Tuesday, April 19th 2016

Invitation

The International 29th VH yeast conference provides you with lectures and presentations on current topics in the fields of markets and quality, applied yeast research and process innovations in yeast production.

You are invited to join the interdisciplinary dialogue with experts and partners from applied science and practical experience. VH members are called to invite their partner companies to enable reduced fees for attendants.

We look forward to welcome you in Oelde.

M. Eng.Sc. Antoine Chagnon — President of VH

Dr.-Ing. Michael Quantz — GM of VH

State of the art yeast production

11:00 a.m. Welcome and opening "GEA fit for 2020"

Executive Vice President Product Management and Sales

KLAUS STOJENTIN, GEA, GER

11:15 a.m. Centrifugal separation machines development – pilot and design

JÜRGEN MACKEL

GEA, GER

The process of centrifugal separation is well known and has its firm place in today's industry.

For more than 120 years GEA is working in the field of centrifugal separation, constantly improving machine design to offer better process solutions to the market. Through this, GEA has become an international market and technology leader in all fields of centrifugal separation.

Centrifuges you can find in more than 2.500 different processes. Many of such processes were significantly influenced by GEA know-how.

Customized or engineered process solutions which apply centrifugal separation require test work. GEA with its CPT (Central Process Technology Centre) offer to our customers different services like product investigations, pilot trials in the test centre or on the customer side and process development.

The most easy and most important test to be done is a spin test, executed inside a lab test centrifuge. By this simple test procedure, which requires only a few hundred millilitre of product, the experienced process expert can gain already lot of information.

E.g. Important question like:
- Can centrifugal separation be applied or not?
- What kind of centrifuge might be needed? Horizontal decanter centrifuge or disc stack centrifuge?
- Requires the product in the industrial scale process later on many centrifuges or not?

Such questions are from big importance when investment volumes need to be judged and estimated.

The preparation of a pilot plant trial requires up front a lot of work. Customers ship their valuable products to our facility, to execute any kind of centrifugation, e.g. solids/liquid clarification, liquid/liquid separation or even solid/liquid/liquid separation.

The selection of the right centrifuge for the trial is the most significant matter. Centrifuge design parameters like g-force, equivalent clarification area, solids holding space, disk stack design, sanitary design, explosion proof design, etc. need to be considered. After such a pilot plant trial a scale up from the pilot to industrial scale is possible. Based on such scale-up a detailed quotation can be prepared.

12:00 p.m. Multi-position monitoring of gradients in the liquid phase of industrial-scale fermenters

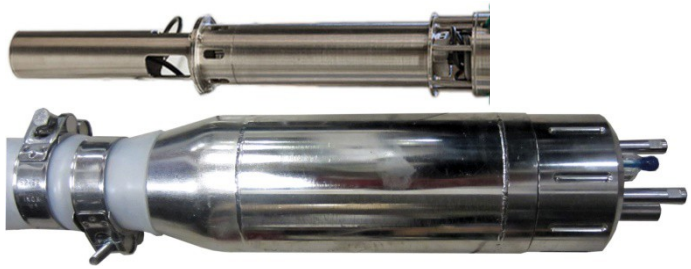
ANIKA BOKISCH [1], JAN BIERING [2], PETER NEUBAUER [1], STEFAN JUNNE [1]

[1] Technische Universität Berlin, Department of Biotechnology, Chair of Bioprocess Engineering, GER

[2] Versuchs- und Lehranstalt für Brauerei in Berlin e.V., GER

In large scale fermentation like the brewing process, process conditions as the local power input and fluid flow might be uneven, and thus gradients occur. Since the available sensor technology, which is usually located at an arbitrarily chosen spot, is not designed for the consideration of heterogeneities, the knowledge about the magnitude of gradients in specific processes is rather low. Computational fluid dynamics do not necessarily contribute to a better understanding, if uncoupled from kinetic models, which consider microbial

consumption and production. Off line samples or on line data from devices installed at one single position of the tank are most likely not representative for the majority part of the liquid phase.



In order to improve process monitoring and to identify critical reactor zones, mobile, sterilizable multi-parameter sensor tools have been developed for in situ and on line monitoring of various process parameters (pH-value, dissolved oxygen, dissolved carbon dioxide, redox potential, conductivity, temperature, pressure) in industrial bioreactors (Fig. 1). These mobile measurements allow for a fast detection of eventual gradients.

