sharing. The Nagoya protocol is a supplementary agreement to the Convention on Biological Diversity and specifies how fair and equitable sharing of benefits arising from use of genetic resources can be ensured. As with any complex issue, 'the devil is in the details'. This is highlighted by efforts in the European Union to prepare guidelines for utilization of genetic resources and associated traditional knowledge within the EU territory. A number of issues have been identified including definitions of common terms like utilization and research. For example, a precise definition of utilization is required to allow an unambiguous determination of when the conditions of the Nagoya Protocol are to be invoked. If these are invoked too early in a project, the regulatory burden may keep companies and academic researchers from exploring resources from (biodiversity rich) provider countries. This is especially relevant in screening campaigns of large numbers of strains and biodiversity assessments. Also, there is the question of *in silico* descriptions of genetic materials which can be analyzed without access to the genetic resource itself but which form the basis of modern biotechnology. An expert workshop, organized by the Lactic Acid Bacteria Industrial Platform (LABIP), was held in Amsterdam in 2017 to discuss these challenges; a number of conclusions and recommendations were formulated and subsequently published (Johansen, 2017; http://rdcu.be/CZxF). These issues and the outcome of the workshop will be described in more detail.

[O19] CHALLENGES FACING THE MICROBIAL FOOD CULTURE INDUSTRY

Paul Tenning[1]
[1]European Food and Feed Cultures Association (EFFCA)

Today, the microbial food culture industry faces the major challenge of securing certainty and clarity in terms of legislation set, both in the EU and worldwide.

This session will elaborate on the key objectives of EFFCA (European Food & Feed Cultures Association) such as, supporting the growth and promotion of use of food cultures at global level. Facilitation of dialogue between economic operators, regulatory bodies and other relevant stakeholders with regards to the applications of food cultures, is just one such way this is accomplished.

This session will also examine other possible solutions to resolve such issues as, shortcomings of the Nagoya Protocol, impact of the Ohne gentechnik legislation in Germany, potential future legislation on protective cultures, expansion of the QPS list and import restrictions of cultures in various jurisdictions.

[O20] DEVELOPING SCIENCE AND TODAY'S REGULATIONS

Lisa Jensen[1]
[1]*Association of Manufacturers and Formulators of Enzyme Products (AMFEP)*

Today, an increasing speed of new scientific advancements is being experienced worldwide. The enzyme industry is witness to the struggle felt by regulatory bodies and safety assessors to keep up with such progress.

This session will elaborate on how AMFEP, the association of manufacturers and formulators of enzyme products, see this issue and what efforts can be made to lessen the natural consequential additional burden of scrutiny met from authorities.

Lisa Jensen, will look to answer questions such as;
• What are the current difficulties for the European Food Safety Authority?
• Development of QPS and what is its impact on the enzyme industry?
• How is the European Commission coping with new science?

[O21] SAFETY DEMONSTRATION OF MICROBIAL FOOD CULTURES. THE IDF EFFCA INITIATIVE

François Bourdichon[1]
[1]*Food Safety and Hygiene Consultant*

Fermentation, as a process for manufacturing foods, has traditionally been used to preserve perishable products and enhance their nutritional value. Fermented foods are typically associated with local and traditional food consumption. They result from the action of microorganisms causing significant and desirable modifications to the food matrix, through biochemical changes. The growing body of evidence with regard to microorganisms and their ecological role in the food matrix has led to industrial applications of processes of fermentation starting in the early twentieth century through the use of specific dedicated microbiota with various levels of characterization.

Until recently, the safety of fermentation processes and the microorganisms employed has neither been questioned nor regulated. In recent decades, the fermentation processes have directly or indirectly come under various regulatory frameworks in many countries. Several of these regulatory frameworks put emphasis on "the history of use", "traditional food", or "general recognition of safety".

The evaluation of "positive" microorganisms cannot be conducted through a "classical" microbiological risk assessment (MRA) as used for the "negative" microorganisms – i.e. pathogenic species. FIL-IDF in collaboration with EFFCA has proposed a specific assessment for microbial species with a known implication in fermentation in all types of food matrices both in traditional and industrial food products worldwide.

[O22] ROLE OF THE QUALIFIED PRESUMPTION OF SAFETY CONCEPT IN THE EFSA RISK EVALUATIONS

Lieve Herman[1,2,3], **Marianne Chemaly**[2,3], **Pier Sandro Cocconcelli**[3], **Pablo Fernandez**[3], **Luisa Peixe**[3], **Miguel Prieto Maradona**[3], **Amparo Querol**[3], **Juan Evaristo Suarez**[3], **Ingvar Sundh**[3], **Just Vlak**[3] and **Sandra Correia**[4]

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[2]*EFSA Panel on Biological Hazards (BIOHAZ)*
[3]*EFSA Working group on QPS (BIOHAZ)*
[4]*EFSA BIOCONTAM Unit*

EFSA is an agency of the European Commission (EC) to provide advice on issues related to safety of food for animals and man. A wide variety of microorganisms are intentionally used at different stages of the food chain and are risk assessed in several EFSA areas e.g. feed, food, pesticides, nutrition, on the basis of an application dossier to the EC. The qualified presumption of safety (QPS) assessment was developed to provide a harmonised generic safety pre-assessment to support the risk assessments performed by EFSA's scientific panels. The safety of unambiguously defined biological taxonomic units and their body of knowledge are assessed. Identified safety concerns for a taxonomic unit (TU) are, where reasonable in number, reflected as 'qualifications'. Possible qualifications of QPS microorganisms need to be evaluated by the EFSA Unit with the information provided in the respective dossier.

The main areas where QPS is applied in the EFSA risk assessment are production organisms for feed and food additives. QPS recommended microorganisms in these areas are still requiring an assessment based on an individual data package but are favoring a fast track evaluation with less requirements in relation to the production organism. The data required in each application has to confirm the unambiguous identification of the organism and the confirmation that the qualifications are met. If the microorganism used in the production of a food enzyme has a QPS status, the food enzyme application does not need to provide specific toxicological test data (Commission Implementing Regulation (EU) No 562/2012).

The lowest taxonomic unit (TU) for which the QPS status is granted is the species level for bacteria and yeasts, and families for viruses. In the QPS Statement published on January 2018 (EFSA Journal, http://onlinelibrary.wiley.com/doi/10.2903/j.efsa.2018.5131/epdf) the possibility for genetically modified microorganisms to get the QPS status was introduced when these microorganisms are used for production of food enzymes, food additives and flavourings and feed additives.

There are important differences between the GRAS system (Generally Recognised As Safe), established by the FDA (Food and Drug Administration) in the United States and the QPS system. An important difference is that a QPS assessment is only triggered by receipt by EFSA of an application dossier seeking a market authorisation of EU regulated products. It is not possible for a putative applicant to ask directly to EFSA for a QPS evaluation of a TU. GRAS system only evaluates at strain level in the case of microorganisms, higher taxonomic units are not considered.

Based on the actual body of knowledge and/or the ambiguous taxonomic position, some TUs are excluded from the current QPS assessment, such as filamentous fungi, bacteriophages, Enterococcus faecium, E. coli, Streptomyces spp. and Oomycetes.

The QPS-granted TUs are evaluated every six months in a Statement published in the EFSA journal, which also contains the results of extensive literature searches (ELS) on the existing QPS granted organisms, which may lead to any possible change in the QPS status or qualifications. Each 3 years a new QPS opinion is published with an update of the QPS process, the QPS list and the results of the 3-year ELS on the QPS TUs. For the update of the QPS opinion in 2019, the role of whole genome sequencing, resistance to antimicrobial and antimycotic agents and the access of third parties to the QPS notification are actually in discussion within EFSA.

[O23] HYDROLYSIS OF MICROBIAL TURNOVER PRODUCTS IN THE GUT: A CASE STUDY IN BROILER CHICKENS

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The microbiota in the gastrointestinal tract lives in a complex ecosystem in equilibrium with the host and the feed components. There, the microbial turnover (replication and death) naturally produces a diversity of microbial cell components that are released into the gut lumen. It was observed that *in vivo* supplementation of a fungal muramidase, that degrades the peptidoglycan component of microbial cell wall fragments (or bacterial cell debris), not only was well tolerated in high amounts, but also improved performance parameters in broiler chickens. This observation prompted us to initiate *in vitro* studies of muramidase hydrolysis of purified peptidoglycans from bacteria with a significant abundance in the gastrointestinal tract of broiler chickens. Both, reducing end assay and mass spectroscopy data, confirmed that the supplemented muramidase could hydrolyze gut relevant peptidoglycan. Interestingly, mass spectroscopy data showed differences in the composition of peptidoglycan hydrolysis products among the different bacteria. Furthermore, a peptidoglycan recognitions protein-based *in vitro* assay demonstrated that the muramidase significantly hydrolyzed the peptidoglycans, observing a reduction in the recognition response.

[O24] REFINING PROTEINS FROM GREEN CROPS USING LACTIC ACID FERMENTATION AND OBTAINING HIGH QUALITY FEED PRODUCTS FOR ANIMALS

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[1] Aalborg University

Green biorefinery concepts have the potential to become a suitable solution for production of organic protein-rich feeds from green crops. Different green crops such as alfalfa, red clover and clover grasses are studied as possible feedstocks for the development of an organic biorefinery system with refined proteins, ensiled press cake to be used as cattle feed and residual juice with potentials for fermentation of amino acids, lactic acid or other uses. A process was developed by which screw pressed juice from the freshly harvested crop was used for lactic acid fermentation to decrease the pH and precipitate proteins. These refined proteins have a favourable amino acid content and are comparable with soy proteins, especially for poultry where methionine is a limiting amino acid. For production of experimental feed for different animal trials, a demo-scale set up was made for a continuous refining process, which included harvesting and processing of 400 tons clover grass. The clover grass was processed into a protein concentrate, a fiber-rich press cake, and a residual stream of soluble nutrients. The protein-concentrate was used in experimental feed formulations for egg-laying hens, broilers and pigs. The fiber-rich press cake was ensiled without additives, and used as feed for dairy cows. The novel protein refining technique using lactic acid fermentation showed robust results in large scale. Furthermore, the feed trials in mono-gastric animals (poultry and pigs) show promising results although the digestibility with increasing amount of green proteins show a decreasing trend. In addition, the ensiled press residues surprisingly resulted in a 5-10% increase in milk production compared with a traditional silage. Further improvements of the technology is expected to lead to feed products with higher digestibility as well as protein products for human consumption.

[O25] LACTIC ACID BACTERIA FOR EFFICIENT DELIVERY OF VITAMIN K2

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Vitamin K is a fat-soluble vitamin that is essential to human health. It acts as an enzyme co-factor required for the activation of proteins involved in vital physiological processes. Vitamin K exists in two forms: vitamin K1 (phylloquinone) which is derived from plants and vitamin K2 (menaquinone) which is of bacterial origin. Intake of vitamin K2 was recently reported to be associated with a reduced risk of coronary heart disease. Vitamin K2 shows advantages over vitamin K1 in terms of health benefit and bioavailability in human body. Therefore, vitamin K2 fortified food products are highly relevant for a healthy human diet.

Lactic acid bacteria (LAB) are key players in various food fermentation processes and some strains produce vitamin K2 which accumulates in the bacterial cell membrane. Optimization of vitamin K2 production in LAB and its delivery to the human host have not yet been explored extensively. Emerging evidence of extracellular membrane vesicle (MV) production in Gram-positive bacteria led us hypothesize that LAB also secrete MVs, which potentially provide a novel method to improve the delivery of the lipid soluble vitamin K2 to the human body.

In this study, we focus on the exploration and exploitation of high vitamin K2 producing LAB which secrete MVs, with the aim to obtain generic knowledge of the MV production process and enable efficient delivery of vitamin K2 to the human host. To this end, we examined a series of *Lactococcus lactis* strains for their natural ability to produce vitamin K2. Different cultivation durations, temperatures, carbon sources, metabolic modes, etc., were applied to *L. lactis* subsp. *cremoris* MG1363 and the effects of fermentation conditions on vitamin K2 production were determined. We demonstrate that strain selection and manipulation of fermentation conditions contribute to natural enrichment of vitamin K2. Furthermore, we provide evidence of MV production in vitamin K2 producing strains, and confirm that vitamin K2 indeed accumulates in the MVs. This finding is a proof of principle that MVs could serve as efficient delivery vehicles of vitamin K2.

