EFS 14

EUROPEAN FUSARIUM SEMINAR

April 8 – 11, 2018
Minoritenkloster
Tulln, AUSTRIA

ORGANIZING COMMITTEE

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(BOKU, AT)

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(BOKU, AT)

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General Information

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Venue address:
Minoritenplatz 1, 3430 Tulln an der Donau, Austria

EFS14 office at the venue:
The registration desk will be occupied throughout the symposium. Please contact us with any congress related queries in person or by e-mail (office@efs14.at).

Short talks:
Please prepare your presentation in power point or pdf format and hand it over to the technician (the latest) during the break before your session via USB-stick. The program is very tight, please stick to the scheduled time! **Best Oral Presentation Award sponsored by World Mycotoxin Journal.**

Poster sessions:
Posters are only accepted in A0 portrait (upright) format and are on display throughout the meeting. The two poster sessions take place on April 9th and 10th from 17:30 – 19:30. Please mount your posters at latest on Monday 9th during the lunch break from 12:30 - 14:00 and remove them on Wednesday morning at the latest. You can find the number of your poster in the poster table on page 22.
Five poster prizes (one in each session) will be sponsored by the MDPI Open Access Journal "Toxins".

Monday dinner hosted by BIOMIN and Romer Labs:
The dinner on Monday, April 8th for all participants will take place at the UFT (Universitäts und Forschungszentrum Tulln) directly after the poster session 1. Busses to and from the UFT will be provided.

Excursion Wachau and conference dinner:
On Wednesday noon April 11th, after the last scientific session participants who have booked the event will be transported by bus to Krems (at the entrance to Wachau valley). We will travel by ship* up the Danube through the Wachau to Spitz, where we will have the possibility of a short walk through the vineyards and apricot orchards, which are expected to be in full bloom at this time. From there we will go back to Dürnstein by bus. Nearby the ruins of Castle Dürnstein with a spectacular view over the Wachau valley we will have the conference dinner at the "Alter Klosterkeller" ("Heurigen" style, warm food, and wine). After dinner busses will return to the hotels in Tulln.
*In case of severe weather conditions this might be subject to change

WiFi is available at the venue with this login data:
Wlan: Atrium
PW: TullinA859

Restaurants in Tulln:
http://erleben.tulln.at/fileadmin/tulln/Dateien/Gastrofolder_Tulln_web.pdf

The **conference language** is English.

Disclaimer: We reserve the right to use any photograph/video taken at the event without the expressed written permission of those included within the photograph/video.

Welcome Address

Dear Fusarium researchers, dear colleagues!

As the chairman of the local organizing committee it is my pleasure to welcome you to the "**14th European Fusarium Seminar**" in **Tulln, Austria.** This meeting is jointly organized by the University of Natural Resources and Life Sciences, Vienna (BOKU) and the Austrian Association of Molecular Life Sciences and Biotechnology (ÖGMBT), in cooperation with the International Society of Mycotoxicology (ISM).

After the opening event with two keynote addresses on Sunday we will have half day sessions covering different aspects of Fusarium research. In each session a keynote address and two invited talks were selected by the chairs. The remaining talks were selected from submitted abstracts by the international scientific committee and the chairs of the respective sessions.

In the session "**Fusarium genomics and virulence mechanisms, population genetics and diversity**" insights into virulence mechanisms of different Fusarium species (with emphasis on secondary metabolite production) will be covered. In the second session "**Host resistance genomics, genetics and plant breeding**" mechanistic insights into resistance in cereals but also rice, maize and banana will be reported. The session "**Fusarium secondary metabolites and metabolomics of Fusarium-host plant interactions"** will besides technological advances deal with biological insights revealed by metabolomics approaches. The session "**Fusarium mycotoxins: toxicology, metabolism and remediation**" will present novel findings regarding classical Fusarium mycotoxins, mycotoxin metabolism and microbial detoxification, and the relevance of combinations of compounds. "**Integrated Fusarium management**" will address topics ranging from forecasting, epidemiology, agronomic practice (including biocontrol and novel control strategies) to avoid Fusarium infection pre-and postharvest, to problems like emerging fungicide resistance. The progress made by the two large EU-funded consortia to develop tools and strategies to minimize health risks and economic impact of Fusarium diseases and mycotoxins will be presented and discussed.

Posters will be on disply throughout this meeting and best poster prizes will be awarded, one for each session based on general voting. Participants will furthermore also be able to select the best oral presentation.

Due to noumerous competing meetings we had to postpone the EFS14 originally planned for last year. The next meeting, EFS15 will take place in Ghent. After the dinner on Monday potential organizers of the EFS16 are invited to make proposals and to advertise their location. If we have competing proposals the decision will be felled by public voting on Tuesday before the second poster session.

After the official closing of the scientific part on Wednesday we will have an excursion to the Wachau Cultural Landscape, listed as UNESCO World Heritage Site. Please bring warm and water repellent clothing – April weather might change very quickly! After the trip by bus and riverboat we will have our conference dinner finalizing the meeting in Dürnstein.

I very much hope you will enjoy the 14th European Fusarium Seminar in Tulln!

Gerhard Adam

Invited Speakers

Vessela Atanasova-Penichon, INRA/Mycology and Food Safety, FR

Vessela Atanasova-Penichon is a research engineer at the French National Institute for Agricultural Research, Mycology and Food Safety unit in Bordeaux. Since 2005, her investigation focuses on the biochemical approaches of mechanisms leading to toxin biosynthesis by *Fusarium spp.* and the regulation of this biosynthesis under biotic and abiotic stresses. Currently, her research is centered on the comprehension of plant resistance mechanisms to the *Fusarium* disease and mycotoxin accumulation, especially, biochemical resistance related to the endogenous metabolites of cereals, using targeted and non-targeted approaches. She also works on the development of biocontrol strategies in order to fight *Fusarium*. Complementary functional approaches including those of reverse and quantitative genetics are also conducted in her team to explain toxin biosynthesis and resistance mechanisms.

Vessela Atanasova-Penichon holds a PhD in Food Science from the National Higher Agronomic Scholl of Montpellier, France. Before joining Mycology and Food Safety research unit, she worked as a postdoc at the Sciences for Enology team of the French National Institute for Agricultural Research in Montpellier, on the subject of the valorization of wine industry by-products by developing new strategies for production of high added-value molecules. Vessela Atanasova is an expert for the French Association for Standardization (AFNOR), biotoxin group. Since 2012, she is an occasional lecturer for the University of Tours, France.

Kris Audenaert, Ghent University, BE

Kris Audenaert is Professor at the Faculty of Bioscience Engineering at Ghent University, Belgium. He is head of the Laboratory of Applied Mycology and Phenomics. As a molecular plant pathologist he is particularly interested in the physiological aspects of secondary metabolite production by plant pathogenic fungi during the interaction with their host plants. His current research focuses on biogenic volatile organic compounds such as green leaf volatiles to prime plants for enhanced defense against toxigenic plant pathogens. In addition, his research group explores the potential of fungal plant endophytes and their metabolites as biocontrol agents.

The research group has recently introduced “Phenomics” in their research. Phenomics uses a combination of RGB, chlorophyll fluorescence, anthocyanin, NIR and GFP/RFP imaging, to visualize the impact of plant pathogens on plants and compute scientifically relevant measures from these images, thus reaching beyond the mere visualization of phenomena.

Franz Berthiller, University of Natural Resources and Life Sciences, Vienna, AT

Franz Berthiller is Associate Professor at the University of Natural Resources and Life Sciences, Vienna (BOKU). He studied chemistry at the University of Vienna and the Vienna University of Technology. In 2006 he received the Brigitte-Gedek-Award from the German Society of Mycotoxin Research for his PhD thesis on masked mycotoxins. Franz continued his research in the lab of Prof. Rudi Krska at the IFA-Tulln, as well as some months abroad at the Danish Technical University and the Food Research Division of Health Canada in Ottawa. He is an expert on liquid chromatography coupled to mass spectrometry and his main research interested are the detection of novel fungal metabolites as well as the determination of mycotoxins and their metabolites. He headed the Christian Doppler Laboratory for Mycotoxin Metabolism from 2011-2017 and researched on the metabolism of mycotoxins in plants, microbes and animals. In 2014, he received the Fritz-Feigl-Award from the Austrian Society of Analytical Chemistry honouring his scientific development. This year, Franz was appointed editor-in-chief of the World Mycotoxin Journal. His scientific output includes more than 140 international peer-reviewed publications, which were cited about 5000 times ever since.

Siska Croubels, Ghent University, BE

Siska Croubels is full professor and head of the Department of Pharmacology, Toxicology and Biochemistry at the Faculty of Veterinary Medicine of Ghent University.
Her research in the field of veterinary toxicology focuses on the toxicokinetics and toxicokinetic modeling of mycotoxins, on the interactions between mycotoxins and pathogens in several animal species, and on the development of *in vitro* and *in vivo* models for efficacy and safety testing of mycotoxin detoxifiers. Her research group is member of the MYTOX and MYTOX-SOUTH association research platforms (www.mytox.be; www.mytoxsouth.org), dealing with the effects of mycotoxins on human and animal health in the North and South.
Her research in veterinary pharmacology focuses on pharmacokinetics, pharmacodynamics, PK/PD modeling and residues of veterinary drugs in several animal species, and on the development of a juvenile animal piglet model for paediatric preclinical drug research (www.safepedrug.eu).
Prof. Siska Croubels is associate member of the European College of Veterinary Pharmacology and Toxicology (ECVPT), and external expert for the Federal Agency for the Safety of the Food Chain and Belgian Superior Health Council. She is also member of the Ghent University Research Council, Industrial Research Fund and of several scientific committees of international symposia, such as the International Symposium for Hormone and Veterinary Drug Residue Analysis (VDRA), European Association for Veterinary Pharmacology and Toxicology (EAVPT) and Antimicrobial Agents in Veterinary Medicine (AAVM) conferences.
Prof. Siska Croubels is author and co-author of more than 220 international peer-reviewed publications and she was promoter of 19 doctoral theses in the field of veterinary pharmacology and toxicology.

Sven Dänicke, FLI Braunschweig, DE

Sven Dänicke is head of the Institute of Animal Nutrition, situated in Braunschweig and one of the institutes of the German Federal Research Institute of Animal health, the Friedrich-Loeffler-Institute. Furthermore, he is lecturer for feed safety at the Agricultural Faculty of the Martin-Luther-University Halle-Wittenberg in Germany.
He studied Animal Production, Nutrition and Veterinary Medicine at the Universities of Leipzig, Halle-Wittenberg and Hannover in Germany and obtained the degrees Dr. agr. habil. and Dr. med. vet.
Currently, he is member of the Society of Nutrition Physiology, German Society of Mycotoxin Research, and the World's Poultry Science Association. Also, he is editorial board member of the journals Archives of Animal Nutrition, Agriculture and Forestry Research, Mycotoxin Research, European Poultry Science, Archives of Animal Breeding and Topical Collection Editor of Toxins (Fusarium toxins). His major research interests are animal nutrition, nutritional physiology, immuno-nutrition, and mycotoxins.

Antonio Di Pietro, University of Cordoba, ES

Antonio Di Pietro is Professor of Genetics at the University of Córdoba. He received a M.Sc. and Ph.D. degree in Biology from University of Basel and was a Swiss National Science Foundation postdoctoral fellow at Cornell University and a visiting scientist at Novozymes Inc. in Davis, CA. He joined University of Córdoba in 1992 as a Marie Curie Fellow. His research is centred around the genetic bases and evolution of pathogenicity in fungi. His group has pioneered the use of trans-kingdom virulence models to study fungal infection on plant and animal hosts. Current interests include molecular fungus-plant signaling and the role of genome plasticity in pathogen adaptation.

Simon Edwards, Harper Adams University, GB

Simon Edwards was appointed Professor of Plant Pathology at Harper Adams University in 2010. He has studied the epidemiology and control of mycotoxigenic *Fusarium* pathogens on cereals for over 20 years. Prof Edwards is a member of JECFA (the FAO/WHO Joint Expert Committee on Food Additives and natural contaminants) and is a member of the EU-funded MyToolBox project and is the leader of the first workpackage focusing on the pre-harvest control of mycotoxins.

Donald Gardiner, CSIRO, AU

Donald joined the Commonwealth Scientific and Industrial Research Organisation, in 2005 and is based in the Agriculture and Food unit. Prior to that he worked at the University of Queensland, Australia, in mammalian genomics. His doctoral research at the University of Melbourne investigated the molecular genetics of toxin biosynthesis in fungal pathogens of both plants and animals.
Since joining CSIRO he has worked with Fusarium pathogens including *F. oxysporum*, *F. pseudograminearum* and *F. graminearum*. For the wheat infecting Fusaria, the groups applies comparative genomics, functional analyses including forward and reverse genetics and biochemical analyses to understand how this group of pathogens is so successful against these important crops. Through these approaches the groups work has uncovered the importance of a pathogens ability to overcome host derived defence compounds. In some cases the ability of pathogens to overcome these defence compounds have been acquired via horizontal gene transfer including across biological kingdom boundaries from bacteria and signatures of these transfer events are evident in the genomes of these pathogens. His team also has a strong interest in understanding how toxins and secondary metabolites in general are synthesised and regulated by these pathogens.

Kim E. Hammond-Kosack, Rothamsted Research, GB

Over 30 years experience in molecular plant pathology and molecular genetics investigating fungal and viral pathogens of wheat, barley, tomato, potato, oilseed rape and arabidopsis. Since 1998 actively engaged in the analysis of newly sequenced fungal pathogen genomes. Main research interests – understanding plant disease resistance mechanisms and pathogen virulence strategies; developing novel approaches to investigate the function of different types of fungal effectors. Her team is currently focusing on the interactions between wheat and the fungi *Fusarium graminearum*, *Fusarium culmorum*, *Zymoseptoria tritici* and *Gaeumanomyces tritici.* She is the author of over 140 refereed papers in international journals. Co-founder in 2005 of the PHI-base (www.phi-base.org) which is a knowledge database now accessed by researchers in over 125 countries. Current positions – Deputy Head of the Department of Biointeactions and Crop Protection at the Institute Rothamsrted Research in the UK, Assocate Editor – Plant Physiology, ELIXIR UK node member.

Petr Karlovsky, University of Goettingen, DE

Petr Karlovsky is professor and head of Molecular Phytopathology and Mycotoxin Research Section at the University of Göttingen in Germany. His lab studies secondary metabolites in interactions of fungi with other fungi, plants and invertebrates, including mycotoxins and their enzymatic transformations. Research highlights include unraveling the biological function of mycotoxin zearalenone; discovering citramalic acid in plant root exudates and its function; elucidating secondary metabolites in the interaction between *Brassica napus* and *Verticillium longisporum*; untangling the role of fusaric acid and its detoxification in interaction between a mycoparasite and its prey; elucidating defense metabolites of *Aspergillus nidulans* targeting fungivores; and identification of volatile metabolites of plants that can serve as biomarkers of fungal infection.

Petr Karlovsky studied biochemistry and obtained his PhD in the Institute of Biophysics in Brno, Czech Republic. He worked as a postdoc at Gesellschaft für Strahlen- und Umweltforschung (GSF) and at the University of Göttingen in Germany before assuming position of a Research Manager at DuPont/Pioneer Hi-Bred in Des Moines (IA) and Wilmington (DE), USA. He was offered visiting professorship by universities in Egypt, Italy and India and accepted visiting professorship awards from the University of Connecticut in Storrs, USA, and Zhejiang University in Hangzhou, China.

Gerrit HJ Kema, Wageningen University and Research, NL

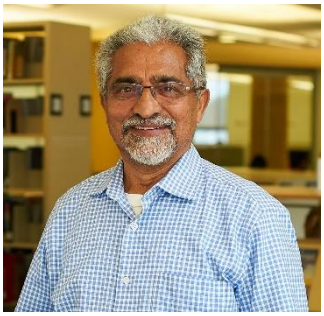
Gert Kema (1957) has 33 years of experience in plant pathology, host and pathogen genetics and genomics, specializing in foliar diseases of cereals and banana. Since 5 years major lead of international programs on Fusarium wilt in banana. Published over 100 peer reviewed scientific articles, holder of patents and international speaker. Interests are plant diseases (in the tropics) and their management, food security. He obtained a BSc degree in agronomy, a MSc in plant breeding. He obtained his PhD in 1996 on research into *Zymoseptoria tritici,* the septoria tritici blotch fungus of wheat, which is still ongoing. Currently he also holds a chair as professor of tropical plant pathology at Wageningen University, The Netherlands, with a focus on banana and the fungal pathogens *Fusarium oxysporum* f.sp. *cubense* and *Pseudocercospora fijiensis*. He is a co-founder of two companies in the field of bioprocessing and genetic engineering of banana.

Rudolf Krska, University of Natural Resources and Life Sciences, AT

Rudolf Krska is full professor for *(Bio-)Analytics and Organic Trace Analysis* at the University of Natural Resources and Life Sciences, Vienna (BOKU). He is head of the Center for Analytical Chemistry at the Department of Agrobiotechnology (IFA-Tulln) at BOKU with some 50 staff. From 2010-2015 Prof. Krska also served as head of the BOKU-department IFA-Tulln with more than 200 staff. He was also member of the Working Group Fusarium of the Scientific Panel on Contaminants of the European Food Safety Authority and worked for one year as A/Chief of Health Canada´s Food Research Division (2009/10) where he was responsible for the research on chemical contaminants in foods carried out within Health Canada´s National Food Chemical Safety Laboratory Network.

Prof. Rudolf Krska has received 10 scientific awards and is (co-)author of more than 300 SCI publications (h-index: 52; citations in 2016: >1250). In 2015 and 2016, Thomson Reuters (Web of Science) identified Prof. Krska as one of the world's most influential contemporary researchers due to his ranking among the top 1% most cited authors in the field of agricultural sciences. His current research interests are in the area of plant-fungi metabolomics, IR-spectroscopy and novel mass spectrometric methods for the determination of multiple mycotoxins including their conjugation and transformation products in food, feed and other biological matrices. As of March 2016, Prof. Krska acts as coordinator of the European Commission funded project MyToolBox (Safe Food and Feed through an Integrated ToolBox for Mycotoxin Management) with 23 partners (including 40% industry participation and 3 partners from China) and a funding volume of more than 6 Mio Euro. Since 2017, he is also Green Area Leader at the Austrian Competence Centre for Feed and Food Quality, Safety and Innovation (FFOQSI) with 18 company partners and a project volume of >5 Mio Euro.