12:30 p.m. Short poster presentation

Poster 1: 3-dimensional holographic and in situ microscopy as novel tools for the detection of population heterogeneity in yeast cultures

ANNA-MARIA MARBÀ-ARDÉBOL, ERIC LORENZ, PETER NEUBAUER, STEFAN JUNNE

Technische Universität Berlin, Department of Biotechnology, Chair of Bioprocess Engineering, GER

In situ microscopy is a promising technique to monitor cell proliferation and morphologic population heterogeneity on a single-cell basis. Although a well-established technique for off line and at line analysis, microscopy, which is applied in situ, remains a challenging procedure, though it provides a lot of benefits for operational control and for bioprocess development.

The real-time measurement of the size distribution in this study is realized by the coupling of a photo-optical probe to an automated image analysis. This is combined with off line analyses of the cells' key metabolites with HPLC and GC methods. The cell physiology is further investigated at line with flow cytometry and 3-dimensional holographic interferometric microscopy (Fig. 1). The latter technique is applied for the determination of the single cell volume and surface heterogeneity as well.

The cell size distribution was measured in *Saccharomyces cerevisiae* scale-down cultivations at oscillating glucose and oxygen availability due to the accumulation of intermediates, osmotic stress and growth reduction. A correlation could be found between the cell size and short-chain fatty acid as well as sterol precursor accumulation. Thus, the determination of morphologic features of a yeast culture provides

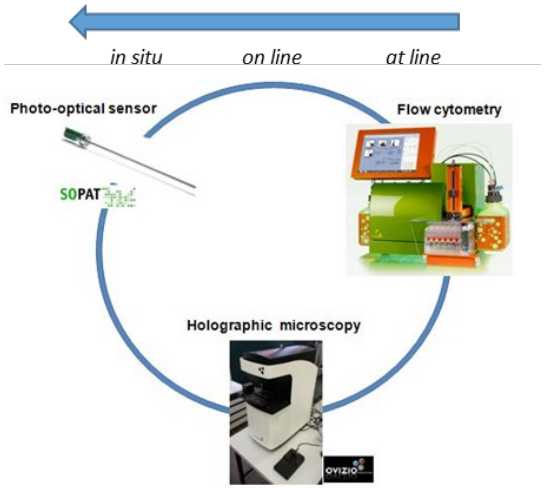


Fig.1: Monitoring techniques for the quantification of cellular morphology on a single-cell level.

In this study, protein which is considered an inner and important component of yeast cells with great beneficial functions to humanity was analysed. This was showcased through disruption of the yeast cell wall mechanically using the high-pressure homogenizer. The diluted homogenized yeast which was released is then compared with samples of undiluted yeast so as to determine the protein concentration yield for both. The design expert v.8 was further used in comparing these analyses to show the variance between the samples.

Poster 2: Process optimization by optical inline analysis

MARKO SCHREIBER

KROHNE Optosens GmbH, GER

The challenge in process analysis technology (PAT) is the on the fly acquisition of precise knowledge about highly complex product compositions. Such knowledge allows for running and control of the process close to specification and to optimize for countless end products.

The poster gives examples of how optical line analysis technology, provided by the OPTIQUAD, enables customers to analyze the product stream inline in almost all stages of production – from raw material to waste water. The concept of the combination of four optical methods (transmission, scattering, refraction and fluorescence) allows adopting the system to an unlimited number of different processes. Used for process control the effectivity and quality of the production can be significantly increased.

Poster 3: FOODSCAN – Development of an automated, novel biosensor platform for pesticide residue detection

FRANK KAGE

IGV FOODTECH, GER

The objective of the FOODSCAN project was to develop a novel and automated biosensor platform for pesticide and other chemical residue detection incorporating membrane-engineered cells with pesticide-specific antibodies. The system is primarily based on the Bioelectric Recognition Assay (BERA)® technology. FOODSCAN is able to detect organophosphate, carbamate and pyrethroid pesticides in cork, wine, cereals, fruits, nuts and vegetables. The portable system is easy-to-handle and can be used for on-spot detection of pesticides residues at the operation sites of agricultural and industrial manufacturing units. The system can be integrated in Quality Assurance Systems.