[O26] DEVELOPING LACTIC ACID BACTERIA FOR THE CONVERSION OF BROWN MACROALGAE TO GREEN CHEMICALS AND FUELS

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Microbial conversion of biomass plays a major role in establishing a bio-based economy, which aims at replacing fossil resources with renewable substrates for the production of fuels and chemicals. Current efforts in using non-edible ('second generation') biomass rather than food-derived sugars focus on lignocellulosic materials such as crop residues and non-edible plants. However, lignin is often toxic to the production organism and hard to eliminate, and economically feasible conversion of cellulose and hemicellulose is still challenging. An attractive alternative includes brown macroalgae or sea weed, which do not contain lignin, do not require fresh water, are not a major food source, and contain a higher sugar fraction. The main sugars include mannitol, laminarin (glucose) and alginate (guluronate and mannuronate). We focus on using metabolic engineering and laboratory evolution of Lactic Acid Bacteria (LAB) for the conversion of brown macroalgae into green chemicals and fuels. To select the best-suited production platform, we screened *Lactobacillus* and *Pediococcus* strains for growth in sea weed hydrolysate and traits like genetic accessibility, substrate utilization and several stress tolerances[a]. The screening resulted in selection of *Lactobacillus reuteri* for further work, in which the first step is the construction of a metabolic model. The constructed metabolic model provides insight in the metabolism of the strain, as well as into the envisioned engineering strategies. Most microorganisms, including LAB and *Lactobacillus reuteri*, do not naturally utilize alginates and hence the introduction of these pathways will be the first step in engineering the selected strain.

[a] E.F. Bosma, J. Forster & Alex T. Nielsen. Lactobacilli and pediococci as versatile cell factories – Evaluation of strain properties and genetic tools. Biotechnol. Adv., accepted.

[O27] A SYSTEMS APPROACH TO GENERATING SUPERIOR INDUSTRIAL YEASTS

Kevin Verstrepen[1]
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The common brewer's yeast Saccharomyces cerevisiae is used in a broad range of industrial applications, from the production of beer, wine and bread to biofuels and pharmaceuticals. Interestingly, there are hundreds of different industrial yeast strains, but their origins and specific characteristics are largely unknown. We combined large-scale phenotyping with genome sequencing to track the genealogy and evolution of today's industrial yeasts. Using this knowledge allowed us to set up large-scale breeding and directed evolution programs to generate superior variants that increase production efficiency and expand the range of yeast-derived products and aroma's, allowing more efficient fermentation, production of superior and novel products.

[O28] THE USE OF PICHIA KLUYVERI YEAST FOR THE PRODUCTION OF LOW ALCOHOL AND NON-ALCOHOL BEER

Sofie Saerens[1]
[1]*Chr. Hansen A/S*

The production of low- and non-alcoholic beer has been increasing worldwide, mainly due to the increasing demand for healthier food and beverages.

At Chr Hansen A/S, a highly innovative patented technology was developed to overcome the drawbacks of the currently used methods, where a normal alcohol beer is produced first and then ethanol is removed afterwards. The technology is based on the findings that a highly aromatic yeast species, Pichia kluyveri, is capable of producing a high level of desirable flavor compounds from the monosaccharides present in the wort with a very limited ethanol production.

[O29] BACTERIOPHAGES FOR THE WINE INDUSTRY: A WAY TO A HEALTHIER WINE

Ifigeneia Kyrkou[1], **Alexander Byth Carstens**[1], **Lars Hestbjerg Hansen**[1]
[1] *Aarhus University; Department of Environmental Science*

The security of inoculating with commercial wine starters instead of allowing spontaneous malolactic fermentation (MLF) to develop is gradually becoming more recognised by winemakers. Nevertheless, wines inoculated with starters may also fail. Bacteriophages are notorious for being able to persist in the processing environs, until a suitable host is available to infect. Thus, phages can cause severe problems in fermentations where the same cultures or culture mixes/rotations are repeatedly applied. In 1985, the first isolation of *Oenococcus oeni* phages from wine with undesirable MLF properties was reported. Since then, only a moderate amount of papers have been published on oenophages. Similarly, no phages of the wine related strains of *Lactobacillus plantarum* have been described so far, given that commercially available wine starters of the species are relatively recent. However, many phages of *L. plantarum* have indeed been isolated from other environments (vegetable juices, silage, etc.).

In our study we have examined must, malolactic fermentation and wine samples originating from various countries for the isolation of phages against *L. plantarum*, a probiotic recently characterized as "the new generation starter of MLF". Other environmental samples, which included wastewater and household waste, were also investigated for phages. Sixteen novel phages, the first ever to be reported against *L. plantarum* wine strains, have already been isolated and sequenced, and are currently being characterized by our group. Sequence comparisons have shown low degree of resemblance between already reported phage genomes and nine of our phages. Moreover, those nine phages clustered together forming two new genera.

We support that our phages could serve as valuable biocontrol tools for the winemakers to prevent growth of *L. plantarum,* when MLF is undesirable or needs to be controlled to get a different organoleptic result. Moreover, addition of those phages could replace the disputed and potentially unhealthy practice of sulfating- used so far to stop the MLF and stabilize the wine- and move towards a 100% organic wine product.

[O30] THE CHALLENGE OF LACTIC ACID BACTERIA TO METABOLIZE PHENOLIC COMPOUNDS IN ELDERBERRY JUICE

Annalisa Ricci[1], **Martina Cirlini**[1], **Luca Calani**[1], **Daniele Del Rio**[1], **Erasmo Neviani**[1], **Gianni Galaverna**[1], **Camilla Lazzi**[1]
[1] *University of Parma*

Lactic acid bacteria (LAB) metabolism may improve the bioavailability and bioactivity of phytochemical compounds putatively involved in human health. Specific strains of LAB, thanks to their enzyme portfolio, were able to metabolize specific phenolics. This ability has a dual significance, as metabolism of phenolic substances represents a strategy to detoxify them, that could affect the integrity of membrane and cell wall, and, it can exert benefits on human health, possibly due to the action of metabolic end-products. In this study, 15 strains belonging to different species of LAB (*Lactobacillus plantarum, Lactobacillus casei* and *Lactobacillus rhamnosus*) were used for elderberry juice fermentation in order to follow the conversion of elderberry phenolic compounds. To this aim, bacterial cell number and phenolic profile were studied before and after fermentation. Due to low pH and high concentration of phenolic compounds, not all strains were able to grow after 48h of fermentation. Most of the tested strains (all *L. plantaraum* and *L. rhamnosus* and three strains of *L. casei*) were able to grow of about two Log cycles, whereas two strains of *L. casei* did not show the same capacity. Total polyphenolic compounds in juice were increased after the addition of the starter, even those unable to grow. Hydroxycinnamic acids were subjected to bacteria metabolism, and, in particular, some strains of *L. plantarum* were able to convert caffeic acid into dihydrocaffeic acid. The highest increase in phenyllactic acids was observed when *L. plantarum* strains were used as starter for fermentation, but also *L. rhamnosus* and *L. casei* showed the ability to produce these metabolites. Quercetin-3-*O*-rutinoside was the flavonoid glycoside that showed the highest increase after fermentation. Anthocyanins, especially cyanidin-3-*O*-glucoside and cyanidin-3-*O*-sambubioside, increased in a strain-specific way, independently of bacterial growth ability. With this study, we described the effect of LAB on phenolics during elderberry juice fermentation, highlighting that compounds could be produced (phenyllactic acids), modified (hydroxycinnamic acids) or increased (flavonoid glycosides and anthocyanins), and showing that this potential is not linked to the growth ability.

[O31] DEVELOPMENT OF NOVEL CHYMOSINS FOR CHEESE PRODUCTION

Christian Jäckel[1]
[1]*Chr. Hansen A/S*

Chymosins initiate milk coagulation in cheese manufacturing by cleaving off the glycomacropeptide (GMP) from the surface of casein micelles. We applied a combination of various protein engineering strategies to generate variants of chymosin from Camelus dromedarius (camel chymosin) with greatly increased specificity for the milk clotting activity over non-specific proteolysis of milk proteins. The newly designed coagulants not only allow the production of cheese with better texture and taste, but also reduce the amount of milk needed for cheese manufacturing.

[O32] ENZYMES IN FRUIT PROCESSING

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[2] *WeissBioTech GmbH*

Enzymatic fruit processing is a typical industrial enzyme application in the food industry. Challenges arrive from the treatment of a natural material on the one hand and the benefits for quantity and quality of a product on the other. The right selection and application of enzymes as processing aid for that application is crucial for successful enzyme use.

Fruit cells are a complex, but well organized, mixture of four well known natural constituents: starch, cellulose, hemicellulose and pectin. Enzymatic treatment during fruit processing offers the opportunity to selectively treat a key component of the fruit at a defined moment in the processing that is appropriate for a desired effect on product quality or yield.

Enzyme application during fruit processing will be discussed and the effect on product yield and product type and quality will be shown in selected examples.

[O33] CASSAVA NON-STARCH POLYSACCHARIDE COMPOSITION & DEGRADATION BY CARBOHYDRASES

Larissa Staack[1], **Eduardo Antonio Della Pia**[1], **Marie Krogsgaard**[1], **Dan Pettersson**[1], **Ninfa Rangel Pedersen**[1]
[1] *Novozymes A/S*

Cassava (*Manihot esculenta*) is a crop native of Amazon that is cultivated mainly for its starchy tubers. Importance of cassava in agriculture is steadily raising; in the last four decades, its global production increased from 114 to 277 million tons per year (FAOSTAT, 2017). A better understanding on cassava cell wall structure could be relevant to decrease its antinutritional effect and improve its application as energy source in monogastric animal feed through treatment with non-starch polysaccharide (NSP) enzymes.

In this project, NSP monosaccharide composition of four cassava samples from different countries in Southeast Asia was studied. Furthermore, the efficacy of RONOZYME® VP (DSM Nutritional Products, Basel, Switzerland) in solubilizing cassava NSP was quantified. RONOZYME® VP is a multicomponent NSP enzyme produced through fermentation of *Aspergillus aculeatus*. It contains different pectinases and hemicellulases, including β-glucanase, which activity is stated to be 120 FBG/ml (fungal β-glucanase units per milliliter).

Analytical chemistry assays showed that the cassava composition varies between samples. The amount of starch fluctuates between 67 and 80%, total protein from 2 to 4% and total fiber from 3 to 6% of total dry matter content. *In vitro* experiments also suggested that, apart from cellulose, pectin is the main polysaccharide present in the cassava cell walls. Neutral sugars determination by gas chromatography indicated that cassava NSP contains an average of approximately 50% glucose, 30% galactose, 9% xylose, 8% arabinose and 3% rhamnose.

After incubation with RONOZYME® VP at 400 ppm (4h, 40˚, pH 5) about 10% of cassava total fiber content was solubilized compared to a control treatment. Despite considerably different fiber composition, RONOZYME® VP performance was alike on different cassava samples, with significant solubilization of galactose, arabinose and rhamnose. Confocal microscopy imaging supports the *in vitro* results. Cassava slices treated with RONOZYME® VP at 200 ppm and stained with Coriphosphine show degradation of cell walls and consequent release of trapped starch granules.

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[O34] CAMEL CHYMOSIN FOR CAMEL CHEESE MAKING PROPERTIES

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[4] *National Food Institute, Technical University of Denmark*

The composition of camel milk protein differ from that of other species and casein micelles from camel milk have a larger average diameter (~380 nm) than bovine casein micelles. Camel milk has a smaller proportion of κ- casein (3.5 % of the total casein) and relatively much more β-casein (65 % of total casein) than bovine milk, where the proportions are 12 % and 33 %, respectively. These differences in casein composition, together with the absence of an enzyme with suitable milk clotting activity has curtailed the opportunity of processing camel milk into various value added products such as cheese. However, camel chymosin is now available as coagulant in the form of Chy-Max®M (Chr. Hansen A/S, Denmark). The effects of pH, temperature, enzyme concentration, and $CaCl_2$ addition have been widely investigated for rennet induced bovine milk gelation process; whereas, the gelation process of camel milk is much less studied. The present work therefore, was undertaken to evaluate the effects of temperature, pH, concentration of camel chymosin and addition of $CaCl_2$ on the hydrolysis of κ-casein and the coagulation kinetics of camel milk. The rate of κ-casein hydrolysis was higher at 40 °C than at 30 °C and with increasing addition of chymosin and decreasing pH. Gelation was initiated at levels of camel milk κ-casein hydrolysis > 95 %. The gelation time (T_g) of camel milk was significantly reduced (from 717 to 526 s) at 30 □ when the concentration of chymosin was increased, but was independent of chymosin concentration at 40 □. Reducing pH also reduced T_g. The gel firmness increased at 40 °C (58 Pa) compared with 30 °C (44 Pa) and effect of $CaCl_2$ addition on the gelation properties of camel milk was found to be dependent on pH; a significant improvement was only found at pH 6.3.

Keywords: *Camel Chymosin, Camel Milk, Gelation, Kinetics*

[035] UPGRADING OF WHEY AND OTHER FOOD SUPPLY CHAIN WASTE

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Food supply chains produce large amount of wastes. Many of them represent an environmental burden, e.g. rice straw or whey. All of them could be updated to valuable bioproducts or biomaterials (not only in the food sector) thus increasing the profit of the farmer or the processor. Moreover the European vision for 2050 strongly pushes towards innovative and circular economic models where nothing is wasted and natural resources are managed sustainably.

To update food supply chain wastes we have to identify the class of chemicals they contain and then to design a valorization pathway by preserving the structure and activity of the molecule or transforming in a better performing one. Vegetable oils and fats are present in very many residues, from rendering fats to spent coffee grounds. They can be used as raw materials for the production of a wide range of products and materials such as biofuels, lubricants, surfactants, additives for cosmetics and functional foods, adhesives and thermoset resins. However, they suffer from low to very high acidity, therefore innovative processes are needed.