Ajjamada Kushalappa, McGill University, CA



Dr. Ajjamada C. Kushalappa, is a professor at McGill University, Quebec, Canada. Current focus of his research is on the identification of resistance genes in plants against biotic stress based on comprehensive transcriptomics and semi-comprehensive metabolomics, and use of these genes/gene segments to replace those that are polymorphic in commercial cultivars based on genome editing using CRISPR-Cas9 system. His team has identified several R-genes in wheat against fusarium head blight and in potato against late blight. The mechanism of resistance is mainly due to resistance related metabolites, polymers and conjugates, that are deposited to reinforce the cell wall, thus containing the pathogen to initial infection. These metabolite biosynthetic genes are regulated by a hierarchy of genes, such as transcription factors, MAP kinases and receptors, following pathogen invasion. He was an invited speaker at several national and international conferences. He has published more than 100 papers and was awarded Dr. and Mrs. Bailey award, by the Canadian Phytopathological Society, for an exceptional and distinguished contribution to plant pathology.

Antonio Logrieco, National Council of Research, IT

Dr. Antonio F. Logrieco, Director of Institute of Sciences of Food Production, National Research Council of Italy (http://www.ispacnr.it/). Scientific responsible and coordinator of various national and international projects dealing on Plant pathology and food safety,with particular attention on mycotoxin problem including the COST action 835 “Agriculturally important toxigenic fungi”; DeTox-Fungi-1999-01380 in the FP V; WP3“Microsystems Technology solutions for rapid detection of toxigenic fungi and mycotoxins in Good Food FP6-IST-1-508774-IP in FP VI; “Novel integrated strategies for worldwide mycotoxin reduction in food and feed chains”-MycoRed-KBBE-2007-2-5-05 in FP VII; MycoKey; “Integrated and innovative key action for mycotoxin management in the food and feed chain) –MycoKey Horizon 2020 (http://www.mycokey.eu/). In addition he participated in the following EU projects: Wine-Ochra Risk-2000-01761; Ramfic-1999-00284; RAFBCA-2000 01391; Myco-globe-7174.

He is the founder of the Agro-Food Microbial Culture Collection "ITEM" (http://server.ispa.cnr.it/ITEM/Collection/). He is the co-founder and former Past-President of International Society for Mycotoxicology (http://www.mycotox-society.org) and acting President of Mediterranean Phytopathological Union. He was President of ISPP “Fusarium Committee”.

Elected as member of Hungarian Academy of Sciences and nominated as Distinguished International Supervisor of Institute of Food Science and Technology, Chinese Academy of Agricultural Sciences. He was member of organizing committee and invited speaker in international conferences and referee and editor of various books and international journals.

He has strong record of publication and papers on international journal (200) and co-editor of 12 books on mycology, molecular/bio-chemical characterization of active secondary metabolites (mainly phyto-mycotoxins) of plant pathogens as well as relevant experties in fungal genetic and fermentation technology.

Antonio Moretti, Research National Council, IT

Researcher at the Istituto tossine e micotossine da parassiti vegetali (ITEM, since 2001 Institute of Sciences of Food Production, ISPA), National Council of Research (CNR), since 1989. Senior Researcher at ISPA-CNR since 2007. He is/has been responsible of several National and International projects on Toxigenic fungi. The area of work are Plant Pathology, Mycology, Mycotoxicology and Fungal genetics with main field of interest in the identification, genetic and toxinogenic characterization of main *Fusarium* and *Aspergillus* species, pathogens of agriculturally important crops and the study of their diversity and pathogenicity. He has been responsible for ISPA of several National and European projects on the toxinogenic fungi. He is now Leader of the Work Package 8 “Communication, Dissemination & Exploitation” of the Horizon 2020 UE Project 678781 Integrated and innovative key actions for mycotoxin management in the food and feed chain “MycoKey”. Research activity of Dr. Moretti is well documented by several papers on International Journals and book chapters ISI and hundreds of abstract and proceedings of National and International scientific meetings. H-index of Antonio Moretti: 31 (ISI Web of Science) and 37 (Scopus).

Paul Nicholson, John Innes Centre, GB

Paul Nicholson leads a group within the Crop Genetics Department of the John Innes Centre. Many years ago, he began working on facultative pathogens involved in disease complexes of the stem base and heads of cereals. His initial work focussed on developing tools to dissect the components of complexes and determine the relative amounts of each species to study the effect of host resistances and/or agronomic practices on individual pathogen species. His group then turned its attention to investigating disease resistance using both crop and model plant species with a focus on FHB. This included studying the role of phytohormone signalling pathways on resistance and potential trade-off between resistance to different diseases and between disease resistance and agronomic traits. This highlighted the importance of host functions in conferring susceptibility to Fusarium pathogens. His group is currently attempting to identify genes associated with resistance and/or susceptibility in order to optimise FHB resistance in wheat.

Isabelle Oswald, INRA, FR

Senior reseach scientsit and deputy head of the INRA-Research Center in Food Toxicology, Toxalim in Toulouse, France.
During the last 20 years she analysed the mechanism involved in the toxicity of mycotoxins, especially Aflatoxins, Fumonisins and Deoxynivalenol. Her work especilly focussed on the effect of mycotoxin on the intestinal and immune responses. Recently her work focussed on the effect of mycotoxin mixtures.
Dr. Oswald has more than 100 peer-reviewed international publications. She is an expert for the European Food Safety agency and for the French Agency for Food, Environmental and Occupational Health & Safety.
She is currently leading a research team with 6 scientists, 4 technicians and 9 doctoral and post-doctoral fellows. This team has two main goals (1) to determine the toxic effects of mycotoxins using both in vitro, ex vivo and in vivo models, (2) to characterize the production of mycotoxins and other secondary metabolites by fungal species.

Robert H. Proctor, US Department of Agriculture, US

Robert (Bob) Proctor is a microbiologist at the United States Department of Agriculture research institute in Peoria, Illinois. He received a bachelor's degree in Biology from the University of Victoria in British Columbia, Canada, and graduate degrees in Plant Pathology from the University of Wisconsin in Madison. After graduate school he moved to the USDA where he began work to elucidate the genetic and biochemical pathways that lead to the formation of mycotoxins in *Penicillium* and *Fusarium*. At the USDA he has also studied the roles that mycotoxins play in the ecology of fungi that produce them, and on other biological processes that contribute to the ecological fitness of *Fusarium*. The results of his collaborative research with chemists and other biologists at the USDA as well as other institutions has provided insights into the genetic bases of variation in mycotoxin production in fungi. More recent work on the genomics and evolution of mycotoxin biosynthesis has unraveled some of the genetic mechanisms that have contributed to the current distribution of mycotoxin production among and within *Fusarium* species. This work has also provided insight in to the evolutionary processes that have contributed to the structural diversity of mycotoxins produced by *Fusarium* and other fungi.

Barbara Steiner, University of Natural Resources and Life Sciences, Vienna, AT

Barbara Steiner is senior scientist in the group of Prof. Hermann Buerstmayr at the BOKU-University of Natural Resources and Life Sciences, Vienna, where she also received a Ph.D. degree in plant breeding. Her research focuses on genetics and genomics of cereal crops and the major disease Fusarium head blight. Thereby the genetic analysis of the quantitative resistance, high-resolution mapping of the resistance loci and resistance gene isolation are main research tasks. She established mutant screens for forward and reverse genetic approaches and phenotyping methods for high-throughput Fusarium and mycotoxin resistance testing. As a plant breeder she is particularly interested in breeding bread wheat and durum wheat for increased Fusarium head blight resistance including germplasm evaluations, characterization and utilization of genetic resources, pre-breeding and the development of methods and tools for genomics-assisted breeding.

Michele Suman, Barilla SpA, IT

Michele obtained his Analytical Chemistry Degree, Summa Cum Laude, at University of Ferrara in 1997. He won the National Prize for Young Researchers promoted by the Italian Chemistry Federation (Federchimica) in 1998.
Then he took a master's degree in 1998 (Master in Science, Technology and Management from University of Ferrara, working at the same time at the "Natta Research Center" of Shell-Montell Polyolefins) and a doctorate in 2005 (PhD in Science and Technology of Innovative Materials from University of Parma), landing the role of Food Chemistry & Safety Research Manager in Barilla Spa company in 2003. Here he has been working in an international contest with public and private research centers\organizations on research projects within the field of food chemistry, food safety-quality-authenticity, food contact materials, sensing and mass spectrometry applications for food products.
He is member of working groups in European Committee for Standardization (CEN) and Vice-Chair of the ILSI Process Related Compounds & Natural Toxins Task Force; Leader in the Food Safety Section of Italian National Cluster Agrifood, member of the Board of Mass Spectrometry Division – Italian Chemistry Society; editorial boards of important peer-reviewed journals (World Mycotoxin Journal, Food Additives and Contaminants).

He has been involved in various National/European Funded Projects and presently he is WP Leader in the EU-FP7 FoodIntegrity Project and in the EU-H2020 MyToolBox projects.
He has developed experience in academic teaching activities, masters\PhD projects supervision, coordination/chairmanship of international conferences (Recent Advances in Food Analysis, World Mycotoxins Forum, FoodIntegrity, MS Food Day, Rapid Methods Europe).
His scientific production is documented by five book chapters, 115 contributions at national and international conferences and 70 papers in international ISI journals.

Silvio Uhlig, Norwegian Veterinary Institute, NO

Silvio Uhlig is senior researcher at the Norwegian Veterinary Institute (NVI) and head of the Chemistry Section. He received his degree in chemistry from the University of Oslo and studied metabolites of *Fusarium avenaceum* for his PhD. He is a toxin chemist and is presently involved in projects dealing with the chemistry of toxins from fungi, plants, marine algae and cyanobacteria. His contribution to these projects is in the development of analytical methods; isolation, purification and structure determination of new metabolites or biotransformation products; bioassay-guided fraction as well as reactivity testing and derivatisation.
Silvio is also a member of the Toxinology Research Group at the NVI, which focuses on toxinology in a one-health perspective. The research group studies the biosynthesis, biological effects and toxicokinetics of various toxins, among them toxins from *Fusarium*, *Aspergillus*, *Penicillium* and *Claviceps*.

Joseph-Alexander Verreet, Christian-Albrechts-University Kiel, DE

Joseph-Alexander Verreet received a diploma degree from the University of Bonn and a Ph.D. degree in agricultural sciences from the Technical University of Munich, where he also acquired his habilitation in plant pathology. Now he is full professor of plant pathology at the University of Kiel, where he still serves as the head of the Department of Phytopathology. In his research, he developed integrated pest management (IPM) models for major crops by analyzing several multi-year supra-regional case studies on epidemic and damage dynamics and extrapolating functional control thresholds. According to the IPM threshold approach, chemical crop protection measures are ideally applied in the epidemiologically sensitive stage of the pathogen at a reduced application rate (transition from accrescence to progression phase). He published models for IPM of wheat, barley, and sugar beet diseases. The sugar beet IPM model is employed across Europe for the control of beet leaf diseases. The models are aimed at optimizing pesticide use by preferentially using cultural controls, including sanitation practices. According to his scientific view, the key for solutions to problems concerning contagious diseases lies in the biology of the pathogens, their physiological abilities, their behavior in the field, and their population dynamics as affected by cultivation and environment. The multi-year collection of biological and meteorological data has now led to the derivation of functional prognosis models in wheat (IPM Model). For example, the prediction accuracy for leaf blotch is 93%, for brown rust and powdery mildew 80%. The multiple regression model for FHB predicts the expected wheat grain contents of DON and ZEA at flowering with an accuracy of 78 to 80%. In addition, the model can be used to carry out a *Fusarium*-specific treatment with fungicides at flowering in case of a prognosticated severe *Fusarium* epidemic with simultaneously high levels of DON and ZEA.

Detailed Program

SUNDAY - April 8

16:00 - 19:00	Registration	
18:00 - 18:30	Welcome Note	**Gerhard ADAM (Chair and local organizer)** **Peter EISENSCHENK (Mayor Tulln)** **Christian OBINGER (Vice rector of research at BOKU)** **Angela SESSITSCH (President ÖGMBT)** **Rudolf KRSKA (President ISM)** **Antonio MORETTI (Chair and local organizer of EFS13)**
18:30 - 19:15	Plenary Lecture	**Antonio di PIETRO (University of Cordoba, ES)** Understanding host adaptation in *Fusarium oxysporum*
19:15 – 20:00	Plenary Lecture	**Petr KARLOVKSY (University of Goettingen, DE)** Ecological functions of toxins produced by *Fusarium spp.*
20:00 - 22:00	Reception	

MONDAY - April 9

9:00 - 12:30	**Session 1**	***Fusarium genomics and virulence mechanisms, population genetics and diversity***
		Chairs: Gerhard ADAM & Kim HAMMOND-KOSACK
9:05 - 9:35	Keynote	**Donald GARDINER (CSIRO, AU)** *Fusarium* pathogenomics: old approaches meet new technology in understanding pathogen virulence
9:35 - 9:55		**Kim HAMMOND-KOSACK (Rothamsted Research, GB)** Investigating the genome of the Quorn fungus *Fusarium venenatum*
9:55 - 10:07		**Linda J. HARRIS (Agriculture & Agri-Food Canada, CA)** Phytotoxic cyclic lipopeptides produced by *Fusarium graminearum*
10:07 - 10:19		**Melvin BOLTON (USDA - ARS, US)** Genomics and effector characterization of the novel sugarbeet pathogen *Fusarium secorum*
10:19 - 10:31		**Francis FABRE (INRA UMR GDEC/UCA, FR)** Comparative genomics of *Fusarium graminearum* strains harboring aggressiveness in bread wheat
10:35 - 11:00	Coffee break	

11:00 - 11:20		**Robert PROCTOR (US Department of Agriculture, US)** Distribution and evolution of genes responsible for biosynthesis of mycotoxins in *Fusarium*
11:20 - 11:32		**Antonio PRODI (University of Bologna, IT)** Role of the *Fusarium tricinctum* species complex in Fusarium Head Blight disease: virulence and mycotoxin production in durum wheat
11:32 - 11:44		**Olga P. GAVRILOVA (All-Russian Institute of Plant Protection (VIZR), RU)** Intraspecific trait variations in *Fusarium langsethiae*
11:44 - 11:56		**Thomas SVOBODA (Universität für Bodenkultur Wien, AT)** IAA-Asp amidohydrolase – a putative effector protein of *Fusarium* species
11:56 - 12:08		**Tapani YLI-MATTILA (University of Turku, FI)** *Fusarium graminearum* diversity in Finland, Norway, and Russia
12:08 - 12:28		**Antonio MORETTI (Research National Council, IT)** Genetic variability, mycotoxin profile and metabolomics of *Fusarium proliferatum* pathogen on different host plants.
12:30 - 14:00	Lunch break	
14:00 - 17:30	**Session 2**	***Host Resistance: mechanisms, genetics, genomics and breeding***
		Chairs: Hermann BÜRSTMAYR & Fiona DOOHAN
14:05 - 14:35	Keynote	**Paul NICHOLSON (John Innes Centre, GB)** Insights into Fusarium-host interactions gained from crop and model species
14:35 - 14:55		**Fiona DOOHAN (University College Dublin, IE)** FHB – resistance genes, genomic hotspots and grain development
14:55 - 15:07		**Thomas MIEDANER (University of Hohenheim, DE)** Genomic approaches for increasing Fusarium head blight resistance in durum and bread wheat
15:07 – 15:19		**Ingerd Skow HOFGAARD (Norwegian institute of Bioeconomy research, NO)** Resistance to *Fusarium langsethiae* in Norwegian oats – Safe Oats
15:19 - 15:31		**Maria BUERSTMAYR (University of Natural Resources and Life Sciences (BOKU), AT)** Anther extrusion, a passive resistance factor of Fusarium head blight, is associated with semi-dwarfing genes *Rht-B1* and *Rht-D1*
15:35 - 16:00	Coffee break	

16:00 - 16:20		**Barbara STEINER (University of Natural Resources and Life Sciences, Vienna, AT)** Classical and genomics-assisted improvement of Fusarium head blight resistance in bread wheat, durum wheat and triticale
16:20 - 16:32		**Davide SPADARO (University of Torino, IT)** Elucidating Bakanae disease resistance in japonica rice
16:32 - 16:44		**Herbert MICHLMAYR (BOKU Vienna, AT)** Trichothecene-conjugating UDP-glucosyltransferases: substrate specificities, kinetics and inhibition by culmorin
16:44 - 16:56		**Karolina Maria SLOMINSKA-DURDASIAK (Leibniz Institute of Plant Genetics and Crop Plant Research (IPK), DE)** Host-induced gene silencing as natural resistance strategy of wheat against Fusarium head blight
16:56 - 17:08		**Lindy ROSE (Stellenbosch University, ZA)** Resistance to *Fusarium verticillioides* and fumonisin accumulation in African maize inbred lines resistant to *Aspergillus flavus* and aflatoxins
17:08 - 17:28		**Gerrit H.J. KEMA (Wageningen University and Research, NL)** Fusarium wilt strikes global banana production, again
17:30 - 19:30		***Poster session 1* – odd poster numbers**
20:00 - 21:30	Dinner	**Dinner hosted by BIOMIN and Romer Labs at UFT**

TUESDAY - April 10

9:00 - 12:30	**Session 3**	***Fusarium secondary metabolites and metabolomics of Fusarium-host plant interactions***
		Chairs: Rainer SCHUHMACHER & Silvio UHLIG
9:05 - 9:35	Keynote	**Ajjamada KUSHALAPPA (McGill University, CA)** Gene discovery and editing to enhance resistance in wheat against fusarium head blight
9:35 - 9:55		**Silvio UHLIG (Norwegian Veterinary Institute, NO)** *Fusarium langsethiae* – the tough fungus from the North
9:55 - 10:07		**Alexandra M. SIMADER (Center for Analytical Chemistry, AT)** Metabolomics of Fusarium head blight: Examining the attack of *Fusarium graminearum* during infection of two near isogenic wheat lines differing in the resistance QTLs Fhb1 and Qfhs.ifa-5A
10:07 - 10:19		**Sylvain CHÉREAU (INRA, FR)** Mycotoxin biosynthesis and central metabolism are two interlinked pathways in *Fusarium graminearum*, as demonstrated by the extensive metabolic changes induced by caffeic acid exposure
10:19 - 10:31		**Justyna LALAK-KAŃCZUGOWSKA (Institute of Plant Genetics of the Polish Academy of Sciences, PL)** How do abiotic factors influence growth, fumonisin biosynthesis and stress response in *Fusarium proliferatum*?
10:35 - 11:00	Coffee break	

