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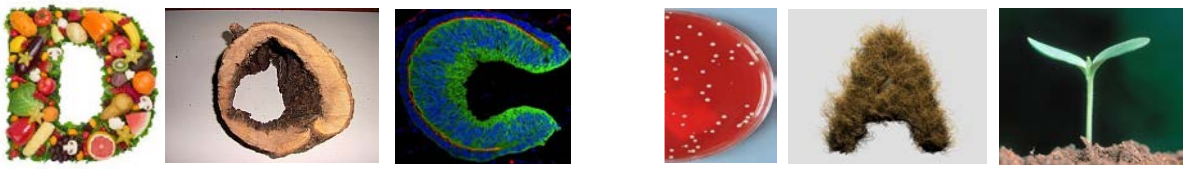

PhD Conference

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BOKU BiRT BioResources & Technologies Tulln

University of Natural Resources and Life Sciences, Vienna

AIT AUSTRIAN INSTITUTE OF TECHNOLOGY

3rd DocDay- Book of Abstracts

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PREFACE

Who is BiRT? And how is BiRT associated with the DocDay?

Such questions may arise if you participate in the DocDay 2015 event taking place at BOKU campus Tulln on October 13, 2015.

BiRT is an acronym for Bio-Resources & Technologies Tulln, an organisational unit at BOKU which thematically integrates all BOKU departments which are fully or partially located at Tulln. BiRT integrates virtually all fields of research and teaching of BOKU´s research groups at Tulln and represents a highly interdisciplinary domain covering the process chains and cycles of sustainable production, processing and utilization of bio-resources for various human demands including food, feed, and renewable, bio-based materials, chemicals and energy. BiRT serves as a platform of discussion, exchange of ideas and development of joint projects in research and teaching and aims at enhancing BOKU´s profile and visibility at campus Tulln.

One of BiRT´s most important achievements has been the annual organisation of the DocDay. The DocDay aims at providing a forum for all PhD students at campus Tulln who want to share their work with their colleagues and supervisors in a highly interdisciplinary environment, and to use the opportunity for networking activities.

On behalf of the BiRT Steering Committee I am glad to welcome you and to wish you a successful and interesting DocDay 2015. If you want to learn more about BiRT please visit our website at http://www.boku.ac.at/wissenschaftliche-initiativen/birt/.

Univ. Prof. DI. Dr. Walter W. Wenzel

Head of BiRT

Univ.Prof. Walter W. Wenzel
Universität für Bodenkultur Wien

Dear colleagues,

Welcome at the 3rd DocDay 2015!

The DocDay is an interdisciplinary conference organized for and by PhD students working at all units (IFA, UFT-BOKU und AIT) on campus Tulln. It is also a unique opportunity for us to present our work, exchange ideas and get to know each other better. This year we can look forward to a rich scientific program comprising of 18 talks and 30 posters. I think we can be proud of that!

I would like to thank the BiRT initiative for the financial support, as well as Prof. Erika Staudacher for opening our conference, Prof. Manfred Grasserbauer for holding the key note lecture, the members of the scientific committee for evaluating the best oral presentation, my colleagues from the VIRIS research group for all the hard work and of course you for participating at this event!

I wish you a great DocDay and look forward to an interesting conference and of course a nice beer together at the Oktoberfest afterwards!

Anastassiya

PROGRAM

8:30	Registration
9:00	Opening - Welcome Words
9:15	**KEYNOTE LECTURE** **Manfred Grasserbauer (Vienna University of Technology)** *Global Sustainable Development: Challenges for Science and Technology*
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10:15	**Reza Omidvar (UFT-BOKU)** *Functional Analysis of Defensin-like Peptide from Arabidopsis thaliana*
10:30	**Noemie Prat (IFA)** *Improvement of Fusarium Head Blight Resistance in Durum Wheat*
10:45	**Stefan Bödi (UFT-BOKU)** *Infection process restores defective DON production in a F. graminearum Heterochromatin protein 1 (Hep1) mutant*
11:00	**Siegrid Widhalm (UFT-AIT)** *Isolation and functional characterization of bacteria associated to common ragweed and their utilization as biocontrol agent*
11:15	Coffee Break
	MICROBIOLOGY AND METABOLOMICS Chair: ANNA WAWRA (UFT-AIT)
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12.00	**Alexandra Simader (IFA)** *Development of a comprehensive metabolomics database system*
12:15	**Hans Yu (IFA)** *Lectin based enrichment of glycoproteins of oviductal fluid samples and subsequent nano-LC-MS/MS analysis*
12:30	**Nora Odabas (UFT-BOKU)** *Pulp cationization in different solvents*

12:45	**LUNCH BREAK & POSTER SESSION**
	BIOREFINERIES AND WOOD TECHNOLOGY Chair: JOHANNES DRAXLER (UFT-BOKU)
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14:45	**Johanna Schritter (IFA)** *Microbial processes in hydrogen exposed porous underground gas storages (UGS)*
15:00	**Josua Timotheus Oberlerchner (UFT-BOKU)** *From Monomers to Polymers across the Oligomeric Region: Advanced Biorefinery Analytics*
15:15	**Cornelia Haas (IFA)** *Utilizing agro-industrial residues with a low carbon content for bioplastic production*
15:30	**Tillmann Meints (UFT-BOKU)** *Wood treatment with bulking and modification agents to reduce the swelling and shrinking behaviour*
15:45	Coffee Break
	ENVIRONMENT AND HEALTH Chair: MONIKA HORSKY (UFT-BOKU)
16:15	**Theresa Rosenkranz (UFT-BOKU)** *Potentials of phytomining from waste incineration bottom ash using hyperaccumulating plants*
16:30	**Abdul Samad (UFT-AIT)** *Microbial communities associated with grapevine and vineyard weeds*
16:45	**Ondrej Hanousek (UFT-BOKU)** *Analysis of sulfate $\delta^{34}S$ in soil by DGT MC ICP-MS*
17:00	**Johannes Draxler (UFT-BOKU)** *Quantitative determination of the elemental distribution of biodegradable Mg alloys in bone tissue by LA-ICP-MS*
17:15	Short Break
17:30	**Awards Ceremony**
18:00	**Oktoberfest**

ABSTRACTS SESSION 1:

PLANT, FUNGI AND BACTERIA

Suitable protectants in formulation of the plant growth-promoting bacterium *Burkholderia phytofirmans* PsJN

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The endophytic, Gram negative bacterial strain *Burkholderia phytofirmans* PsJN is known to be an effective plant growth-promoter and may thus be applied as an alternative to agrochemicals. Beneficial influences have been observed in a variety of host plants, including maize and wheat. However, transfer of these positive results from controlled conditions to the field has been largely unsuccessful. This is due to the low shelf life of these bacteria and stress factors such as desiccation. For this reason, it is crucial to develop a suitable formulation that maintains bacteria in a stable, “dormant” state and protects them during handling and application.

However, also the formulation process poses stress on the bacterial cells. Therefore, a range of additives was evaluated regarding their protective effect during lyophilisation (freezing + desiccation stress), air drying (desiccation stress) and exposure to heat as it may occur during spray drying. The additives were chemically heterogeneous and included trehalose, sucrose, galactose, lactose, sorbitol, mannitol, glycerol, ficoll, corn starch, carboxymethylcellulose, maltodextrin, gum arabic, alginate, skimmed milk, yeast extract, gelatine, PEG 6000, DMSO, humic acid and LB medium. Furthermore, the bacteria were processed in form of a biofilm (including self-produced exopolysaccharides). Bacterial viability was measured after drying/heat stress exposure by drop-plating and colony counting. The storage stability was evaluated at different temperatures (48 °C, 37 °C, 22 °C, 4 °C).

We demonstrated that specific additives dramatically improved the survival rate of bacteria during drying by up to 100,000-fold and also supported a high shelf life. These observations help identifying a suitable product composition, which is a major prerequisite for successful application of plant growth-promoting bacteria in the field.

The authors thank the Austrian Research Promotion Agency (FFG) for financial support of the FEMtech project “EndoCaps” (No. 839341).

Functional Analysis of Defensin-like Peptide from *Arabidopsis thaliana*

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Plant defensins are small, basic peptides that have a characteristic three dimensional folding pattern that is stabilized by four disulphide bridges. In addition to the previously known defensin genes, more than 300 DEFLs (defensin-like) genes coding for small cysteine-rich peptides (CRP) have been discovered in the Arabidopsis genome. We are studying a subgroup with 9 defensin-like (*PdfL*) genes with special emphasis on *PdfL2.1*. We produced the PDFL2.1 peptide for *in vitro* tests in the cytoplasm of the *E. coli* strain C3030 as a His-tagged thioredoxin fusion protein with a TEV recognition sequence. The purified peptide showed strong inhibitory effect against two tested *Fusarium* species. Microscopic analysis showed that in case of *F. graminearum* distinct morphological changes including hyperbranching and swelling of fungal cells, that are indicative of antimicrobial activity, could be observed. This peptide is only slightly basic while the majority of known antimicrobial peptides, including plant defensins, are usually basic with a pI above 8. It was therefore interesting to further study the function of this PDFL peptide. The structure of PDFL2.1 was determined using ^{13}C-^{15}N solution NMR with Bruker 850-MHz system. The structure is very similar to the canonical plant defensins with a common core structure that consists of an α-helix and an anti-parallel triple-stranded β-sheet.

Acknowledgements
All NMR data were acquired at MNMR Center, University of Minnesota. Mass spectrometry was done at the University of Minnesota Center for Mass Spectrometry and Proteomics. The authors thank Dr. Charles Schwieters for the discussion of the use of XPLOR-NIH.

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Improvement of Fusarium Head Blight Resistance in Durum Wheat

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Fusarium head blight (FHB) is one of the main diseases affecting cereal cultivation worldwide, causing direct harvest losses and affecting food quality and safety through accumulation of hazardous mycotoxins in contaminated grains. Cropping of resistant varieties plays a key role in integrated disease control. In durum wheat (*Triticum durum*) the development of resistant cultivars remains a challenge for breeders due to its extreme susceptibility and the small variation available for FHB resistance in durum wheat gene pool.

In this study we aimed to enhance durum wheat resistance level by using resistance derived from bread wheat (*Triticum aestivum*), a cultivated related species. Three populations were developed crossing respectively three susceptible European durum wheat varieties with a resistant donor line carrying *Fhb1*, a major FHB-resistance QTL derived from *T. aestivum*. The lines derived from these crosses were evaluated during three seasons in field experiments using artificial spray-inoculation of *Fusarium* conidia. The populations showed large spectrum of response for FHB resistance ranging from highly resistant to susceptible. In order to identify genomic regions associated with enhanced resistance, the lines were genetically fingerprinted using molecular markers. Several chromosome locations were associated with improved resistance notably on chromosome 3B, corresponding to *Fhb1*, and on chromosome 4B.

Our results are promising for future resistance breeding as, for the first time, successful introgression of *Fhb1* in the background of durum wheat is reported, providing stable and enhanced resistance. Morphological characters appear to play an important role in the observed variation for resistance as the FHB-resistance QTL identified on chromosome 4B overlaps with QTL for plant height.