Moreover, an integrated approach can be adopted in order to valorize not only one, but all the component of a waste and to obtain products with different added value. As an example from rice bran we can extract the oil and valuable phytosterols, we can use the oil to produce monoglycerides or to obtain esters with the phytosterols. The last one are very useful in the formulation of functional foods to lower the level of cholesterol in blood but are also promising as agents against multidrug resistance. After extraction of oil and phytosterols we still have proteins in defatted bran. They have very good nutritional properties and can also be hydrolyzed to obtain products with biological, functional and sensorial properties. A similar approach could be used to valorize brewery wastes.

As far as whey is concerned, lactose in the permeate after separation of the proteins can be converted in one step to sorbitol and dulcitol, two sugar alcohols that can be used as sweeteners but also as monomers for the production of Bio-plastics. Delactose permeate has shown to be effective in antimicrobial treatment to increase the shelf life of fruit and vegetables.

[036] VALUE ADDED PRODUCTS FROM DAIRY WASTE

Peter Ruhdal Nielsen[1]
[1] *Technical University of Denmark*

Dairies producing cheese and certain other fermented milk products generate side streams rich in lactose and other nutrients, such as whey, whey permeate, etc. Depending on size and location, different dairies have different solutions and applications of their side streams, but still today many dairies have underutilized, low value side streams containing significant amounts of sugar.

In our research group we have developed a range of cell factories based on modified lactic acid bacteria which can efficiently use these resources for production of value-added products. In this talk I will discuss examples of our work including production of high value chemicals and butter flavor.

I will also discuss a newly started spin-out company from DTU Food – AlcoWhey. The aim of this company is, together with dairies and distilleries worldwide, to implement a new bacterial technology for producing high quality potable alcohol from residual whey side streams.

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[037] PHAGE THERAPY AGAINST SOFT ROT ENTEROBACTERIACEAE IN POTATOES UNDER SIMULATED STORAGE CONDITIONS

Amaru M. Djurhuus[1], **Alexander Byth Carstens**[1], **Lars Hestbjerg Hansen**[1]
[1] *Aarhus University; Department of Environmental Science*

Soft Rot *Enterobacteriaceae* (SRE) (*Pectobacterium spp.* & *Dickeya spp.*) are important pathogens of many important crops. Amongst these crops, is the world's most important non-grain crop potato (*Solanum tuberosum L*). Around 22% of potatoes are lost annually, due to viral, bacterial, fungal and pest attacks, with SRE alone estimated to account for 30-50% of this loss[1]. The losses caused by SRE take place both in field, where SRE gives rise to the disease potato blackleg, and post-harvest during storage, as soft rot. Here, we emphasize the potential of bacteriophage (phage) therapy as a means of biological control of SRE in potatoes. Phages have shown great potential to be used against plant pathogens, both to treat or prevent diseases in the field but also to increase shelf-life of green produce[2]we have learned that phages can constitute a promising alternative in the food industry to eliminate bacterial pathogens from seedlings in greenhouse and field environments, as well as from fresh-cut food products. The fruit and vegetable industry requires quite a different approach than the meat or dairy industry. Several factors can inhibit efficacy of phage treatment such as plant watering or washing ready-to-eat products (water may dilute therapeutic doses. Phages are inherently non-toxic for humans and the environment. Consequently, phages can be used in organic agriculture as well as post-harvest both in storage and on retailer shelfs, unlike conventional treatments such as pesticides and copper sprays.

As there are currently no effective ways of combating SRE, we sought to develop an approach, which could easily be applied in the potato production pipeline. To this end, more than 70 phages were isolated infecting prominent SRE pathogens and thoroughly characterized using next-generation sequencing and relevant biological attributes including growth dynamics, host range and virulence genes. A subset of phages was selected and included in a phage cocktail, which was applied in a proof-of-principle experiment, to treat Soft Rot in potatoes under *in vivo* conditions mimicking potato storage conditions.

Elucidating the interactions occurring between phages and their bacterial host in this *in vivo* setting can contribute to our knowledge of these interactions in a more authentic environment and could help to pave way for more a more informed approach to phage therapy in general.

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[038] PRODUCTION OF NUTRIENT-RICH YEAST FROM WOOD-DERIVED SUGARS

Ricardo Ekmay[1], Alex Berlin[1]
[1] *Arbiom Inc.*

The expected growth in the world's population and the middle class will result in a doubling of the global protein demand over the next 30 years. However, natural resources are being strained in the current food production systems. To meet society's future protein demand, we must gain efficiencies by producing more food requiring fewer natural resources, as well as tap into previously under-utilized resources for food creation. Historically, torula yeast products were produced utilizing spent liquor from sulfite pulp mills. However, this approach fell out of favor as these mills closed and as the price of sugars, such as molasses, dropped. Presently, in North America and Europe, wood stocks are increasing faster than demand for wood, representing an abundant and sustainable source of carbon. Additionally, the global forest products & paper industry has core competencies in sourcing and processing a long-term, renewable biomass supply, with mature supply chains and sizeable asset base, presenting an opportunity to apply technology to unlock new value creation opportunities, such as the production of feed and food.

Arbiom's processing and fermentation technology enables an economically viable production pathway from wood to food. Arbiom's technologyutilizes wood biomass, e.g. sawdust, to create distinct 5- and 6-carbon sugar-rich streams for the production of torula yeast, or other fermentation applications. Further, Arbiom has developed a novel *Candida utilis* strain that has high overall nutrient bioavailability and increased levels of key essential amino acids. The amino acid profile of the enhanced torula yeast makes it a favorable substitute for conventional high-protein sources, such as fish meal or soy protein concentrates based on digestible indispensable amino acid score. The result is a natural, traceable and ecologically-friendly solution to meet global protein production challenges.

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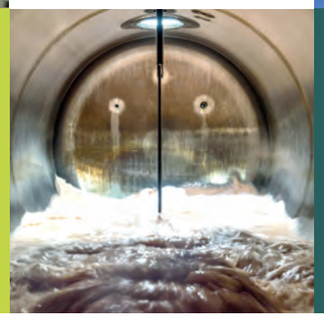

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[P1] A LINK BETWEEN GUT MICROBIOTA COMPOSITION AND ALCOHOL CONSUMPTION HABITS

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The dynamics and diversity of human gut microbiota are constantly changing through the host's lifetime in response to various factors. Among those, diet is considered as one of the key determinants in altering the microbial composition. Due to its geographical location, a dietary pattern of most Slovenian people follows the guidelines of the Mediterranean diet. Such dietary pattern consists of a high variety of vegetables and fruits, legumes and whole grains, olive oil as a main source of fat and is coupled with a moderate consumption of red wine. The aim of our study was to find a linkage between the gut microbiota composition and the alcohol consumption habits of participants.

A cross-sectional study of gut microbiota composition was performed on 141 healthy individuals from Slovenia, Europe. The collected fresh stool samples were diluted in PBS, homogenized and shipped on dry ice to Spanish Institute of Food Science, Technology and Nutrition, where they were stored at -80°C until analysis. For the assessment of the microbial community composition, group specific primers for accurate quantification of several major bacterial groups from faecal samples were assayed using quantitative PCR.

By gender, there was 33,3 % men (n=47) and 66,7 % women among the participants. The data showed a significant difference ($p \leq 0,05$) in the microbiota composition between genders; with a greater abundance of Bacteroidetes and Proteobacteria phyla in women and a greater abundance of Firmicutes phylum, *Ruminococcus*, *Caloramator* and *Clostridium* genera in men. On the basis of questionnaire, there were 13,5 % individuals (n=19) that did not consume alcohol and 86,5 % (n=122) individuals that consumed alcohol occasionally (n=24) or habitually (n=98). A significant difference ($p \leq 0,05$) in the abundance of *Bifidobacterium* and *Lachnospira* genera was shown, with a lower proportion of both in the individuals with habitual drinking habits.

In summary, gut microbiota composition was linked to the gender and alcohol consumption habits of the participants from Slovenia. Based on these results, a further research work of microbiota composition and its connectivity to dietary pattern will be established.

[P2] A PROTOCOL WITH INCREASED THROUGHPUT FOR EXTRACTION OF INFECTIVE VIROMES FROM LOW VOLUME SAMPLES SUITABLE FOR METAVIROME SEQUENCING

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During the last decade it has become evident that the gut microbiome (GM) has profound influence on human health and disease. The GM contains approximately as many viral particles as prokaryotic cells and the majority of these viruses are bacteriophages (bacterial viruses, in short phages). The role of phages in shaping the GM remains poorly investigated but there are strong indications that they play important roles in shaping the GM early in life and in the gut bacterial dysbiosis associated with inflammatory bowel disease flares.

Based on the Copenhagen Prospective Studies on Asthma in Childhood 2010 (COPSAC2010) cohort it has recently been found, that the development of the GM during the first year of life can impact the development of childhood asthma[1]. With the aim of characterizing the gut virome of the COPSAC2010 cohort children at 1 year age, we have developed a protocol with increased throughput enabling extraction of infective viruses from low volumes of fecal sample input yielding viromes of a purity well-suited for down-stream high throughput sequencing based investigations.

The extraction protocol is based on size selection of the phages through different filters, which minimized the biases as well as the loss of phages that would be introduced by other treatments during extraction. Phages T4 (*Myoviridae*), c2 (*Siphoviridae*), Φ29 (*Podoviridae*) and ΦX174 (*Microviridae*) were spiked into fecal samples (from other 1-year infants) and their recoveries at the different steps of the protocol were confirmed by plaque-assays. The average recovery rates for infective phages were approx. 37%, 63%, 59% and 29% respectively. After the virome isolation, the viral DNA was extracted, amplified by MDA to include the sequencing of ssDNA virus and finally shot gun sequenced. The project leads to the construction of the biggest gut metavirome dataset generated to date. Further, the phages extracted are still infective and able to infect and lyse their bacterial hosts therefore enabling establishment of phage-host pairs, which might prove useful for future phage-based modulation of gut microbial communities.

1 Stokholm, J. *et al.* Maturation of the gut microbiome and risk of asthma in childhood. *Nature communications* **9**, 141, doi:10.1038/s41467-017-02573-2 (2018).

[P3] MICROGUT: AN IN VITRO MODEL OF THE HUMAN GASTROINTESTINAL TRACT

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Objective: To design, develop and validate an automated microfluidics-based dynamic model of the human gastrointestinal tract allowing comprehensive recapitulation of the human gut microbiota along with compatible downstream high resolution molecular omic analyses.

Method and Results: The microGUT model is an automated desktop-sized microfluidics-based model of the human gastrointestinal tract (GIT). Contrary to the traditional fermentors-based *in vitro* gut model, microGUT provides unique features ensuring comprehensive recapitulation of the gut microenvironment as well as downstream compatibility with omic analyses. The microGUT allows cultivation of complex gut microbiota encompassing mucosal and luminal strains in conditions representative of the GIT physiology. Salient features of microGUT model are personalized passage times, automated sampling and control, continuous real-time oxygen and pH monitoring and control across the GIT and volatile gas sensing, among others. The microGUT control board was capable on controlling the GIT transit time from 12-72 hours via a control software interface. The sampling ports integrated in luminal bioreactors allowed continuous sampling of the luminal microbiota. The adherent mucosal microbiota was sampled as an end-point analysis. The microGUT model was validated by cultivation of pooled fecal inocula of 3 healthy donors including over 600 strains for 2 weeks. Resultant microbial community dynamics across the GIT were analysed using 16S rRNA gene amplicon high-throughput sequencing. Resultant mixed microbial community contained 592 strains sustained over 2 weeks (~92%) and showed region specific variations in the microbial community composition.

Conclusion: The automated microGUT platform provides a significant advancement to existing *in vitro* gut fermentation models. microGUT enables comprehensive recapitulation to the human GIT microbiota *in vitro*.

[P4] MODULATION OF THE GUT MICROBIOTA BY PECTINS IS RELATED TO THEIR STRUCTURAL PROPERTIES

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Background. Pectins are indigestible polysaccharides produced from fruits and vegetables. Like other dietary fibers, pectins can be digested by the gut microbiota exhibiting beneficial properties for the host. Knowledge of the impact of pectins on microbiota composition is insufficient and limited only to a few types of pectins. This study investigates the structure-function relationship of pectins by studying their potential to modulate the gut microbiota in a beneficial way. For this purpose, dynamics of microbiota composition and activity was studied in fermentations of nine structurally diverse pectins and a pectic derivative rhamnogalacturonan I (RGI) using a TIM-2 colon model. Composition of the fecal microbiota was assessed by 16S rRNA gene sequencing using Illumina NextSeq technology.