11:00 - 11:20	**Vessela ATANASOVA-PENICHON (INRA/Mycology and Food Safety, FR)** Metabolomics to decipher biochemical defence of cereals against *Fusarium* and mycotoxin accumulation
11:20 - 11:32	**Michele C. LOEWEN (National Research Council of Canada, CA)** 'Omics profiling of *Clonostachys rosea* strain ACM941 highlights unique genetic and metabolic features potentially contributing to its bio-control of *Fusarium graminearum*
11:32 - 11:44	**Safa OUFENSOU (University of Sassari, IT)** *FCRAV2*, a gene involved in significant changes in the physiological and metabolic profiles of *F. culmorum*
11:44 - 11:56	**Christoph BUESCHL (University of Natural Resources and Life Sciences, Vienna (BOKU), AT)** Annotation of *Fusarium graminearum*'s dark matter: Clustering of unknown, structurally similar fungal metabolites during wheat infection by molecular networking
11:56 - 12:08	**Nadia PONTS (INRA, FR)** Nucleosome dynamics in the toxin-producing plant pathogen *Fusarium graminearum*
12:08 - 12:28	**Kris AUDENAERT (Ghent University, BE)** Uncovering the priming potential of Z-3-hexenylacetate in the tripartite interaction between wheat, *Fusarium graminearum* and the English grain aphid *Sitobion avenae*
12:30 - 14:00	Lunch break

14:00 - 17:30	**Session 4**	***Fusarium mycotoxins - Toxicology, Metabolism and Remediation***
		Chairs: Dieter MOLL & Siska CROUBELS
14:05 - 14:35	Keynote	**Isabelle OSWALD (INRA, FR)** Cocktail effect of trichotechenes on the intestine
14:35 - 14:55		**Siska CROUBELS (Ghent University, BE)** Comparative toxicokinetics and oral bioavailability of (emerging) *Fusarium* mycotoxins in pigs and poultry, in relation to species dependent sensitivity and selection of biomarkers for exposure and efficacy testing of mycotoxin detoxifiers
14:55 - 15:07		**Kerry O'DONNELL (USDA, ARS, US)** Marasas' et al. (1984) “Toxigenic *Fusarium* Species: Identity and Mycotoxicology” revisited
15:07 - 15:19		**Carol Verheecke VAESSEN (University of Cranfield, GB)** Impact of Climate Change interacting factors on growth and T-2 and HT-2 production by *Fusarium langsethiae* strains
15:19 - 15:31		**Daren W. BROWN (USDA, US)** New regulatory tricks for an old toxin cluster
15:35 - 16:00	Coffee break	
16:00 - 16:20		**Franz BERTHILLER (University of Natural Resources and Life Sciences, Vienna, AT)** Masked Fusarium mycotoxins: an overview on recent discoveries
16:20 - 16:32		**Heidi SCHWARTZ-ZIMMERMANN (University of Natural Resources and Life Sciences, Vienna, AT)** A foray into the metabolization of deoxynivalenol by different animal species
16:32 - 16:44		**Doris MARKO (University of Vienna, AT)** Impact of co-occurring xenoestrogens on the endocrine disruptive potential of zearalenone and α-zearalenol
16:44 - 16:56		**Laura De METS (Ghent University, BE)** Microbial degradation of zearalenone by Actinobacteria: Mind the toxicity
16:56 - 17:08		**Jason CARERE (Agriculture and Agri-Food Canada, CA)** The enzymatic detoxification of deoxynivalenol (DON): Identification of the DON epimerization pathway
17:08 - 17:28		**Sven DÄNICKE (FLI Braunschweig, DE)** Inactivation of *Fusarium* toxins: Implications for health and performance of pigs
17:30 - 19:30		***Poster session 2* – even poster numbers**

WEDNESDAY - April 11

9:00 - 12:30	**Session 5**	***Integrated Fusarium management (pre/postharvest, epidemiology and forecasting, fungicide resistance, utilization of contaminated batches)***
		Chairs: Rudolf KRSKA & Antonio LOGRIECO
9:05 - 09:35	Keynote	**Rudolf KRSKA (University of Natural Resources and Life Sciences, AT)** A novel integrated management strategy to tackle mycotoxins along the entire food and feed chain
9:35 - 10:05	Keynote	**Antonio LOGRIECO (National Council of Research, IT)** Integrated and innovative MYCOKEY actions for *Fusarium* mycotoxin management in the food and feed chain
10:05 - 10:25		**Simon EDWARDS (Harper Adams University, GB)** Integrated *Fusarium* management
10:25 - 10:37		**Susanne VOGELGSANG (Agroscope, CH)** Healthy and safe oats? Dominant *Fusarium* species, influencing cropping factors, epidemiology and susceptibility
10:40 - 11:05	Coffee break	

11:05 - 11:25	**Michele SUMAN (Barilla SpA, IT)** Synergic potential of pre-milling and milling strategies to minimize mycotoxins and increase fiber content of wheat-based products
11:25 - 11:37	**Rumiana RAY (University of Nottingham, GB)** Interactions between *Fusarium* and *Microdochium* species, fungicides and host resistance: consequences for Fusarium head blight disease and mycotoxin production in wheat
11:37 - 11:49	**Cheng LIU (RIKILT, NL)** Cross validation of forecasting models for DON in wheat in Europe
11:49 - 12:01	**Gary A SECOR (North Dakota State University, US)** Integrated Fusarium management of potatoes in the USA
12:01 - 12:13	**Päivi PARIKKA (Natural Resources Institute Finland (Luke), FI)** Does zero-tillage increase mycotoxins in oats and barley in cool climate?
12:13 - 12:33	**Joseph-Alexander VERREET (Christian-Albrechts-University Kiel, DE)** Forecasting model for the control of *Fusarium* diseases and the mycotoxin content of wheat and maize
12:33-12:40	**Closing remarks and announcements**
12:40 - 14:00	Lunch break
14:00 - 22:00	**Social program: Excursion WACHAU + CONFERENCE DINNER**

Posters Table

Poster presentation times: Poster session 1: odd poster numbers, Poster session 2: even poster numbers.
All posters are on display from Sunday until Wednesday

Poster #	Poster presenter
PP-01	Giovanna Delogu
PP-02	Klaus R. Westphal
PP-03	Eugènia Miró Abella
PP-04	Reinhard Wimmer
PP-05	Vijai Bhadauria
PP-06	Jana Chrpová
PP-07	Gerlinde Wiesenberger
PP-08	Tsutomu Arie
PP-09	Gerhard Adam
PP-10	Slavica Stanković
PP-11	Thomas Svoboda
PP-12	Matias Pasquali
PP-13	Tomke Musa
PP-14	Nadia Ponts
PP-15	Marine Ollier
PP-16	Stephane Bieri
PP-17	Virgilio Balmas
PP-18	Aparna Haldar
PP-19	Rasmus Wollenberg
PP-20	John F. Haidoulis
PP-21	Heidi Udnes Aamot
PP-22	Seong Mi Jo
PP-23	Lydia Woelflingseder
PP-24	Maarten Ameye
PP-25	Nora A. Foroud
PP-26	Josue Diaz
PP-27	Christo J. Botha
PP-28	Akvile Jonaviciene
PP-29	Alejandro Gimeno
PP-30	Guro Brodal
PP-31	Daniel Palmero
PP-32	Jiang Tan
PP-33	Romain Valade
PP-34	Juan M. Palazzini
PP-35	Mohamed Fathi Abdallah
PP-36	Xiben Wang
PP-37	Gerlinde Bichl
PP-38	Joseph Crosby
PP-39	Ludwig H. Pfenning
PP-40	Ganesh Thapa
PP-41	Sobia Ajaz
PP-42	Simone Bachleitner
PP-43	Marko Maricevic
PP-44	Miguel Costa E Silva Dos Santos
PP-45	Samira Chekali
PP-46	Ilse Dohnal
PP-47	Akos Mesterhazy
PP-48	Doris Hartinger
PP-49	Dianevys Gonzalez-Pena Fundora
PP-50	Renata Cantoro
PP-51	Kane D'Arcy Cusack
PP-52	Heidi Udnes Aamot
PP-53	Alessandra Lanubile
PP-54	Ana Butrón
PP-55	Dian Schatzmayr
PP-56	Silvia Labudova
PP-57	Theresa Schlederer
PP-58	Évelin F. Wigmann
PP-59	Benjamin Hales
PP-60	Sonja Tančić Živanov
PP-61	Seweryn Frasinski
PP-62	Paulina Georgieva
PP-63	Krisztian Twaruschek
PP-64	James R. Tucker
PP-65	Nadezhda Gogina
PP-66	Hege H. Divon
PP-67	Amal Kahla
PP-68	Rita Zrenner
PP-69	Valérie Cadot
PP-70	Hye-Seon Kim
PP-71	Rachel Goddard
PP-72	Camila P. Nicolli
PP-73	Katrin Rehak
PP-74	Ruth Dill-Macky
PP-75	Roma Semaskiene
PP-76	Rogelio Santiago
PP-77	Samia Gargouri
PP-78	Jens Laurids Sørensen
PP-79	Aleksandra Orina
PP-80	Hiroki Saito
PP-81	Asja Ceranic
PP-82	Derrick Mayfield
PP-83	Lukasz Stepien
PP-84	Florence Forget
PP-85	Dimitrios Drakopoulos
PP-86	Ingerd S Hofgaard
PP-87	eya khemir
PP-88	Jonas Vandicke
PP-89	Felix Hoheneder
PP-90	Pi-Fang Linda Chang
PP-91	Keshav Bahadur Malla
PP-92	Torsten Schöneberg
PP-93	Maria Teresa Senatore
PP-94	Maria Doppler
PP-95	Ilse Vanhoutte
PP-96	Tatiana Gagkaeva

EFS14 - EUROPEAN FUSARIUM SEMINAR

APRIL 8 – 11, 2018, TULLN, AUSTRIA

Lectures

EFS14 - EUROPEAN FUSARIUM SEMINAR

APRIL 8 – 11, 2018, TULLN, AUSTRIA

Welcome Note & Plenary Session

Plenary Lecture

PS-01 Understanding host adaptation in *Fusarium oxysporum*

Cristina López-Díaz[1], Dilay Hazal Ayhan[2], David Turrà[1], Li-Jun Ma[2], **Antonio Di Pietro[1]**

[1] Genetics, University of Cordoba, Spain
[2] University of Massachusetts, Amherst, USA

Filamentous plant pathogens pose a severe threat to global food security. These organisms often show exquisite host adaptation, but also undergo rapid evolution leading to shifts or expansions in the host range. The genetic mechanisms of pathogen-host adaptation remain poorly understood. In the soil-inhabiting vascular wilt fungus *Fusarium oxysporum,* individual isolates tend to exhibit high specificity towards a given plant host, while the species complex collectively attacks more than a hundred different crops. In addition, *F. oxysporum* is also an emerging human pathogen that provokes lethal systemic infections in immunocompromised individuals. Remarkably, a single field isolate of this fungus can kill tomato plants, immunodepressed mice and insects. By following a combination of reverse genetics and experimental evolution approaches, we found that *F. oxysporum* uses multiple strategies to adapt to different host environments. These include recruitment of conserved fungal signaling pathways or hijacking of host regulatory mechanisms for new virulence mechanisms. Strikingly, fungal populations evolved after serial passaging through different environments displayed large-scale chromosomal reorganizations in transposon-rich accessory regions of the genome, suggesting that chromosome plasticity could act as a major evolutionary driver in *F. oxysporum*. Understanding the genetic mechanisms that govern virulence evolution and host adaptation may reveal new ways to control diseases caused by filamentous pathogens and improve plant health.

Plenary Lecture

PS-02 Ecological functions of toxins produced by Fusarium spp.

Petr Karlovsky

Molecular Phytopathology and Mycotoxin Research, University of Goettingen, Germany

Secondary metabolites of Fusarium spp. possess amazing structural diversity and exert interesting biological activities, inspiring the industry to use them as lead structures for new drugs and plant protection chemicals. Four drugs based on Fusarium metabolites made it up to clinical trials and one metabolite was on the market as a drug till 2016. Many Fusarium metabolites are toxic and some of these mycotoxins are regulated. In spite of the impact of metabolites of Fusarium on food and feed safety, their application potential as biologically active compounds and extensive research on their toxicity, biosynthesis and detoxification, only limited efforts have been devoted to understanding their biological function. Why do Fusarium spp. produce these amazing structures? How do mycotoxins enhance the fitness of their producers? What selection pressures maintain their expensive biosyntheses? It is astonishing that these fundamental and fascinating questions have been largely ignored by the reserch community. The impact of mycotoxins on food and feed safety might partly account for this situation: practitioners ask how to reduce mycotoxin exposure and the society pays for empirical solutions rather than for the gain of knowledge. Restricted view of specialists working in narrow fields of research may account for the remaining part of the neglect. Plant pathologists only test their standard hypothesis that a fungal metabolite is involved in a plant disease; other options are not considered. Toxicologists study the effects of mycotoxins on mammalian cells, ignoring the fact that toxicity to vertebrates is a side effect and thus an artifact that does not bring us any closer to fundamental questions. Biological chemists generated a wealth of data on the toxicity of fungal metabolites to insects, postulating that mycotoxins are fungal defense agents, but their hypotheses have seldom been tested in ecologically relevant settings. To make the situation worse, unsupported assumptions ("mycotoxins are stress metabolites") and hypothesis that have been disproved decades ago ("zearalenone is a fungal pheromone") continue to be copied from old to new literature. The goal of this contribution is to draw attention to the bias of current research on secondary metabolites of Fusarium and suggest how fundamental questions can be addressed, based on the results generated by concerted efforts of ecologists and biological chemists in the last years.

EFS14 - EUROPEAN FUSARIUM SEMINAR

APRIL 8 – 11, 2018, TULLN, AUSTRIA

Session 1: Fusarium genomics and virulence mechanisms, population genetics and diversity

Chairs: Gerhard ADAM & Kim HAMMOND-KOSACK

Keynote Lecture

FV-01 Fusarium pathogenomics: old approaches meet new technology in understanding pathogen virulence

Donald Gardiner

Agriculture and Food, CSIRO, Australia

Fusarium pseudograminearum is the predominant wheat infecting Fusarium in Australia, although *F. graminearum* is certainly present. Both species cause Fusarium crown rot and Fusarium head blight. We have a research program focusing on pathogen virulence (FCR and FHB) and host resistance to FCR. A number of projects in the group will be presented including a forward genetics approach to mapping a FHB virulence locus in *F. pseudograminearum*, the recent development of Cas9-mediated genome engineering in *F. graminearum* as well as a brief update on our efforts to clone host resistance loci.

FV-02 Investigating the genome of the Quorn fungus *Fusarium venenatum*

Kim E. Hammond-Kosack

Biointeractions and Crop Protection, Rothamsted Research, United Kingdom

The soil dwelling saprotrophic non-pathogenic fungus *F. venenatum*, routinely used in the commercial fermentation industry, is phylogenetically closely related to the globally important cereal and non-cereal infecting pathogen *F. graminearum*. We have recently sequenced, assembled and annotated the completed *F. venenatum* (strain A3/5) genome, and compare this genome with *F. graminearum* (King et al. (2018) BMC Genomics, resubmitted).

Using shotgun sequencing, a 38,660,329 bp *F. venenatum* (*Fv*) genome was assembled into four chromosomes, and a 78,618 bp mitochondrial genome. In comparison to *F. graminearum* (*Fg*) the predicted gene count of 13,946 was slightly lower. The *Fv* centromeres were found to be 25 % smaller compared to *Fg*. In *Fv* there is an increased abundance of repetitive elements and transposons, but not transposon diversity. On chromosome 3 a major sequence rearrangement was found, but its overall gene content was relatively unchanged. Unlike homothallic *Fg*, heterothallic *Fv* possessed the *MAT1-1* type locus, but lacked the *MAT1-2* locus. The *Fv* genome has the type A trichothecene mycotoxin *TRI5* cluster. From the *Fv* gene set, 786 predicted proteins were considered to be species- specific versus NCBI. The annotated *Fv* genome was predicted to possess more genes coding for hydrolytic enzymes and species-specific genes involved in the breakdown of polysaccharides than *Fg*. A comparison of the *Fg* versus *Fv* proteomes identified 15 putative secondary metabolite gene clusters (SMC), 114 secreted proteins and 39 candidate effectors not found in *Fv*. Five of the *F. graminearum* specific 15 SMCs that were either absent or highly divergent in the *F. venenatum* genome showed interesting patterns of *in planta* expression.

This study has identified differences between the *Fv* and *Fg* genomes that may contribute to contrasting lifestyles, and highlights the repertoire of *Fg* specific candidate genes and SMCs potentially required for pathogenesis.

FV-03 Phytotoxic cyclic lipopeptides produced by *Fusarium graminearum*

Linda Harris[1], Adilah Bahadoor[2], Elizabeth Brauer[1], Whynn Bosnich[1], Danielle Schneiderman[1], Yves Aubin[3], Jeremy Melanson[2], Steve Gleddie[1], Barbara Blackwell[1]

[1] Ottawa Research & Development Centre, Agriculture & Agri-Food Canada, Canada
[2] Measurement Science and Standards, National Research Council Canada, Ottawa, ON, Canada
[3] Centre for Biologics Evaluation, Biologics and Genetic Therapies Directorate, Health Canada, Ottawa, ON, Canada & Department of Chemistry, Carleton University, Ottawa, ON, Canada

Fusarium graminearum is a broad host pathogen causing FHB in diverse cereal crops. Investigating host-preferential gene expression, we identified a nonribosomal peptide synthetase in *F. graminearum* responsible for the biosynthesis of two novel cyclic lipopeptides, named gramillin A and B. A combination of LC-HRMS as well as 1D and 2D-NMR experiments on ^{15}N-enriched compounds were performed to determine the structure of the gramillins. Gramillins are phytotoxins, causing cell death in select plants including maize and Arabidopsis, but not wheat. These labile compounds can linearize under particular conditions and consequently lose their bioactivity. *F. graminearum* mutants unable to produce gramillins are less able to colonize maize silks but just as virulent as wildtype *F. graminearum* on wheat spikes.