The results are directly applicable for durum wheat breeding and may pave the way to improved FHB resistance level by combining resistant alleles from diverse sources into durum wheat cultivars.

We gratefully acknowledge financial support from the French Ministry of Higher Education and Research, CIFRE funding 2012/1405

Infection process restores defective DON production in a *F. graminearum* Heterochromatin protein 1 (Hep1) mutant

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Heterochromatic marks are known to be involved in the regulation of secondary metabolite gene clusters in saprophytic fungi. To test the influence of chromatin-level regulation on metabolite production and virulence in a plant pathogenic fungus, we deleted the heterochromatin protein-1 homologue, called Hep1, in *F. graminearum*, a species causing head blight disease of wheat. In axenic culture the *hep1* deletion mutant showed a strongly altered secondary metabolite profile including significant reduction in toxin levels of deoxynivalenol (DON) and its acetylated derivative 15ADON. Interestingly, the addition of a simple plant-derived DON/15ADON inducer (ornithine [1]) to synthetic media could not restore the defective trichothecene production in the mutant. In contrast, on dead wheat heads, the most similar saprophytic substrate for the pathogenicity assays, DON/15ADON production already was higher in the *hep1* deletion mutant compared to the Ph-1 wild type. This indicates that pre-existing plant metabolites are present in dead wheat heads which are able to counteract the negative effect on DON/15ADON production in the heterochromatin mutant. In the following infection assays on living wheat heads, the *hep1* deletion strain significantly exceeded the wild type levels of DON/15ADON production. Consistently the mutant also showed stronger disease symptoms and a ~ 1.5 fold higher overall infection rate on the wheat cultivar *Remus*. These results indicate that the plant response to infection is upregulating the production of metabolites able to overdrive a repressive genetic network responsible for suppression of DON/15ADON production in the *hep1* deletion strain. RNA-seq based transcriptome analysis of the saprophytic and pathogenic growth states was done to identify genes with potential function in chromatin-related pathogenicity control.

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Isolation and functional characterization of bacteria associated to common ragweed and their utilization as biocontrol agent

Siegrid Widhalm, Angela Sessitsch, and Friederike Trognitz

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Common ragweed represents a potent allergenic species that has greatly expanded on a global scale. Being a native plant of North America, the wind-pollinated annual ragweed plant has been repeatedly introduced into Europe as a contaminant in crop and birdseeds from the 19th century onwards. However, it started to invade large parts of SE and SW Europe only when changing conditions in land use and climate allowed it to spread. It is prospected that by 2050, airborne ragweed pollen concentration will be four times higher than they are today.

The use of herbicides against common ragweed in agriculture crops is not available against this noxious plant. As no plant pathogens are known for ragweed, the exploitation of the plant-associated microbes of the plant may yield new biocontrol agents.

As a first step cultivable bacteria were isolated from common ragweed, following by a rigorous screening for deleterious and beneficial microbes. The federal state Burgenland was selected as sample site, as the state with the earliest introduction of common ragweed in Austria. From this region three ragweed infested sites were chosen, with 3-4 plant per site. From three different plant parts the bacteria were isolated. Endophytic bacteria were isolates from stem and root and ectophytic from the rhizosphere. Taxonomic classification was done using the 16S rRNA gene. Furthermore, the isolates were selected for functional characteristic studies depending on the sequence similarity using the Neighbor-Joining method. Additional the bacteria were functional characterized for beneficial and biocontrol properties.

In total 46 different genera were found. *Bacillus*, *Microbacterium* and *Pseudomonas* could be detected in all 3 locations and in all 3 plant parts. Genera specific for a sub-sample site as well as for a plant compartment could be distinguished. Five *Pseudomonas* were chosen based on their characteristics to test of herbicidal effect on in vitro plants. The results will be discussed.

ABSTRACTS SESSION 2:

MICROBIOLOGY AND METABOLOMICS

Formulations of chitosan incorporating Cellobiose Dehydogenase: antimicrobial systems for the treatment of wound infection

Gregor Tegl, Barbara Thallinger, Bianca Beer, Gibson S Nyanhongo and Georg M Guebitz

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The infection of wounds caused by bacterial contamination still constitutes a major issue in health care. An accumulation of organisms promotes biofilm formation that consequently increases resistance and hampers therapy. In order to prevent microbial colonization, a timely and efficient antimicrobial therapy is of great importance. Only a short list of invented antimicrobial agents is integrated in routine therapeutic items, facing several issues like retarding wound healing processes and antibiotic resistances [1].

Within this work formulations with enhanced antimicrobial activity were prepared. Therefor the natural polymer chitosan in combination with cellobiose dehydrogenase (CDH) was employed. Chitosan is a prominent biopolymer exhibiting antimicrobial activity that is mainly caused by its high content of primary amines facilitating the attachment on the bacterial cell surface [2]. The flavoheme enzyme CDH is known to produce the antimicrobial agent hydrogen peroxide. Recent studies utilized this enzyme for antimicrobial and antibiofilm functionalization [3]. In the presented study, different strategies for the preparation of CDH/chitosan precipitates were implemented. The precipitates were grinded to particles and investigated towards their CDH content, leaching of enzyme and its antimicrobial effect. Different leaching behaviors were observed within the different particle systems. An antimicrobial assay was developed that enables simultaneous detection of the optical density and colony forming units. Thereby hydrogen peroxide production by CDH was induced by the addition of cellobiose in solution as well as by the addition of cellobiose containing particles. Significant differences in bacterial growth were observed when applying CDH-chitosan formulations.

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Development of a comprehensive metabolomics database system

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Metabolomics aims at the determination of the complete low molecular weight metabolite complement of a biological system.

Most metabolomics studies produce huge amounts of data and require the documentation of a wide range of experimental metadata. Manual data evaluation as well as administration of metadata would be unfeasible or at best extremely time consuming and make the use of custom-tailored databases necessary.

As part of my PhD thesis I have started to extend an existing metabolomics database which enables easy and well-organised access to all data in a combined manner. The database system shall be suitable to handle, store and combine big data from different experiments or organisms, which can be derived from literature or experimentally. Currently the system is able to differentiate between GC-MS and LC-HRMS data and stores a literature derived catalogue of compounds occurring in wheat plants, which is continuously extended by the metabolomics group. Until now, it is possible to query and manage general information for compounds like m/z ratio, retention time, retention index, different substance identifiers with hyperlinks to other databases, as well as compound classes, compound structure and reference MS spectra, if available.

The presentation will exemplify the successful use and benefit of the developed database with metabolomics data from different wheat studies. It will be demonstrated that the already existing and continuously developing catalogue of compounds managed within a database system can greatly promote metabolite annotation and biological interpretation. Moreover, an outlook on the future developments of the database system will be presented.

The authors thank the Austrian Science Fund (project SFB Fusarium F3706-B11) for the financial support.

Lectin based enrichment of glycoproteins of oviductal fluid samples and subsequent nano-LC-MS/MS analysis

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Secreted proteins and metabolites from cells exhibit important functions, e.g. in the formation of the extracellular matrix (ECM) and in signal transduction. Secreted proteins are often glycosylated and it is known that glycan structures confer essential features for cell-cell recognition [1]. Glycoproteins also exert a crucial role in mammalian reproduction [2]. Secreted glycoproteins from the oviductal epithelium are assumed to be involved in modulating the function and fertilizing ability of male gametes [3]. However, so far no detailed comprehensive analysis of the glycoproteome of the oviductal fluid exists.

Thus, the focus of the present study was the enrichment of glycoproteins of oviductal fluid samples from rabbits (*Oryctolagus cuniculus*). Glycoproteins were enriched using a tandem approach with both Concanavalin A (ConA) and Wheat Germ Agglutinin (WGA) lectin beads. The specificity of the enrichment step was evaluated by 1D and 2D gel electrophoresis combined with glycospecific staining. Protein spots were further subjected to vacuum MALDI-TOF/TOF based identification. In addition the reproducibility of the enrichment was analysed by nano-LC-MS/MS. The obtained protein classes of both fractions were compared as well.

Preliminary results indicate a clear separation of proteins into eluate and flow through fractions. A variety of lectin enriched proteins, including oviductal glycoprotein 1 were identified. It is also noteworthy that proteins from the complement system, ECM components, and proteins related to the immune system were identified.

Taken together, we present an applicable enrichment method for glycoproteins. Further, this method will enable the analysis of the role of glycoproteins in reproductive processes in the oviduct.

This project is funded by Niederösterreichische Forschungs-und Bildungs GmbH, LSC13.

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Pulp cationization in different solvents

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Fibers resulting from pulping of wood are negatively charged by nature. To overcome the electrostatic repulsion among the fibers, papermakers usually add cationic starch. In this study, the same cationic epoxide that is used for the cationization of starch, 2,3-epoxypropyltrimethyl-ammonium chloride, was applied to fibers. We used different solvent systems to investigate their impact on the reaction efficiency.

A bleached Kraft pulp was subjected to cationization in water, dimethyl sulfoxide (DMSO), isopropanol and tetrahydrofuran (THF) under otherwise identical conditions (liquid-to-solid ratio, time, temperature profile, amount of alkali and amount of reagent). In parallel “blank” assays, the epoxide reagent was replaced with sodium chloride. The resulting materials were analyzed by conductometric titration and elemental analysis, infrared spectroscopy and gel permeation chromatography.

The degree of substitution determined by conductometric titration was around 0.05 when using water or DMSO, 0.15 for isopropanol and 0.4 for THF. The success of the cationization also showed in the samples’ infrared spectra – both the introduced functional group itself and the resulting increase in hydrophilicity were visible. The impact of the reaction conditions on molar mass distribution and supramolecular structure was studied with the aid of the materials derived from the blank reactions. A severe decrease in molar mass was found for the assays with DMSO; the reactions in water, isopropanol and THF caused no notable degradation.

This series of experiments showed that using isopropanol or particularly THF considerably improves the reaction efficiency. We conclude that from all tested systems, a THF-water-mixture is the best choice for controlling the degree of substitution without inducing molar mass loss or major structural changes.

An abstract with similar content was submitted for an oral contribution to the 18th International Symposium on Wood, Fiber and Pulping Chemistry, 09.-11.09.2015, Vienna, Austria.

ABSTRACTS SESSION 3:

BIOREFINERIES AND WOOD TECHNOLOGY

Continuous anaerobic digestion of steam-exploded maize straw for biogas production

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The utilization of crops for biofuel production competes directly with their use as food. Using lignocellulosic residues as a raw material for the production of biofuels such as biogas can alleviate this competition. One of the most widely available lignocellulosic residue is maize straw. It consists of 35−40 % cellulose and 20−25 % hemicellulose. Lignocellulosic biomass like maize straw requires a pretreatment step prior to anaerobic digestion in order to allow the hydrolysis of polysaccharides into fermentable sugars. One efficient pretreatment method for hard degradable biomass is steam explosion. It consists of heating the biomass at high temperatures under high pressure achieved by direct steam injection, followed by a sudden pressure drop, which leads to mechanical disruption of the biomass fibers. This pretreatment allows an increase of methane yields and degradation speed. The aims of this study were to optimize the utilization of steam-exploded maize straw in a continuous system.