Poster 4: Application of flow cytometry for bioprocess development of yeast fermentation

MAXIMILIAN SCHMACHT[A] , ERIC LORENZ[B], MARTIN HAGEBÖCK[A], MARTIN SENZ[A]

[A] Research and Teaching Institute for Brewing in Berlin (VLB), DepartmentBioprocess Engineering and Applied Microbiology, GER

[B] TU Berlin, GER

By the application of flow cytometry (FC), a large amount of cells can be analyzed via light scattering as well as specific fluorescence emission, whereby detailed data of each single cell is generated leading to broad information of cell physiological data in a short period of time. Advantageously, this technique is independent of cultivability, which makes it possible to detect cells in different physiological states (e. g. so-called viable but non-culturable (VBNC) cells, dormant cells or death cells) that may considerably account to the overall metabolic activity in fermentations. Therefore, FC can be used as an at- or on-line technique in fields of e. g. food industry, pharmaceutical industry, water management or biotechnology.

FC is not only useful to control product quality and safety but also in terms of process development. The VLB Berlin therefore uses this powerful technique e. g. to monitor viability, fatty acid content or flocculation properties in microbial processes. This contribution gives a short overview of different applications in the development of yeast fermentations.

Poster 5: RPP Raising Power Probe

UDO SCHMALE, HOLGER MÜLLER

BlueSens gas sensor GmbH, GER

The use of yeast in a variety of baked goods requires a constant improvement and monitor of the quality of bakers' yeast, both in production and in the craft and industrial bakery. The specific yeast characteristics and the requirements in accordance with the dough formulations must be discussed and thoroughly characterized. One important parameter is the raising power of baker's yeast (ml CO2 / h). Another important value to be known is the capability of the dough to hold the produced CO2. Both together define the volume of the baked dough.

BlueSens, in corporation with the VH Berlin, has developed a new measurement system to comply with these needs. With the new system it is now possible to quickly get comparable values for the raising speed of different recipes. For quality control the system produces transferable values for the dough volume. In addition it also is possible to determine the total produced volume and the fraction of the produced

volume that is responsible for the dough expansion, even if the dough cracks during the experiment. The time when the dough cracks can also be determined precisely.

Poster 6: Soft-sensor based fermentation control

Udo Schmale [1],Holger Müller

BlueSens gas sensor GmbH, GER

Pharmaceutical industries try to establish PAT-conform processes. PAT means "process analytical technologies" and is an initiative of the FDA. The idea is to run a controlled process instead of analyzing the product afterwards. To ensure that the process runs in his "specs" reliable sensors and analyzer are necessary. Unfortunately not every analytes are online measureable.

Therefore mathematical models based on existing measurements are used to predict process behaviors or calculates interesting parameters like biomass, growth rates, metabolic rates like CER/OUR or specific yield rates.

If these models are directly (online) usable, we are talking about "soft-sensors".

Naturally these soft-sensors are not only interesting for pharmaceutical companies but also for companies that are interested in continuous productions e.g. yeast industry.

In the following we show some soft-sensors that could be used to control continuous yeast fermentation.

01:00 p.m. Lunch break

Clients & Markets

02:00 p.m. Yeast diversity of sourdoughs and associated metabolic properties and functionalities

Luc De Vuyst, Henning Harth, Simon Van Kerrebroeck, and Frédéric Leroy

Research Group of Industrial Microbiology and Food Biotechnology, Department of Bioengineering Sciences, Faculty of Sciences and Bioengineering Sciences, Vrije Universiteit, BE

Yeasts play a key role in the production process of sourdough, together with the acidifying lactic acid bacteria. They are either naturally present or are added as a starter culture. A diversity of yeast species is encountered worldwide. *Saccharomyces cerevisiae*, *Candida humilis*, *Kazachstania exigua*, *Pichia kudriavzevii*, *Wickerhamomyces anomalus*, and *Torulaspora delbrueckii* are among the most common ones. Sourdough-adapted yeasts are able to withstand the stress conditions encountered during their growth. Those include nutrient starvation as well as the effects of acidic, oxidative, thermal, and osmotic stresses. From a technological point of view, their metabolism primarily contributes to the leavening and flavour of sourdough products. Besides ethanol and carbon dioxide, yeasts can produce metabolites that specifically affect flavour, such as organic acids, diacetyl, higher alcohols from branched-chain amino acids, and esters derived thereof. Additionally, several yeast strains possess functional properties that can potentially lead to nutritional and safety advantages. These properties encompass the production of vitamins, an improvement of the bioavailability of phenolic compounds, the dephosphorylation of phytic acid, the presence of probiotic potential, and the inhibition of fungi and their mycotoxin production. Several strains of diverse yeast species are new candidate functional starter cultures, as they offer opportunities beyond the conventional use of baker's yeast.