FV-04 Genomics and effector characterization of the novel sugarbeet pathogen *Fusarium secorum*

Melvin Bolton[1], Ronnie de Jonge[2], Gary Secor[3]

[1] USDA - ARS, United States of America
[2] Utrecht University, Utrecht, NL
[3] North Dakota State University, Fargo, ND, USA

Sugar beet is an important source of sucrose for human consumption throughout the world. A new disease of sugarbeet, Fusarium yellowing decline, was recently found in the Red River Valley of MN and ND, USA. This disease is caused by a novel pathogen named *Fusarium secorum*, a member of the *Fusarium fujikuroi* species complex. Although symptoms are generally similar to the more typical sugarbeet disease known as Fusarium yellows caused by *F. oxysporum*, *F. secorum* is more aggressive than *F. oxysporum*. Fungal pathogens such as *F. secorum* secrete molecules during infection called 'effectors' that help to establish disease and contribute to pathogenicity to their host. Having a better understanding of the effectors produced by *F. secorum* during infection is necessary to pursue host resistance as a long-term means of managing Fusarium yellowing decline in sugarbeet. Using genome sequencing, transcriptome analysis, and sugarbeet xylem sap mass spectrometry, we identified 30 candidate effectors for *F. secorum*. To functionally characterize these effectors, we developed a transformation system to knock-out genes of interest. Mutant analysis of 11 candidate effectors identified one with a significant role in virulence on sugarbeet. This work provides an important genomic resource not only for *F. secorum* but also for other closely related *Fusarium* spp. and may provide a framework for developing host resistance as a management tool for this important disease.

FV-05 Comparative genomics of *Fusarium graminearum* strains harboring aggressiveness in bread wheat

Francis Fabre[1], Manon Vignassa[2], Hélène Rimbert[3], David Bornaque[4], Caroline Chebance[5], Joerg Bormann[6], Thierry Langin[7], Ludovic Bonhomme[8]

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By targeting host cellular processes, fungal effectors promote growth and spreading of pathogenic fungi in plant tissues. During the wheat/*Fusarium graminearum* (*Fg*) interaction causing the *Fusarium* head blight (FHB) disease, the nature and the function of these fungal molecular components which control plant susceptibility factors, remain largely unknown. Two *Fg* strains (*Fg1* and *Fu10008*) contrasting for their aggressiveness in field trials were sequenced using Sequel technology. *Fg1* and *Fu10008* displayed 11,171 CDS (37,2Mb) and 10,629 CDS (36,1Mb), respectively. The comparative analysis of the predicted proteomes revealed 1,320 specific proteins in Fg1 and 693 in Fu10008, comprising 205 and 81 candidate putative effectors, respectively. A thorough pathotyping of the two *Fg* strains on three wheat genotypes of different susceptibility to FHB was assessed to test their mycotoxin production and their infection dynamics, including symptoms development and fungal biomass progress in point-inoculated spikelets (PI) and in the uninoculated peripheral ones (Up or Dn). Fg1 strain induced systematically the most severe symptoms in the PI, Up and Dn parts of each wheat genotypes. Whatever the infected genotype, Fg1 initiated symptom development 24 h earlier than Fu10008. Spreading of both strains in Up and Dn spikelets appeared preferentially towards the top of the spike as early as 5 days post-inoculation, displaying symptoms very close to those of the PI parts suggesting that they are related to fungal migration in healthy plant tissues. qRT-PCR and mycotoxin analyzes (DON and ZEA) of the different spike zones are currently performed to refine the early stages of the infection process of both strains. This will allow for connecting these *Fg* data to recent work investigating the wheat susceptibility factors (Chetouhi et al., 2016) and will contribute to shape an integrated picture of the molecular events piloting FHB.

Chetouhi, C., Bonhomme, L., Lasserre-Zuber, P., Cambon, F., Pelletier, S., Renou, J.-P., & Langin, T. (2016). Transcriptome dynamics of a susceptible wheat upon *Fusarium* head blight reveals that molecular responses to *Fusarium graminearum* infection fit over the grain development processes. *Functional & Integrative Genomics*, 16(2), 183-201.

FV-06 Distribution and evolution of genes responsible for biosynthesis of mycotoxins in *Fusarium*

Robert H. Proctor

Mycotoxin Prevention and Applied Microbiology, US Department of Agriculture, United States of America

Fusarium secondary metabolites (SMs) include some of the mycotoxins of greatest concern to food and feed safety. In fungi, genes directly involved in synthesis of the same SM are typically located adjacent to one another in gene clusters. To better understand the distribution and evolution of mycotoxin biosynthetic gene clusters in *Fusarium*, we examined genome sequences of ~250 *Fusarium* species and 24 other fungi. By mapping the presence and absence of mycotoxin clusters on a phylogeny inferred from housekeeping gene sequences mined from the genome sequences, we discovered that some mycotoxin clusters are present in fusaria in which they were not expected to occur, and some clusters are absent in species previously reported to produce the corresponding mycotoxins. Our analyses have also provided evidence that horizontal gene transfer (HGT) has contributed to the current distribution of some mycotoxin clusters. However, there is also evidence that phylogenetic signals for some HGT events have been lost over time, because SM biosynthetic genes tend to diverge more quickly than housekeeping genes. Comparison of genome sequences from multiple fungal genera has provided evidence for how *Fusarium* mycotoxin clusters have diverged from homologous clusters in other fungi. For example, the *Fusarium* trichothecene biosynthetic gene (*TRI*) cluster has likely acquired six and lost up to six genes since it diverged from homologs in other genera. In addition, the functions of at least four *Fusarium TRI* genes have changed substantially since they diverged from homologs in other fungi and/or as *Fusarium* homologs have diverged from one another. These findings help explain the structural differences of *Fusarium* mycotoxins and provide tools to better assess the causes of and measures to prevent mycotoxin contamination of crops.

FV-07 Role of the *Fusarium tricinctum* species complex in Fusarium Head Blight disease: virulence and mycotoxin production in durum wheat

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Fusarium head blight (FHB) is a worldwide-occurring cereal disease caused by several *Fusarium* species. They often cause indistinguishable symptoms and are responsible for yield losses, grain quality reduction and mycotoxin accumulation in the grains. The most frequent species causing FHB in Italy are: *F. graminearum* (Schwabe), *F. culmorum* (W. G. Sm.) Sacc., *F. poae* (Peck) Wollenw, *F. avenaceum* (Fr.) Sacc. The pathogenic role of the major FHB agents, *F. graminearum* and *F. culmorum*, is well known but little information is available about some of the less virulent species such as *F. acuminatum* and *F. tricinctum*, belonging to the *Fusarium tricinctum* Species Complex (FTSC), which are generally considered of secondary importance. The aim of the present study was to investigate the pathogenic role of the FTSC in greenhouse and field experiments and to evaluate their mycotoxin production. A population of ten FTSC strains isolated from Italian durum wheat grains was characterized both morphologically and molecularly, by partial sequencing of the *translation elongation factor 1-alpha* (*tef 1α*) gene. In addition, the strain capacity to produce secondary metabolites such as enniatins (A, A1, B, B1) and beauvericin both *in vitro* and *in vivo* was also investigated by HPLC-MS/MS.Artificially inoculated durum wheat heads, both in the greenhouse and in the field, allowed for the first time the detection of specific symptoms caused by *F. tricinctum sensu stricto* strains, which caused necrotic "peacock eye" shaped and necrotic spots at the glume level. *In vitro* beauvericin biosynthesis was absent, while enniatins were produced in significant amounts, independently of the phylogenetic group. In the greenhouse experiments, the enniatin amounts detected in durum wheat heads were higher than those of beauvericin. These results are useful to clarify the role of *F. tricinctum sensu stricto*, but further studies are necessary to better understand the role of each species of the FTSC in different climatic and environmental conditions, since their occurrence is increasing in different European areas.

FV-08 Intraspecific trait variations in *Fusarium langsethiae*

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Fusarium langsethiae, a strong producer of T-2 toxin, is distributed over the entire territory of Europe. In Russia, this species is found throughout the European part of the country. We have maintained a collection of 189 strains of *F. langsethiae* isolated from small grain cereals. Most of them (154 strains) were detected in the territory of Russia, and 35 strains originated from Germany, Greece, Finland, Italy, Latvia, Norway, Sweden, and the UK. Our studies considered the variation in diverse traits of *F. langsethiae*, including traditional phenotypes like size or morphology, sensitivity to abiotic factors (the temperature, fungicides application), and molecular-genetic differences.

langsethiae strains have various features, which indicated their high variability. These strains can be classified into prototrophs and auxotrophs (Gavrilova et al., 2017). Our investigations revealed that 24 strains of *F. langsethiae* were spontaneous auxotrophic mutants for biotin, and 4 strains required thiamine as a growth factor. In our collection of *F. langsethiae* strains, the auxotrophic mutants were detected only among strains originating from northern Europe. In addition, the genetic diversity of *F. langsethiae* has been established, which makes it possible to distinguish two subgroups (I and II) within the species (Konstantinova, Yli-Mattila, 2004). Interestingly, *F. langsethiae* strains belonging to subgroup II were detected only among those strains distributed in the northern part of Europe. Moreover, it was found that all strains of subgroup II stop growing at a temperature 30 °C. The development of different intraspecific responses to habitat is a remarkable property of fungi, conferring their broad adaptive capability and adaptation to the environment.

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FV-09 IAA-Asp amidohydrolase – a putative effector protein of Fusarium species

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Synthesis of tryptamine derived hydroxycinnamic acid amides (HCAAs, such as coumaroyl- and feruloyl-tryptamine) is induced in cereals during *Fusarium* infection. While these HCAAs are considered to be antifungal, *F. graminearum* can overcome initial inhibition by inducing extracellular amidohydrolase activity. *In vitro*, cleavage of the HCAAs leads to release of tryptamine, which is then converted into auxin (IAA, indole-3-acetic acid) at high yield. Massively elevated levels of IAA and the inactivation products IAA-glucoside and IAA-aspartic acid (IAA-Asp) were detected in infected 'Apogee' wheat. We have started to characterize the large family of amidohydrolases of *Fusarium* by heterologous expression in yeast and *E. coli*. While the amidohydrolase(s) responsible for cleavage of the tryptamine-derived HCAAs are still unknown, we were able to confirm that one member of a branch of six amidohydrolases with similarity to plant IAA-amino acid conjugate hydrolases indeed has activity with IAA-Asp. *Fusarium* can hydrolyse both IAA-Ala and IAA-Asp *in vitro*. However, the recombinant enzyme in yeast is only able to hydrolyze IAA-Asp, which is considered to be a dead-end inactivation product of IAA in Arabidopsis because no corresponding hydrolase is known. The highly conserved *Fusarium* amidohydrolase has a predicted signal peptide and a highly positively charged C-terminal extension, which is dispensable for enzymatic activity of the protein when expressed in *E. coli*. We hypothesize that the C-terminus may act as cell penetrating peptide mediating uptake of the protein. Knock out of the amidohydrolase gene did not lead to reduced virulence, which may be due to redundancy. Alternatively, the concentrations of auxin might be sufficiently high for defense suppression even in the absence of the amidohydrolase effector, which is presumably converting back inactivated IAA-Asp to IAA inside the plant cell.

FV-10 *Fusarium graminearum* diversity in Finland, Norway, and Russia

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The 3ADON chemotype of *F. graminearum* predominates in northern Europe, whereas the 15ADON chemotype is predominant in Central and southern Europe. In the present work, molecular chemotypes and variable number tandem repeat (VNTR) markers were used to assess population structure and diversity among *Fusarium graminearum* isolates from four regional locations: Finland and northwestern Russia (F+NWR; N = 40), south Russia including central European Russia (SR; N = 54), Russian Far East (RFE; N = 96), and Norway (NOR; N =106). Trichothecene genotype composition was significantly different across the sampling locations. The 3ADON type was predominant in F+NWR+NOR, the 15ADON type was predominant in SR, and RFE had a balanced composition of these two trichothecene genotypes. The NIV genotype was not observed among the studied collection of *F. graminearum*. Analyses of population structure and relatedness indicated that the *F. graminearum* population in F+NWR are closely related to the NOR population and they can be considered as a unified population. However, significant differentiation was observed between the F+NWR+NOR population and those from the other sampled regions. The F+NWR+NOR population had substantially less genetic diversity than in the other regions. The observed high genetic diversity of populations in the Russian Far East suggests that it may be a source population for the other locations. Combined analyses of isolates from all sampled locations using a Bayesian clustering method indicated that there were two major genetic clusters in the sample, although additional substructuring was observed. Each of the two genetic clusters contained about 50 % of the isolates, and all but nine isolates (3.0%) were assigned to one of the two genetic clusters with high probability ($q \geq 0.8$). Significant ($P < 0.001$) regional differences in genetic population frequencies were observed. Cluster 1 including F+ NWR and most Norwegian isolates had less genetic diversity than cluster 2 including most SR and RFE isolates. Cluster 1 may be more specialized to oats, which is supported by the fact that only the 3ADON genotype has been found in oats in Europe.

FV-11 Genetic variability, mycotoxin profile and metabolomic of *Fusarium proliferatum* pathogen on different host plants.

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Fusarium proliferatum, a member of the *Fusarium fujikuroi* species complex, is a plant-pathogenic fungus able to both colonize a wide range of plant hosts and produce a relevant number of mycotoxins, among which fumonisins are the most important in terms of their toxicity and worldwide occurrence on several crops. *Fusarium proliferatum* is considered as the major fumonisin-producer worldwide, as we have proved for strains isolated from several other hosts such as maize, asparagus, onion, rice and ornamental and date palm. However, strains isolated from fig in Southern Italy and Turkey and from date palm in Iran appear to lack the capacity to produce fumonisins. In the last decade, many studies, devoted to investigate the genetic variability of *F. proliferatum* occurring on different hosts, have been performed. These investigations were done in order to understand the capability of this fungus to adapt in a widely different array of ecological niches, and therefore exerts its pathogenicity on different plants. Along with genetic studies, many efforts have been addressed to evaluate the possible variability of the mycotoxin profile of *F. proliferatum* strains isolated from the different plants, mainly to evaluate a possible specificity in the secondary metabolite production according to the crop origin. Recently, we gained more insight into the gene content and organization of the fumonisin gene (*FUM*) cluster, and data revealed no significant differences in gene occurrence, orientation and genome location among producing and non-producing fumonisin *F. proliferatum* strains. Also, phylogenetic analyses were performed for identifying possible clades within the species based on distinction using DNA barcodes, but only slight differences were provided. Thus, a more in-depth investigation of both the genetic variability and metabolic production of *F. proliferatum*, by generating and analyzing genomes and by metabolic profiling was performed. This research underpinned that a poly-omic approach is a powerful tool for unraveling the genetic, mycotoxin profile variability and pathogenicity of *F. proliferatum* from different host plants,
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EFS14 - EUROPEAN FUSARIUM SEMINAR

APRIL 8 – 11, 2018, TULLN, AUSTRIA

Session 2: Host resistance: mechanisms, genetics, genomics and breeding

Chairs: Hermann BÜRSTMAYR & Fiona DOOHAN

Keynote Lecture

HR-01 Insights into Fusarium-host interactions gained from crop and model species

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While most research into Fusarium head blight (FHB) is understandably focussed on resistance we have become interested in susceptibility. Investigations of wheat, barley and *Brachypodium distachyon* have highlighted the potential involvement of phytohormone signalling in both susceptibility and resistance to FHB. The relationships between particular pathways and susceptibility are not always clear-cut because of the hemi-biotrophic nature of the interaction between *Fusarium graminearum* and wheat. It appears that *F. graminearum* may be exploiting certain pathways to prevent the plant from mounting an effective defence. This view is supported by the finding that isolates of *F. graminearum* are capable of producing some of the core phytohomones and these may be used by the pathogen to force/persuade the plant to maintain growth at the cost of mounting a full defence.

While most wheat varieties lack the ability to prevent the spread of the fungus once it enters the spike, barley varieties have high levels of so-called Type 2 resistance and the fungus is generally restricted to infect individual spikelets. We examined barley chromosome addition/substitution lines of wheat to determine whether the addition of particular barley chromosomes could provide Type 2 resistance. The most potent effect, however, derived from the substitution of chromosome 4D suggesting that the lack of type 2 resistance in wheat is due to the presence of a susceptibility factor(s) rather than the absence of resistance factor(s).

In other studies, we have been examining how particular aspects of plant architecture and morphological structures influence resistance to FHB. The core question is whether any observed effects are due to linkage or pleiotropy. Our evidence suggests that the answers are not always obvious but may highlight avenues for further investigation.

While resistance to FHB and DON mycotoxin accumulation in agronomically adapted varieties can undoubtedly be enhanced by the introduction of resistance from various sources it is also possible that resistance can be increased through the elimination of susceptibility factors. The challenge in both cases is to provide robust FHB resistance without compromising other important agronomic characteristics required by breeders and growers.

HR-02 FHB – resistance genes, genomic hotspots and grain development

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One of the main foci of our team is to identify genes that are of benefit to breeders in terms of marker-assisted selection for FHB resistance, and at the same time elucidating the signaling mechanisms involved in the host-pathogen interaction. We used a functional genomics approach to identify genes up-regulated during the FHB resistance response associated with the barley ‘uzu’ mutation and we thereafter determined than an up-regulated receptor gene and its homologs enhance wheat and barley resistance to FHB disease. Functional genomics of a population segregating for FHB resistance led us to the *TaFROG* gene and further studies validated its role and that of its interacting proteins, SnRK1 and a NAC transcription factor, in disease resistance. Ongoing studies are determining the allelic diversity of candidate FHB resistance genes and their promoters and developing markers for gene selection. Using a bioinformatics approach, we have identified genomic hotspots for FHB resistance, combining functional genomics, genome data and gene validation studies. One of the interesting trends emerging from our studies is that many of the FHB resistance genes we identified also positively affect grain development and new research programmes aim at better understanding the impact of specific genes on yield and the relationship between grain development and FHB resistance.