For this purpose, maize straw was pretreated in the steam explosion unit designed by Biogas Systems GmbH (Austria) at 173 °C with a residence time of 15 minutes. The continuous fermentation test was done at mesophilic (40 °C) and thermophilic (55 °C) conditions. Different organic loading rates of 1, 1.5, 2.5 and 3.5 kg of volatile solids per m^3 per day were tested. The only nutrient which was added to the system was urea, in order to reduce the C:N ratio for a better performance. Different parameters such as dry matter, volatile solids, COD, pH, ammonium, total nitrogen, micronutrients or residual biogas potential was regularly measured. Results showed that the utilization of steam exploded maize straw in a continuous anaerobic digestion system is feasible under the tested organic loading rates (OLR). Higher organic loading rates should be tested to identify the point of instability.

The optimization of the process can help decrease current competition between biofuels and food production for raw material, by ensuring good methane yields from lignocellulosic residues.

Microbial processes in hydrogen exposed porous underground gas storages (UGS)

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Considering the effects of fossil energy consumption on climate change, it is of utmost importance to increase the share of energy produced from renewable resources. However, energy produced from wind or solar power is fluctuating in response to availability of the energy source. Resulting peak production of electrical power is just partly in line with peak consumption. As a consequence, storage possibilities for large quantities of energy are required. “Power-to-Gas” (conversion of excess electricity to hydrogen) is already state of the art but efficient storage facilities for hydrogen are still a matter of investigation. The Underground Sun Storage Project focuses on the introduction of hydrogen blended with natural gas into porous underground gas storage facilities. Potentially, hydrogen could be decreased by biogeochemical transformation processes possibly accompanied by a loss in pressure, well clogging, acidification and MIC. Therefore, the objective is to study microbial and geochemical processes associated with the exposure of hydrogen to underground gas storages.

For simulating an underground gas storage facility at lab scale, UGS drilling cores were inoculated with UGS formation water and then placed in 10 corrosion resistant bioreactors (including two abiotic controls) and operated at reservoir conditions of the testbed (45°C, 48 bar). In a first step, UGS conditions were simulated with only methane being stored for 2 months. Following, the cores were exposed to various gas mixtures (hydrogen 4-10%, carbon dioxide 0,3-2,5%, methane). Prior to and after hydrogen exposure (6 months), formation water and cores were analysed with respect to hydrochemical and microbiological characteristics. During hydrogen exposure, the partial pressure of hydrogen, methane and carbon dioxide was monitored. A loss in pressure and consumption of hydrogen and carbon dioxide were observed in biotic reactors. Molecular biological analysis revealed eubacterial and archaeal communities why microbial processes were concluded to be responsible for hydrogen depletion. Potential microbial hydrogen consumption reactions at UGS conditions comprise homoacetogenesis[Diekert G. etal 1994], sulphate reduction[Magot M. etal 2000] and methanogenesis[Garcia J-L. etal 2000, Whitman W.B. etal 2006] with the latter being rather a conversion than a loss of energy. Data suggest that in the presence of carbon dioxide the majority of hydrogen was converted to methane indicating that the introduction of hydrogen into porous UGS is a promising approach to integrate renewable energy into state of the art storage techniques.

From Monomers to Polymers across the Oligomeric Region: Advanced Biorefinery Analytics

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In contrast to a petrol refinery a lignocellulosic biorefinery uses woody materials as feedstock. The main products are the structural polymers cellulose, hemicelluloses and lignin, which are used directly or in a more degraded form after different treatments. As the composition of natural feedstock varies, fast and robust screening methods are required to determine the raw material quality and processing conditions. The objective of this project was to establish analytical methods for the different carbohydrate fractions found in biorefinery product streams: polymers, oligomers and monomers. For the separation and quantification of commonly found sugars in wood different methods can be used [1]. The established methods often suffer from poor recovery rates and thus require correction factors to obtain meaningful results. Additionally, samples containing a major fraction of interfering matrix (e.g. plant hydrolysates) often cause problems during analysis. Here we introduce a High Performance Thin Layer Chromatography (HPTLC) method which is capable to separate and quantify monomeric carbohydrates found in wood and can overcome matrix related problems. Analytical methods for the oligomeric fraction in lignocellulosic analysis are quite rare [2]. In general, oligomeric fractions are more or less neglected in biorefinery concepts. However, these compounds can be a valuable resource. To close this analytical gap we took different analytical approaches. After the production of oligomeric standards we established quantitative HPTLC method; a quantitative HPLC method; and extended an existing Size Exclusion Chromatography (SEC) method. For cellulose, which represents most of the polymeric part in lignocellulosic samples, SEC-MALLS (Multi Angle Light Scattering) is the method of choice yielding absolute molar mass averages (M_w) without the need of calibration [3]. However, some constants need to be known for gaining molar masses (averages) with high accuracy, first of all the refractive increment (*dn/dc*). The determination of this value is extremely difficult and cumbersome. To resolve contradictions concerning the *dn/dc* we compared M_w values obtained by two different SEC-MALLS systems.

Financial support by BASF SE and Lenzing AG is gratefully acknowledged.

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Utilizing agro-industrial residues with a low carbon content for bioplastic production

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Agricultural residues with high carbon content such as molasses are already intensively used as fermentation substrates. Residues with lower carbon content, such as whey, stillage, wastewater from plant oil mills, etc. are not suitable for fed-batch fermentations, the most frequent fermentation mode for Polyhydroxybutyrate (PHB)-production. This problem can be circumvented by either concentrating the carbon in the feed stream or retaining the cells during the fermentation in the bioreactor. The latter strategy has the advantage that it can also be used for substrates containing low concentrations of inhibitors, which would get co-concentrated along the desired carbon source. Excretion products during the fermentation are continuously removed from the bioreactor. Furthermore, as PHB accumulates intracellular, the product is up-concentrated during the fermentation.

In order to prove this concept, we have developed this membrane process. Using synthetic media we achieved already high productivities (1.2 g PHB/Lh), reach high cell densities (52 g/L) and got good yields (0.27 g PHB/g glucose).

Wood treatment with bulking and modification agents to reduce the swelling and shrinking behaviour

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With changing air humidity the moisture content of wood changes. An increase in wood moisture content results in swelling of the wood. Shrinking on the other hand is caused by wood moisture content decrease. Since many technical applications demand dimensional stability, this natural material has a slight disadvantage compared to other construction materials. To improve its properties, different kinds of wood modification were developed to reduce its moisture depended swelling and shrinking behaviour. The positive influence of nontoxic polyethylene glycol (PEG) on the dimensional stability of wood is well described and investigated. The common method to impregnate wood with PEG is to store the samples for several weeks in the PEG-solution and let the PEG diffuse into the material. This method is used to stabilise archaeological wood, but isn't suitable for industrial application so far. For this investigation a short time vacuum-pressure cycle was used to impregnate oak with different PEG-solutions. Using the same procedure, oak-wood was also impregnated with PEG-functional silane, which provides nearly the same benefits known from PEG, except leaching out in aqueous environment, like PEG does. The PEG impregnation resulted in weight-percent-gain-values (WPG) comparable to the common impregnation method by diffusion only. According to dimensional stability, a radial anti-shrinkage-efficiency (ASE) of up to 70 % was found. The leaching test reveals less leaching for the PEG-functional silane compared to the PEG impregnation, while still providing a certain ASE. These results show the potential for the usage of PEG (-functional) wood treatment in more critical climate conditions or even outdoor use.

ABSTRACTS SESSION 4:

ENVIRONMENT AND HEALTH

Potentials of phytomining from waste incineration bottom ash using hyperaccumulating plants

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Facing a predicted shortage of resources [1], different strategies to deal with a decreasing availability of raw materials have been proposed, involving the use of so-called anthropogenic resources. In 2012, around 580.000 tons of waste incineration residues were produced in Austria, with 80% of that material being landfilled [2]. In that way a great resource potential is lost every year. The use of phytomining in combination with waste incineration residues is a novel and innovative approach and could offer an environmentally sound and cheap technology to recover valuable metals from waste incineration residues.

A pot experiment was set up in an experimental greenhouse in December 2014. The experimental setup consisted of a full factorial design involving five plant species (*Brassica napus*, *B. juncea*, two different clones of *Nicotiana tabacum*, *Sedum plumbizincicola* and *Alyssum serpyllifolium*), two different substrates and an unplanted control. The substrates consisted of a mixture of waste incineration bottom ash, residues from mechanical biological treatment of municipal solid waste and biochar.

The two hyperaccumulator species *S. plumbizincicola* and A. *serpyllifolium* were growing slowly and seemed to have problems to cope with the difficult substrate. Nevertheless, they showed elevated concentrations of nickel and zinc, respectively, in the above ground biomass. Half of the tobacco plants showed moderate growth, whereas the other half died off, independent of the substrate mixture and clone. *B. napus* and *B. juncea* were growing well and showed good potential to cope with the substrate.

With the knowledge gained from the first pot experiment it was clear that not every plant species is growing well on the waste incineration material. Thus, further substrate mixtures are under investigation. Moreover, a pot experiment using metal accumulating species in combination with microbial inoculants in order to improve plant growth and health was carried out and is currently under investigation.

Microbial communities associated with grapevine and vineyard weeds

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Vineyards harbor a variety of weeds, which are usually controlled since they compete with the grapevines for water and nutrients. However, weeds may host a wide variety of microorganisms that interact with grapevine, having beneficial, neutral or phytopathogenic effects. Therefore, the objective of this study was to extensively characterize the natural microbiome of grapevine plant and associated weeds grown in the vineyard to assess the role on microbial communities in vineyard soil.

Using cultivation 500 rhizosphere bacteria and root endophytes were isolated, characterized and identified from hoary cress (*Cardaria draba)* and grapevine *(Vitis vinifera*). Isolates showed a rich structural and functional diversity. A total of 38 different genera were found with *Pseudomonas, Arthrobacter* and *Bacillus* being most dominant. Some genera were common in both plants, while others were plant specific. All isolates were tested for various functional characteristics like ACC deaminase, HCN production, IAA production, siderophores production, phosphorus Solublization. In hoary cress more isolates with the ability to produce HCN, IAA and siderophores were found, while the grapevine isolates were better able to solubilize phosphorus and to utilize ACC.

We further investigated the microbiomes associated with roots and rhizospheres of grapevine and four different weeds (*Lamium amplexicaule, Veronica arvensis, Cardaria draba* and *Stellaria media*) from five independent sites within a vineyard using Illumina-based 16S rRNA gene sequencing. Microbiome data analysis revealed that the plant type and the site were the main drivers of rhizosphere microbiomes, whereas the root microbiomes were only influenced by the plant type. Weed microbiomes were more similar and showed low similarity to that of grapevine. Roots hosted less diverse and differently structured bacterial communities than the rhizosphere. This study suggests that vineyard weeds and grapevine host different microbial communities, which are potentially characterized by different functional characteristics. More research is needed to determine, whether the presence of weeds can influence the structural and functional diversity of grapevine-associated microbiota.