02:30 p.m. The first World Sourdough Library

Maria De Angelis [1], Stefan Cappelle [2], Fabio Minervini [1], Guylaine Lacaze [2], Anna Lattanzi[1], Bernard Genot[2], Raffaella Di Cagno [1], Marco Gobbetti[1]

[1] Dipartimento di Scienze del Suolo, della Pianta e degli Alimenti, Universita degli Studi di Bari Aldo Moro, Bari, IT

[2] Puratos NV, Groot-Bijgaarden, BE

Traditional sourdoughs represent an immense source of microbial diversity, resulting from the combination of ingredients, protocols of propagation and house microbiota. Such microbial diversity strongly affects the peculiar qualities of the derived baked goods(1). The microbial community of traditional sourdough is often subjected to unpredictable fluctuations that may be not always beneficial to the quality of the product (2). Based on these premises the first World Sourdough Library was conceived and created in Saint Vith (Belgium). It aims at preserving all the components of sourdough: protocols of propagation, pure microbial cultures, and the sourdoughs themselves, in a fresh or frozen state.

Currently the Sourdough Library is composed of more than 90 sourdough samples collected in different countries and microbiologically and biochemically characterized. *Lactobacillus sanfranciscensis* and *Saccharomyces cerevisiae* were the red tread for most of the sourdoughs. Overall, bacterial diversity was higher than yeasts. Besides *L. Sanfranciscensis, L Plantarum* and *Leuconostoc sp.* were identified, with a frequency

varying from country to country and from sample to sample. Unusual bacterial species (*Lactobacillus xiangfangensis* and *Lactobacillus diolivorans*) were found in sourdoughs collected in France. In some sourdoughs *S. cerevisiae* was replaced by *Candida humilis* or by species of *Kazachstania genius* . Three sourdough collected in Hungary showed the presence of *Saccharomyces uvarum, Candida zemplinia* and *Metschnikowia sp.*, which are rarely encounter in the sourdough ecosystem.

03:00 p.m. Moderated audience discussion "Health aspects of yeast and sourdough in bread meaking"

Moderator: Klaus Lösche

Audience contributions are welcome!

03:15 p.m. Coffee break

Analytics

03:30 p.m. Raising Power Probe (RPP)

Ann-Maria Dinse [1]; Klaus Lösche [2]

[1] Versuchsanstalt der Hefeindustrie e. V., GER

[2] NFT GmbH, GER

The application of yeast in a variety of different bakery products involves a permanent improvement and monitoring of the yeast quality for bakery products, in yeast production as well as in the application in traditional and industrial bakeries. Yeast as a baking ingredient influences a number of properties of the bakery goods (for example crumb and flavor). But the most obvious effect is the lifting of the dough by production of CO_2, the raising power of the yeast. To ensure a constant product quality of bakery products, it is necessary to determine that raising power. Another important value to determined is the capability of the dough to hold the produced CO_2. Both together define the volume of the final baked dough.

In cooperation with BlueSens gas sensor GmbH, the VH Berlin e. V. has developed a new measurement system to combine the measurements to satisfy those needs. The analysis software will be presented together with evaluation measurement results in comparison to the established SJA Fermentograph/VolScan Profiler. The lecture will also show the current and future trend of quality assurance in the bakery production and what challenges result from this.

04:00 p.m. Identification of organic ingredients in complex food samples by Next Generation Sequencing NGS approaches

Karsten Liere , Martin Meixner

SMB GmbH, GER

Metagenomics is the study of genetic material recovered from mixed samples. In order to identify the different species within those samples, a specific genomic region is exclusively amplified and sequenced, generating so called 'barcodes' for each analyzed species. E.g., bacterial species can be identified by their rDNA (genes for ribosomal RNA) sequences, with each specific variant serving as the species' fingerprint. Similarly, eukaryotic species (plants as well as fungi) are identified by amplification and sequencing of their rDNA internal transcribed spacer (ITS) sequences or their mitochondrial cytochrome c oxidase 1 gene. Individual species from a mixed sample are then identified by comparison of these 'barcodes' to databases with known sequences.

Using a high throughput technique such as Next Generation Sequencing (NGS), one can identify not only the most prominent organism, but also other candidates within a sample. Depending on sample and organisms, even rough amounts or ratios can be estimated.