HR-03 Genomic approaches for increasing Fusarium head blight resistance in durum and bread wheat

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Fusarium head blight (FHB) in cereals is a serious concern in Europe caused by the mycotoxin contamination of food and feed. Although fungicides exist, breeding resistant cultivars is the most effective and environmentally friendly measure against FHB. The availability of high-density SNP arrays lead to the main question how to use genomic approaches most efficiently. For analyzing a specific cross and/or a specific locus a quantitative trait locus (QTL) analysis is still the best option as shown recently by our group for a newly detected reduced height gene (*Rht*) and its impact on FHB resistance in bread wheat. However, in elite wheat populations only small-effect QTL for FHB resistance have been found making marker-assisted selection (MAS) expensive and complex. Genomic prediction (GP) or genomic selection (GS) have been more successful in bread wheat than MAS. We explore now the realized selection gain for a GS approach in elite bread wheat. In contrast, MAS and GP were similar effective in a winter durum wheat population. Phenotypic selection had always the highest genetic progress being, however, also most expensive and slower. In conclusion, substantial progress in FHB resistance can be made by both, phenotypic and genomic selection. Nowadays, the main challenge is to breed FHB-resistant cultivars that are accepted by the market, a scenario that might be resolved best by GS.

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HR-04 Resistance to *Fusarium langsethiae* in Norwegian oats – Safe Oats

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Over the recent decades, the Norwegian cereal industry has had major practical and financial challenges associated with the occurrence of *Fusarium* and mycotoxins in cereal grains. From 2011, payment reductions to farmers were implemented for oat grain lots with high levels of deoxynivalenol (DON). However, according to preliminary results by NIBIO, NMBU and Graminor, certain oat varieties with generally medium or low DON contamination, may contain high levels of HT-2 and T-2-toxins (HT2+T2). These mycotoxins, formed by *Fusarium langsethiae,* are considerably more toxic than DON. Resistance to *F. langsethiae* is not included in the variety screening in Norway.

The SafeOats project was initiated in 2016. An important objective of this project is to develop resistance screening methods in order to facilitate the phase-out of Fusarium-susceptible oat germplasm. Furthermore, SafeOats will give new insight into the biology of *F. langsethiae* and HT2+T2 accumulation in oats, and thus facilitate the choice of relevant control measures. The results from SafeOats will benefit consumers nationally and internationally by providing tools to increase the share of high quality grain into the food and feed industry.

The relative ranking of oat varieties according to *F. graminearum*/DON versus *F. langsethiae*/HT2+T2 content has been explored in field and greenhouse trials. In the greenhouse studies we have analysed the content of Fusarium DNA and mycotoxins in grains of selected oat varieties inoculated at different development stages. Furthermore, we plan to study the transcriptome during *F. langsethiae* infestation of oats. The project will also focus on the occurrence of *F. langsethiae* in oat seeds and possible influence of the fungus on seedling development in a selection of oat varieties.

SafeOats is coordinated by NIBIO and is a collaboration between NIBIO, NMBU, Kimen Seed Laboratory, and the main Norwegian and Swedish breeding companies, Graminor and Lantmännen. Harper Adam University (UK) and Julius Kühn-Institute (Germany) are international collaborators. The project is financed by The Foundation for Research Levy on Agricultural Products/Agricultural Agreement Research Fund/Research Council of Norway with support from the industry partners Graminor, Lantmännen, Felleskjøpet Agri, Felleskjøpet Rogaland & Agder, Fiskå Mølle Moss, Norgesmøllene, Strand Unikorn/Norgesfôr and Kimen Seed Laboratory.

HR-05 Anther extrusion, a passive resistance factor of Fusarium head blight, is associated with semi-dwarfing genes *Rht-B1* and *Rht-D1*

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Fusarium head blight is a complex quantitative trait and interacts with morphological characters of the plant. Typically, taller plants and plants having a high degree of anther extrusion are more resistant. While a rapid and complete anther extrusion has no known negative impact on yield and is even highly desired in hybrid breeding for improved cross-pollination, breeding for increased plant height is inapplicable because medium to short cultivars are required for modern crop management practices. The semi-dwarfing alleles for reduced height *Rht-D1b* and *Rht-B1b* are widely deployed in wheat breeding. Both alleles have similar effects on plant height but studies showed that FHB severity was remarkably higher for *Rht-D1b* than for *Rht-B1b* genotypes (Miedaner & Voss 2008; Srinivasachary et al. 2009). Our work aimed to determine the relationship of *Rht-B1b* and *Rht-D1b* alleles to plant height, anther retention and FHB through testing two populations in field experiments, each segregating for both alleles, allowing a side-by-side comparison in different genetic backgrounds. Although *Rht-B1b* and *Rht-D1b* lead to approximately the same plant height reduction, they differed markedly in their effect on anther extrusion. The stronger negative impact of *Rht-D1b* on FHB resistance can be partially explained by the higher rate of retained anthers. *Rht-B1b* should be preferred in breeding whenever FHB is an important issue (Buerstmayr & Buerstmayr 2016).

Acknowledgements

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HR-06 Classical and genomics-assisted improvement of Fusarium head blight resistance in bread wheat, durum wheat and triticale

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Fusarium head blight (FHB) is a fungal disease of worldwide importance to small grain cereals that may lead to severe losses in both yield and quality. The major issue associated with FHB is the contamination of the crop with mycotoxins such as deoxynivalenol (DON). The development of resistant varieties is the most effective approach for managing FHB.

Genetic mapping of FHB resistance in wheat resulted in the discovery of numerous quantitative trait loci (QTL). The most prominent among these is *Fhb1*, located on chromosome 3B, mainly involved in resistance to fungal spread. Using marker-assisted backcrossing we introgressed *Fhb1* into susceptible durum and triticale elite background and generated mapping populations for genotypic and phenotypic characterisation. In this plant material markers linked with *Fhb1* were clearly associated with FHB severity, confirming its successful implementation into durum wheat and triticale.

Our previous data revealed the association of *Fhb1* with DON resistance and toxin metabolization of DON to DON-3-Glucoside (Lemmens et al. 2005). In order to identify the DON detoxifying gene behind *Fhb1* we have established the genomic sequence of the resistant line CM-82036 (*Fhb1 carrier*) and fine-mapped the QTL interval to 860 kb comprising 28 genes. For functional validation a mutant population (EMS) of CM-82036 was generated. Mutant screening using forward and reverse approaches is ongoing and should highlight candidates for the gene causing DON resistance.

The second major QTL *Qfhs.ifa-5A* is predominantly associated with resistance to initial infection. Fine-mapping of this QTL is hampered due to its location in the low-recombining pericentromeric region of chromosome 5A. Radiation mapping allowed characterizing the QTL region in much higher resolution.

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HR-07 Elucidating Bakanae disease resistance in japonica rice

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Fusarium fujikuroi, causal agent of Bakanae disease, is the main seedborne pathogen on rice. Profiles of defense-related phytohormones and phytoalexins were investigated on two rice cultivars, inoculated or not with *F. fujikuroi* (Siciliano et al., 2015). In the resistant genotype Selenio, the pathogen induced high production of phytoalexins, mainly sakuranetin, and symptoms of Bakanae were not observed. In the susceptible genotype Dorella, the pathogen induced the production of gibberellin and abscisic acid, inhibited jasmonic acid production, phytoalexins were very low and Bakanae symptoms were observed. A RNA-seq transcriptome study was performed (Matic et al., 2016). The basic rice resistance machinery against *F. fujikuroi* involved PR genes, glucanases and peroxidases, since they were upregulated in both the resistant and susceptible cultivars. The specialized and evolved resistance mechanisms in the resistant cultivar included WRKY transcriptional factors, MAPK cascades, and some cytochrome P450 genes. These mechanisms were further confirmed by KEGG identification of Ca2+-dependent protein kinase gene, JASMONATE ZIM-DOMAIN-like genes, CEBiP, CERK1, and MYC2 genes, found only in Selenio. These genes participate in one of the molecular patterns: response to chitin, jasmonic acid biosynthesis, and plant hypersensitive response. When the gibberellin production was controlled, Selenio plants activated the jasmonic acid metabolic pathway. The fungal pathogen in the resistant cultivar acts locally, at lower concentrations, and probably it causes a rice hypersensitive response without any further damage to the plants. A germplasm collection of japonica rice was screened for *F. fujikuroi* resistance, allowing the identification of accessions with high-to-moderate levels of resistance to bakanae (Volante et al., 2017). A GWAS approach uncovered two genomic regions highly associated with the observed phenotypic variation for response to bakanae infection. A search for candidate genes with a putative role in bakanae resistance was conducted considering all the annotated genes and *F. fujikuroi*-related DEGs included in the two genomic regions highlighting several gene functions that could be involved in resistance, thus paving the way to the functional characterization of the resistance loci.

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HR-08 Trichothecene-conjugating UDP-glucosyltransferases: substrate specificities, kinetics and inhibition by culmorin

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Fusarium species infest cereal crops and produce several toxic metabolites, the most problematic being trichothecene-toxins such as deoxynivalenol (DON). Trichothecenes inhibit eukaryotic protein synthesis and are relevant virulence factors promoting disease spread. Plants are able to detoxify trichothecenes by glucosylation. This reaction is catalysed by members of a large and functionally versatile family of UDP-glycosyltransferases (UGTs). Transcriptome analysis of barley upon *Fusarium* infection and DON application led to identification of the first crop plant enzyme, HvUGT13248, capable to glycosylate DON.

We have recombinantly expressed, affinity purified and characterized HvUGT13248 and homologous UGTs from rice (OsUGT79) and *Brachypodium* (Bradi5g03300, Bradi5g02780) regarding their abilities to glucosylate DON and other trichothecenes. Interestingly, the type A trichothecenes HT-2 toxin and T-2 triol are the kinetically preferred substrates in all cases. Furthermore, substitutions at C-4 clearly affect substrate specificities. HvUGT13248 and Bradi5g02780 prefer nivalenol as substrate and are able to conjugate C-4 acetylated trichothecenes to some degree. OsUGT79 and Bradi5g03300 display higher affinity to DON than for nivalenol and are completely inactive with C-4-acetylated derivatives such as T-2 toxin and fusarenon X. While both Bradi5g03300 and Bradi5g02780 glucosylate nivalenol, the latter is inactive with DON. Generation of hybrid proteins and site directed mutagenesis is used to identify the relevant amino-acid differences. We utilized OsUGT79 and HvUGT13248 to glucosylate several trichothecenes (DON, nivalenol, T-2 toxin, HT-2 toxin, T2 triol, neosolaniol, 4,15-diacetoxyscirpenol) in preparative scale. NMR analysis showed that exclusively β-D-glucosides were formed regioselectively at position C3-OH of the molecules.

Culmorin is another *Fusarium* metabolite co-produced with DON, its function is still unknown. We present evidence that culmorin inhibits trichothecene-conjugating UGTs in a cooperative binding mode *in vitro*, with about equal or 10-fold higher binding affinity compared to DON as estimated in the case of OsUGT79 and HvUGT13248, respectively. Yet, it has no effect on a zearalenone-conjugating enzyme. Increased toxicity of DON in the presence of culmorin was observed in a root elongation assay with *Arabidopsis thaliana*. We therefore hypothesize that culmorin may interfere with plant defence by targeting DON-detoxification.

HR-09 Host-induced gene silencing as natural resistance strategy of wheat against Fusarium head blight

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Plant pathogenic fungi such as *Fusarium culmorum* causing fusarium head blight (FHB) are a constant threat to cereals crops. Host-induced gene silencing (HIGS), reported to act against many pathogens, might be used as a tool to control invasive fungi by RNA interference-based mechanisms. Although the mechanism of HIGS is still not well understood it is proposed that fungus-specific small interfering RNAs (siRNAs) are produced by host plants and subsequently transported to the pathogen where they induce silencing of the target genes.

The bidirectional cross-kingdom dsRNA or siRNA trafficking between fungi and plants was already shown e.g. between tomato and *Botrytis cinerea*. Our interest is to explore HIGS as a naturally occurring phenomenon in a cereal crop. To this aim, from a list of natural sRNA molecules accumulating in *Fusarium*–infected barley 10 candidates, which might have harmful effects in *Fusarium,* were chosen for functional validation in wheat and tested by Virus-induced Gene Silencing using the Barley-Stripe Mosaic virus (BSMV) as a HIGS trigger. The corresponding target genes seemed to be important for fungal development and/or virulence, based on their proposed function. HIGS of two out of 10 candidates genes revealed partial protection of wheat and will be further tested. Next steps include sequence analysis of the genomic loci producing the sRNAs, their potential to silence genes in multiple fungal species, and their allelic differences as well as co-localization with disease resistance QTL of wheat.

HR-10 Resistance to *Fusarium verticillioides* and fumonisin accumulation in African maize inbred lines resistant to *Aspergillus flavus* and aflatoxins

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Fusarium ear rot (FER) and Aspergillus ear rot (AER) of maize is caused by *Fusarium verticillioides* and *Aspergillus flavus*, respectively. These mycotoxigenic fungi represent the most common pathogens associated with maize grain in Africa and are of concern to producers as they reduce grain yield and affect quality. *F. verticillioides* and *A. flavus* contaminate maize grain with the mycotoxins fumonisins and aflatoxins, respectively, which has been associated with noxious health implication for humans and livestock. Common resistance mechanisms to FER, AER and associated mycotoxin contamination have been reported. Therefore, ten Kenyan inbred lines resistant to AER and aflatoxin accumulation were evaluated for resistance to FER, *F. verticillioides* colonisation and fumonisin accumulation; and compared to nine South African lines resistant to FER and fumonisin accumulation. Inbred lines were evaluated in field trials, conducted at three localities in South Africa and two localities in Kenya. FER severity was visual assessed, while *F. verticillioides* colonisation was determined as the amount of target DNA quantified by real-time PCR. Fumonisin content was measured by liquid chromatography tandem mass spectrometry. Significant genotype x environment interactions was determined at each location ($P \leq 0.05$). Kenyan inbred CML495 was most resistant to FER and *F. verticillioides* colonisation, and accumulated the lowest concentration of fumonisins across localities. It was, however, not significantly more resistant than Kenyan lines CML264 and CKL05015, and the South African line RO549 W, which also exhibited low FER severity ($\leq$5%), fungal target DNA ($\leq$0.025 ng L^{-1}) and fumonisin levels ($\leq$2.5 mg kg^{-1}). Maize inbred lines resistant to AER and aflatoxin accumulation appear to be promising sources of resistance to *F. verticillioides* and fumonisin contamination and represent the potential to develop varieties with resistance to multiple mycotoxigenic fungi.

HR-11 Fusarium wilt strikes global banana production, again

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Banana is a major staple food, a primary fruit crop in many domestic markets and a commodity supplying global retail stores. However, banana is also a typical orphan crop. Global inputs in research and development are minimal and the number of banana improvement programs is negligible compared to other crops. To complicate matters, many issues in banana production are complex and require multidisciplinary approaches. Traditionally, the sector is inclined to short-term actions and solutions. Hence, there is a threshold to develop and accept long-term strategies for sustainable and fair global production. Therefore, banana production is one of the remaining global monocultures. This comes with a risk and the sector suffers from immense threats, including plant diseases. Panama disease or Fusarium wilt is one of them and represents a key problem that requires short-term action and long-term strategies grounded in scientific data. Initially, we largely focused on projects revolving around genome analyses, genetic diversity and plasticity of the causal agent *Fusarium oxysporum* f.sp. *cubense* (Foc). We built a large global collection of Foc strains, including approximately 200 isolates form the center of origin, the Indonesian archipelago, and characterized them through genotyping-by-sequencing technologies as well as phenotyping assays. Later, we screened hundreds of banana accessions with the Foc strains that caused the previous and the current epidemic. Furthermore, we completed genetic mapping studies to identify genes for resistance to Foc in wild banana germplasm. Short-term research focused on the epidemiology of Foc. This included the development of technologies to diagnose the so-called Tropical Race 4 strain that threatens the global Cavendish-based banana industry. In addition, we investigated the survival of Foc in its natural habitat and under different management practices, including anaerobic soil disinfestation to slow down the epidemic by reducing the inoculum load in the soil. Together with soil and social scientists we placed this information in a multidisciplinary context that calls for action to support a fragile local and global industry that provides food, fruit and employment to millions of people.

EFS14 - EUROPEAN FUSARIUM SEMINAR

APRIL 8 – 11, 2018, TULLN, AUSTRIA

Session 3: Fusarium secondary metabolites and metabolomics of *Fusarium*-host plant interactions

***Chairs*: Rainer SCHUHMACHER & Silvio UHLIG**

Keynote Lecture

MB-01 Gene discovery and editing to enhance resistance in wheat against fusarium head blight

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Fusarium head blight (FHB) is a devastating disease of wheat around the world, causing severe yield and grain quality losses. The fungal pathogen, *Fusarium graminearum*, spores mainly enter through florets at anthesis and spreads to other spikelets through rachis. Several resistance R genes have been identified based on RNA seq and semi-comprehensive metabolome profiling, and the gene resistance functions have been proved. The mechanism of resistance is mainly due to the accumulation of resistance related metabolites, especially in the phenylpropanoid pathway, where the conjugated metabolites are deposited to reinforce the cell walls around the infected area to contain the pathogen spread to the initial infection area. The RR metabolite biosynthetic genes were also found to be regulated by transcription factors that are induced following recognition of invading pathogen by the host receptor genes. Some of these R genes are found to be polymorphic in susceptible commercial cultivars. These polymorphic gene segments are being replaced with functional segments from resistant genotypes, based on genome editing using CRISPR-Cas9 system, to enhance the resistance in commercial cultivars.

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MB-02 *Fusarium langsethiae* – the tough fungus from the North

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F. langsethiae is a toxigenic *Fusarium* species and an important contaminant of small grain cereals, especially oat, in Northern Europe. Morphologically, it shows similarities with *F. poae*, but its mycotoxin profile is similar to that of *F. sporotrichioides*. *F. langsethiae* is thus an important producer of type-A trichothecenes and likely the main causal agent for the occasional high contamination of oats with T-2 and HT-2 toxin in Norway.*langsethiae* is a rather slow-growing species and produces much less aerial mycelium than other *Fusarium* species causing head blight. Nevertheless, *F. langsethiae* persists in the field, and when grown in co-culture with other faster-developing fungi it may resist for some time before it is finally overgrown. This is manifested by formation of an inhibition zone around the *F. langsethiae* colony. In order to study the chemical interaction between *F. langsethiae* and *F. graminearum* in a pilot study, we grew 14 strains of *F. langsethiae* on YES agar together with either a *F. graminearum* 3-acetyl-DON or a 15-acetyl-DON genotype strain. Eleven of the 28 co-cultures showed a distinct inhibition zone around the *F. langsethiae* culture. The strains were genotyped, and all *F. langsethiae* strains forming an inhibition zone belonged to the *F. langsethiae* “subgroup II”. This subgroup harbors a 140 bp deletion within the intergenic spacer (IGS) region of the rDNA cluster, and yields a 650 bp amplification product using primers CNL12/PulvIGSr. The ability of subgroup II strains to form an inhibition zone thus indicates possible differences in metabolite expression between the two subgroups. Agar plugs were sampled from the edges of the *F. langsethiae* cultures facing the inhibition zone and from the corresponding strains growing in pure culture, as well as from *F. graminearum* monocultures. Extracts from the agar plugs were subjected to untargeted analyses based on reversed-phase HPLC and HILIC coupled to HRMS. The resulting data set was evaluated using multivariate analyses (PCA and OPLS-DA). OPLS-DA revealed three *F. langsethiae*-related metabolic features that were significantly upregulated in the co-cultures relative to the corresponding pure cultures (mean peak area ratios co-culture/pure culture 9.5–82). The identity of these metabolites with molecular weights of 276, 321 and 659 Da is still unknown and currently under investigation.