Analysis of sulfate $\delta^{34}S$ in soil by DGT MC ICP-MS

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Sulfur is an essential plant nutrient and can be found as part of amino acids, proteins or lipids. The main sulfur form taken up by plants is sulfate. Sulfur may undergo various biogeochemical processes in soil, many of which are affecting its bioavailability. Moreover, these processes are known to influence the isotopic composition of sulfate S. Therefore, analysis of stable sulfate S isotopes is a tool to follow biological activity and geochemical processes, which affect the availability of sulfate to plants in soils.

A classical method to analyze the content and isotopic composition of bioavailable sulfate S in soil is a soil extraction (e.g. using NH_4NO_3) followed by precipitation ($BaCl_2$) and mass spectrometric analysis using isotope ratio mass spectrometry (IRMS). However, sulfate precipitation is sample consuming and/or demands high contents of sulfate in the sample. Multi collector inductively coupled plasma mass spectrometry (MC ICP-MS) allows for the direct measurement of S isotopes in a solution even in the sub-mg g^{-1} range (limited by the S background concentration only). The measurement is, however, limited due to effects of co-extracted matrix elements (K, Ca etc.) on the measurement precision and accuracy. Therefore, an alternative method is required combining the extraction of sulfate from soil and the high sensitivity and isotope ratio measurement accuracy of MC ICP-MS.

We present a DGT method for direct sampling of bioavailable sulfate from soil, even at low concentrations and small amounts of sample. The sampled sulfate can be leached from the resin gel easily in 1 mol L^{-1} HNO_3. The eluate is then directly measured by MC ICP-MS without the need for further matrix element separation. Method parameters (diffusion coefficient in APA2 gel, elution efficiency from resin gel) were determined using ^{35}S as radiotracer. Isotope ratio analysis was optimized using the high resolution capabilities of MC ICP-MS and effects of the sampling procedure on the isotope ratio were investigated in order to ensure that an unbiased isotopic composition of bioavailable S can be assessed.

An abstract with a similar content was submitted for DGT Conference 2015, San Sebastian, Spain

Quantitative determination of the elemental distribution of biodegradable Mg alloys in bone tissue by LA-ICP-MS

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Chemical imaging using laser ablation – inductively coupled plasma – mass spectrometry (LA-ICP-MS) has evolved into a powerful tool for the investigation of the spatial distribution of elements and isotopes in biological tissues. However, specific challenges concerning the spatial resolution and the accurate quantification are considered as critical. Moreover, the distinct spatial matching of chemical data gathered by LA-ICP-MS with information from other sources still requires development. There is significant potential for improving data interpretation by linking LA-ICP-MS results with data of other sources such as elemental distribution pattern along with particle size distribution, chemical information on organic compounds, crystallization, or tissue densities.

This study focuses on the adoption of chemical imaging with LA-ICP-MS with respect to laser parameters and suitable quantification strategies in combination with advanced spatial data reduction and analysis in a geographical information system software in order to understand the degradation behavior of Mg-based biodegradable implants in rat bones as model system. This has been accomplished by investigating the spatial distribution of alloying elements (Mg, Mn, Yb, Zn, and Zr) along with the hydroxyapatite matrix (Ca, P) and organic content (C) in the cortical bone.

A method was developed for the direct interpretation of the quantitative images in relation to structural bone information using ArcGIS® as tool for the spatially distinct matching and analysis of chemical data with microscope pictures. As a result, this technique allowed to properly identify the mineralized bone tissue areas as specific regions of interest (ROIs) allowing for the spatio-temporal interpretation of the degradation behavior of the alloys. The distribution patterns of the elements could be nicely shown in the chemical images. The method was further applied to investigate the Mg degradation by using ^{26}Mg isotopically enriched implants. First results will be shown on the example of ^{24}Mg/^{26}Mg isotopic patterns within bone material.

POSTER PRESENTATIONS

P1: Biochar and compost: soil amendments altering root exudation and tomato-soil fungus interactions

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The tomato wilt caused by Fusarium oxysporum f.sp. lycopersici (Fol) pose a serious threat to tomato production both in the greenhouse and the field. The application of organic matter inputs or arbuscular mycorrhizal fungi (AMF) has been shown to suppress a wide range of soil borne diseases. In the present study, the main objective was to elucidate the Fol development and the alteration in root exudates of tomato plants grown in soil substrate compositions including compost alone as well as in combination with wood biochar (WB), and/or green waste biochar (GWB) along with or without AMF. The tomato plants were grown under greenhouse conditions for 6 weeks and processed for extraction of root exudates. The AMF incorporation into the soil substrate containing compost alone and in combination with GWB positively affected the tomato plant growth even under disease stress. There was significant reduction in disease severity in tomato plants grown in compost associations with GWB and WB in the presence of AMF. The AMF root colonization was enhanced in Fol inoculated plants grown in the compost alone and in combination with WB unlike the plants in GWB containing soil substrate. The root exudates were analyzed for their effects on Fol growth and development in vitro. The microconidia germination rate was highest in root exudates from the tomato plants grown in the soil containing GWB followed by the root exudates from AMF colonized plants. In contrast, root exudates of tomato plants from soil containing WB had a reducing effect on in vitro growth and development of Fol. From our results, it is concluded that organic soil amendments with additional AMF may influence the quality and composition of root exudates differently with respect to their effects on soil borne fungi.

P2: Converting a polyesterase into a polyamidase

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Enzymes from the α/β-hydrolase family have been used for hydrolysis of polyesters [1,2]. In contrast, an effective enzyme for the biodegradation of man-made polyamides is currently lacking. Recently it has been shown that a hydrogen-bond donated by the scissile NH group of the amide substrate is a key to efficient enzyme-catalyzed amide bond hydrolysis. The active site of esterases lacks this interaction which significantly reduces the rate of hydrolysis of the C-N bond. A hydrogen-bond acceptor residue can be designed in the active site which increases transition state affinity and the rate of hydrolysis of amide bond [3]. Herein, this strategy was persued to create a polyamidase based on using the cutinase from *Thermobifida cellulosilytica* Thc_Cut1, a natural polyesterases. The structure of Thc_Cut1 was studied in order to identify residues which could afford the formation of the hydrogen-bond after engineering. The Ile179, in the active site, was found to be the key residue. Analysis of the specificity against ester and amide soluble substrates revealed that designed variants possess a higher specificity for amide bond hydrolysis compared to the wild-type. Hydrolysis of bulky amide substrates, oligomers and polymers would be highly beneficial in biocatalysis applications.

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P3: Inbreeding Effects in the Predatory Mite *Phytoseiulus Persimilis*

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Predatory mites such as *Phytoseiulus persimilis* Athias-Henriot (Acari: Phytoseiidae) are widely used in biological pest control by killing and eating unwanted pests and maintaining pest populations at non-damaging levels. *P. persimilis* are sexually reproducing and, because of their specialization on herbivorous spider mites, living in groups. Hence, choosing a compatible mate of the same species is important to ensure production of viable healthy progeny. Individuals of closely related but different species, or sometimes even of separated populations of the same species, do not make good mates. On the other side, excessive inbreeding, that is, mating with too close relatives, commonly also results in fitness loss. Therefore, mate choice should be based on an optimal balance between in- and out-breeding. We tested this hypothesis using two populations of *P. persimilis*, one from Sicily and the other from Greece. We examined the fecundity of females, and viability and sex ratio of their offspring, that mated with either a sibling male, a male from the same population or a male from the other population. Additionally, we recorded two important components of mating behavior, mating latency and duration of copulation.

P4: Sugar and anthocyanin modulation in Berry Shrivel grapes

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Berry Shrivel (BS) is a shrinking disorder of high economic importance affecting the most widely grown red grapevine variety in Austria, Zweigelt [1]. Although the symptoms of BS are well described (loss of turgor, high acidity, low pH, low anthocyanin and sugar contents and cell death at mesocarp and rachis) its causes are still unknown and the timescale of symptom induction is still obscure.

Grapes accumulate high amounts of glucose and fructose through ripening. There are two growth phases in grape, first the sugars are transported through plasmodesmata and cell division occurs, and later on an apoplast transport is established and sugars are rapidly accumulated [2]. During apoplasmic phloem unloading mainly cell wall invertases and monosaccharide transporters are highly active for the uptake of sugars. Anthocyanins accumulate coinciding with the fast accumulation of sugars. These components have a natural role in seed dispersion, defense to UV light and towards abiotic and biotic stress. Different internal factors influence anthocyanin biosynthesis, as gene expression at the anthocyanin pathway and presence of important triggering factors as sugars and ABA [3].

BS berries are characterized by low sugar and anthocyanin contain. In the presented study the key players of the sugar and anthocyanin metabolism were analyzed and compared in healthy and BS grapes on a timescale from pre-veraison to full-ripe with the goal of understanding mechanisms and illness induction timescale.

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P5: Growing Synechocystis Salina in Digestate Fractions for producing Poly(hydroxybutyrate)

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Environmental pollutions caused by non-biodegradable plastics based on petroleum are increasing. Due to this reason it is important to do research on biodegradable biopolymers like poly(hydroxyalkanoates) (PHA), including poly(hydroxybutyrate) (PHB). In the current PHB production process, sugar is used as carbon source for heterotrophic bacteria. As an alternative, CO_2 from exhaust gas can be used in photoautotrophic PHB-production which reduces the global demand for crops (food, feed, biofuel, etc.). In this study a further step to increase the ecological and economic efficiency of the photoautotrophic PHB production was investigated by using anaerobic digestate as nutrient source. This digestate could be derived e.g. from anaerobic digestion of the residual biomass (after PHB extraction).

In the first experimental phase, focusing on biomass growth of cyanobacteria on digestate fractions, the strains *Aphanothece microscopica* (CCALA 19) and *Synechocystis salina* (CCALA 192) were tested. Thin stillage digestate (residue from bioethanol production) was used as a nutrient source. Different pre-treatment options were tested: centrifugation with or without prior addition of precipitating agents, as well as subsequent ultrafiltration. Finally different dilutions of the digestate fractions were tested (1:2, 1:3, 1:5, 1:7, 1:10). The highest biomass growth was shown by *S. salina* in centrifuged digestate with precipitating agents diluted 1:5 (OD_{485} = 11.9, OD_{750} = 8.5 in 12 days), followed by the dilution 1:3 and 1:2. The best digestate pre-treatment (centrifuged digestate with precipitating agents) and the strain *S. salina* were used in the second experimental phase, focussing this time on PHB production. Again different dilutions (1:3, 1:5, 1:20) were tested. The highest PHB concentration was observed in the dilution 1:3 (95 mg/L in 14 days). Finally, the most promising experiment, using centrifuged digestate with precipitating agents diluted 1:3 and *S. salina*, was scaled up and carried out in a tubular photobioreactor at pilot scale (200L) (Figure 1). The achieved results are comparable with the results at laboratory scale (dry matter: 1.6 g/L, PHB concentration: 90 mg/L, which equals 4.7 %/TS, in 40 days).