We will present some interesting examples from our day-to-day work of using NGS to identify the content of organisms in/on different samples.

05:00 p.m. Shuttle transfer to GEA

07:30 p.m. Conference dinner and convial evening at GEA (until 11 p.m.)

Wednesday, April 20th 2016

Applied Research

9:00 a.m. Patent review – genomic enineering of yeast for food applications

Erik Pollmann

Versuchsanstalt der Hefeindustrie e. V., GER

Genomic engineering of microorganisms to facilitate improved or even new metabolic products is becoming increasingly common in many industries. The field of 2nd generation bio fuel production sees many genomically engineered strains of S.cerevisiae in the patent process, but engineered yeasts for food applications are not as prevalent.

This patent review from the VH Berlin Refworks database shows some of the current developments in regards to genetical modification of S.cerevisiae for food processes to see where industry trends may lead.

09:30 a.m. Genomic analysis of the wine yeast *Hanseniaspora uvarum* / *Kloeckera apiculata* - what can we learn about glycolysis and aroma production?

Christian von Wallbrunn [2], Jürgen J. Heinisch [1], Anne-Kathrin Langenberg [1], Melanie Wieschebrock [3], Hans-Peter Schmitz [1]

[1] Universität Osnabrück, GER

[2] Hochschule Geisenheim, GER

[3] Deutsche Institut für Lebensmitteltechnik e. V. (DIL), GER

The yeast *Hanseniaspora uvarum* (commonly known as *Kloeckera apiculata*) is the predominant fungal species on grapes, apples and other fruits. It constitutes more than 80% of the yeast population on grapes and in the must prior to wine fermentation. Due to its high capacity for aromatic ester production, it is believed to have a major influence on wine quality. Even if starter cultures of *Saccharomyces cerevisiae* are employed, *H. uvarum* dominates the first day of must fermentation. We have sequenced >90% of the genome and identified several homologs of genes encoding enzymes involved in alcoholic fermentation and ester production. A systematic analysis of the specific activities of glycolytic enzymes indicated that key steps in the lower part of glycolysis are responsible for the diminished capacity for alcohol production in *H. uvarum* as compared to *S. cerevisiae*. The presentation will address these results as well as data obtained from the heterologous production of genes encoding alcohol esterases from *H. uvarum* in other yeasts.

10:00 a.m. Production of glutathione enriched yeast: Opportunities for process optimization.

Martin Senz [1], Eric Lorenz [2], Maximilian Schmacht [1]

[1] Research and Teaching Institute for Brewing in Berlin (VLB), Department Bioprocess Engineering and Applied Microbiology, GER

[2] Berlin University of Technology, Institute of Biotechnology, Chair of Bioprocess Engineering, GER

Based on its reducing characteristics, preparations of glutathione (GSH) enriched yeasts can be used in food industry as dough modifier. Resulting products have no need for declaration, as it would be the case for instance by applying cysteine (E 920). There are various strategies for the fermentation process of GSH production, the most common being fed-batch procedure with the yeast *Saccharomyces cerevisiae*. For an efficient production process, factors like medium costs and process robustness at various scales, as well as achieving high cell density with high intracellular GSH content, are of high relevance.

This presentation provides a compilation of different process optimization strategies for the production of GSH-enriched yeast cells. Different studies addressing the fermentation format, feeding strategy, medium composition, precursor for GSH transformation and its type of supplementation, are presented. Promising process formats were varied concerning the applied yeast strain and process scale. Thus, insights and prospects for current and further process developments in GSH production via yeast are offered.

10:30 a.m. Coffee break

11:00 a.m. Analysis of ion mobility spectrometry data to monitor metabolic processes in yeast and detect change points

Sven Rahman

University Hospital Essen, University of Duisburg-Essen, GER

The yeast S. cerevisiae is widely used in the food and pharmaceutical industry for diverse applications, such as food conservation, ethanol production, and synthesis of pharmaceutically relevant proteins. Efficient production (at high rate) depends on an optimal (as high as possible) glucose concentration, yet avoiding unfavorable conditions, such as diauxic growth, pseudohyphal growth or glucose repression induced fermentative metabolism under aerobic, that could lead to the loss of an entire fermentation run.

At present, these unfavorable conditions are difficult to detect before it is too late and the whole batch transits to a different state (i.e., ethanol production) and is lost. In the project YeastScent, we aim to establish volatile marker metabolites that can be used as inputs for a feed controller in order to attain robust fermentation of yeast.