MB-03 Metabolomics of Fusarium head blight: Examining the attack of Fusarium graminearum during infection of two near isogenic wheat lines differing in the resistance QTLs Fhb1 and Qfhs.ifa-5A

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The fungus Fusarium graminearum (*Fg*) is able to infect wheat and other small grain cereals. Major problems arising from this so called Fusarium head blight (FHB) disease comprise yield and quality losses as well as mycotoxin contamination of the affected crop. In the present study a greenhouse experiment has been carried out with the aim to investigate fungal attack of two near isogenic wheat lines differing in the FHB resistance QTLs Fhb1 and Qfhs.ifa-5A. To this end, we have developed an isotope-assisted, untargeted metabolomics approach to distinguish fungal metabolites from those produced by the affected plants.

Within a time course experiment encompassing 3, 6, 12, 24, 36, 48, 72 and 96 hours after infection (hai), about 100 metabolites were determined to be explicitly deriving from *Fg*. For another approximately 250 metabolites, all of which were found to be induced by fungal infection, we were able to classify as being either produced by *Fg* to support infection, the plant in defence or by both interaction partners. Ca. 50% of the fungal metabolites could be annotated including those belonging to the type B trichothecene pathway. Many of the *Fg* assigned metabolites were found to be produced in both genotypes. Towards the end of the time course, most of the detected fungal metabolites were more abundant in the susceptible wheat genotype, lacking the two resistance QTLs. Interestingly, at an earlier stage during infection (about 48 hai) some of these fungal compounds were found to be more abundant in the resistant compared to the susceptible genotype. Our data suggest that in the resistant cultivar, the fungus is either “forced” to produce more and higher amounts of secondary metabolites at an early infection stage or alternatively, higher toxin formation by *Fg* in the resistant wheat genotype may help the plant to induce a quicker/stronger response against the pathogen including the detoxification of mycotoxins via glycosylation. Towards the end of the monitored time window (96 hai) we observed that *Fg* is not only able to produce higher amounts of well-known toxins, but also to produce a much higher number of metabolites in the infected plants, as was particularly observed for the susceptible wheat line.

MB-04 Mycotoxin biosynthesis and central metabolism are two interlinked pathways in *Fusarium graminearum*, as demonstrated by the extensive metabolic changes induced by caffeic acid exposure

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Fusarium graminearum is a major plant pathogen that causes Fusarium Head Blight in wheat and Gibberella Ear Rot in maize and produces type B trichothecene mycotoxins (TCTB) in infected grains. A comprehensive understanding of the molecular and biochemical mechanisms underlying the regulation of TCTB biosynthesis is required for improving or developing novel strategies to efficiently manage the risks posed by F. graminearum. Elucidation of the association of TCTB biosynthesis with other metabolic processes, central and specialized ones was the focus of this study. Combined ^{1}H-NMR and LC-QTOF-MS analyses were used to investigate and compare the exo- and endo- metabolomes of *F. graminearum* grown in toxin-inducing and repressing caffeic acid-conditions. Ninety-five metabolites were putatively or unambiguously identified including 26 primary and 69 specialized metabolites. Our data demonstrated that the inhibition of TCTB production induced by caffeic acid exposure was associated with significant changes in secondary and primary metabolism of F. graminearum although the fungal growth was not affected. The main metabolic changes were an increase in the accumulation of several polyketides including toxic ones, alterations in the tricarboxylic organic acid cycle and modifications in the metabolisms of some amino-acids and sugars. While these findings allow improving the knowledge on the mechanisms that govern the inhibition of TCTB production by caffeic acid, they also demonstrate the interdependence between the biosynthetic pathway of TCTB and several additional metabolic pathways including primary and specialized ones. These outcomes provide further evidence of the multifaceted role of TCTB in the life cycle of *F. graminearum*.

MB-05 How do abiotic factors influence growth, fumonisin biosynthesis and stress response in *Fusarium proliferatum*?

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Fusarium proliferatum is a *major* agricultural *plant pathogen* that synthesizes toxic secondary metabolites, known as mycotoxins or fusariotoxins. Identification of the factors affecting basic metabolism of *F. proliferatum* allows for better understanding of the regulation of mycotoxin biosynthetic pathways during the process of infection and fungal spread across the host plant organism. Here we examined the effect of abiotic factors, namely osmotic stress, salinity and extreme temperatures, on growth and fumonisin biosynthesis by diverse *F. proliferatum* strains.

Four *F. proliferatum* strains originating from garlic, asparagus, maize, and pineapple plants were grown *in vitro* in 100 ml flasks containing 50 ml fumonisin-inducting liquid medium (malt extract 0.5 g/l, yeast extract 1 g/l, mycological peptone 1 g/l, KH_2PO_4 1 g/l, $MgSO_4 \cdot 7H_2O$ 0.3 g/l, KCl 0.3 g/l, $ZnSO_4 \cdot 7H_2O$ 0.05 g/l, $CuSO_4 \cdot 5H_2O$ 0.01 g/l and D-fructose 20 g/l). Abiotic stress factors were applied on the fifth day of culture in different culture variants: temperature of 35°C, sorbitol (0.5-2M), and sodium chloride (0.2-1.5M). Subsequently, samples of liquid media were collected in 2-day intervals and subjected to fumonisin quantification. After 14 days of still incubation, the cultures were transferred into the pre-weighted falcons, the mycelia were centrifuged and freeze-dried for dry weight measurements. In all variants fumonisins B (FBs) were analyzed using UPLC/MS/MS method.

All strains yielded amounts from 42 to 286 mg of dry weight of mycelia after 14 days of culture. Strains biomass showed similar changes under experimental conditions. The sorbitol-induced osmotic stress had the highest biomass increase effect of the three abiotic factors tested (top values of over 4.3-fold increase for 0.5M sorbitol). NaCl was also strong inductor for most of the stains (top values of over 1.4-fold increase for 1.2M NaCl). Culturing of strains at 35°C had a repressing effect on growth; a 1.3-fold decrease in biomass amount was recorded for two strains. Concentrations of FBs varied and depended on type and level of stress factor, strain and time of incubation. Our results demonstrated that abiotic factors might play important roles in the development of diseases caused by *F. proliferatum* by increasing fungal biomass, altering fumonisin synthesis as well as influencing the expression of genes involved in pathogenesis.

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MB-06 Metabolomics to decipher biochemical defence of cereals against *Fusarium* and mycotoxin accumulation

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Fusarium graminearum is the main causal agent of *Gibberella ear rot* (GER) and *Fusarium* Head Blight (FHB), two important fungal diseases affecting maize and wheat, respectively. GER and FHB lead to significant economic losses and serious health issues due to the ability of *F. graminearum* to produce mycotoxins such as type B trichothecenes. Many factors including environmental, agronomic or genetic ones may contribute to high levels of accumulation of mycotoxins in the grains and there is an urgent need to implement efficient and sustainable management strategies to reduce mycotoxin contamination. Fungicides are not far enough efficient to control the mycotoxin risk and, in addition, because of negative effects on human health and environment, their use will be seriously restricted in the near future. To durably solve the problem of mycotoxin accumulation, the breeding of tolerant genotypes is one of the most promising strategies for cereals. However, this objective cannot be achieved without a better understanding of plant resistance mechanisms to both *Fusarium* and mycotoxin accumulation. *Fusarium* resistance depends on the plant ability in preventing initial infection and containing the development of the toxigenic fungi. Resistance to mycotoxin is also related to the capacity of plant tissues in reducing mycotoxin accumulation. This capacity rests on two mechanisms: metabolic transformation of the toxin into less toxic compounds and inhibition of toxin biosynthesis. This last mechanism involves host metabolites able to interfere with mycotoxin biosynthesis. Several non-targeted, and targeted studies have highlighted biochemical rearrangements in both primary and secondary metabolisms, that cereals employ to counter *Fusarium* and its mycotoxin production. These latest metabolomics advances will be detailed in the present communication. In addition, recent studies performed in our research group that aimed to characterize new sources of resistance to both FHB in durum wheat and GER in maize through the combination of targeted and non-targeted metabolomics approaches will be developed. Lastly, the ability of *Fusarium spp.* to metabolize plant metabolites potentially involved in resistance will be discussed.

MB-07 'Omics profiling of *Clonostachys rosea* strain ACM941 highlights unique genetic and metabolic features potentially contributing to its bio-control of *Fusarium graminearum*

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Clonostachys rosea strain ACM941 is patented as a biocontrol agent against the Fusarium Head Blight causative agent *Fusarium graminearum*. Although the molecular and biochemical basis are yet fully resolved, previous studies have suggested that *C. rosea* secretes *Fusarium* growth inhibitors when grown in liquid medium. To gain insight into the genetic and metabolic factors contributing to this, we have established a research program investigating ACM941's responses under a variety of different conditions that mimic natural environments such as treatment with deoxynivalenol (DON) or Fusarium spent media, during plate confrontation and in comparison to other less potent strains of *C. rosea*. We are currently applying transcriptomic (RNAseq) and secreted metabolomic (NMR and MS) profiling methods to identify target genes, gene clusters, pathways and metabolites of interest. To date we have established our RNAseq library revealing 24,112 *C. rosea* unigenes, of which 5,605 and 6,285 were differentially regulated by treatments with DON and *Fusarium* spent media, respectively. More than half of these unigenes (3,167 and 3,121, respectively) were up-regulated notably with annotations suggesting enhancement of putative polyketide (PK) and non-ribosomal peptide (NRP) biosynthesis gene clusters. These findings are being validated by qPCR and metabolomics profiling, while additional RNAseq libraries are being developed to investigate other conditions. We will provide an update on our findings highlighting unique aspects of the genetic and metabolic repertoire utilized by *C. rosea* to control *F. graminearum* growth.

MB-08 *FCRAV2*, a gene involved in significant changes in the physiological and metabolic profiles of *F. culmorum*

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A pathogenicity mutant was identified within the course of an extensive transposon-mediated mutagenesis survey in the wheat pathogen *F. culmorum*. In order to identify new *F. culmorum* gene (*FcRav2*) involved in both FHB and FRR on durum wheat diseases, a *mimp1/impala* double component approach was adopted. the *mimp1* transposable element was integrated within the last exon of the *FcRav2* gene, encoding a hypothetical protein of 318 amino acids containing a ROGDI-like leucine zipper domain including a region of 30 amino acids with leucine repeats every seven or eight residues, known to have a regulatory role. The putative role of *FcRav2* was determined in *F. culmorum* by the analysis of the effect of deletion on the fungal phenotype. Based on the analysis of phenotype, expression and function, the present study revealed that the *FcRav2* has a key role on affecting the resistance to various stresses particularly the ability to grow on different sugars as well as the sensitivity to pH and tebuconazole fungicides. Indeed, ∆*FcRav2* inactivation results in increased sensitivity to osmotic and oxidative stress, and to the sterol biosynthesis inhibitor tebuconazole. In addition, weak acids, such as bromosuccinic acid and b-hydroxy-butyric acid, may represent sources of stress on an impaired deleted mutant. The phenotype of the *F. culmorum* wild-type treated with bafilomycin, which is known to inhibit specifically the V-ATPase complex, reproduced the same morphological features as the *FcRav2* deletion mutants allowing confirmation that at least part of the phenotypic effect of the *FcRav2* mutation can be mimicked by a V-ATPase-inhibiting drug. Furthermore, pathogenicity observed in *F. graminearum* and *F. culmorum* confirms indirectly the putative function of *FcRav2/FgRav2* gene as a controller of vacuolar and endosomal processing. Interestingly, *FcRav2* appears to be deeply involved in a number of morphological and physiological traits. The present work represents the first report in term of the role of *Rav2* in filamentous fungi, and further exploration of the *mimp1* reinsertion mutant library in the future will possibly allow the identification of new genes involved in the fitness of the pathogen.

MB-09 Annotation of *Fusarium graminearum*'s dark matter: Clustering of unknown, structurally similar fungal metabolites during wheat infection by molecular networking

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The fungus *Fusarium graminearum* is a potent pathogen for wheat and other important agricultural crop plants. It causes the devastating *Fusarium* head blight disease thereby contaminating crop yield with mycotoxins and rendering the yield unsafe for animal and human consumption. While there are great efforts in investigating the roles of the currently known major *Fusarium* compounds involved in the disease (e.g. the mycotoxin deoxynivalenol), both structure, function and involvement of many other fungal metabolites during the infection process are currently unknown.

To shed light on these unknown compounds, which are referred to as dark matter in untargeted metabolomics applications, we have carried out a greenhouse experiment involving susceptible and resistant wheat plants (QTLs Fhb1 and Qfhs.ifa-5A) and infected them with *Fusarium graminearum*. All samples have been measured with liquid chromatography-high resolution mass spectrometry and the generated data have been processed with an untargeted data processing approach in order to detect all wheat- and *F. graminearum*-derived metabolites. A sophisticated statistical and biological analysis of these mostly unknown compounds has been carried out to separate fungal and wheat compounds thereby reporting a total of 107 fungal metabolites of which the majority remained unknown without any information about their structure.

Subsequently to the statistical analysis, fragmentation spectra of the fungal metabolites have been acquired.

These fragment spectra of the fungal unknowns have then been organized in a graph with the aim to group compounds with similar MS/MS fragment spectra as compounds with common fragments and/or neutral losses can be assumed to have structural similarities (molecular networking [1]). Clusters of similar MS/MS spectra and therefore tentatively related chemical structures have been correlated with their abundance patterns obtained from the statistical analysis. Results of applied molecular networking approach will be presented for *F. graminearum*'s so far unknown chemical constituents.

[1] Watrous et al. Mass spectral molecular networking of living microbial colonies, PNAS 2012 109 (26) E1743–E1752, doi:10.1073/pnas.1203689109

MB-10 Nucleosome dynamics in the toxin-producing plant pathogen *Fusarium graminearum*

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Nucleosome dynamics are the first level of regulation of all eukaryotic molecular processes that use genomic DNA as a template, including gene expression. Abundant literature on various organism, notably in yeasts, reports that the position of nucleosomes and their relative stabilities are major parameters influencing gene expression. Changes in nucleosome positioning during cell differentiation and growth are commonly observed in eukaryotes in general. In the yeast, *Candida albicans*, such mechanisms were shown to be involved in morphological plasticity that plays a role in virulence. Here, we report the investigation of nucleosome dynamics during the development *in vitro* of the mycotoxin-producing phytopathogen *Fusarium graminearum*. Nucleosome landscapes were investigated using MNase-Assisted Isolation of Nucleosomal Elements coupled to deep sequencing, or MAINE-seq. The general nucleosomal organization extensively described in various organisms appears conserved in *F. graminearum*, with most nucleosomes arrayed and well-positioned relative to start and stop codons of genes. In the details, nucleosome positioning at promoters and gene expression are well correlated. Strong nucleosome rearrangements are observed in culture conditions when significant metabolomics changes are observed, including regarding toxin production. The observed events regard mostly differences in nucleosome stability, sometimes referred to as occupancy. Additional transcriptomics data provide leading information regarding the significance of these observations.

MB-11 Uncovering the priming potential of Z-3-hexenylacetate in the tripartite interaction between wheat, *Fusarium graminearum* and the English grain aphid *Sitobion avenae*

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In wheat ears, *Fusarium graminearum* and the English grain aphid *Sitobion avenae* are important aggressors. Depending on the type of aggressor, complex interacting regulatory signaling networks mount an appropriate defense response. One of these defense responses comprises the production of volatile compounds such as green leaf volatiles (GLVs). In a first approach, we focused on the interaction between *F. graminearum* and wheat. In previous research, we already demonstrated that wheat ears infected with *F. graminearum* emit the GLV Z-3-hexenyl acetate (Z-3-HAC) which primes other wheat ears for enhanced defense against subsequent infection with *F. graminearum*. However, not much is known about the underlying mechanisms. Using an untargeted metabolomics approach, we identified changes in the metabolome of wheat after exposure to Z-3-HAC and/or a subsequent infection with *F. graminearum*. Changes in amino acid metabolism, plant hormones and the plant's glycosylation status upon wheat exposure to GLVs were detected. As introduced above, *F. graminearum* is not the only aggressor on wheat ears. It often co-occurs with English grain aphids. Therefore, we wanted to investigate the (combined) effect of aphids and *F. graminearum* on wheat. Wheat ears infected with *F. graminearum* showed more disease symptoms and higher trichothecene levels when pre-exposed to aphids relative to a sole inoculation with *F. graminearum* which suggested that pre-exposure of wheat ears to aphids affects the plant response against *F. graminearum*. However, it was remarkable that aphids survived well in the presence of trichothecenes. In search for an explanation, we demonstrated for the first time the conversion of deoxynivalenol (DON) to DON-3-glucoside (DON-3G) in insects. This biotransformation mechanism is normally reported in plants. As the DON-3G metabolite was less toxic than DON for aphids, this conversion is considered a detoxification reaction. Finally, we demonstrated that English grain aphids, which evolutionary co-occur with the DON producer *F. graminearum* on wheat, tolerate DON much better, and convert DON to DON-3G more efficiently than pea aphids (*Acyrthosiphon pisum*), the latter being known to feed on legumes which are no host for *F. graminearum*. Using a non-targeted metabolomics approach, we detected on top of DON-3G, DON-diglucosides in aphids as a result of sequential glucosylation reactions. This observation points to a type of co-evolution of *F. graminearum* and grain aphids.