Results. Relative abundances of numerous bacterial taxa were affected by pectins, with major shifts observed within the prevalent families *Ruminococcaceae*, *Lachnospiraceae*, *Prevotellacea* and *Bacteroidaceae*. Besides, pectins promoted growth of rare taxa, belonging to *Enterococcaceae* and *Enterobacteriaceae* families. High similarity in microbiota profiling was determined for a group of high methylated pectins from citrus fruits, low methylated amidated pectins and between sugar beet pectin and RGI. Production of short chain fatty acids (SCFA) propionate and butyrate was largest in fermentations of sugar beet pectin and RGI. The levels of SCFA were not significantly different between the pectins from citrus fruits, indicating functional redundancy of microbiota. The main structural features linked to pectin-specific effects on the composition of microbiota and production of SCFA included degree of methylation, composition of neutral sugars, distribution of homogalacturonan and rhamnogalacturonan fractions, degree of branching and the presence of amide groups. These structural parameters correlated significantly with abundances of various bacteria associated with human health. Among them, *Faecalibacterium prausnitzii*, *Coprococcus*, *Ruminococcus*, *Dorea*, *Prevotella copri*, *Sutterella*, and *Bacteroides* spp. were either increased or decreased depending on the substrate, suggesting that these populations can be controlled using structurally different pectins.

Conclusions. This study contributes to the understanding of structure-function relationship of pectins in the gut. The knowledge can be used to develop pectin-based diets, targeting beneficial gut bacteria and favoring more balanced microbiota profiles.

[P5] DO DIFFERENT DOSES OF A NOVEL BACILLUS SUBTILIS CHCC16872 PROBIOTIC STRAIN INFLUENCE PERFORMANCE DATA?

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The objective of this study was to investigate the effect of different doses of a novel probiotic strain on performance parameters in broilers recovering from necrotic enteritis (NE) caused by *Clostridium perfringens*. The experiment consisted of 1920 Day of hatch Cobb 500 male chickens distributed in 48 pens starting with 40 male broiler chickens per pen. The treatments were replicated in eight blocks, randomized. Feed and watering were performed *ad libitum*. Means for live weight, weight gain, feed consumption, feed conversion ratio (FCR), NE lesion scores and % NE mortality were calculated.

On days 19, 20, and 21 all pens except group 1 were challenged with a field isolate of *C. perfringens* originating from a commercial broiler and known to cause NE. Fresh inoculum (approximately 10^9 CFU/pen) was used and each pen received the same amount administered by mixing *C. perfringens* into the feed. On day 21, three birds from each pen were selected, sacrificed, group weighed and examined for the degree or presence of NE lesions. Statistical calculation was made using a regression model; the level of statistical significance was set at a p-value of <0.05.

		Performance results:		
			Day 42	Day 21
			Weight	NE
Group	*C. perfringens*	FCR	Gain (kg)	mortality (%)
1. Non-medicated	Healthy	1.721 d	2.241 a	0.0 c
2. Non-medicated	+	1.848 a	2.140 b	5.7 a
3. CHCC16872 5x10^4CFU/g	+	1.792 b	2.174 ab	2.1 b
4. CHCC16872 1x10^5CFU/g	+	1.774 bc	2.170 ab	1.1 bc
5. CHCC16872 5x10^5CFU/g	+	1.759 c	2.152 ab	2.2 b
6. CHCC16872 3.2x10^6CFU/g	+	1.747 cd	2.206 ab	1.4 bc

In conclusion, the novel Bacillus CHCC16872 probiotic strain decreased the degree of NE mortality, and performance significantly improved in treated groups as compared to the non-treated group. The different doses of Bacillus CHCC16872 probiotic strain included in the feed gave raise to differences in performance results.

[P6] PREBIOTIC ACTIVITY OF BETA-GLUCANS EXTRACTED FROM PLEUROTUS OSTREATUS

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Over the last few years, polysaccharides of fungi, namely beta-glucans captivated researchers' attention for they hold many significant properties; the most important ones are immunomodulating and antitumor properties. The most biologically active form of beta-glucans is beta-1,3 / 1,6-glucan. Most often, this type of beta-glucans is found in some yeast, bacteria, and micellial fungi (in particular, in oyster mushrooms - *Pleurotus ostreatus*).

In an attempt of developing a new type of functional food, we have studied the influence of beta 1,3 / 1,6 glucans --which have the status GRAS (Generally recognized as safe) by the American Food and Drug Administration-- on the activity of dairy bacteria. Beta glucan containing extracts from *Pleurotus ostreatus* mushroom were added in different concentrations to the milk inoculated with probiotic bacteria. The prebiotic influence of Beta-glucans was studied using *Lactobacillus acidophilus*, *Streptococcus salivarius subsp. thermophiles*, *Lactobacillus bulgaricus*, *Lactococcus lactis subsp. lactis*, *Lactococcus lactis subsp. cremoris*, *Lactococcus lactis subsp. diacetylactis* and *Leuconostoc mesenteroides* strains. Studies of the influence of beta glucan supplementation on the pH value, electrical conductivity, red-ox potential, titratable acidity and the viscosity of milk clots were conducted.

The results demonstrated that the addition of beta-glucans significantly increases the electrical conductivity of the liquid culture of lactic acid bacteria. This indicates a more active growth of microorganisms in comparison with control samples. Lower pH values and high value of titratable acidity may indicate greater glycolytic activity of lactic acid bacteria in test samples compared to control samples. Also, the higher the proportion of beta glucan added the greater was the viscosity of milk clots. This can be an important factor in improving the rheological properties of dairy products.

[P7] INVESTIGATING THE EFFECT OF PROBIOTICS ON REGULATING EPITHELIAL BARRIER FUNCTION IN AN AUTOMATED SYSTEM

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Intestinal health is a key factor in poultry production and positively affects growths performance. Many factors, including disease, feed composition and stress can compromise gut health by causing a loss of the integrity of the intestinal epithelial lining, leading to a breach of barrier function with increased intestinal permeability. This results in increased inflammatory responses and reduced performance. As antibiotics as growth promotors are phased out, probiotics are emerging as a novel alternative that reduce pathogen outgrowth, improve barrier function and overall improve gut health and immune response in chickens.

We investigated the effect of pathogens and spore and none-spore forming probiotics on the barrier function of Caco-2 cells. The barrier function was assessed by using an automated real time device (cellZscope®, nanoAnalytics GmbH, Germany) measuring trans epithelial electrical resistance (TEER). The cellZscope was able to differentiate strains within each category of bacteria. Within spore-forming probiotics both increase and decrease of TEER could be shown, while none-spore-forming probiotics varied widely in their positive effect on TEER. The cellZscope therefore emerged as a unique tool for studying interaction between pathogens, probiotics and cells, and for improving the selection of probiotic candidates.

Functional tight junctions ensure barrier function in epithelial intestinal cells, and changes in TEER often correlate with altered tight junction proteins localization. Key protein of these intercellular junctions, such as Zonula occludens-1 (ZO-1) and occludins, can be visualized by laser scanning confocal microscopy. Therefore, we studied the effect of probiotic on TEER measurement with altered subcellular localization of ZO-1 and occludins for understanding the effect of pathogens and probiotics on the barrier function in epithelial intestinal cells in more depth.

[P8] MYCOTOXINS - BIOLOGICAL WAYS OF FOOD & FEED DETOXIFICATION

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Mycotoxins are secondary metabolites produced by different fungal species. These toxins are important feed and food contaminants. Upon ingestion in sufficient amounts, the effects on human and animal health are detrimental. In 2004, a survey program was started to assess the mycotoxin contamination of feed and food commodities. During the last years, > 60000 samples of different origin were analyzed. More than 80% of all samples contained detectable amounts of one or more members of the main toxin groups: trichothecenes, aflatoxins, fumonisins, or zearalenone. Thus, mycotoxins are a serious health threat and effective detoxification strategies are required. For control of mycotoxin in feed, specific microbial additives were developed that encompass different mode of actions for the different mycotoxins. Microbial decontamination of mycotoxins is safe and allows at the same time preservation of the nutritional value of the feed. To date, numerous toxin-degrading microbes were identified and characterized, but only a few were further developed into products. An example for such a biological detoxifying agent is the strain BBSH 797. It acts on several type A- and B-trichothecenes, such as deoxynivalenol that are produced by different *Fusarium* species. Other microbes such as, *Nocardioides* WSN05-2 or *Devosia mutans* 17-2-E-8 can convert deoxynivalenol to less or nontoxic metabolites.

Aflatoxin B1, a highly cancerogenic toxin produced by *Aspergilli* species, is degraded by *Corynebacterium rubrum, Rhodococcus erythropolis and Nocardia corynebacterioides. Another prominent mycotoxin occuring in feed is the estero-genic acting substance zearalenone, which can cause infertility and abortion. Zearalenon can be successfully detoxified via binding and enzymatic cleavage by* a unique yeast strain named *Trichosporon mycotoxinivorans.*

Besides the use of microbes as additives, the development of toxin-specific enzyme products for decontamination is of increasing interest for the industry. For example, a fumonisin esterase from *Sphingopyxis* **sp. MTA 144** is the functional component of the product FUM*zyme*® that converts fumonisin B1 into the non-toxic hydrolysed FB1 and thus helps to detoxify feed. Microbial detoxifiers and their corresponding enzymes hold a highly valuable potential for direct feed applications and are on the rise in the food & feed industry.

[P9] VAGINAL PROBIOTIC ADMINISTRATION IN THE MANAGEMENT OF PRETERM PREMATURE RUPTURE OF MEMBRANES

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Background: Preterm premature rupture of membranes (PPROM) occurs in approximately 2% of pregnancies and is a major cause of preterm delivery. The etiology of PPROM is not clearly understood, yet infections are believed to be implicated in PPROM, both as a causative agent but also as a consequence of PPROM. Standard care of PPROM involves antibiotic treatment, which reduces the rate of chorioamnionitis and improves perinatal outcomes. However, antibiotic treatment adversely affects the vaginal microbiota leading to an increased risk of infections. Thus, a restoration of the vaginal microbiota, as indicated for probiotic usage, after antibiotic treatment might reduce infection risk and consequently improve perinatal outcomes.

Objective: To examine the influence of vaginal probiotic administration as an adjunct to standard antibiotic treatment on perinatal outcome in women with PPROM.

Materials and Methods: This was a prospective randomized trial of cases with PPROM (24–34 weeks) that were admitted to 1st department of obstetrics and gynecology at Alexandra Hospital in Athens between 2011 and 2015. Forty-nine cases received vaginal probiotics for 10 days in combination with antibiotic prophylaxis and were compared to 57 subjects that received only antibiotics for the same time period.

Results: The mean gestational age at birth (35.49 vs. 32.53 weeks), the mean duration of the latency period (5.60 vs. 2.48 weeks), and the mean birth weight (2,439.08 vs. 2,004.81 g) were significantly higher in the study group in comparison to the controls. Moreover, the neonates of the study group had a lower chance to enter the neonatal intensive care unit or the neonatal special care unit, shorter total hospitalization time, and lower need for oxygen administration and mechanical ventilation, as well as reduced length of oxygen administration.

Conclusions: Vaginal probiotics as an adjunct to antibiotic prophylaxis in women with PPROM prolonged the latency period and improved the perinatal outcome.

[P10] DEVELOPMENT OF HYBRID PROTEIN-BASED COATING FOR PROTECTION OF ALGINATE MICROBEADS IN ACIDIC CONDITIONS

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Probiotic bacteria cannot withstand acidic conditions in the stomach[1] without a successful delivery system. One of the most studied probiotics encapsulation systems are based on alginate[1]. However, alginate hydrogel matrix is not physically stable, especially in low pH of the stomach[2].

We report a hybrid coating, multilayers of proteins and biopolymers, which improves the protection efficiency of alginate microbeads at low pH for probiotics encapsulation. A successful multilayer formation was confirmed using zeta potential measurements and quartz crystal microbalance with dissipation. The microscopy observations demonstrated physically stable coated microbeads after simulated gastric fluid (SGF) treatment. A sequential treatment in simulated gastric and intestinal fluids led dismantling of hybrid protein-coated microbeads while biopolymer-coated microbeads were still intact. The uncoated alginate microbeads did not protect bacteria (L. rhamnosus GG as model) after SGF treatment, whereas the both biopolymer and hybrid protein coatings protected bacteria with a decrease in culturability (5.8 and 4.8 log CFU/g, respectively compared to initial load). However, considering microscopy observations, the culturability results are most likely underestimated. Therefore, this work provides a concept for future protein-based coatings to improve the limitations of bare alginate microbeads for probiotics encapsulation.

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[P11] METABOLIC FOOTPRINTING OF TWO LACTOBACILLUS PLANTARUM STRAINS IN SOLID AND LIQUID MATRICES AND THE IMPACT ON ANTIFUNGAL ACTIVITY AGAINST PENICILLIUM BREVICOMPACTUM

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The antifungal strain *Lactobacillus plantarum* Ind2013 and *Lactobacillus plantarum* Ind3478, which lack antifungal activity, were cultivated in both liquid and solid matrices made from a chemically defined medium. The antifungal activity of the samples at the end of fermentation was assessed against *Penicillium brevicompactum* DCS1540 with a spot assay. Results from the spot assay showed that growth of *Penicillium brevicompactum* DCS1540 was prevented for at least ten days on solid ferments of *Lactobacillus plantarum* Ind2013 compared to the reference where mould growth was detected after three days of storage. In contrast, no inhibition of *Penicillium brevicompactum* DCS1540 was observed on the liquid ferments of *Lactobacillus plantarum* Ind2013 and neither on liquid and solid ferments of *Lactobacillus plantarum* Ind3478.