Establishing a novel original fermenter monitoring technology based on ion mobility spectrometry, YeastScent will contribute analytical techniques and biological insights required for fermentation process operation.
In my talk, I will highlight the bioinformatics approaches that we are using to detect and quantify peaks in multi-capillary-column-coupled ion mobility spectrometrymeasurements in order to scan fermenter off-gas for volatile metabolites and detect concentration changes as early as possible, alerting us to changes in yeast metabolic state. To avoid the presentation becoming too technical, I will focus on principles behind methods instead of too many implementation details.

11:30 a.m. Impact of oscillating oxygen and substrate concentrations on the sterol synthesis in *Saccharomyces cerevisiae* fed-batch cultivations

STEFAN JUNNE, ANNA-MARIA MARBÀ-ARDÉBOL, PETER NEUBAUER

Technische Universität Berlin, Department of Biotechnology, Chair of Bioprocess Engineering, GER

Gradients of substrate and oxygen occur during the course of a typical nutrient-limited fed-batch cultivation in industrial scale. Thus, cells are exposed to oscillating conditions, which can disturb the process performance, e.g. by a reduced growth rate, unwanted side-product formation or a decreased product yield. Since usually lab-scale bioreactors do not offer suitable conditions for a realistic approach to mimic heterogeneous conditions of the large scale, scale-down reactors can be applied.

In this study, a two-compartment reactor comprising of a stirred tank reactor and a plug flow reactor and an extended version of a scale-down reactor, which comprises of a second plug flow module (see figure 1), is used to study the effects of oscillating conditions on the metabolite accumulation of the main carbon and sterol biosynthesis in the yeast *Saccharomyces cerevisiae*. The latter metabolism requires oxygen as co-factor for multiple conversion steps. Thus, oxygen depletion as it occurs near the addition of the feeding solution in a fed-batch cultivation due to substrate excess becomes crucial for sterol synthesis.

In the presented study, regulation in the sterol pathway as observed earlier in yeast when shifted from oxygen-limited to oxygen excess, is investigated under scale-down cultivation conditions. Surprisingly, the accumulation of sterol pre-cursors leads to increased sterol formation, while under certain conditions the accumulation of the first sterol-pathway specific compound squalene indicates a rate limitation caused by the lack of oxygen supply.

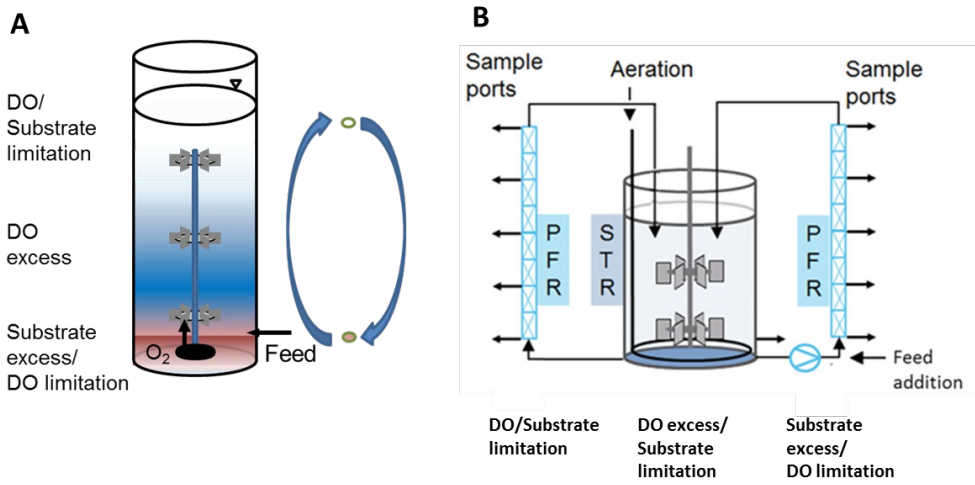


Fig 1.: Schematic draw of substrate and dissolved oxygen gradients in large-scale fed-batch cultivations (A); Three-compartment scale-down reactor comprising of a well-mixed stirred tank reactor (STR) and two plug flow reactor (PFR) modules (B).