Figure 1 Tubular photobioreactor at pilot scale for photoautotrophic PHB-production by *Synechocystis salina*

P6: Stable isotope assisted evaluation of extraction solvents for LC-HRMS based untargeted metabolite profiling of wheat

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For untargeted metabolomics approaches the evaluation of suitable extraction protocols is difficult due to the high number of non-specific MS signals and features. We have recently developed a novel stable isotope-assisted workflow for untargeted LC-HRMS based plant metabolomics [1], which allows for the first time the direct consideration of every detected feature with respect to workflow evaluation including the comparison of extraction solvents. In the current work, the efficiency and complementarity of commonly used extraction solvents, namely 1:1 (v/v) mixtures of water and selected organic solvents (methanol, acetonitrile, methanol/acetonitrile 1:1 (v/v) or acetone), with and without addition of 0.1% (v/v) formic acid were compared. Four different wheat organs were sampled, extracted and analysed by LC-HRMS. Data evaluation was performed with the in-house developed MetExtract software and R. With all tested solvents a total of 888 metabolites were extracted in ear samples, 800 in stem, 758 in leaf and 535 in root respectively. Surprisingly the number of extracted metabolites was similar across solvents. This effect could be observed in all organs where 650-730 metabolites were extracted in ear, 530-610 in stem, 520-580 in leaf and 350-400 in root samples. More than 45% of the detected substances were shared by all solvents. Although the total number of extracted metabolites is similar for every organ, there are substantial differences in the resulting extracts. Using hierarchical cluster analysis, the comparison of relative extraction efficiencies resulted in separation according the revealing methanol content of solvents as the major discriminating factor, followed by separation according the acidification. Matching m/z value and number of carbon atoms against an in-house database resulted in the annotation of 117 metabolites. Analysis of different annotated substance classes showed that choice of the right extraction solvent can be crucial in sample preparation and furthermore basis for successful analysis.

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P7: A complementary set of methods to characterize phosphorus fertilizers of different origins and solubility

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Several standard methods exist to characterize P solubility and availability from fertilizer and from soils. These methods are mainly batch extractions of P at pseudo-equilibrium at a specified substrate:solution ratio. Different procedures are in use depending on the country and the substrate to be analyzed: *fertilizer* P evaluation methods are commonly batch extractions using e.g. water, ammonium citrate, or formic acid solutions, while for *soil* P, batch extractions using calcium acetate lactate (CAL), sodium bicarbonate (Colwell, Olsen), or combinations of acetic acid, ammonium nitrate, ammonium fluoride, nitric acid, hydrochloric acid, and/or EDTA (Bray II, Mehlich 3) are in use. Although such methods are well established, (1) the mechanism by which P is extracted is generally of little relevance in regard to plant root activity, and they neither (2) quantify the total amount of soluble fertilizer P, nor (3) its diffusional behavior in soil.

In the frame of the FERTI-MINE project we use a set of complementary approaches to characterize the behavior of P from fertilizers of different origins and solubility. With process-based methods (e.g. depletion-induced P desorption to mimic plant root uptake, imaging of the diffusional behavior of fertilizer P in soil) more information can be gained about the characteristics of P fertilizers than with standard extracts. Moreover, some of our methods can be universally implemented either for fertilizer or for soil and are therefore highly suitable for studying soil-fertilizer interactions.

This poster presents three methods which are currently under evaluation in our laboratory. The results of preliminary tests with single superphosphate and a biochar derived from municipal sewage sludge indicate the suitability of these complementary approaches to characterize the behavior of P from fertilizers of different origins and solubility.

P8: Evaluation of Biotypisation assays of Grape Phylloxera (Daktulosphaira vitifoliae Fitch) on Vitis ssp.

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Grape phylloxera (*Daktulosphaira vitifoliae* Fitch) biotypes became well-known after the outbreak of an unknown strain, which was able to infest the rootstock hybrid AXR#1 (*Vitis vinifera* x *V. rupestris*) in the late 1980s. This biotype caused substantial economic losses in California. When referring to biotype performances on, or preference for, a particular host (e.g. feeding on a rootstock accession) is implied. The term “biotype” has been used for insect strains that vary in their response to the hosts. Their specific genetic background is often not clear. However phenotypic variation could be based on allelic genotypes, individuals or population levels. Over the last 30 years, numerous studies have phenotyped phylloxera, particularly regarding their performance on various hosts. The results are difficult to interpret and compare because of the lack of homogenous nomenclature and standardized phenotyping assays. Our goal was to compare screening methods on their respective efficacy to provide stringent biotype definitions by performing a comparison of the applied methods. Three commonly used biotyping assays were tested employing two phylloxera strains: Biotype A adapted to *V. vinifera*) and biotype C (adapted to *V. vinifera* and rootstock hybrids *V. riparia x V. berlandieri*) on the two tolerant rootstock cultivars Teleki 5C (*V. berlandieri x V. riparia*) and Fercal (B.C.n°1B x 31 Richter) as well as *V. vinifera* L. cv. Riesling. 1) Simple isolation chambers [1] consisting of rooted cuttings in a perlite:peat substrate were cultivated and infested in climate chambers (25±5°C, 40% rH and 16h light). 2) Excised root bio assays [3] were performed employing the two phylloxera biotypes on Teleki 5C and Riesling roots in petri dishes located in lightened incubators at 24°C±1°C. 3) Sterile tissue culture vines of Riesling and Teleki 5C were infested employing the two phylloxera biotypes according to the aspetic dual culture system [3] in a growth chamber with 22°C. Life table parameters of phylloxera as well as plant based response (root galling) were recorded. The results of the three applied screening systems were not consistent showing that the existent biotyping assays are not comparable among them and lead to divergent biotype assumptions.

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P9: GC-MS based metabolomics of Borneo's "exploding ants": headspace solid phase microextraction of mandibular gland content

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Carpenter ants belonging to the *Camponotus* (*Colobopsis*) *cylindricus* complex ('COCY', or exploding ants), that are colonizing the rain forest on Borneo, reveal a so-far unique and remarkable behavior called "autothysis": In one-to-one confrontations with other ants acting as potential enemies, they sacrifice themselves by rupturing their intersegmental abdomens. Hereby they release a sticky and irritant secretion from their mandibular glands, which leads to the death of the affected opponent.

To widen the current, very limited knowledge about the chemical composition and the nature of tentatively toxic constituents of the mandibular gland content, 50 ants of the YG (yello goo) species were dissected and the contents of their mandibular glands were measured by headspace solid phase microextraction-gas chromatography-mass-spectrometry (HS-SPME-GC-MS). Data analysis, compound annotation and identification were carried out with the current version of the *MetaboliteDetector* software. To this end, a combination of retention indices, the Wiley Registry/NIST-, as well as an established in-house library and, if available- authentic standards were used.

In total, 54 substances were detected in the mandibular gland content of the investigated ants. They mainly belonged to the structure classes of alkanes, alkenes, terpenoids, carboxylic acids, ketones, and phenols. Many of these substances have also been found in defensive secretions of other ants/insects, where they have been described as antimicrobials, antifungals, or as adjuvants to support the effect of the other compounds. This indicates that the bioactive metabolites found in the 'COCY' ants may not only function to directly kill enemy arthropods, but may also act against certain bacteria and fungi and therefore may also serve to defend their own nests against detrimental microorganisms.

P10: Effects of hydrophobin-cutinase fusion proteins on enzymatic hydrolysis of polyethylene terephthalate

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Poly(ethylene terephthalate) (PET) is one of the most widely used synthetic polymers worldwide. Despite its many outstanding properties, PET has a very hydrophobic character, which makes processing and functionalization real challenging. A wide variety of hydrolases have been described to be able to hydrolyse PET [1,2]. Mild hydrolysis of the upper most layer of the polymer and extensive hydrolysis to the monomers are two approaches that can be used to functionalise or recycle PET in a highly selective and environmentally friendly way. Among the different enzymes able to hydrolyse PET, cutinases have been described to be the most efficient, however, the fact that PET is a non-natural, hydrophobic substrate the hydrolysis-rates are rather low.

Hydrophobins (Hfbs) are small cysteine-rich proteins expressed by filamentous fungi that form amphipathic monolayers on hydrophobic/ hydrophilic interfaces and are supposed to stimulate the hydrolysis rate of PET [3]. In nature they occur on the outer surfaces of cell walls of hyphae and conidia where they play a major role for interactions between the fungus and the environment.

In this study the effect of the fusion of two class II Hfbs (Hfb 4 and Hfb7) and the pseudo-class I Hfb9b to Cutinase 1 from *Thermobifida cellulosilytica* (Thc_Cut1) on PET hydrolysis was investigated. Furthermore the effect of free and covalently bound Hfbs on PET hydrolysis was compared [4]. The soluble release products of PET hydrolysis namely terephthalic acid (TA), mono-(2-hydroxyethyl) terephthalate (MHET), and bis-(2-hydroxyethyl) terephthalate (BHET) were measured by HPLC with a UV detector.

This study shows that enzymatic PET hydrolysis can be considerably enhanced by fusion of Hfbs to cutinases and is therefore an important step towards increased rate of PET modification and recycling.

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P11: Biodegradation of aromatic polyesters in wastewater treatment plants

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Polymers are used in many articles for daily use products. After usage, polymers can be released into different environments like wastewater treatment plants (WWTP), soil [1] rivers and oceans. [2] In the WWTP the microbial communities have an essential part of the biodegradation processes of polymers. The extracellular enzymes produced by the microorganism are important for the first hydrolysis steps to produce smaller oligomers easier for the microorganisms to degrade. Therefore, is a more detailed knowledge about enzymatic and microbial degradation of polymers in e.g. WWTP is crucial for a basic understanding of biodegradation processes.

In order to study microbial and enzymatic degradation of polymers, a group of structurally different aromatic polyesters was synthesized. The polymers contain 5-sulfoisophthalic acid sodium salt (NaSIP), terephthalic acid (TA) and different diols and glycols. Our strategy to find potential polymer degrading enzymes and microorganisms in WWTP was to screen the polymers with already known polyesterases. The screening identified a cutinase from Thermobifida cellulosilytica (Thc_Cut1) [4] to successfully hydrolyze the polyesters of interest. Based on the sequence of Thc_Cut1 we performed an in-silico search for similar enzymes expressed by wastewater microorganisms. A cutinase from Pseudomonas oliovoranse (CutA) [5] was identified. CutA was expressed in E.coli and purified for further investigations. To investigating the enzymatic hydrolysis of the aromatic polyesters, the polymers were incubated with CutA for 7 days at 28 °C. The expected release products, NaSIP and TA, were separated and analyzed with help of HPLC-UV. TA was detected for most of the investigated polymers while NaSIP only could be detected for a few of them. For investigation of the microbial degradation capacity of the polymers the strain P. oliovorans was cultivated following the recommendation in the DMSZ and incubated with a selected group of the aromatic polyesters. An overnight culture was prepared and used to inoculate the media containing the aromatic polyesters. The polymers were incubated for 7 days at 28 °C with the strain. The culture was harvested and the supernatant were used for further analysis of the expected release products. NaSIP and TA were separated and analyzed with the help of HPLC-UV. TA was detected for most of the investigated polymers.

An enzyme from P. oliovorans, named CutA, has successfully been identified by in-silico search as potential polyester degrader. The enzyme has been proven to degrade a variety of aromatic polyesters. We could also show that P. oliovorans successfully can degrade a selected group of the aromatic polyesters. The improved knowledge is an important step to a more detailed knowledge about the enzymatic and microbial degradation of polymers in WWTP.