In parallel to the spot assay, the exometabolomic footprints of the two strains were studied throughout the fermentation period by employing a comprehensive suite of non-targeted analytical protocols, namely LC/MS, headspace-SPME-GC/MS and methoxymation-silylation GC/MS.

Multivariate data analysis enabled monitoring the different trajectories for the four different fermentations. The four types of end-point fermentations could readily be classified, reflecting the relative metabolism of the strain differences, but most interestingly the stress induced by the solidified medium. It was evident from the detected compounds that the initial nutrients were partly metabolized differently as some metabolites were found in higher or lower concentrations in solid fermentations. While the nutrients in the solid matrix were diluted by the addition of agar as compared to the liquid fermentations, it is more likely that other factors influenced the metabolism of the two *Lactobacillus plantarum* strains, such as limited accessibility of nutrients in solid matrix, faster growth rate in liquid or different availability of oxygen. Therefore, the cells may have been exposed to diverse types of stresses resulting in different metabolic pathways, which again resulted in the observed, altered antifungal efficacy of *Lactobacillus plantarum* Ind2013. The combination of extensive chemical profiling and antifungal spot test enabled the identification of a number of metabolites positively correlating with antifungal effect. Further biological assays could establish any causal relation between these candidates and antifungal potency.

[P12] COULD THE ADDITION OF SUGARS BOOST BACTERIAL INTERACTION AND ACCELERATE CHEESE RIPENING TIME?

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The dynamics involved in bacterial interaction during cheese ripening is not fully understood. The complexity as well as the variability related to cheese production are challenges witch need to be investigated by researchers and industry. Significant cost saving could be achieved by accelerating the lysis of the starter lactic acid bacteria (SLAB) and by increasing the growth of non-SLAB. This study, using broth models and cheese trials, was performed to investigate the growth of non-SLAB in presence of added N-acetylglucosamine, Ribose or N-acetylgalactosamine with or without amino acids addition. The eight produced cheeses were analyzed using standard methods for salt-in-moisture, sugars and lysed cells concentration as well as pH, water activity and microbial counts throughout ripening at 10 °C for six months. Sensory analysis and sequencing of DNA from microbial colonies were performed in ripened cheeses. Results indicated no differences in bacterial interaction between the cheeses with addition of sugars and/or amino acids compared to control cheese. Despite the levels of salt-in-moisture and pH being within the range for high quality cheese, sensory analysis indicated that all ripened cheeses were not harmonious. Non-SLAB were under the detection limit of the applied method (2 $\log_{10}$ cfu g^{-1}) throughout the whole ripening period. In the broth model, SLAB were not able to use the tested sugars as source of energy. However, in the cheese matrix SLAB were able to utilize some of the added sugars, N-acetylglucosamine was firstly depleted while N-acetylgalactosamine was less preferable source of energy. The nature of non-SLAB is speculated to be an important factor and its concomitant use as adjunct culture is suggested, especially when cheeses are manufactured with pasteurized milk.

[P13] ABSTRACT WITHDRAWN

[P14] PROBIOTIC POTENTIAL AND IMMUNOMODULATORY EFFECTS OF ENTEROCOCCUS FAECIUM BGPAS1-3 ISOLATED FROM ARTISANAL CHEESE

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Enterococci are genus of lactic acid bacteria (LAB) inhabiting the gastrointestinal tracts of a wide variety of animals adapted to living in complex environments and surviving adverse conditions. Enterococcal strains can be found in a variety of fermented foods contributing to the ripening and aroma development of certain cheeses, as well as probiotics to improve human or animal health. *Enterococcus faecium* is one common species used as probiotic in animal feed mainly used to treat or prevent diarrhea, for immune stimulation or to improve growth. Immunomodulatory properties are important in the mode of action of probiotics. Their ability to specifically modulate the host's immune responses to pathogens was demonstrated as well as ability to enhance nonspecific immunity *in vitro* and *in vivo*.

Enterococcus faecium BGPAS1-3 is isolate from semi-hard homemade cheese produced from no pasteurized cow's milk. The strain BGPAS1-3 exhibited an antimicrobial activity against *Streptococcus aureus*, *Listeria monocytogenes* and *Enterococcus faecalis* strains. *En. faecium* BGPAS1-3 showed good adhesion ability to the HT29-MTX epithelial intestinal cell line (75%) and mucin (73%). In the presence of BGPAS1-3 adhesion of *Salmonella* Enteritidis 654/7E to HT29-MTX was reduced 3%. Depending on the duration of UV treatment of BGPAS1-3 cells, obtained products exhibited different effects on mesenteric lymph node (MLN) cells stimulated with Concanavalin A (ConA) *in vitro*. Short-UV treated BGPAS1-3 stimulated proliferation of ConA-treated MLN cells and IFN-g, IL-17 and IL-10 cytokines production. At the other hand, prolonged UV-treatment of BGPAS1-3 inhibited proliferation of ConA-treated MLN cells and production of pro-inflammatory IFN-g and IL-17 cytokines but stimulated production of immuno-regulatory cytokine IL-10. These results pointed on the stronger UV-resistance of some immuno-suppressive molecules expressed by BGPAS1-3 in comparison to its immuno-stimulatory factors. Such properties could be used to produce postbiotic with opposite effects from same strains by controlling duration of UV-treatment.

[P15] EXOPOLYSACCHARIDE ISOLATED FROM LACTOBACILLUS PARAPLANTARUM BGCG11 STRAIN ALLEVIATES INFLAMMATORY PAIN IN WISTAR RATS

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Probiotics and their products are widely used for fermented foods formulations, offering different organoleptic, technological, nutritional or health advantages. Among the plenty of compounds, bacteria from *Lactobacillus* genus possess the ability to produce different kind of polysaccharides attached to the cell surface (exopolysaccharides, EPS) which are responsible for improvement of the viscosity and texture of the fermented products during milk fermentation. However, beside good technological properties of EPS and well established role in maintenance of bacterial homeostasis, the number of studies reporting health promoting potential of EPS has increased extensively. Therefore, the aim of this study was to test the potential of high molecular weight exopolysaccharide produced by probiotic strain *Lactobacillus paraplantarum* BGCG11 (EPS CG11) to reduce inflammatory pain in Wistar rats. The antihyperalgesic and antiedematous effects of the EPS CG11 were examined in the carrageenan-induced paw inflammatory hyperalgesia model in rats using Von Frey anesthesiometer and plethysmometer, respectively. Simultaneously, inflammation mediators in rat's paw tissue were monitoring by qPCR analysis of *IL-1β*, *TNF-α*, *IL-6* and *iNOS* mRNA expression. Results revealed that intraperitoneal administration of EPS CG11 produced significant decrease in mechanical hyperalgesia and paw swelling in dose-dependent manner. These effects were followed by decreased expression of *IL-1β* and *iNOS* mRNAs in rat's paw tissue implicated that the antihyperalgesic and antiedematous effects of the EPS CG11 are related to the suppression of inflammatory response. To the best of our knowledge, this is the first study reporting antihyperalgesic effect as novel property of bacterial exopolysaccharides. Thus, food enrichment with probiotic-derived compounds like EPS may be considered as a new therapeutic strategy for prevention of inflammatory diseases and other conditions accompanied by pain discomfort.

Keywords: exopolysaccharide, *Lactobacillus paraplantarum*, hyperalgesia, inflammation, IL-1β, iNOS

[P16] PROTECTIVE EFFECT OF EXOPOLYSACCHARIDES-PRODUCING LACTOBACILLI AGAINST CADMIUM INDUCED TOXICITY ON CACO-2 CELLS

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Contamination of foods by heavy metals is a growing problem. One of the most toxic heavy metals is cadmium which was classified as a human carcinogen in 1993. The harmful effects of cadmium include a number of acute and chronic disorders, such as renal diseases, as well as damage to the cardiovascular system and the liver. Some reports have shown that species belonging to *Lactobacillus* sp. present in the human gastrointestinal tract and in fermented foods have the ability to bind and detoxify heavy metal ions. Also, microbial exopolysaccharides (EPS) can be used as tools for removing heavy metal ions by detoxication, which prevents their absorption from the gastrointestinal tract by host cells. Adsorption of heavy metals by EPS is a metabolism-independent process which is attributed to the interaction between metal cations and the negatively charged acidic functional groups of EPS.

The aims of this study were to determine the ability of 12 EPS-producing lactobacilli strains from laboratory collection to bind cadmium and increase survival of human intestinal epithelial cell (IEC) line Caco-2.

The cadmium binding ability was measured by *Inductively Coupled Plasma Mass Spectrometry* (ICP-MS). Based on the obtained results three lactobacilli strains *Lactobacillus paraplantarum* BGCG11, its EPS-CG11^{-} derivative NB1 and *Lactobacillus plantarum* BGAN8, with good cadmium binding ability, were evaluated for its protective effects against cadmium toxic effect on Caco-2 cell line by cytotoxicity assay (LDH), metabolic assay (MTT) and enzyme assay (SOD).

The results of this study showed that all tested lactobacilli strains decreased Cd-induced cytotoxicity in the human IEC line and improved their survival. In conclusion, the probiotics with both good Cd-binding and antioxidative capacities can be used as daily supplements for the prevention of oral Cd exposure.

Keywords: *cadmium, Lactobacillus, exopolisacharide (EPS)*

[P17] CHARACTERIZATION OF PROBIOTIC POTENTIAL OF GABA-PRODUCING NATURAL ISOLATE LACTOBACILLUS BREVIS BGZLS10-17

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Intestinal microbiota may influence host immunity via g-aminobutyric acid (GABA). The evidence that the GABA signaling system is involved in maintenance of immune system homeostasis is increased extensively. GABA production and release were found to occur at very high levels in lactobacilli originally isolated from traditionally fermented foods. Considering expanded thinking about using natural nutriceuticals instead of synthetic additives there is an increased usage of metabolites produced by lactic acid bacteria isolated from craft products. For this study we used natural isolate *Lactobacillus brevis* BGZLS10-17 from Zlatar cheese, an artisanal cheese produced in isolated parts of natural reserve mountain Zlatar.

In our previous work, we characterized strain *Lactobacillus brevis* BGZLS10-17 with high GABA-producing ability, isolated from artisanal Zlatar cheese. Since, GABA could be a good strategy to modulate immunological response in various inflammatory diseases we investigated the effects of BGZLS10-17 strain's supernatant containing GABA on metabolic activity and cytokine production of concanavalin A (ConA) stimulated lymphocytes. At first, the cytotoxic effect of two selected concentrations of supernatant (1.25% and 2.5%) was tested and the results revealed that none of the treatments displayed the cytotoxic effect on lymphocytes. However, both treatments significantly reduced metabolic activity of lymphocytes compared to non-treated cells. Production of proinflammatory cytokines, IL-17A, IFN-γ and TNF-α, was significantly lower in cultures of lymphocytes treated with tested supernatants, compared to non-treated cells. Also, GABA containing supernatants significantly stimulated the production of anti-inflammatory cytokine IL-10 in comparison to non-treated cells. Additionaly, treatment with supernatants in both concentrations remarkably stimulated the expression of *Foxp3*$^{+}$ mRNA in comparison to non-treated cells.

In conclusion, *in vitro* tests indicated that BGZLS10-17 is good probiotic candidate that could be eventually used in modulation of immunological response and formulation of functional starter cultures for production of innovative foods.

Keywords: γ-aminobutyric acid (GABA), immunomodulation, probiotic, functional food.

[P18] IMPEDANCE MICROBIOLOGY: A STEP FORWARD IN EXOPOLYSACCHARIDES DETECTION IN FERMENTED MILK

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Lactic acid bacteria (LAB) are able to secrete exopolysaccharides (EPS) of great interest for food manufacturing as they show a positive effect on texture, mouthfeel, taste perception and stability of fermented foods, opening up diverse commercial applications as alternatives for plant polysaccharides commonly used in the food industry. Additionally, the *in situ* production of polysaccharides by LAB has strongly been associated also with the diverse functional and health-promoting properties displayed by these microorganisms.

Considering the great technological interest there are still many unknown facts when working with EPS- producing LAB strains, due to their strain-dependence, environmental factor as well as the difficulties to evaluate the amount and the moment of production especially in food.

There are different direct or indirect analytical methods available to evaluate the EPS production, but, to the best of our knowledge there isn't a method available for a rapid detection of EPS producing LAB.

In this work, impedance microbiology was proposed as an alternative method for the evaluation of LAB EPS production in milk. The impedance curves of the EPS+ strains showed a different shape when compared to the conventional curves (Bancalari et al., 2016). To prove the hypothesis that this differences in the curves were due to the synthesis of EPS during growth, the ability of 24 LAB EPS+ strains to modify the impedometric curves was investigated. Furthermore, to deeper understand this behavior, two *Lactobacillus delbrueckii* subsp. *bulgaricus* strains were studied by means of a multidisciplinary approach; comprising the use of TEM, confocal microscope, quantification of EPS amount and the study of the gene expression.