12:30 p.m. Fast, direct and high-sensitivity profiling of microbe volatome: the potential of PTR-ToF-MS for on-line determination, automatized analysis, and massive screenings

VITTORIO CAPOZZI [1], LUCA CAPPELLIN [2], FLAVIA GASPERI [2], ANA SANCHEZ-JIMENA [3], MATTEO SCAMPICCHIO [4], GIUSEPPE SPANO [1], AND FRANCO BIASIOLI [2]

[1] University of Foggia, IT

[2] Research and Innovation Centre, Fondazione Edmund Mach, IT

[3] Lallemand Baking Solutions, FR

[4] Free University of Bolzano, IT

The volatile subset of the metabolome of eukaryotic microbes, often indicated in an -omics perspective as 'volatome', is of fundamental and applicative interest in several fields as food science and technology, environmental sciences, biotechnology and

medical studies. In addition, its monitoring represents a powerful tool to develop and confirm new hypotheses in biology, breeding sciences, and yeast products innovation.

Among the possible approaches to volatile compounds analysis and monitoring, Proton Transfer Reaction Time of Flight Mass Spectrometry (PTR-ToF-MS) represents a valid compromise between sample throughput and analytical insight, with the advantage of on-line process monitoring and non-invasive analysis. In more details, PTR-ToF-MS allows the rapid (an entire spectrum is acquired in a split second), high-sensitivity (sub ppt) and direct analysis of the sample headspace without any treatment or pre-concentration. We further increased these performances coupling PTR-ToF-MS with an auto-sampler and tailored data analysis tools.

We demonstrated the applicability of our comprehensive methodology (automatic sampling, rapid PTR-ToF-MS analysis and tailored data handling and analysis) in the study of volatile organic compounds released during alcoholic fermentations. In particular, we used this analytical approach i) to differentiate the volatome of bakery yeast starter cultures in the doughs, ii) to assess the effect of different bakery yeast starter cultures/flour combinations, ii) to evaluate the interaction between *Saccharomyces cerevisiae* and *Lactobacillus sanfranciscensis* as model microorganisms in the sourdough environment, iii) to distinguish different traditional sourdoughs, iv) to characterize bakery commercial aromatic yeast starter of wine and beer origin, v) to study the yeasts/hops combinations during beer production.

More generally, PTR-ToF-MS can be used for the high-throughput and high-sensitivity profiling of samples volatome and for the direct on-line/off-line monitoring of any bioprocess.

12:30 p.m. Lunch break

Process

01:30 p.m. Results of the study of the sugar mills ESST-Working Group „Nitrite in Feed“

STEFAN FRENZEL

Südzucker AG, GER

In 2010 the nitrite content of feed materials in general and especially of those from sugar industries gained new interest due to a European legislation setting maximum limits to some materials not limited so far. Directive 2010/6/EU amending Directive 2002/32/EC, a nitrite limit of 15 mg/kg, expressed as sodium nitrite at 12% moisture content, became applicable to feed materials, including those of the sugar industry, with the exception of silage. From a quick scan, it became clear in the beginning of 2010 that this nitrite limit could cause major problems, at least for some feed products. More data and better comprehensive knowledge of the situation was needed. Therefore, a discussion was started within the European sugar industry and EU authorities and it was decided to establish an expert group to push ahead investigations on this matter. This Study Group “Nitrite in Feed” was commissioned at the 2nd Conference of the European Society for Sugar Technology (ESST) in Bratislava in 2011. It is composed of scientists and technologists from several sugar companies and research centres in Europe. The presentation includes monitoring results of the companies participating in this ESST expert study group on “Nitrite in feed”. It will be obvious that the complex behaviour of nitrite in the sugar extraction process overlaps with external effects such as growth condition of the beet which are not under the control of the process owner. Among this as for the time being there is no standardized and validated method available for the determination of nitrite in the complex matrix of molasses or other feed products of the sugar industry.

02:00 p.m. Investigations in self-digestion and separation of yeast extract

JANA MÜLLER, STEFFEN HRUSCHKA, JOACHIM WEINEKÖTTER

GEA, GER

Autolysis (Self-digestion) of yeast saccharomyces cerevisiae means that the cell wall structure is destroyed and inner cell liquid passes through into the outside continuum. This is a biological process induced by enzymes. These enzymes can be natural enzymes (endogenous) of the cell or added enzymes. Finally a liquid-solid separation forms a liquid phase, the extract and a solid phase formed by the empty cell walls and the other remaining undestroyed yeast cells.