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P12: Towards a Sr isoscape of Austria for the determination of growth regions of prehistoric wood

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Wood artefacts from a prehistoric salt mine in Hallstatt, Austria, present a unique archive of information on Bronze and Iron Age mining. For instance, the geochemical signatures of growth locations are stored in the finds, though masked by contaminating minerals attributable to the storage conditions. Trade is assumed for certain archaeological finds. Consequently, analysis of the radiogenic strontium isotope amount ratio $n(^{87}Sr)/n(^{86}Sr)$ has been applied to investigate the geographic origin of these artefacts, in order to allow conclusions on trade routes.

In order to reveal the biogenic signatures of the prehistoric finds, a decontamination method based on acid leaching was developed. It enabled the separation of biogenic from diagenetic Sr. A mixing model was adopted to account for possibly incomplete removal of the latter. In addition to Hallstatt, seven regions in Austria were selected for sampling of modern trees based on known settlements in the time period of interest. The geological bedrock variability was considered within all regions for the definition of sampling spots, which resulted in a total of 26 locations. Drill cores from four tree species represented in the archaeological finds (i.e. *Picea abies*, *Abies alba*, *Fagus sylvatica* and *Quercus sp*.) were sampled upon availability. Sr isotope ratios were measured in wood digests after Sr/matrix separation using multicollector inductively coupled plasma-mass spectrometry (MC ICP-MS).

The isotopic signature of bioavailable Sr that was obtained from modern trees reflects the geological heterogeneity in Austria, which challenges the creation of an isoscape and its applicability to distinct provenance determination. Different bedrock types can be distinguished by their $n(^{87}Sr)/n(^{86}Sr)$. Furthermore, the data indicate that the isotope ratios of bio-available Sr within one geological substrate also vary strongly. The results highlight the importance to consider even small scale geological and environmental variability in a comprehensive sampling strategy for a reliable application of Sr isotope ratio analysis to the determination of origin of biogenic material.

P13: Oxygen: A limiting factor for enzymatic oxidation of lignin

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Lignin is one of the most abundant biopolymers worldwide and is a by-product of pulp and paper industry [1]. It is a highly complex, heterogenic polymer with high interfacial tension and low reactivity, which limits its industrial application to below 10 % in polymer blends [2]. In order to improve the properties of lignin many physico-chemical, biological, and enzymatic modification processes are actively being investigated. Therefore, the enzyme laccase, which is a multi-copper enzyme, offers an environmentally friendly way to oxidize lignin with the help of molecular oxygen as an electron acceptor. By oxidation, the reactive phenolic species formed in lignin constitute the reactive sites of the lignin and provide ideal sites for cross-linking, grafting of a variety of functional molecules or synthesis of novel materials [3].

In this study, Mg-lignosulfonate lignin was used for determining the efficiency of laccase-catalyzed oxidation (isolated from Myceliophthora thermophila) in the presence of either external air or oxygen supply. It was demonstrated that supply of oxygen as an essential electron acceptor can be limiting in laccase mediated oxidation of lignin processes and can be overcome by supplying pure oxygen instead of simple aeration. Fluorescence, which is an intrinsic property of lignin, decreased 40 times faster in sample mixtures supplied with oxygen than shaking samples with external air supply or simply shaking in aerated atmosphere. The supply of oxygen also caused a 17-fold increase in molecular weight which was confirmed with aqueous size exclusion chromatography (SEC). Concluding the obtained results we can postulate that oxygen is one of the important and also limiting factors in the enzymatic oxidation of industrial lignins.

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P14: Modulation of Aspergillus flavus NRRL 3251 oxidative status by fullerenol C60(OH)24: the interplay of TBARS and aflatoxin production

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Aflatoxins (AFTs) are highly carcinogenic secondary metabolites of fungi genus Aspergillus which are produced under the oxidative and/or drought stress conditions of environment [1]. Thiobarbituric acid-reactive substances (TBARS) [2] are produced if the oxidative/antioxidative balance of the fungus cell is perturbed by reaction of free fatty acids and reactive oxidative species (ROS) [3]. One of the possible environmentally stressors are fullerenes C_{60} and their hydroxylated derivate fullerenols $C_{60}(OH)_{24}$ which are part of widespread commercial products [4,5]. Oxidative stress, a pre-requisite for AFTs production, could be modulated by fullerenol $C_{60}(OH)_{24}$ nanoparticles, as possible antioxidants. Fullerenol $C_{60}(OH)_{24}$ nanoparticles caused statistically significantly decrease of AFTs production ($p=0,001$). There was statistically significantly correlation between TBARS production ($r=0.3213$, $p=0.0085$) and detected AFTs in YES medium. Thereby, there is significant effect of fullerenol $C_{60}(OH)_{24}$ nanoparticles on modulation of oxidative status of Aspergillus flavus NRRL 3251 after 168 hrs growth in YES microbiological medium.

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P15: Investigation of possible peptide markers for 5 allergenic food commodities via LC-MS/MS within the EU-project iFAAM

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In the European Union the food labelling of 14 food allergen groups is mandatory. As there is little published about dose thresholds, total avoidance of contaminated food is the only safe opportunity for allergic people. During the last decades, ELISA and PCR techniques became the methods of choice for allergen analysis [1]. However, in comparison to MS/MS methods, they can lack the precision and rigor required [2], heading false positives and wrong negatives.

In this approach a pool of marker peptides for an MRM-multimethod for the simultaneous detection of milk, egg, peanut, hazelnut and walnut in a chocolate dessert, which serves as a food challenge model matrix in the iFAAM-project for determining dose thresholds, was investigated. Performing an in-silico digest with the protease trypsin of the relevant proteins to set up the LC-MS/MS method resulted in a theoretical pool of 200 marker peptides. After a refinement including BLAST Search in the Uniprot Database to prove specificity of the peptides for the allergic commodity and further LC-MS/MS measurements to check the peptide fitness for MRM analysis a number of 73 peptides will be further investigated.

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P16: Identification of volatile organic compounds in different grapevine genotypes after inoculation with *Plasmopara viticola*

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The grapevine Vitis vinifera cv Pinot noir is susceptible to several pathogens including Plasmopara viticola that is the causal agent of downy mildew [1]. Hybrids of V. berlandieri and V. riparia (SO4 and Kober 5BB) and hybrids of Muscadinia rotundifolia and V. vinifera (BC4) and others such as Solaris are resistant or tolerant to downy mildew. It has been demonstrated recently [2] that resistant in vitro hybrids SO4 and Kober 5BB emit volatile organic compounds (VOCs) in response to P. viticola infection. In particular, the most interesting class of VOCs constitutes terpenoids (mono- and sesquiterpenes) emitted by the resistant cultivars, whereas for Pinot noir no terpenes have been detected under the tested conditions.

In the present study we have used gas chromatography coupled with mass spectrometry (GC-MS) to study in more detail the chemical identity of the compounds produced by selected plants of the five genotypes Pinot noir, Kober 5BB, SO4, BC4 and Solaris. All the genotypes were cultured in the greenhouse and leaves were harvested immediately (0 dpi) and six (6 dpi) days after the inoculation with P. viticola. All samples were immediately frozen and homogenized under cooled conditions. VOCs were extracted by using solid phase microextraction (SPME) and analyzed by GC-MS. Mass spectral deconvolution and annotation / identification of volatile compounds was based on comparison of mass spectra and retention indices with reference values and performed by Metabolite Detector software [3].

Preliminary results showed increased levels compared to day zero of sesquiterpenes in resistant cultivars six days after inoculation, demonstrating that terpenes could play an important role in plant resistance against downy mildew in resistant genotypes.

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P17: High strength nanopaper from Miscanthus biogas production residue

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Cellulose nanofibers (CNF) were extracted from Miscanthus biogas digestion with two different treatment temperatures (37.5 °C and 55 °C). A non-treated Miscanthus material was used as reference. The samples were initially Soxhlet extracted to remove any soluble components. Thereafter, lignin and hemicelluloses were widely removed by applying a pulping process consisting of an oxidative step using an acidified sodium chlorite solution followed by an alkaline extraction step. The remaining Miscanthus pulp was rinsed with distilled water and diluted to a concentration of 0.5 wt% and then separated to nanofibers by several passes through a high-pressure laboratory homogenizer (APV 1000) for the desired fibrillation effect. Sheets of nanopaper were prepared from the resulting CNF dispersions by casting and evaporation. The finished sheets were cut into strips with a width of 6 mm and a length of 60 mm which were subsequently used for tensile tests. The results showed that nanopaper prepared from the fraction of 37.5 °C delivered higher values for tensile strength and modulus of elasticity than the reference specimens. This shows that even after biogas production the remaining residues still have a potential for material applications before composting concludes the biogas biorefinery process.

P18: Evaluation of 3 winter triticale populations and one panel for Fusarium Head Blight resistance and related traits

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Fusarium Head Blight (FHB) is considered worldwide as a disease of economic importance able to attack all the classes of wheat and other small grains [1]. If the basis of the genetic resistance is now well described for the wheat [5], almost nothing has been done for some other cereals as triticale.

Triticale (×Triticosecale Wittm.) comes from a crossing between wheat and rye. Today, most of the grown varieties are hexaploid, meaning that they are carrying the genome RR from rye (Secale cereale) and AABB from durum wheat (Triticum durum).

In the last few years, a FHB resistant triticale line: G8.06, has been selected from a back-cross population of the highly FHB resistant wheat line CM-82036 with the triticale cultivar Santop. This line carried two FHB resistance QTL coming from CM-82036: Fhb1 and Qfhs.ifa-5A [7]. In order to identify the genetic basis of the FHB resistance in triticale, and to evaluate the potential influence of the rye genome on the efficiency of resistance QTL coming from wheat, G08.06 has been used as resistant parental line in 3 bi-parental crossings. A panel of cultivars and breeding lines has also been constituted to increase the genetic diversity in this project. The 3 populations and the panel were evaluated in 2014 and 2015 in a disease nursery through artificial FHB inoculation at BOKU University in Tulln (Austria). FHB, mildew, strip rust disease symptoms were visually scored, plant height and flowering date were recorded.

The aim of this presentation will be to give a statistical overview of these phenotypic data thanks to a multidimensional analysis (PCA) and a set of variance and co-variance analysis (ANOVA / ANCOVA).

We gratefully acknowledge financial support from the French Ministry of Higher Education and Research, through CIFRE funding (Conventions Industrielles de Formation par la Recherche).

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P19. Provenancing of Strawberries using Sr Isotopes and Multielement Pattern: Investigation of Elemental Sources in the Bioavailable Fraction of Soils

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The investigation of local - specific elemental and $^{87}Sr/^{86}Sr$ - isotopic fingerprints is a powerful tool for the determination of provenance of food. It is known that the fingermark of the bioavailable fraction in soils is well reflected in the plant and consequently in the fruit. Nonetheless, a major concern is the influence of potential additional sources of strontium and other elements, such as fertilizers, rain water or irrigation. In case of Sr, liming was suspected to add a major contribution. Lime is used in conventional agriculture as a buffer substance to ensure optimal pH conditions for the growth of plants and is usually rich in Sr (25 µg g^{-1}). Therefore, it is of importance to know the impact on the elemental and Sr isotopic composition. As a consequence, the fingerprints of strawberry plants and strawberry fruits grown in limed and non-limed soil were determined, using ICP - QMS and multicollector ICP - MS. Additional influence parameters, such as the irrigation water and rain water, were taken into account, as well.