The data obtained confirmed ours hypothesis and suggests that impedance measurement, turned out to be an useful method for a rapid and economic screening of a large number of sample at the same time. Furthermore, since EPS production is a very unstable feature, this method could also be useful for the evaluation of the best *in situ* producing-conditions that are linked to the different characteristic of each species.

[P19] USE OF TECHNOLOGIES BASED ON EFFECTIVE MICROORGANISMS IN AGRICULTURE

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People prefer ecologically safe food products from organic farming. Organic products do not contain genetically modified organisms, chemically synthesized conservants, dyes and fragrances. Microbiological preparations may used in producing environmentally friendly agriculture products. EM-preparations include effective microorganisms selected from nature and cultured..

Experiments on the use of a microbiological preparation in plant growing were carried out on vegetable crops and spring barley crops. The use of EM-1 (produced by the EM Research Organization) in the cultivation of lettuce allowed to increase the yield of the crop relative to the control by 38%, and the application of EM-1 in mixture with bocashi - by 67%. The greatest increase in yields obtained under the use of EM-1 with potassium chloride, the yield of lettuce increased by 87% relative to the control and the accumulation of ^{137}Cs in production decreased by 13%. The application of EM-1 in production crops of spring barley made it possible to obtain an increase in yield by 20%, relative to the traditional technology of cultivation.

Experiments on the use of EM technology in livestock breeding were carried out on calves of the Holstein black-and-white breed of cattle aged 2.5-4.0 and 4.0-6.0 months. The first test group daily get EM-1 with milk in ammount 30 ml, and the second older culfs got 40 ml of the preparation. The daily weight gain of each animal was 11.2% higher than the previous period. The daily weight gain of the experimental group relative to the control group was 0.115 kg or 10.3%. For the milk production experiment, groups of cows with similar performance and age indicators were selected.

The experiments prove that the use of EM technologies in the production of food products is economically justified and allows improving the quality of the products.

[P20] COMPARATIVE EFFECTS OF VARIOUS TYPES OF THE FOOD INCLUDING PROBIOTICS ON PERIODONTITIS IN RAT MODEL

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Objective: The studies to prevention and treatment for infectious oral diseases using probiotics have been carried out. Our previous study showed *L. lactis* and *S. thermophilus* have antimicrobial activity against *Porphyromonas gingivalis* as a periodontal pathogen. However, it is not easy to apply probiotics to the oral environment because the food and the food supplements do not last long in oral cavity. Therefore, in this study, the various types of the food supplemented probiotics were fed to rats and investigated their effects on periodontitis induced rats.

Methods: The periodontitis of rats was induced by *P. gingivalis*-adhered ligatures onto the molars, and two types of the feed with or without probiotics, which are slurry type like a yogurt and freeze-dried type like a cracker, were given to rats for 3 weeks. The rats were then investigated the levels of inflammatory cytokines by ELISA kit and clinical index by probe and micro CT. Also, the level of P. gingivalis and probiotics in the oral cavity of the rats was measured by quantitative real-time PCR.

Results: Freeze-dried feed including probiotics significantly reduced the induction of periodontitis by *P. gingivalis*-ligature than the feed without probiotics, and the slurry type slightly reduced the induction of periodontitis. Also, the levels of P. gingivalis were more decreased by the freeze-dried feed than the slurry type, and the probiotics were more detected in the rats fed the freeze-dried feed compared to the slurry type.

Conclusions: In the basis of the results, the freeze-dried food including probiotics may a candidate food to apply for prevention of the periodontitis of human.

[P21] ASSEMBLY AND COMPARISON OF WHOLE GENOMES OF DIFFERENT BACTERIA USING THE NEXTERA DNA FLEX LIBRARY PREP KIT

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Analyses of bacterial genomes provide information on genomic composition and organization. Nucleic acid sequence is mandatory to study virulence factors, antibiotic resistance, epidemiology, and nosocomial outbreaks.

Over the last 15 years, NGS (Next Generation Sequencing) has become the most common approach for bacterial genome sequencing. While the handling time to perform sequencing and analysis has been reduced, the number of samples to be tested has dramatically increased.

The objective of the study was to perform an evaluation of a kit for rapid DNA library preparation, the first step of the entire sequencing workflow.

The Nextera Flex Library Prep Kit protocol is compatible with DNA inputs ranging from 1ng–500ng. And can be successfully used across both small and large complex genomes. Here we tested bacterial cultures and colonies as input. To do so, the following bacterial species were selected based on different features, including Gram negative or Gram positive bacteria, aerobic and/or anaerobic strains: *Escherichia coli*, *Staphylococcus aureus*, *Bordetella pertussis* and *Clostridium tetani*. Three different amounts of DNA (100 ng, 300 ng, and 500 ng) were used as input.
We analyzed the data through different pipelines (STRT2, Bacterial analysis Pipeline, and others), all available on the BaseSpace Hub.

Methods: The Nextera DNA Flex Prep Kit utilizes bead-based "tagmentation" chemistry by combining DNA fragmentation and adapter tagging in a single reaction on a solid support, reducing the need for accurate quantification at inputs greater than 100ng. From the sequencing data of those bacteria, multiple analysis tools are required to examine the data: Spades for assembling the genomes, and STRT2 for an MLST comparison of the strains sequenced.

Results & Discussion: In this study, we have combined key features of a new innovative workflow to analyze whole bacterial genomes. Nextera DNA Flex provided a method for rapid extraction of DNA from colonies on a plate, to a normalized library, ready for sequencing. This study evaluated multiple species of bacteria and demonstrated the suitability of the kit on projects where bacterial genomic composition and organization are required. This kit can be applied to other bacterial species without any additional development needed.

[P22] PRODUCT-YIELD SELECTION IN WATER-IN-OIL EMULSIONS

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Many fermentation industries aim for the production of extracellular products, such as flavor compounds. These processes can be improved by using strains with a higher product-yield. To have non-GMO strains, it would be ideal to directly select high product-yield mutants. A cultivation method suited for extracellular product-yield selection is compartmentalization of single cells in water-in-oil emulsions. Compartmentalization ensures that each cell has its own substrate pool and it allows for coupling of the phenotype (extracellular product concentration) to the corresponding producer cell. This coupling of extracellular product to producer is only possible when cells and products stay inside the microdroplet. However, quite some interesting extracellular products with hydrophobic properties, such as diacetyl, ethanol or acetaldehyde, leak into the oil-phase.

We here analyzed the effect of product leakage on high product-yield selections in water-in-oil emulsions. A *Lactococcus lactis* strain was used as producer and fluorescent indicator strains were used as read-out of the product-concentration in microdroplets. We created conditions which allow or prevent product leakage and we analyzed mixtures of droplets with indicator strains only and droplets with both producer and indicator strains. The fluorescence of microdroplets was analyzed using flow cytometry.

Under conditions of product leakage all droplets with indicator strains show a measurable signal response, indicating that under these conditions we cannot discriminate between droplets with and droplets without producer strains. Upon incubation under conditions without product leakage only droplets with both indicator and producer strains showed a response, which should allow FACS sorting of high product-yield mutants. The obtained results indicate therefore that coupling of the product to the corresponding producer is essential to product-yield selections in water-in-oil emulsions.

[P23] INSIGHTS INTO THE NUTRITIONAL AND REGULATORY ROLE OF YEAST EXTRACT COMPONENTS ON STREPTOCOCCUS THERMOPHILUS

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The industrial fermentation of commercial lactic starters is usually performed in semi-defined or complex media whose composition is of utmost importance in order to reach high levels of biomass quantity and quality. Several nutrient sources are available for the industrial production of starters and probiotics. In particular, yeast extracts (YEs) are an excellent source of a wide variety of nutrients such as nitrogen, vitamins, nucleotides or minerals. The main purpose of using YEs in industrial culture media is to provide readily available nitrogen compounds to the cultivated bacteria as they contain a complex mixture of free amino acids and peptides. In this study, we specifically aimed at exploring the nitrogen-based content of YEs and evaluating their nutritional and regulatory role during the culture of an industrial strain of *Streptococcus thermophilus*. We therefore established a complete analytical pipeline that notably combines, on one hand, a peptidomic approach for mapping the YE peptides, and, on the other hand, bottom-up proteomics and RNAseq in order to measure the strain metabolic and physiological responses to YE-based cultures. So far, this work led to the identification of a substantial number of peptides composing the analyzed YEs. Subsequently, we were able to determine which types and classes of peptides were preferentially consumed and infer the basis of their utilization by *S. thermophilus*. Finally, our results also contribute to pave the way of a putative regulatory role of a very specific class of YE peptides that may influence different regulatory networks linked to cell fitness. The results obtained in this study provide some hypotheses to explain how YEs can differentially modulate cellular programming.

[P24] MICROFLORA AND AROMA PROFILE OF TRADITIONAL SLOVAKIAN RAW EWES' MILK-BASED CHEESES

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Traditional cheeses such as Slovenska bryndza PGI, Slovenska parenica PGI, Slovensky ostiepok PGI or Oravsky korbacik PGI are very popular in Slovakia. Although their industrial versions are produced and marketed in large amounts, the versions produced in a recently standardized traditional way are organoleptically superior. The latter are produced mainly from raw ewes' milk without adjunct cultures. We studied the microflora responsible for the unique organoleptic properties of these cheeses, mainly focusing to ewes' lump cheese in various ripening stages, which is an intermediate product being processed to various products. Lactic acid bacteria were isolated, strains clustered using Fourier transform infrared spectroscopy, classified by 16S rDNA sequencing, analysed for the presence of selected genes related to proteolysis and interesting strains underwent whole genome sequencing. The selected strains were used as adjunct cultures in preparation of model lump cheeses, whose properties were followed by high-throughput sequencing of metagenome and by analysis of aroma-active volatiles. The characteristics of model cheeses were compared to those of farm-produced lump cheeses. Dominant bacteria were *Lactococcus* spp. and *Lactobacillus* spp., dominant fungi were *Dipodascus* spp. (including the former *Galactomyces*/*Geotrichum*). Aroma profiles contained 20-30 key compounds including 3methylbutanal, 3-methylbutanol, butanoic acid, pentanoic acid, ethylhexanoate, 2phenylethanol and octanoic acid. Certain strains used as adjunct cultures facilitated modulation of aroma profiles, keeping them compatible with the traditional characteristics.

[P25] ENCAPSULATION OF BENEFICIAL PROBIOTIC BACTERIA IN EXTRACELLULAR MATRIX PRODUCED NATURALLY DURING BIOFILM FORMATION BY BACILLUS SUBTILIS

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Live beneficial probiotic bacteria are often supplemented into foods and beverages to provide putative health benefits. To ensure their beneficial effects, these organisms must survive processing and storage of food, its passage through the upper gastrointestinal tract (GIT), and subsequent chemical ingestion processes until they reach their target organ. However, there is considerable loss in viability of probiotic bacteria in the acidic conditions of the stomach and the high bile concentration in the small intestine. Bacillus subtilis, a spore-forming non-pathogenic bacterium, recently has gained interest in its probiotic properties; it effectively can maintain a favorable balance of microflora in the GIT. In addition, B. subtilis produces a robust extracellular matrix that protects it from stressful environments. We thus hypothesized that the extracellular matrix produced by B. subtilis could protect other probiotic bacteria and therefore potentially could be used as a vehicle for delivering viable probiotic cells to humans. Subsequently, we have developed a novel cultivation system that enables co-culturing of B. subtilis along with probiotic lactic acid bacteria (LAB) by increasing production of the extracellular matrix by *B. subtilis* cells. Moreover, we showed that *B. subtilis* improved survivability of LAB during food preparation, storage and ingestion. Consequently, we believe that the results of our study will provide a novel technique of using a natural system for preservation and delivery of probiotics to humans.

[P26] DEFINED CO-CULTURES OF YEAST AND BACTERIA MODIFY THE GLUTEN, AROMA, CRUMB AND SENSORY PROPERTIES OF BREAD

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Yeast and bacterial communities inhabit a sourdough starter to make artisanal bread. This bread is increasingly sought after by consumers because of its distinctive flavour, attractive aroma and health properties of reduced gluten sensitivity. It is likely that microbial interactions provide some of these positive outcomes, and if characterised could be used to produce bread of superior health qualities. Our donated sourdough starters identified *Saccharomyces cerevisiae* and *Kazachstania exigua* yeasts. When these yeasts were used to ferment wheat flour in an extended time fermentation, the bread had a heterogeneous crumb structure, a deeper colour and a distinctive chemical aroma profile than those made with commercial baker's yeast. When bread was made combining these yeasts individually and in combinations with lactic acid bacteria also isolated from these sourdough starters, including *Lactobacillus plantarum*, *Lb. brevis*, *Lb. rossiae*, *Lb. casei*, the bread aroma profiles and crumb structure were more distinctive, with compounds associated with *sour* aromas produced, and preferred by sensory panels. We assayed the protein structure of these breads by LC-MS and found that the low-molecular weight range of gliadin and glutenin were more diverse in mixed culture fermentations. The interactions of the yeasts and bacteria are likely to influence the production of enzymes which lead to the modification of gliadin and lead to an altered gluten content. This may mean that the bread is suitable for consumption for people on a low-gluten diet. The use of defined mixed cultures in commercial breadmaking, by exploiting the microbial diversity of artisanal starters, can produce bread with distinctive and attractive aromas, crumb structure which is appreciated by sensory panels and has altered and potentially enhanced health properties.