If the process is highly efficient, the higher the extract volume and the dry matter in the extract are, but without remaining cells. The task of this investigation was to apply a Nano Reactor® for this purpose. The inventor of the Nano Reactor® informed accordingly:

A Nano Reactor® is a static device with no moving parts. Its name is due to the creation of nanometer sized bubbles while the fluid is pumped through the Nano Reactor® under high pressure. The Nano Reactor® creates intense hydrodynamic cavitation with shockwaves that can disrupt the yeast cell walls.

The self-digested yeast cells are pumped through a series of scientifically designed geometries. At each stage, there is a dramatic pressure drop. The water molecules in the cells vaporize and recompress to a liquid at each stage and create shockwaves that may possibly break cell wall structure.

Process flow

In an experimental set-up, the Nano Reactor® was installed behind an agitator tank. The tests were performed with yeast cells from different levels of the standard and the enzymatic autolyze process by a producer in Germany. A high pressure pump is used to push the yeast cells from the agitator tank into the Nano Reactor®. The high pressure pump elevates the pressure of the yeast cells-suspension to a pressure of 40 -80 bar. This pressure is dissipated as the yeast cells passes through the Nano Reactor®. The high pressure is regulated by means of a frequency variator acting on the high pressure pump motor as a function of the pressure differential.

The sampling of the outgoing yeast cells is transferred to conical flasks in lab-scale and processed as in the real process until the separation of yeast extract from cell walls.

In addition, a multiple step in the Nano Reactor® was applied. Afterwards, the outgoing yeast cells were recycled into the stirring tank and finally passed again through the Nano Reactor®. The circulation was repeated during the entire reaction time.

The presentation shows the results of these tests. It is shown that there is an influence of the Nano Reactor® on the process. But the results do not give a clear correlation between investigated influencing parameters like pressure, implementation steps of the Nano Reactor® of the process and different pretreatment of yeast.

02:30 p.m. Phosphorus Recovery via Hydrothermal Carbonization (HTC) - Prospective Applications

DOMINIK WÜST, GERO BECKER, ANDREA KRUSE.

University of Hohenheim – Institute of Agricultural Engineering, GER

Availability of phosphates as fertilizers for providing food stability and security becomes worse, because natural resources are finite within a foreseeable period of 300 to 400 years (IFDC 2010). However, quality will decrease by increasing heavy metal contents (Sabiha et al. 2009). Additionally, availability and thus mining of phosphate rock is limited to few countries in Africa and Arabia (Röhling 2012). In these countries miners are working under worst conditions with health risks (Mening 2015). Due to these circumstances the European Commission has admitted phosphorus (P) to the list of Critical Resources recently. Therefore, implementation of P recovery in established agricultural, municipal and industrial processes becomes more and more important.

As a first step to recover P from organic waste Hydrothermal Carbonization (HTC) has to be conducted. Water as reaction medium affects the functional groups of the biomass. These chemical reactions induce decomposition of cellular structures and water is released (Kruse et al. 2012). Further chemical reaction chains such as condensation and polymerization of dissolved compounds result in formation of a solid product, termed hydrochar (Kruse et al. 2013, Kruse und Dahmen 2015). Thereby, the main reactants are dissolved organic carbon compounds (DOC), nitrogen compounds, salts and metals. In the end, the liquid product, termed process water, contains the organic compounds such as acids, alcohols, ketones, aldehydes, phenols, and inorganic compounds such as ammonia and phosphates which have not reacted (Dead-Ends).

Before hydrochar, which contains most of the phosphates, can be leached with organic or mineral acids it has to be separated from the process water and be dewatered. This procedure has already been applied for P recovery from sewage sludge and its ashes (BAFU 2013, Weidelener 2010). Finally, there is a P-reduced hydrochar with increased carbon content and thus a higher heating value. Either it can be used as energy carrier or as active carbon due to their oxygen-containing functional groups and high specific inner surface after physical or chemical activation.

The process water which is well-known as a valuable by-product of HTC or hydrothermal conversion processes (Biller et al. 2012) can also be used within this process combination. By mixing P-leachate and process water, enhancing the pH to 9 by sodium hydroxide addition and an addition of magnesium chloride Magnesium-Ammonium-Phosphate (MAP) can be crystallised. In this way, approximately 80 % of the phosphates can be recovered from sewage sludge.

P-recovery via HTC can be seen as applicable for agricultural, municipal and industrial processes, whenever it is possible to close nutrient cycles by utilizing P-rich and wet organic waste.

03:00 p.m. Feedback and final remarks

03:30 p.m. End of the conference