Small strawberry plants were purchased and planted on two patches with soil from Waldhausen (Waldviertel, Lower Austria) which exhibits a $^{87}Sr/^{86}Sr$ ratio of 0.71663 ± 0.00018 (*U* (*k*=2)). After a growth period of about four months, plants and strawberries were harvested. Sample preparation and analysis followed established protocols [1]. Soils were extracted using ammonium nitrate solutions to obtain the bioavailable metal fraction.

Although the $^{87}Sr/^{86}Sr$ isotope ratios of the lime (0.70808 ± 0.00032, *U* (*k*=2)), the irrigation water (0.71002 ± 0.00014 *U* (*k*=2)) and the rain water (0.71001 ± 0.00014, *U* (*k*=2)) were significantly lower than the isotope ratio of the soil, the $^{87}Sr/^{86}Sr$ isotope ratios of the plants (0.71331 ± 0.00014, *U* (*k*=2)) and the strawberries (0.71363 ± 0.00014, *U* (*k*=2)) showed a clear pattern: Plant and fruit Sr isotope ratios were shifted from the original signature at purchase towards that of the soil. Further, differences of the elemental pattern between limed and non-limed soil were insignificant.

According to these results, no significant influence of lime, irrigation water and rain water to the elemental and Sr isotopic fingerprint was observed. These results confirm that the elemental and isotopic information in the bioavailable fraction in soils is not altered by the investigated agricultural practices under the present conditions. Thus, Sr isotopes and elemental pattern are a robust and powerful method combination for the investigation of food provenance.

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P20: Environmentally friendly enzymatic synthesis of bio-based polyesters

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The interest for novel bio-based functionalized polyesters is incredibly rising. New synthetic biocatalyzed routes can concretely respond to this challenge by combining benign conditions with efficiency and selectivity of enzymes. Bio-based chemistry has recently demonstrated the feasibility of the synthesis of value-added polyesters derived from renewable monomers, both polyols and dicarboxylic acids, obtainable via fermentation. Aliphatic polyesters are of particular interest for the biomedical field due to their biodegradable and non-toxic properties and are raising the interest of researchers in the development of new polymers. Hydrolases in particular are nowadays the most studied enzymes because they are attractive and sustainable alternatives to toxic catalysts like tin or other metals normally used in polycondensation reactions. Despite the wide number of studies addressing in vitro enzymatic polycondensation catalyzed by lipases, insufficient progress has been documented in the last decades towards the preparative and industrial application of this methodology. In recent years several new fungal cutinases have been described for the hydrolysis of commercial polyesters such as poly(lactic acid) (PLA) and poly(ethylene terephthalate) (PET). [1] Moreover, the cutinase from *Humicola insolens* (HiC) has been also employed in the synthesis of aliphatic polyesters. In this work we illustrate consistent advances in the field of enzymatic synthesis of bio-based polyesters. The application of a cutinase from *Thermobifida cellulosilytica* (Thc_cut1) was assessed within a comprehensively “renewable context”. Enzymatic polycondensations have been validated in solvent-free systems implemented in thin-layer reactors. [2] Additionally, regarding the use fully bio-based building blocks and catalysts, novel immobilization methods have been developed by exploiting agro-food biomasses/residues, thus overcoming the dependence on petroleum-based resins for enzyme immobilization purposes.

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P21: Two novel anaerobic polyesterases from *Clostridium* hydrolyze synthetic polyesters

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Several enzymes including cutinases, esterases and lipases from aerobic bacteria and fungi with hydrolytic activity on man-made polyester were identified in the recent years. However, there is a lack of information about enzymes from anaerobic organisms involved in polyester degradation.

Here, we present two new esterases from the anaerobic strain *Clostridium botulinum* (Cbotu_EstA and Cbotu_EstB) that were heterologous expressed in *E. coli* BL21-Gold(DE3). Cbotu_EstA was discovered to be active on the synthetic polyester poly(butylene adipate-co-butylene terephthalate) (PBAT), while the activity of Cbotu_EstB on the polymer was considerably lower. The two esterases show their maximum activity around pH 7.0 and exhibit a sequence identity of approximately 40 %. Comparison of the Cbotu_EstA crystal structure and Cbotu_EstB model revealed several structural differences that might explain the lower activity of Cbotu_EstB on PBAT.

The findings obtained in this study will improve the understanding of synthetic polyesters hydrolysis processes in both natural and artificial anaerobic environments. Moreover, the new esterase Cbotu_EstA has a potential for polyester hydrolysis, recycling of building blocks and functionalization of polymer surfaces.

P22: Toolbox for the specific detection and quantification of the plant growth promoting bacterium B. phytofirmans PsJN

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Despite the enthusiastic expectations of the scientific community and a growing interest of industries in the use of microbial inoculants with the aim of improving plant growth, health and nutrition, beneficial microbes are still hardly implemented in agricultural practice. Only a very limited number of microbes have been moreover commercialized. One reason is that effects observed in controlled conditions are very often not reproducible in the field or show undesirably strong variations. In this context it is very important to better understand microbial competitiveness and plant colonization under natural conditions.

We here present a toolbox allowing selectively monitoring and quantifying cells of B. phytofirmans PsJN in plant tissues as well as rhizosphere, independent from genetic modifications. The toolbox consists of a species-specific TaqMan qPCR based on the amplification of selected house-keeping genes and fluorescence in-situ hybridization (FISH) employing probes targeting the 23S rRNA of B. phytofirmans. The applicability of the developed tool box was not only shown in the lab and greenhouse but also in field trials.

P23: Human mobility along the Nile: Preliminary strontium isotope analyses for migration studies in ancient Nubia

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The “Across ancient borders and cultures”-project (FWF START Y615-G19) focuses on the establishment of a settlement on Sai Island (Nile) in Upper Nubia (modern Sudan) as a part of the southwards directed Egyptian expansion in the New Kingdom (c. 1539-1077 BC). In the course of the investigations, new insights on the life style, the living conditions and especially the coexistence and merging of cultures in the 2nd Millennium BC of Egypt and Nubia are expected [1].

During the last decades, strontium (Sr) isotopic analyses in human and animal skeletal remains as indicator of the locally specific fingerprint taken up from the environment via diet has evolved into a key tool in anthropology and archaeology for tracing residential changes and living conditions of ancient humans. Beside the Egyptian ‘colonialist’ (allochthonous), Nubian indigenes (autochthonous) were probably part of the ancient population of the Egyptian settlement on Sai Island. Therefore, Sr isotopic analysis in tooth enamel of individuals buried on Sai island will be performed and compared to Sr isotopic signatures assessed in the local proximity (via the establishment of so-called ‘isoscapes’).

The preliminary analytical work covered the multi-element and $^{87}S/^{86}Sr$ isotopic analyses of soil extracts (bioavailable Sr fraction), water, as well as recent and ancient animal tooth samples (hot-plate assisted acid digests) from the northern part of Sai Island. Multi-element analyses were performed with an ICP-QMS and the Sr isotope ratio were measured on a multi collector ICP-MS using standard procedures [2].

The Sr isotope ratio of the environmental samples from Sai Island showed a narrow range from 0.70660 to 0.70808. Herein, the paleo-sediments showed a higher Sr isotope ratio than the younger Nile silts and alluvial deposits. The Sr isotope ratio in enamel and dentin of the recent animal samples lay in the range of the younger sediments and that of the ancient animal samples overlapped with the excavation site. The isotopic map (isoscapes) of Sai Island will be used as basis for the further interpretation of autochtony or allochtony of the skeletal remains of excavated individuals.

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P24: Challenges to detect key-chromophores by DESI-MS on cellulosic material

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Yellowing of paper is highly relevant in preservation of historic paper documents as well as in brightness reversion in industrial paper mills. Degradation products generated from cellulose and hemicellulose which cause this effect are of strong interest. In the present study key chromophores were measured directly from cellulosic surfaces under ambient conditions. Therefore Desorption Electrospray Ionization (DESI) coupled with mass spectrometry was applied as an analytical tool allowing such analysis. In addition a derivatization step was added to enhance the detectability of the target analytes. Girard's reagent T proofed to be the agent of choice due its reactivity and chemical properties with respect to DESI-MS analysis. Overall, it could be demonstrated that DESI-MS is a versatile tool for the analysis of key chromophores directly from cellulosic surfaces.

Please note: This abstract was submitted before in similar form for the 18th International Symposium on Wood, Fibre, and Pulping Chemistry (ISWFPC 2015)

P25: Elemental and 87Sr/86Sr isotope pattern as a tool for provenancing sturgeon caviar

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Sturgeon caviar is one of the most expensive food commodities in the world. While farming of sturgeons for caviar production is emerging, there is a decrease in the natural populations due to illegal fishing. As a consequence, sturgeon caviar trade has been put under international regulations. However, unambiguous origin determination is still a basic necessity: Sound analytical methods are required to discriminate farmed from wild caviar, in order to control illegal trade and to foster sustainable farming. Therefore in this pilot study, untreated caviar, processed caviar (i.e. salted), water, fish feed and salt from five sturgeon farms in Central Europe (one in Austria and four in Italy) were investigated for their elemental and $^{87}Sr/^{86}Sr$ isotopic composition using (multi collector) inductively coupled plasma mass spectrometry ((MC) ICP-MS). Sample preparation was performed according to optimized standard protocols.

The elemental pattern and the $^{87}Sr/^{86}Sr$ isotope ratio of water of a fish farm are reflecting the local water chemistry that is expected to be transferred into the caviar, as well. Therefore, the chemical information can be used as a fingermark for provenancing. Nonetheless, the potential effect of fish food on the chemical signature of caviar needed to be considered, as well as the effect of caviar processing (e.g. salting).

Firstly, fish farms from geologically different areas could be differentiated by the elemental and isotopic signature of the water. Moreover, the chemical fingerprint of these fish farms was different from the water signature of the natural living habitat of the sturgeon in the river Danube. Secondly, the information of the water could also be found in the caviar allowing for the distinction between caviar produced in the investigated fish farms from caviar of wild fish from the Danube. However, a shift in the elemental and isotopic composition by the fish feed and salt used in the fish farms was observed.

Further investigations will use mixing models to evaluate the contribution of different sources to the final isotopic an elemental pattern in sturgeon caviar. The fully validated analytical protocol for identifying the original $^{87}Sr/^{86}Sr$ isotope ratio and elemental composition of raw and processed caviar has great potential to act as a new tool in caviar provenancing.

An abstract with a similar content was submitted for RAFA 2015, Prague, Czech Republic

P26: Molecular Detection of Airborne Mold by Microfluidic qPCR

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Indoor mold enlarges the potential risk to develop respiratory symptoms and infections, skin symptoms, asthma or allergies (1, 2). These adverse health effects are caused by viable as well as non-viable fungal spores and mycelia fragments (3). The majority of detection systems are based on cultivation to determine the amount of fungal propagules in settled dust or air.