[P27] INFLUENCE OF PROBIOTICS AND SYNBIOTICS ON INTESTINAL MICROBIOTA OF PIGS, TURKEYS AND CHICKEN BROILERS

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Synbiotics are combination of probiotics and prebiotics with the synergetic benefits for a host health via intestinal microbiota, where probiotics are defined as living microorganisms which have an advantageous effect if administrated in the right amount, whereas prebiotics are nondigestible food ingredients that selectively stimulate growth or/and activity of bacteria in colon of the host.

The aim of this research was to designate an impact of newly designed probiotics and synbiotics on intestinal microbiota of pigs, turkeys and chicken broilers.

The trial was conducted on finishers divided into six study groups. Animals belonging to groups A, B and C were fed forage with newly designed synbiotics. Tested synbiotics contained *Lactobacillus* sp. bacteria, *Saccharomyces cerevisiae* yeast, inulin as a prebiotic and they were created at the Institute of Fermentation Technology and Microbiology. Feed for animals assigned to groups D and E included commercially available probiotic products, which was BioPlus and Cylactin®. Control group were accounted for reference animals for which forage wasn't enriched witch any additives. During the experiment, dominant microflora in contents of intestines and faeces samples were analysed. Intestinal microbiota was determined by the standard Koch's plate method, using selective microbiological media. During experimental work total number of anaerobic bacteria (Plate Count Agar), *Lactobacillus* (MRS agar), *Bifidobacterium* (RCA Agar), *Clostridium* (TSC agar), *Enterobacteriaceae* (VRBD agar), *coliform* bacteria (TBX agar), *Enterococcus* (BAA agar), *Bacteroides* (VL agar) and yeasts (SDA agar) were examined in samples.

Studies have showed that synbiotics have beneficial influence on intestinal microbiota of pigs, turkeys and chicken broilers. The decrease of number of potentially pathogenic microorganisms was observed with simultaneous increase number of beneficial species of bacteria. Usage of synbiotics resulted in more significant changes than in case of using probiotics as an additive to forage.

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[P28] POTENTIAL IMMUNOMODULATORY ROLE OF LAB INDUCING NFKB AND IRF-3 ACTIVATION AND PHAGOCYTOSIS UNDER EXPERIMENTAL CONDITIONS

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Lactic acid bacteria (LAB) comprising various genera including *Lactobacillus* and *Pediococcus,* are common inhabitants of the mammalian intestinal tract and confer a health benefit on the host. The gut immune system recognizes LAB via Toll-like receptors (TLRs) that are expressed on immune cells and epithelial cells. These TLRs, through the activation of intracellular biochemical pathways, trigger the activity of cells involved in the innate immune response. For example, IRF3 and NF-κB pathways are crucial in maintaining immune homeostasis and regulating immunological and antimicrobial responses in barrier tissues such as the intestine. The aim of this work was to assess whether a set of LAB isolated from wild boar are able to modulate both IRF3 and NF-kB pathways and phagocytosis in monocytes and macrophages under experimental conditions. 12 LAB isolated from wild boar were identified as *Pediococcus* spp., *Lactobacillus* spp. and *Enterococcus* spp. Immunomodulatory capabilities were assessed examining the induction of IRF3 and NF-κB signal transduction in M1-like macrophages by luciferase reporter assay. Some of the isolates have induced NF-κB and IRF-3 activation in M1-like macrophages, either dead or alive, suggesting a potential role as vaccine adjuvants or boosters of Th1 immune response; whereas other isolates have showed potential anti-inflammatory properties as they suppress activation in the macrophages. Also, PKH2 labelled LAB were exposed to whole blood and analysed by FACS after 1 h incubation. Quantification of the percentage of PMN and monocytes positive for labelled LAB showed that whereas monocytes efficiently bound and phagocytosed selected strains, PMNs being associated with higher tissue damage, remained largely unresponsive. These results revealed that LAB selectively interfere with the innate immune system inducing phagocytosis and apoptosis in monocytes and macrophages while sparing neutrophils. Further studies such as whole-genome sequencing and antibiotic resistance tests will confirm the potential use of our LAB isolates as probiotics in wild boar or other animal species before any *in vivo* confirmatory experimental study. Nevertheless, our current *in vitro* data suggest that these isolates could be used as an early therapy and/or vaccine adjuvants to reduce or prevent infection but also as a tool to reduce inflammation and boost immunity.

[P29] DEXTRAN SULFATE SODIUM-INDUCED COLITIS IN PIGLETS

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Inflammatory bowel disease (IBD) results from complex interactions between genetic and environmental factors. Although a genetic contribution has been proven, dietary factors have also shown to play a role in the development of IBD.

This study aims to investigate the effect of adding red meatto the diet of piglets in a dextran sodium sulfate (DSS)-induced colitis model.

This study is based on a previous (unpublished) study where a piglet DSS-induced IBD model was developed. Colitis was induced in piglets (n=24) by adding DSS and 15% beef meat to the diet. The treatments were: Control (standard weaner diet); DSS (Control + 1,25 DSS/kg·BW^{-1}); Meat (Control with 15% beef meat); Meat+DSS (Meat+ 1,25 g DSS/kg·BW^{-1}). The impact of the diet was studied by analyzing both fecal material and gut content. From the hypothesis that colitis development was related to an increased number of sulphate reducing bacteria (SRB), resulting in higher intestinal concentrations of hydrogen sulfide, qPCR was conducted. Gut microbial composition was also analyzed using 16S rRNA gene amplicon sequencing. Metabolites reflecting both carbohydrate and protein microbial fermentation was measured including short fatty acids, and biogenic amines. The number of *dsrB* gene copies (as a fraction of total 16s rRNA) showed a tendency to be lower in the Control and Meat treatment (0,084% and 0,047%, respectively) compared to the DSS and Meat+DSS treatments (0,183% and 0,175%, respectively)(P=0,434). These results were in line with the 16s rRNA amplicon sequencing data, where the Control and Meat treatments clustered together and DSS and Meat+DSS clustered together. The data indicate that DSS resulted in an increased number of SRB in the large intestine of piglets; and that DSS had a bigger impact on the microbiota composition than the addition of 15% beef meat to the diet.

[P30] SAFE STRAIN LINEAGE - SUPPORTING SAFE ENZYME PRODUCTS WITH LESS ANIMAL SACRIFICE AND LESS DATA TO EVALUATE BY REGULATORS AND INDUSTRY

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Safety assessment of food and feed enzymes includes an evaluation of the safety of the enzyme component, the production organism, including genetic engineering aspects, the manufacturing process, and the dietary exposure.

A comprehensive decision tree that encompasses all these aspects has been in use by industry for over 15 years. The decision tree includes the concept of Safe Strain Lineage, which allows for the extrapolation of existing toxicology data obtained for earlier members of a microbial lineage to support the safety of products produced by newer members of the same lineage. A Safe Strain Lineage can be established after repeated assessment via the decision tree evaluation procedure. This includes the safety of the host organism (including existing toxicological information for the host or for enzymes produced by production strains in the lineage), all of the introduced DNA, and the methods used to genetically modify the host.

The enzyme industry successfully introduced the concept of Safe Strain Lineage to regulators. This concept applies not only to enzymes, but can be used for many other industrial biotechnology products, and has the potential to minimize unnecessary animal sacrifice, reduce the amount of data that needs to be assessed by industry and regulators, yet preserve the ability for industry and regulators to thoroughly evaluate product safety.

As always, establishing and maintaining a science-based, risk-focused regulatory oversight framework for industrial biotechnology products requires relevant data and effective dialog with all stakeholders (expert groups, regulators and consumer groups) on the technology, its safety, and its benefits.

[P31] ABSTRACT WITHDRAWN

[P32] EXTRACELLULAR PHYTASE FROM LACTOBACILLUS FERMENTUM SPP KA1: OPTIMIZATION OF ENZYME PRODUCTION AND ITS APPLICATION IN IMPROVING THE NUTRITIONAL QUALITY OF SOY MILK

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Research Objectives:

Phytases are phytate specific phosphatases catalyzing the step-wise dephosphorylation of phytate, which act as an anti-nutritional factor in food due to its strong binding capacity to minerals. In recent years microbial phytases have been explored for improving nutritional quality of food. But the major limitation is acceptability of phytases from these microorganisms. Therefore, efforts are being made to isolate organisms which are generally regarded as safe for human consumption such as Lactic Acid Bacteria (LAB). Phytases from these organisms will have an edge over other phytase sources due to its probiotic attributes. Only few LAB have been reported to give phytase activity that too is generally seen as intracellular. LAB producing extracellular phytase will be more useful as it can degrade phytate more effectively. Moreover, enzyme from such isolate will have application in food processing also. This study reports isolation of a probiotic strain of *Lactobacillus fermentum* spp KA1 which produces extracellular phytase. Only few species of *Lactobacillus* producing extracellular phytase have been reported so far.

Findings: Lactobacillus fermentum spp KA1 which produces extracellular phytase has been isolated. The strain meets all the characteristics of probiotic. Conditions for the optimal production of phytase have been optimized and resulted in approximately 13-fold increase in yield. The phytate degradation potential of extracellular phytase in soymilk has been explored and conditions for optimal degradation were optimized. Under optimal conditions, there was 2.5-fold increase in release of inorganic phosphate.

Research Outcomes: Although previous research findings suggest that phytate content is reduced upon soaking of soybean seeds but is not completely removed. Still 0.20-0.50 per cent of phytate has been reported to be found in soymilk prepared from soaked soybeans. Phytate removal using *Lactobacillus fermentum* spp KA1 phytase will be significant for mineral availability in soymilk.

Future Scope: *Lactobacillus fermentum* spp KA1 can be used as a probiotic in phytate rich foods where it will help in the removal of phytate along with its probiotic effects.

[P33] THE INVESTIGATION OF STRTEGIES OF ARABINOSIDASE AND XYLANASE SYNERGY FOR XYLAN BIO-DEGRADATION

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The complex structure of xylan requires variety of enzymes to breakdown the main and side chain. The complex mechanisms, characteristics and cooperation of various hemicelluloses are still being studying. The aim of this study is to investigate the synergy strategy of Arabinosidase and Xylanase for more effective bio-degradation of xylan and biomass conversion.

In this study, three α-L-arabinofuranosidases from different origins were tested with xylanase from *Thermocyces lanuginosus*. Pure wheat arabinoxylan and oat hull were treated with α-L-arabinofuranosidases and xylanase mixture, respectively under the condition of pH 6.0 and 50°C for 240 minutes. Three strategies have been evaluated A) two-step synergy that α-L-arabinofuranosidases were added first and xylanase was added 120 minutes afterward; B) two-step synergy that xylanase was added first and α-L-arabinofuranosidases were added 120 minutes afterward; C) one step synergy α-L-arabinofuranosidases and xylanase were added at the same time. Samples were analyzed by HPLC and 3,5-dinitrosalicylic acid (DNS) method. The results show that the equivalent xylose release is B>A>C.

In this work, it is assumed that xylanase could cleave the main chain and reduce the polymerization, which might increase the possibility of the side branch exposure and the chance for side-chain enzymes to attack. Therefore, arabinosidases might work more effectively to remove the arabino-side chains and enhance the general performance of the xylanase.

[P34] EXPANDING THE BIODIVERSITY OF THE WINE BACTERIA OENOCOCCUS OENI

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In wine, alcoholic fermentation is usually followed by a malolactic fermentation, which is driven almost exclusively by the *O. oeni* lactic acid bacterium. It is highly adapted to the wine environment, resisting both alcohol and acidity, and rarely detected in other environments. However it has been reported in cider, which has similar physicochemical properties as wine, and recently in kombucha. Due to its importance in performing a robust malolactic fermentation and effects on wine flavor, selection of efficient *O. oeni* strains is a priority for winemakers.

Phylogenetics based on nearly 300 genomes of mainly wine strains, as well as a few from cider, showed two major phylogroups within the *O. oeni* species, named groups A and B. In this work we expand the phylogenetic tree of *O. oeni* with two new groups, C and D, and explore their genetic repetoires.

The genomes of 8 strains from cider and 5 from kombucha were sequenced using Illumina Paired-End technology, and two complete sequences were generated by complementing the sequencing with Mate-Pair reads. Phylogenetic clustering based on whole-genome or core SNP aligments placed the cider and kombucha strains as separate branches – Group C and D – with the addition of some australian wine strains in group C. The complete genomes were used to show overall synteny to the only public complete genome reported to date from strain *O. oeni* PSU-1, showing no major genome rearrangements. Whole genome comparison performed with Mauve and BRIG indicated that the cider and kombucha strains differ in a few small regions.