EFS14 - EUROPEAN FUSARIUM SEMINAR

APRIL 8 – 11, 2018, TULLN, AUSTRIA

Session 4: Fusarium mycotoxins - Toxicology, Metabolism and Remediation

Chairs: Dieter MOLL & Siska CROUBELS

Keynote

MT-01 Cocktail effect of trichotechenes on the intestine

Isabelle Oswald

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Fusarium species produce several trichotechenes simulataneously, it is thus important to determine the effect of these mycotoxins when present together. Following the ingestion of mycotoxin-contaminated food, the intestine may be exposed to high concentrations of toxicants. TCT has been shown to impair can affect several intestinal functions such as the nutrient uptake, cell proliferation, the barrier function and the intestinal immune response.

The toxicity of combinations of mycotoxin cannot always be predicted based upon their individual toxicities. Most of the studies concerning the toxicological effect of contaminant have been carried out taking into account only one compound. Multi-exposure may lead to additive, synergistic or antagonist toxic effects. Data on the combined toxic effects of trichotechenes mycotoxins are limited and therefore, the health risk from exposure to a combination of these mycotoxins is incomplete. A synergistic effect between trichothecenes mycotoxins was observed for both their intestinal cytotoxicity and their local inflammatory response. The synergy was already seen at low doses.

In addition, besides mycotoxins, raw material, especially cereals can be contaminated with other contaminant such as heavy metals. The combined exposure to DON and Cadmium was also studied in several human cell lines and interactions were specific to the target organ.

The importance of microbiota in intestinal health is gaining interest. In this aim the interaction between DON and microbiota was investigated. We demonstrated that DON exacerbated the intestinal DNA damages induced by *Escherichia coli* stains producing colibactin raising questions about the synergism between food contaminants and gut microbiota.

All together, these data demonstrated that the simultaneous presence of trichothecene can lead to synergistic interaction and that mycotoxin contamination should be taken in the global context of all food contaminants and the host intestinal microbiota.

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MT-02 Comparative toxicokinetics and oral bioavailability of (emerging) *Fusarium* mycotoxins in pigs and poultry, in relation to species dependent sensitivity and selection of biomarkers for exposure and efficacy testing of mycotoxin detoxifiers

Siska Croubels

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Several *Fusarium* mycotoxins are frequently detected food and feed contaminants on a worldwide basis. Some of these compounds are regulated in food and feed, others are considered more emerging mycotoxins. Besides mycotoxins as such, food and feed may be co-contaminated with modified forms thereof, such as deoxynivalenol-3-β-D-glucoside and T-2 toxin-3α-glucoside.

To adequately assess internal exposure and *in vivo* toxicity, knowledge on the *in vivo* oral bioavailability, disposition and toxicokinetic properties in humans and animals is mandatory. Moreover, comparative insights in differences in absorption, distribution, metabolism and/or excretion (ADME) processes, might reveal species-specific differences in sensitivity towards the toxic effects. Toxicokinetic modeling, i.e. mathematical descriptions of the ADME processes, therefore finds its application in mycotoxin research.

The first goal of this presentation is to provide an overview of major species-specific differences in oral bioavailability and toxicokinetic properties between pigs and poultry. Also specific differences in toxicokinetics between bird species such as broiler chickens and turkeys, will be presented. Selected examples of trichothecenes, zearalenone and enniatins will be given. Secondly, the role of toxicokinetic modeling in the assessment of biomarkers for exposure and efficacy testing of candidate mycotoxin detoxifiers, will be demonstrated.

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MT-03 Marasas' et al. (1984) "Toxigenic *Fusarium S*pecies: Identity and Mycotoxicology" revisited

Kerry O'Donnell[1], Susan McCormick[1], Mark Busman[1], Robert Proctor[1], Gail Doehring[1], Todd Ward[1], David Geiser[2], John Rheeder[3], Hanneke Alberts[3]

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Fusarium ranks as one of the world's most economically destructive and taxonomically challenging genera of mycotoxigenic plant pathogens. Due to the significant threat that fusarial toxins pose to agricultural biosecurity, food safety, and plant and animal health, several compendia have attempted to catalog the diversity of toxigenic fusaria and the toxins they produce. Marasas' et al. (1984) compendium occupies a special place in the *Fusarium* mycotoxicology literature because the authors tested a number of the strains in this treatise for toxins, and because the strains are archived in the South African Agricultural Research Council (ARC; formerly Medical Research Council - MRC) and *Fusarium* Research Center (FRC) Culture Collections. Given the transformative impact genealogical concordance phylogenetic species recognition (GCPSR)-based studies have had on *Fusarium* systematics over the past two decades (reviewed in Aoki et al., 2014), we initiated the present study to: (i) reevaluate the species identity and phylogenetic diversity of 156 MRC strains via GCPSR, (ii) predict mycotoxin potential of any putatively novel *Fusarium* species discovered within the MRC collection by mining their whole genomes for biosynthetic pathways that encode mycotoxins, and (iii) test the MRC strains for mycotoxin production in solid grain cultures and liquid media using high performance liquid chromatography-mass spectrometry and gas chromatography-mass spectrometry.

Acknowledgments

We are pleased to acknowledge the skilled technical assistance of Christine Hodges, Debbie Shane, Nathane Orwig and Amy McGovern in various aspects of this study. Mention of trade names or commercial products in this publication is solely for the purpose of providing specific information and does not imply recommendation or endorsement by the U.S. Department of Agriculture. USDA is an equal opportunity provider and employer.

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MT-04 Impact of Climate Change interacting factors on growth and T-2 and HT-2 production by *Fusarium langsethiae* strains

Carol Verheecke-Vaessen, Alejandro Lopez-prieto, Esther Garcia-cela, Angel Medina, Naresh Magan

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Fusarium langsethiae is an increasing problem in small grain cereals in Northern Europe including the UK/Ireland and Norway. The infection process of *F. langsethiae* is not yet well understood but this can lead to high amounts of T-2/HT-2 contamination, especially in oats. T-2/HT-2 toxins are type A trichothecenes and considered to be the most potent ones which inhibit proteins synthesis. There is little information on whether interacting Climate Change (CC) factors (+2-4°C, 400 vs 1000 ppm CO_2, drought stress) will affect growth and T-2/HT-2 toxin production by strains of *F. langsethiae* on oat-based matrices.

We have thus examined the effect of two way and three-way interacting environmental factors on growth and toxin production by three strains of *F. langsethiae*. Two way interactions between a_w (0.995-0.90) and temperature (10-30°C) showed much shorter lag times and faster growth rates at 30°C. Regarding toxin production, T-2 was produced in higher concentrations (e.g. 67 ppm after 10 days at 0.995 a_w, 25°C) and the ratio of HT-2/T-2 was much lower compared to previous studies.

This data was used to develop two dimensional contour maps of growth including boundary conditions for these two life cycle parameters. In three-way CC related interacting factors of water stress (0.98-0.95 a_w), CO_2 (400 vs 1000 ppm) and temperature (25-30-34°C) we found that there was a significant impact on the ecology of the *F. langsethiae* strains. Overall these results showed that both growth and toxin production are modified by exposure to such CC scenarios. The results are discussed in the context of a better understanding of the impact of CC on *Fusarium* pathogens and the development of appropriate control strategies.

A new RT-qPCR approach has been developed for key *TRI* genes involved in toxin biosynthesis by *F. langsethiae* and have been applied in the CC studies. These data are the first to examine efficacy of CC factors on *F. langsethiae* gene expression.

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MT-05 New regulatory tricks for an old toxin cluster

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Trichothecenes are among the mycotoxins of greatest concern to food and feed safety and are produced by at least two lineages of *Fusarium*: the *F. sambucinum* (FSAMSC) and *F. incarnatum-equiseti* (FIESC) species complexes. Trichothecene biosynthesis begins with the formation of a cyclic sesquiterpene from the isoprenoid farnesyl pyrophosphate (FPP) followed by up to eight oxygenation and four acylation reactions. Most trichothecene biosynthetic genes (*TRI*) are co-regulated and located in a cluster. All *Fusarium TRI* clusters characterized to date include two regulatory genes: *TRI6* and *TRI10*, which encode a C_2H_2 zinc finger and a fungal transcription factor domain, respectively. Functional analysis of FSAMSC members indicate that *TRI6* is required for wild-type expression of all genes involved in synthesis of the trichothecene deoxynivalenol (DON), except *TRI10*, and eight genes involved in FPP synthesis while *TRI10* regulates *TRI6* expression, and to a lesser degree, all eight FPP genes. Here, we characterized a new gene, *TRI21*, located in the *TRI* cluster of FIESC but not FSAMSC, that encodes a Zn_2Cys_6 transcription factor protein. Gene deletion and precursor feeding studies indicate that *TRI21* is required for function of two *TRI* cluster genes (*TRI11* and *TRI13*) responsible for the last two oxygenation reactions required for synthesis of the trichothecene 4,15-diacetoxyscripenol (DAS), but not for function of other cluster genes. RNA-Seq analysis indicated that *TRI21* is required for wild-type expression of 10 of 14 FIESC *TRI* cluster genes as well as six of the eight FPP genes. These results indicate fundamental differences in the regulation of the *TRI* cluster in FIESC and FSAMSC. Furthermore, the presence of a pseudogenized *TRI21* in some members of FSAMSC indicates that the evolution of *TRI* cluster regulation in FSAMSC has included loss of *TRI21* and extension of *TRI6* function.

MT-06 Masked Fusarium mycotoxins: an overview on recent discoveries

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"Masked mycotoxins" are plant metabolites of mycotoxins. Fungal infection on the field results in formation of mycotoxins, which can be partly metabolized by crops. Often, glucose or other polar substances are conjugated to Fusarium mycotoxins, such as deoxynivalenol, HT-2 and T-2 toxin, nivalenol or zearalenone. Several of these metabolites, such as deoxynivalenol-3-glucoside [1] or zearalenone-glucosides [2], have been tested using both *in vitro* [3] and *in vivo* models. For others, their toxicological fate and relevance is still unknown.

So far, tested masked mycotoxins show in common a diminished absorption rate and no intrinsic biological activity. However, all tested compounds can also be (partly) cleaved by the gut microflora and release their toxic aglycons. The European Food Safety Authority [4] therefore pragmatically assumed that all those modified forms should be regarded having the same toxicity as their parent mycotoxins. The aim of this presentation is to highlight recent discoveries regarding masked mycotoxins and to discuss their potential impact on food safety.

Very recently, we conducted a toxicokinetic experiment by orally administering nivalenol as well as nivalenol-3-glucoside to rats. First results, including the first described formation of a nivalenol-glucuronide will be presented.

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MT-07 A foray into the metabolization of deoxynivalenol by different animal species

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The mycotoxin deoxynivalenol (DON), a frequent contaminant of food and feed, is readily metabolized by animals and humans. The manner and extent of metabolization is highly species dependent. Currently, 16 DON metabolites have been identified in animals, including deepoxy-DON (DOM), five DON derived glucuronides, three DOM derived glucuronides, DON-3-sulfate and DOM-3-sulfate as well as three DON- and two DOM-sulfonates. All of the formed metabolites have (presumably) far diminished toxic effects on the animal, highlighting the importance of metabolisation. However, the multitude of metabolites also leads to poor biological recoveries in excreta of DON exposed animals unless all major metabolites are considered in biomarker analysis. Whereas deepoxydation and glucuronidation of DON have been known for over a decade, sulfation and sulfonation have only lately been discovered as main (sulfation in chickens) or substantial (sulfonation in rats) metabolization pathways of DON (Schwartz-Zimmermann *et al.*, 2014; Schwartz-Zimmermann *et al.*, 2015; Wan *et al.*, 2014). In addition, novel DON-, DOM-, iso-DON and iso-DOM glucuronides have only very recently been identified in urine of rats, mice, cows and pigs (Schwartz-Zimmermann *et al.*, 2017). This presentation gives an overview of metabolization of DON in humans and different animal species and highlights how unexpected metabolism pathways and novel metabolites were discovered. In addition, implications of the novel findings for DON biomarker analysis in solid and liquid excreta are discussed.

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MT-08 Impact of co-occurring xenoestrogens on the endocrine disruptive potential of zearalenone and α-zearalenol

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The Fusarium mycotoxin zearalenone (ZEN) is well known for its estrogenic properties. This endocrine disruptive effect is substantially enhanced by reductive metabolism to α-zearalenol (α-ZEL). The advances in mycotoxin analysis in the last decade demonstrated impressively that contaminated food and feed might contain not only single mycotoxins but a spectrum of secondary fungal metabolites, which might occur in different concentrations and ratios. For example, ZEN and ZEL have been found in several food commodities together with the non-regulated weak mycoestrogen alternariol, formed by *Alternaria alternata*. But mycoestrogens are not the sole hormonally active constituents in food of plant origin. Beside other endocrine distruptive contaminants, food of plant origin might contain naturally occurring phytoestrogens e.g. isoflavones in soy-based products or xanthohumol and 8-prenylnaringenin in beer (hops).

We addressed the question whether the presence of non-regulated mycoestrogens or the presence of phytoestrogens affect the endocrine disruptive potency of ZEN and α-ZEL. The estrogenic activity of potential combinations were studied in Ishikawa cells, an endometrial carcinoma cell line, expressing both isoforms of the estrogen receptor. An estrogenic stimulus activates the expression of alkaline phosphatase, which can be quantified photometrically. In this test system, the presence of nanomolar concentrations of alternariol were sufficient to significantly enhance the estrogenic impact of ZEN and α-ZEL. Over a broad range of concentrations and ratios, synergistic effects were observed (Vejdovszky et al., Arch Toxicol (2017), 91: 1447-1460). Combinations with the soy isoflavon genistein showed either synergism or antagonism, depending on the ratio and concentration range. In contrast, the presence of the hop flavonoids 8-prenylnaringenin and xanthohumol decreased the estrogenic impact of ZEN and α-ZEL. These data demonstrate that co-occurring xenoestrogens affect the endocrine disruptive potential of ZEN and α-ZEL. But the interaction is hard to predict and the actual impact cannot be derived by simple addition of the effects of the single compounds. Thus, further studies are needed on the toxicological relevance of respective estrogenic mixtures in food.

MT-09 Microbial degradation of zearalenone by Actinobacteria: Mind the toxicity

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Mycotoxins are a worldwide problem that continues to challenge the whole food chain. An integrated crop protection system is needed to sufficiently manage fungal and mycotoxin contamination. Microbial and enzyme-based detoxification of contaminated food and feed is a promising bioremediation technique and has many advantages over the use of chemical treatments or physical binding agents. Actinobacteria are known to display a wide catabolic versatility for degradation of several environmental aromatic pollutants, including structurally related mycotoxins. In this research, a collection of *Streptomyces sp.* and *Rhodococcus sp.* is screened for the detoxification of zearalenone (ZEN), a non-steroidal estrogen harbouring a benzene ring.

Degradation experiments were carried out in rich growth medium containing 5 mg/L ZEN during 72 hours. Microbial degradation was monitored through HPLC-FLD and confirmed through LC-MS/MS, whereas microbial detoxification is monitored through the BLYES assay which determines the estrogenic properties of possible metabolites.

Screening of the Actinobacteria showed many strains capable of degrading ZEN, with degradation up to 91,20 % under the used experimental conditions. Remarkably, these high degradation rates coincided with both a significant decrease (-97,96%) and increase (+42,4%) in estrogenic properties of the resulting culture medium, highlighting the importance of toxicity monitoring in the screening process. Through a poly-omics approach combined with toxicity screening, we hope to unravel the degradation pathway of ZEN by the most performant strains and to depict which steps are crucial for detoxification.

MT-10 The enzymatic detoxification of deoxynivalenol (DON): identification of the DON epimerization pathway

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The enzymatic detoxification of mycotoxins, including deoxynivalenol (DON), represents a promising approach for addressing the problem of cereal grain contamination. DON has negative consequences on human and animal health; acute exposure leads to emesis associated with a total loss of appetite while long-term exposure leads to reduced weight gain and increased sensitivity to disease. *Devosia mutans* 17-2-E-8 (*Devosia* spp. 17-2-E-8), a bacterial isolate capable of transforming DON to the non-toxic stereoisomer 3-*epi*-deoxynivalenol, has generated interest in the possible mechanism(s) and enzyme(s) involved. An understanding of these details could pave the way for novel strategies to manage this widely-present toxin. It was previously shown that DON epimerization proceeds through a two-step bio-catalysis; DON is transformed to 3-keto-DON and subsequently transformed to 3-epi-DON. Recently, our team has identified the enzymes responsible for this transformation. The first enzyme, DepA is a PQQ-dependent dehydrogenase responsible for the oxidation of DON at the C3 position. It was shown to completely convert DON to 3-keto-DON, a less toxic intermediate in the DON epimerization pathway. The second enzyme in the pathway, DepB, a NADPH dependent dehydrogenase, is capable of transforming 3-keto-DON to 3-*epi*-DON, a compound with at least 50-fold less toxicity than DON. By utilizing these enzymes during the animal feed production process or directly on feed, a feasible strategy for DON mitigation may be developed within the near future.

MT-11 Inactivation of *Fusarium* toxins: Implications for health and performance of pigs

Sven Dänicke

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Compliance with the guiding values for critical *Fusarium* toxin concentrations for complete feedstuffs for pigs ensures that no adverse effects occur under normal production conditions. Deviations from these assumptions might result in adverse effects at mycotoxin levels lower than the corresponding guidance values. Therefore, preventive measures should include both minimization strategies to decrease feedstuff contaminations and all measures to optimize the housing conditions for pigs. Limitations of such a procedure especially arise if on-farm-mixing of farm-harvested cereal grains is practised and unfavourable agronomic conditions have resulted in an area-wide high contamination of the whole cereal harvest. Thus, seek for effective inactivation procedures is an ongoing issue. The *in vivo* inactivation implies that contaminated feed is fed together with feed additives supposed to manage the toxin inactivation during the passage through the digestive tract whereby local effects and absorption shall be minimized. Such potential feed additives are supposed to act mainly through two different mechanisms, namely *via* adsorption by e.g. mineral clays and degradation by enzymes and micro-organisms. In 2009 a new functional group was established by the European Commission (286/2009/EC) and placed within the main group of technological additives and termed as “substances for additional reduction of contamination of mycotoxins: substances that can suppress or reduce the absorption or promote excretion of mycotoxins”. Technical treatments of contaminated feedstuffs are performed prior to feeding. Especially alkaline treatments have been proven to successfully degrade several mycotoxins. However, most of these procedures are expensive or were only lab-tested and require the use of technical equipment normally only available at commercial feed mills. However, a high proportion of total cereal grains for animal feeding are utilized as feed directly at the farm without being processed by a commercial feed mill. Thus, cost effective inactivation procedures which could be used on a farm level would be helpful. Such a potential procedure could include the use of sodium metabisulfite/sulfite when added to wet stored deoxynivalenol (DON) contaminated cereal grains together with propionic acid. Under such conditions DON is converted to DON sulfonates (DONSs) to a large extent. As DONs are less toxic than DON this procedure is considered as inactivation measure.