Aim of this study is to establish a molecular detection system for airborne mold to provide qualitative and quantitative information of indoor fungal particle exposure. Biodiversity together with a comparison of indoor and outdoor fungal communities are essential parameters for evaluation of indoor mold situations. The analytical method is based on quantitative PCR (qPCR) which is able to reveal the fungal load independently from viability and therefore permits a more precise determination of the biological air quality. The microfluidic system combines conventional quantitative PCR with partial automation and high throughput analysis.

So far ~20 qPCR assays have been developed to detect and quantify relevant indoor molds as well as important fungal outdoor reference species. Complex sample mixtures (DNA, spores, bioaerosols) are examined in detail to obtain a robust basis for the analysis of indoor air samples. Bioaerosols are collected at different sampling sites (visible mold growth, suspected mold growth, no signs of mold growth), at differing locations (rural vs. urban) and at different seasons. The assays are designed to cover mainly higher phylogenetic levels such as orders or genera. By this approach we expect a complete coverage of the airborne fungal biodiversity.

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P27: Cutinase-catalyzed functionalization of synthetic poly (ethylene terephthalate) fabrics

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Polyesters are ubiquitous in our daily life, used in a wide variety of products, such as textiles, plastic bottles, coatings and medical devices. The modification and functionalization of these polyesters is usually performed under harsh conditions, whereas in the last years, enzymes have been proven to be capable of replacing harsh and toxic chemicals for modifying a wide range of polyesters.

In this study we successfully made use of cutinases to partially hydrolyze PET fabric, creating new carboxylic (-COOH) and hydroxyl (-OH) groups on the polymer surface. We quantified the formation of new groups with Toluidine Blue (TBO), as well as the soluble release products, namely terephthalic acid (TA) and mono-(2-hydroxyethyl) terephthalate (MHET) via HPLC-UV. Different cutinases (Thc_Cut1, Thc_Cut2, Thc_Cut2_DM, Thc_Cut2_TM) together with NaOH as control were compared.

The enzymatic treatments resulted in new reactive polar groups which increased the hydrophilicity (WCA) and reactivity of the fabric. SEM and EDX techniques were carried out to characterize the morphological differences after enzymatic and chemical treatments, and to confirm complete protein surface removal.

Despite the fact, that synthetic polyesters are not natural substrates for the enzymes, it could be shown, that enzymatic hydrolysis indeed has a potential for modification, functionalization or degradation of different kinds of polyesters.

P28: Effects of co-cropping on lead uptake in native potential phytoextractor species (*Bidens pilosa*, *Tagetes minuta*) and *Lactuca sativa*

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Agricultural soils polluted with toxic metals are a serious problem for the food safety due to the potential accumulation of heavy metals in edible crops. Phytoremediation is generally time-consuming and requires the cessation of agriculture. Therefore, the purpose of this study was to evaluate if co-cropping of native species (*Bidens pilosa*, *Tagetes minuta*) and lettuce (*Lactuca sativa*) is suitable to produce safe crops while soils being remediated. In this work we performed a pot experiment in order to compare co-cropping effects on lead (Pb) uptake vs. mono-cropping. Plants were grown in agricultural lead polluted soils (low: 20; medium: 300; and high: 1200 µg g^{-1} Pb pseudo-total concentrations) collected nearby to a former Pb smelter in Cordoba, Argentina. Plants were separated into roots, shoots and leaves and digested for heavy metal determination. Lead and other metals were analyzed by total reflection X-ray fluorescence at the Brazilian Synchrotron Light Laboratory.

Results indicate that the interaction between rhizospheres increased the phytoextraction of Pb, which was accompanied by an increase in the biomass of the native species. The highest values of translocation, accumulation and bio-concentration of Pb were found in co-cropping conditions for *T. minuta*. In addition, the accumulation of Pb in roots of native species was closely related to other metals (Fe, Cu, Mn and Zn), especially in co-cropping. Thus, our results suggest that roots interaction promote metals mobilization, producing the entry of various metals alongside Pb in these species, being competition for nutrients and combined root exudates most likely related to these observations. Furthermore, the native species revealed to be tolerant to elevated levels of Pb in soils as opposed to lettuce plants. Lettuce plants showed an important capacity to uptake Pb that can be enhanced in co-cropping. Lettuce accumulated Pb exceeding the maximum permitted levels for consumption for all soil treatments in both co-crop and mono-crop. Thus, the consumption of lettuce represented a toxicological risk for human consume. This study showed that both crop yield and metal concentrations can be affected by rhizosphere interaction in lead contaminated soils.

P29: Dry, hydrophobic, and re-dispersible micro-fibrillated cellulose powder for polymer reinforcement obtained in a simple procedure using alkyl ketene dimer

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In order to improve the hydrophobicity of micro-fibrillated cellulose (MFC) and its dispersibility in the polymer, alkyl ketene dimer (AKD) was used to modify the MFC. In this study, a dry hydrophobic MFC powder was produced in a simple and straightforward way from a mixture of AKD and organic solvent. Scanning electron microscope (SEM) images showed that no significant differences in fibril morphology were observed for the solvent-exchanged and AKD-modified MFC. Contact angle measurements revealed that highly hydrophobic MFC was obtained by treatment of AKD, and even after extraction it remained hydrophobic which may due to strong anchoring of alkyl chains on the cellulose surface via ketoesterification. Attenuated total reflection infrared spectroscopy (ATR-IR) analysis further proved the formation of β-ketoester linkages. X-ray analysis showed that AKD was only reacting with the surface of MFC and not being detrimental to the cellulose crystal structure. Further, composites of PLA reinforced with AKD-modified MFC were prepared. Light-microscopic images of the films shown much more even and homogeneous distribution of MFC filler in PLA were achieved with the AKD-modified variant compared to the solvent-exchanged, however which did not mean an increased mechanical reinforcement revealed by static tensile testing. Dynamic mechanical analysis further proved the certain but not very significant reinforcement effect of AKD modification on MFC.

P30: Characterization of lignin model compound based on enzymatic polymerization

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Lignins are produced by all plants and they represent one of the most abundant groups of biopolymers in nature. In nature the lignin is a three-dimensional, highly cross-linked branched macromolecule composed of three types of substituted phenols, which include: coniferyl, sinapyl, and p-coumaryl alcohols that generate the guaiacyl (G), syringyl (S) and hydroxyphenyl (H) lignin subunits, respectively.

In the present study a coniferyl alcohol as a common lignin monomer was polymerized by oxidation with horseradish peroxidase (Amoracia rustiana) and H_2O_2. The polymerized lignin is also the so-called dehydrogenative polymer (DHP) was purified by CH_2Cl_2 extraction of low molecular weight compounds. DHP was characterized by FTIR and size exclusion chromatography (SEC), the structure was determined by NMR. The results have demonstrated that monolignols like coniferyl alcohols can form polymers with general the same types of inter-monolignols bonds as in natural lignin. On the other hand, today there is no isolation available to produce quantitatively and chemically non-modified lignin, therefore its true molecular weight unknown. The reality is that molecular weight determinations by SEC are performed using calibration on linear standards, which is not correct in case of branched lignin. The aim of this study was to analyze the polymerization process of lignin and to characterize lignin as a branched macromolecule. The idea of this study is to get real able standards for calibration of SEC system. The ability to enzymatically engineer new lignin structures and advances in analytical chemistry can ensure new powerful methods to control of molecular weight of lignin and analyzing its polymer structure.

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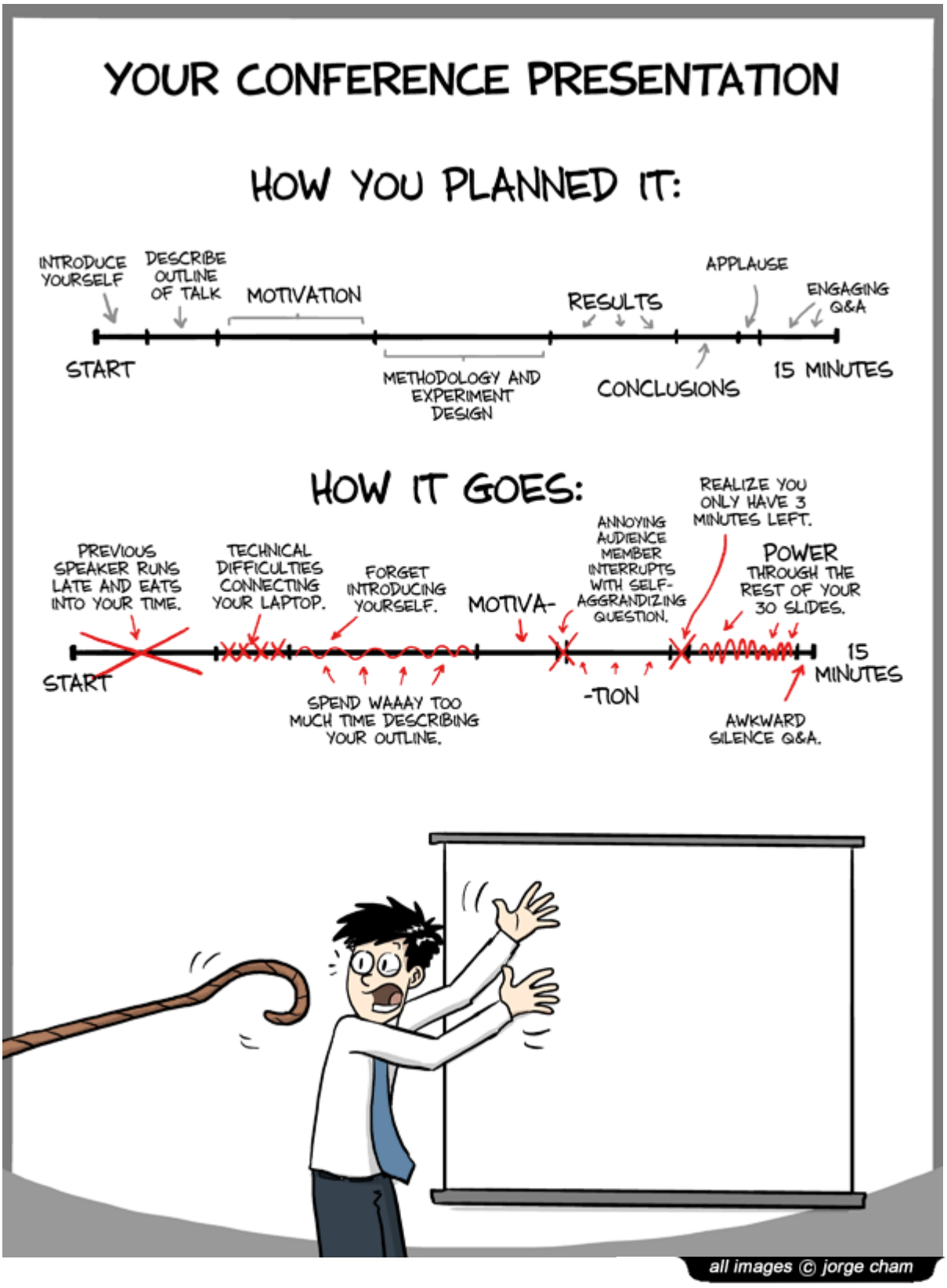
NOTES



Thank you for your participation at 3rd DocDay!