Using the pangenome to compare the newly described groups, we found differences in amino acid biosynthesis, sugar transporters and resistance genes, both separating the new groups from A and B, as well as C from D.

We speculate that these changes represent adaptation to the different environments of cider and kombucha. These changes are exemplified in the finding of mutations in the malolactic operon of the kombucha strains, indicating that they are unable to perform the malolactic fermentation. This results suggests that this trait it is not as important for *O. oeni* in this environment.

[P35] INFLUENCE OF DIFFERENT TEA SUBSTRATES ON MICROBIAL ECOLOGY, ANTIOXIDANT ACTIVITY, POLYPHENOLS AND FLAVONOID CONCENTRATION IN KOMBUCHA BEVERAGE

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Kombucha is a traditional tea-based beverage fermented by a symbiotic colony of acetic acid bacteria and yeasts recognized since the ancient time for his nutraceutical properties. The starter culture consist in a cellulose matrix floating on the top of the batch and a small part of liquid from the fermentative process. The aim of the study was to characterize the microbial ecology and antioxydant activity in Kombucha beverage using one starter culture and the same sugar concentration (80 g/l) on three different substrates: Black tea "Ceylon" (*C. sinensis*), Green tea "Sencha" (*C. sinensis*) and Rooibos (*Aspalathus linearis*). After 14 days of fermentation at 27 ±1 °C, the diversity and abundance of bacteria and yeasts in the different substrates were studied through culture-dependent method. Bacteria isolates were grouped and identified using RAPD PCR and 16S rRNA gene sequencing; yeasts were identified by amplification of their ITS1-5.8S rDNA-ITS2 regions and by sequencing of the D1/D2 domain of the 5' end of the large subunit (26S) rDNA.

Clustering analysis results and sequencing led to the identification of 4 bacterial species (*Komagataeibacter intermedius, Komagataeibacter swingsii, Komagataeibacter rhaeticus* and *Gluconoacetobacter entanii)* and 2 yeasts species (*Zygosaccharomyces parabailii* and *Brettanomyces bruxellensis).* The abundance of these microorganisms was significantly different in each substrates. The antioxydant activity of the three fermented substrates was evaluated with DPPH and FRAP methods. Polyphenols and flavonoid concentration was quantified spectrophotometry based on different wavelength absorbance. The obtained results revealed that the substrates strongly influence the microbial ecology and the antioxidant activity and give a chance to improve the nutraceutical effects of Kombucha beverages using different teas. Furthermore, the characterization of polyphenols during and after the fermentative process may give useful data on Kombucha therapeutic effects.

[P36] TRANSFUCOSYLATION CATALYZED BY GH29 A-L-FUCOSIDASES FOR ENZYMATIC PRODUCTION OF HUMAN MILK OLIGOSACCHARIDES

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Human milk oligosaccharides (HMOs) constitute a unique family of bioactive lactose-based molecules present in human breast milk. HMOs are complex, heterogeneous oligosaccharides made of glucose, galactose, *N*-acetylglucosamine, fucose, and sialic acid. They are of major importance for infant health and development, but also virtually absent from bovine milk used for infant formula. Controlled synthesis of HMOs is a novel field of food ingredient research, and several strategies are currently pursued with the aim of producing HMOs for infant formula supplementation. Among the HMOs, the fucosylated species are the most abundant. Transfucosylation catalysed by retaining α-L-fucosidases is a new route for manufacturing biomimetic, fucosylated HMOs. In a recent study, seven α-L-fucosidases from glycosyl hydrolase (GH) family 29 were recombinantly expressed, characterized in terms of substrate specificity and thermal stability, and all were shown to catalyse transfucosylation with varying donor specificity and regioselectivity. *Cp*Afc2 from *Clostridium perfringens* (EC 3.2.1.111; GH29B) efficiently catalysed the formation of the complex HMO structure lacto-*N*-fucopentaose II (LNFP II) using the simpler HMO structures 3-fucosyllactose (3FL) as fucosyl donor and lacto-*N*-tetraose (LNT) as acceptor with a 39% molar yield on the donor substrate. Thus, mixtures of 3FL and LNT (e.g. obtained from fermentation of engineered *E. coli* which is efficient for producing simpler HMO structures) could be enriched with LNFP II by enzymatic transfucosylation, while mixtures of 3FL and lacto-*N*-neotetraose (LNnT) could be enriched with the HMO structure LNFP III. Furthermore, α-L-fucosidase *Fg*FCO1 from *Fusarium graminearum* (EC 3.2.1.51; GH29A) was able to catalyse transfucosylation of lactose using citrus xyloglucan as fucosyl donor. This transfucosylation resulted in formation of 2'-fucosyllactose (2'FL), which is the most abundant HMO, reaching a molar yield based on the donor substrate of 14%. Recent results on α-L-fucosidase engineering for improved transfucosylation efficiency are also presented.

[P37] BIOCATALYSIS FOR BIOMASS VALORIZATION: PEPTIDES AND FATTY ACIDS FROM RICE BRAN

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Waste upgrading practises have attracted a significant attention in recent years with the aim of managing agrofood by-products in a gainful and sustainable way.

We describe here how biocatalysis can assist rice bran valorization, according to the biorefinery concept. [1]

Rice is the staple food for over half the world's population. Rice milling generates a massive amount of waste, namely rice bran (70 kg/ton of rice) and rice husk (200 kg/ton of rice). Rice bran (RB), containing fibers (7-11%), proteins (10-16%), lipids (15-22%), carbohydrates (34-52%), micronutrients, represents a second-generation biomass. [2]

Rice bran proteins (RBP) have a high nutritional value and optimal digestibility and are gluten-free, hypoallergenic and rich in essential amino acids. However, the first hurdle to be overcome for RBP production and large scale application is their extraction. Structural complexity, poor solubility, and strong aggregation make RBP hardly available.

The sequential treatment of RB with carbohydrases and proteases was used to prepare mixtures of water-soluble peptides (RBPHs, RBP Hydrolysates) to be tested as antibacterial, antioxidant and anticholesterol agents, as well as flavour enhancers. [3] Interestingly, sensory analysis revealed that the obtained RBPHs exert only sweet and umami taste.

Rice bran oil (RBO) is one of the most underutilized agricultural commodities. We investigated the use of RBO as a feedstock for the production of FFA-derived chemicals (*e.g.* sugar fatty acid esters). [4] To this aim, RBO was submitted to a preparative lipase-catalyzed hydrolysis to obtain pure FFA. [5] The high acidity of RBO, so far considered as a bottleneck in the exploitation of RBO (*i.e.* biodiesel production) was here turned into an advantage, making available FFA mixtures as synthetic precursors for high added value products.

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[P38] PROLONGED SHELF LIFE AND NOVEL BIO PACKAGING (WILD FERMENTATION EXPERIMENT)

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The aim of the present study was to demonstrate and test a wild fermentation approach to prolong shelf life of surplus biomass. The ambition was to find simple, solid, and easy implementable management tool to control and implement a fermentation technology into the production line at Aarstiderne A/S, and further to produce a new product from a potential waste or surplus biomass.

Today, up to a third of world food and beverages are produced by fermentation and consumers are looking for products that stand out through locality, taste and novelty. The basic idea is that nature has developed what we need, and therefore a wild fermentation approach was suitable in this case. Further Aarstiderne experience an increased focus on quality, storytelling and gastronomy from the customers but nobody produce or deliver a healthy fermented product that still is biological active at the end user level e.g. in a private household. To satisfy that need two different semi permeable bio packaging was tested that both allowed CO_2 to pass from the product to the surrounding environment. With the novel semi permeable bio packaging no heat treatment or additional curing was needed. The results showed that the product contained vast numbers of LAB and that ph. remain low throughout the entire experiment. Further ph. is an easy and fast quality test-method implementable in the production line at Aarstiderne A/S. The applied wild fermentation where successful and both produces products with an appealing sensory gastronomical experience, and at the same time satisfy the guidelines and recommends from the Danish Veterinary and Food Administration. Finally it looks like the vent bags is the best choice due to lower drying-out capability

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Starrenburg	Marjo	O12
Stefanie	Falque	P21
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Stokholm	Jakob	P2
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Staack	Larissa	**O33**
Suarez	Juan Evaristo	O22
Sulakvelidze	Alexander	**O14**
Sundh	Ingvar	O22
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Tenning	Paul	**O19**
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Veljovic	Katarina	P14
Veng	Charlotte Gianna	P7
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Vestergaard	Mike	O10
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Vogensen	Finn Kvist	O4
Vuillemin	Marlene	P36
Weber	Barbara	P8
Wiersma	Anne	O12
Wiese	Maria	**P3**
Williams	Tiffany	O12
Wilmes	Paul	P3
Winters	Michela	**P26**
Witold	Kot	O4
Wong	Connie H. Y.	O3
Xu	Limin	**P33**
Yahav	Sagit	P25
Zenhausern	Frederic	P3
Zentek	Juergen	O17
Zeuner	Birgitte	**P36**
Zhu	Zhigang	O8
Zivkovic	Milica	P16

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Clinical Microbiomics is a contract research organization specialized in microbiomics and innovative bioinformatics concepts. We offer end-to-end microbiome research services including DNA-extraction, library preparation, sequencing and bioinformatics. We base our shotgun metagenomics analyses on our metagenomic species (MGS) concept, which facilitates high-resolution metagenomics analysis, far beyond classical reference-based approaches.

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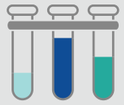
- ISO/IEC 17025:2005 accreditation for laboratory work related to microbiological testing
- End-to-end microbiome lab services
- DNA extractions, library preparation and sequencing

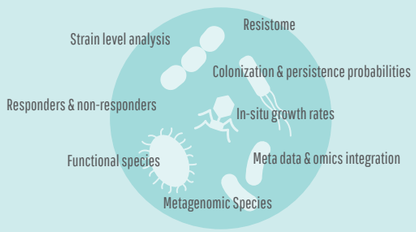


DNA extraction & library preparation

Sequencing (16S or Shotgun)

METAGENOMICS PLATFORM

- Metagenomic Species Concept (MGS) based sample profiling
- Taxonomical and functional microbiome comparisons
- Systems biology & advanced analyses



APPLICATIONS IN FOOD AND FEED

- Analyzing effects of prebiotics
- Detecting probiotic strains
- Distinguishing closely related strains

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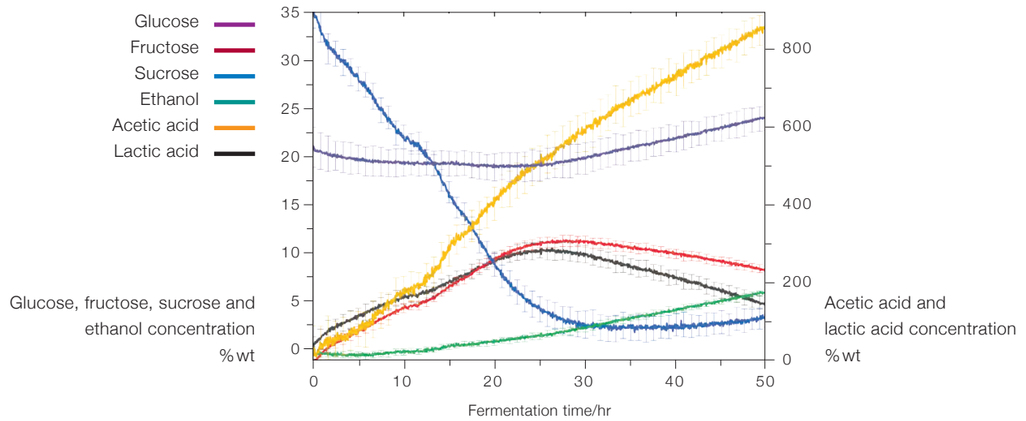

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FERMENTATION SOLUTIONS

TOOLBOX FOR MICROBIAL CULTURE APPLICATIONS

		YEAST EXTRACTS							YEAST AUTOLYSATES	PEPTONE
		100	100 ag	150	105 ag	103 ag	125 ag	110	315	800 ag
CULTURES	*Bacillus*	●	●	●		●				
	Bifidobacteria	●	●	●	●		●	●		
	Fungi				●	●				
	Lactobacillus	●	●	●	●	●	●	●	●	●
	Lactococcus	●	●	●	●		●	●		●
	Leuconostoc	●	●	●	●		●	●		
	Propionibacteria	●	●	●		●	●	●		
	Streptococcus	●	●	●	●		●	●		●
	Others	●	●	●	●	●	●	●	●	●
NUTRITION	High B vitamins				●					
	High nucleotides									●
	High minerals					●				
	High *alpha*-amino N	●	●	●	●	●	●	●		
FORMS	Liquid			●						
	Powder	●	●		●	●		●	●	●
	Agglomerated powder		●		●	●	●			●
PURIFIED	Microfiltered							●		
	SOLUBILITY:	GOOD						HIGH	VERY LOW	GOOD

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