EFS14 - EUROPEAN FUSARIUM SEMINAR

APRIL 8 – 11, 2018, TULLN, AUSTRIA

Session 5: Integrated Fusarium management (pre/postharvest, epidemiology and forecasting, fungicide resistance, utilization of contaminated batches)

Chairs: Rudolf KRSKA & Antonio LOGRIECO

Keynote Lecture

FM-01 A novel integrated management strategy to tackle mycotoxins along the entire food and feed chain

Rudolf Krska

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There is a pressing need to mobilise and integrate the wealth of knowledge from the international mycotoxin research conducted over the past 25-30 years, and to perform cutting-edge research to close still existing knowledge gaps. The European Commission pointed out that 5 – 10% of global crop production is lost annually due to mycotoxin contamination. The total costs of losses due to mycotoxin contamination, such as reduced yields, food and feed losses, increased costs for inspection and analyses, and others, may easily reach billions of Euros annually, as estimated by Mitchell and colleagues. Mycotoxin-contaminated cereals and derived products, such as dried distillers´ grain solubles (DDGS), in animal feed impact livestock production as well.

To tackle these issues, existing knowledge must be combined with novel findings to bridge gaps on mycotoxin reduction along the food and feed chain. By using mainstream information and communication technology (ICT), losses and waste along the food and feed chain can be prevented, and traceable information to the supply chain and consumers can be provided. This is the mission of MyToolBox (www.mytoolbox.eu), a four year project, which is funded by the European Commission (EC) and launched in March 2016. The overall objective of the MyToolBox project is to develop a series of integrated measures that reduce different kind of losses due to mycotoxin contamination. The project applies a multi-actor and multi-disciplinary approach throughout the food and feed chain with 40% industry participation including five end-users and three well-known institutions from China, who collaborate closely with farmers and stakeholders from the industry.

MyToolBox specifically addresses the most prevalent *Fusarium* mycotoxins (Deoxynivalenol [DON], T-2 toxin, HT-2 toxin, Zearalenon [ZEA] and fumonisins) in wheat, oats, maize and animal feed, ochratoxin A (OTA) in wheat, and aflatoxins in maize, peanuts and dried figs. Besides a field-to-fork approach, MyToolBox also considers safe use options of mycotoxin-contaminated batches to efficiently produce biogas and bioethanol, thus considering alternative use options of otherwise wasted cereal badges. This paper will present latest results achieved within the MyToolBox project with a special focus on the reduction of *Fusarium* mycotoxins.

Keynote Lecture

FM-02 Integrated and innovative MYCOKEY actions for *Fusarium* mycotoxin management in the food and feed chain

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The management of good agricultural practices in the pre-harvest is a key issue for minimizing the risk of *Fusarium* mycotoxin accumulation in the crops before the harvest. Such practices can involve crop rotation, tillage, proper fertilization and fungicide or biological control distribution, variety selection, timely planting and harvests and the control of the insects which often facilitate the *Fusarium* species infection. On the other hand, the reduction of *Fusarium* mycotoxins along the agro-food chains is also highly depending from a correct post-harvest management that must aim firstly at the separation of the infected crop products from the healthy material. Therefore, the use of different tools such as manual sorting or optical sensors is also a crucial point for reducing the level of mycotoxin contamination of a given crop. Moreover, it is extremely important to prevent post-harvest contamination and develop practical and effective post-harvest procedures for mycotoxin reduction in the food and feed supply chains and to provide alternative and safe use options for contaminated batches. An update review will be given on integrated management of pre-and post harvest practices aiming at the minimizing the risk of mycotoxin contamination of the main crops of agro-food importance and main effective solutions, including the development of a MycoKey app, proposed and reached by EU project MycoKey (http://www.mycokey.eu/) .

Keywords: toxigenic fungi, good agricultural practices, sorting, storage

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FM-03 Integrated *Fusarium* management

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There are numerous strategies to control *Fusarium* pathogens resulting in reductions in the mycotoxin contamination of cereals. However, no one strategy is consistently effective at reducing mycotoxins to below legislative limits when disease pressure is high. To minimise the risk of fusarium mycotoxins, it is therefore necessary to combine control strategies. By this integrated approach, *Fusarium* may be targeted at different points throughout its lifecycle resulting in additive or even synergistic effects. The application of multiple strategies to control *Fusarium* conforms to European legislation in the application of “Good Agricultural Practice” (GAP) to reduce fusarium mycotoxins in cereals. The European Union project MyToolBox is compiling GAP based on available literature and building on this knowledge by the study of novel control strategies. These include the use of biopesticides and the control of fusarium inoculum within crop debris via biocontrol agents and via biofumigation. Output from the project will be available within an interactive web-based platform that will include static information on GAP as well as mycotoxin risk prediction tools and safe storage monitoring tools.

The MyToolBox project has received funding from the European Union's Horizon 2020 research and innovation programme under grant agreement No 678012.

FM-04 Healthy and safe oats? Dominant *Fusarium* species, influencing cropping factors, epidemiology and susceptibility

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In contrast to other small-grain cereals, oats represent a low-input crop, including lower susceptibility to foliar diseases and higher tolerance to abiotic stresses. Furthermore, oats possess a unique nutritional profile, containing high amounts of β-glucans as well as phenolics with antioxidant activity. Nevertheless, to be healthy, cereals need also to be free of health threatening mycotoxins.

Between 2013 and 2015, over 300 samples of commercially grown oats were collected in Switzerland along with data on respective cropping factors. Among all *Fusarium* species, *F. poae* (FP) was found to be dominant whereas T-2/HT-2 toxins, produced by *F. langsethiae* (FL), were the major mycotoxins, with an average of 304 µg/kg in hulled grains. Samples from fields with small-grain cereals as previous crop showed the highest concentrations of T-2/HT-2. Higher amounts of nivalenol (NIV) and T-2/HT-2 were detected in samples from fields with reduced tillage compared with samples from ploughed fields (Schöneberg et al. 2018).

To understand the ecological requirements of FP and FL and to predict the occurrence of their mycotoxins, climate chamber experiments were conducted to investigate the influence of different temperatures and high humidity durations on the infection of oats with FP and FL. In addition, to discover the most susceptible period of oats, artificial FL infections were conducted at different growth stages. The climate chamber experiments revealed a higher contamination with NIV and T-2/HT-2 in the 10°C treatments and with a prolonged humidity duration of 12h 99% rH. Inoculation of the oats at early and full anthesis led to a higher FL infection and T-2/HT-2 accumulation in the grains compared with growth stages before anthesis.

Furthermore, seven oat genotypes with different amounts of beta-glucans were grown at three different sites in Switzerland and artificially inoculated with FP or FL during anthesis. High humidity was critical for FL infection whereas FP infected over a wider range of climatic conditions. No significant differences were found between these genotypes, however, the hull-less variety Samuel showed lower NIV and T-2/HT-2 concentrations. Results from the current study are highly valuable to develop recommendations for optimised cropping systems that reduce the risk of mycotoxin contaminated oat grains.

Schöneberg T et. al (2018) European Journal of Agronomy 92, 123-132. https://doi.org/10.1016/j.eja.2017.09.004

FM-05 Synergic potential of pre-milling and milling strategies to minimize mycotoxins and increase fiber content of wheat-based products

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Several clinical studies from around the world show that a daily consumption of whole grain components can reduce the risk of cardiovascular disease and the development of diabetes, in addition also to a reduction in the risk of cancer and mortality.

Producers must commit to guarantee a high level of safety of whole grain food products, while developing creative recipes and food palatable proposals that encourage the public to use them.

In fact, at the same time food based on grains (e.g. pasta, bread, bakery products) account for the largest contribution to mycotoxin exposure in all age classes, in particular due to the mycotoxins produced by *Fusarium spp.*

Within post-harvest interventions devoted to minimize mycotoxins impact with respect to diet intake, one of the first effective actions is to integrate novel down-stream processing approaches.

Cleaning, debranning, peeling, soaking, dry- or wet- fragmentation, air separation etc. are efficient, proven and versatile, cost effective methods for companies to achieve high quality particle size reduction results and to allow more accurate separation of grain tissues with characterized different mycotoxin contamination levels.

This presentation will illustrate how the synergic potential of these pre-milling and milling strategies permit to achieve an accurate separation of grain tissues with different mycotoxin levels, detailing the composition of the internal tissues, minimizing the mycotoxin concentration and increasing contemporaneously the overall fiber contents in raw material selected fractions to be then (re-)combined for final safer wheat products destined for consumers.

In particular, in a close future scenario, bread and pasta could be then added with increased fiber content, up to 10% according to technological requirements.

FM-06 Interactions between *Fusarium* and *Microdochium* species, fungicides and host resistance: consequences for Fusarium head blight disease and mycotoxin production in wheat

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Fusarium head blight (FHB) is an economically important disease of wheat worldwide caused by a complex of *Fusarium* and *Microdochium* species. Epidemics can result in significant losses in grain yield and quality, and mycotoxin accumulation in grain. FHB pathogens naturally co-exist, however host resistance and/or fungicide application can potentially alter species predominance and relationships within the pathogen complex thus influencing disease outcome and mycotoxin accumulation. To determine the consequences of interactions between FHB pathogens, host resistance and chemical control on disease severity and mycotoxin production, a series of glasshouse and field experiments using *Fusarium* spp. or mixed inoculations with *Microdochium* spp. of wheat genotypes with combinations of quantitative trait loci (QTL) located on chromosomes 3B, 5A and 6B and fungicide applications were performed. QTL effects were consistent for deoxynivalenol accumulation and fungal biomass of *F. culmorum*, *M. nivale* and *M. majus* irrespective of inoculum diversity, however the effects of QTL combinations on disease development, DNA of *F. graminearum* and zearalenone accumulation in grain differed under *Fusarium* or mixed inoculations with *Microdochium* spp. All genotypes benefited from fungicide application to reduce deoxynivalenol except the genotype with single QTL on 3B. In field experiments, individual fungicide efficacy against *Fusarium* or *Microdochium* spp. significantly influenced pathogen DNA concentrations and mycotoxin accumulation in grain at harvest, however the environment affected more the direction of the relationship between *Fusarium* and *Microdochium* species and the production of deoxynivalenol and zearalenone in grain. The results show that QTL or fungicides used alone are inconsistent in controlling FHB and mycotoxins associated with *Fusarium* or mixed inoculations with *Microdochium* species and therefore future control programmes should consider integrating resistance with chemical control to achieve greater efficacy against diverse pathogen complexes.

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FM-07 Cross validation of forecasting models for DON in wheat in Europe

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In the course of the H2020 project MyToolbox, a forecasting modelling envelope for the mycotoxin Deoxynivalenol (DON) in wheat in Europe will be developed. Forecasting models for DON in wheat intend to predict, around the flowering period, the toxin in wheat kernels at harvest. Such forecasts can be used in decision making on the application of fungicides, particular storage or processing of batches and/or more intensive checking of batches with a high predicted level of the mycotoxin level.

First, all available forecasting models for DON in wheat in individual European countries, as published in scientific literature, were evaluated. Second, in communication with the authors, the models for the particular countries were retrieved. This resulted into the availability of six different models for four countries/areas in Europe. A cross-validation study of the available models was then performed. This implies that one particular model is applied to data of another country, and the performance of the particular model in the other country is evaluated. The cross-validation will provide insights into the extent to which a forecasting model is applicable in wider regions as the country for which it was initially developed.

Last, a model envelope will be designed in which all available models will be included, for predicting DON in wheat on the European basis.

As a preliminary result from this study, cross validation between Dutch model and the Italian model will be presented at the conference.

FM-08 Integrated Fusarium management of potatoes in the USA

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Fusarium spp. that cause storage dry rot, field wilt and seed decay of potatoes continues to be a major problem throughout the world despite concerted efforts to manage this disease complex. Fusarium is both a seed-borne pathogen and a soil inhabitant where it can persist for many years. Soil inoculum causes wilt and can infect tubers through harvest injuries that can develop into dry rot during prolonged storage. Fusarium from infected seed potatoes in storage spreads during handling, cutting and planting operations and can result in slow seed decay after planting. Seed infection by Fusarium can predispose seed to secondary infection by Pectobacterium resulting in rapid soft rot seed decay and poor stands, so managing Fusarium indirectly manages bacterial soft rot. An integrated approach using crop rotation, clean seed, cultural practices, fungicides and resistant cultivars is important to manage Fusarium diseases of potato. Management relies most heavily on cultural practices and fungicides. Because Fusarium needs an injury to infect potatoes, the most important cultural practice to limit Fusarium is reducing injury. Fusarium response to fungicides is highly variable for many reasons. The QoI class has limited activity, and most of the SDHI class does not have efficacy, but the new carboxamide SDHI compound adepidyn appears to have excellent activity. Resistance to thiabendazole in *Fusarium sambucinum* continues to be widespread, and isolates with resistance to fludioxanil is present in North American states and provinces. Mancozeb has good field activity for managing Fusarium, and two DMI fungicides, difenoconazole and prothioconazole effectively manage Fusarium. Some biological compounds and disinfectants can suppress Fusarium, but are not commonly used for Fusarium management. The importance of managing Fusarium by fungicides to reduce both dry rot and soft rot seed decay in the potato crop will be discussed. Commercial cultivars with resistance to Fusarium is not common.

FM-09 Does zero-tillage increase mycotoxins in oats and barley in cool climate?

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Reduced tillage has been reported to increase mycotoxin producers and mycotoxins, especially DON in European cereal production. *F. graminearum* is the most important DON producer and in Finland oats has the highest infections and DON contaminations. *F. langsethiae* is the main T-2/HT-2 producer infecting oats and barley.

Zero tillage and reduced tillage are increasing due to environmental reasons. The effects of reduced tillage on *Fusarium* infections and mycotoxin contaminations in Finnish cereal grain have been studied during the past ten years. Unlike generally found elsewhere, the results indicate that, zero tillage combined with direct drilling has not increased incidence of deoxynivalenol (DON) producing *Fusarium* species on grains in controlled long term tillage experiments. With spring barley and oats the zero tillage + direct drilling growing system has been compared with the ploughing in autumn + harrowing in spring + sowing in spring. For the analyses, data from 19 field experiments in seven years, 2004 to 2007 and 2011 to 2014, at eight locations were pooled. The field experiments allowed separating the effects of weather and the tillage method. The association of the incidence of *Fusarium* species with the tillage method was studied by computing Kendall rank correlations of incidences with the tillage method at three different development stages: at heading, two to three weeks later and at harvest. Both for barley and oats, the incidence of the DON producers, *Fusarium graminearum* and *F. culmorum,* in grains after heading and at harvest was reduced significantly by employing direct drilling instead of tilling the soil. However, the incidence of *F. langsethiae*, a producer of T-2 and HT-2 toxins, was higher in the direct drilled plots in barley and oats. *F. langsethiae* was more prevalent on barley under direct drilling at heading and two weeks after it, and for oats at all sampling stages including harvested grain. The incidence of *F. sporotrichioides* was reduced by ploughing in barley. *F. poae* was more common under ploughing.

The results show that zero tillage and direct drilling can be used without increased risk of ear infection by *F. graminearum* but it can increase the presence of *F. langsethiae*, especially in oats. As a hypothesis it can be suggested that in the present climate in Finland reduced tillage in oat and barley production does not increase risk for DON but it can be a risk for increased T-2/HT-2 contaminations.

FM-10 Forecasting model for the control of *Fusarium* diseases and the mycotoxin content of wheat and maize

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Winter wheat grain (3 susceptible cultivars from 2008 to 2017) and maize silage (4 susceptible cultivars from 2011 to 2015) from ten locations in Schleswig-Holstein were analyzed for the occurrence of *Fusarium* species and for DON and ZEA content and the effect of weather, cultivar susceptibility, crop rotation and soil cultivation was determined.

The same seven *Fusarium* species were detected in both wheat and maize. The DON and ZEA producers *F. graminearum* and *F. culmorum* dominated in all years, followed by *F. avenaceum* and *F. poae*.

The DNA amounts of *F. graminearum* and *F. culmorum* and DON and ZEA contents of wheat grain differed significantly between individual years and within each year between locations due to weather at flowering. Significant correlations were found between precipitation and temperature at flowering and DON and ZEA contents in wheat grain at harvest. Three multiple regression models were developed for forecasting DON and ZEA contamination of wheat grain at harvest. Their predictive performance was evaluated with new data on weather at flowering and DON and ZEA contents in wheat grain at the same locations. Regressing the observed versus predicted DON and ZEA values showed that 82 % and 83 % of the variation in the observed DON and ZEA values is explained by the variation in the predicted values. The models predicted correctly in 96.9 % of the cases whether the DON content was either lower or higher than the maximum threshold of 1250 µg/kg. For ZEA, the models provided 93.8 % correct predictions. Cultivar-specific reductions of DON and ZEA contents were observed in all years. In the low to moderately susceptible cultivar, DON and ZEA contents were reduced by 53 % and 52 % and in the moderately to high susceptible cultivar by 18 % and 22 % compared to the high susceptible cultivar. Triazole fungicides applied at flowering reduced DON and ZEA contents by 47 % and 50 %, respectively.

Ploughing reduced DON contents in maize silage by 78 % and ZEA contents by 61 % compared to pre-crop maize and conservation tillage. The cultivation of maize with crop rotation and ploughing reduced mycotoxin contents by 87 % (DON) and 65 % (ZEA). More tolerant cultivars had lower mycotoxin contents than more susceptible ones.

Weather conditions at flowering and DON contents of maize grains at harvest significantly correlated at ten Bavarian locations, each with six cultivars, from 2010 to 2014. To exactly determine the period of flowering, the temperature sum model of Rath et al. (2005) was used. Precipitation and temperature during that period correlated with DON content of maize grains (e.g. R^2 = 0.91 for cultivar "Susann"). Based on this correlation, a multiple regression model was developed for the weather-based prediction of DON contents in maize grains. The model predicted DON contents in Austria in 2014 86 % and in 2015 89 % correctly.

EFS14 - EUROPEAN FUSARIUM SEMINAR

APRIL 8 – 11, 2018, TULLN, AUSTRIA

Poster Presentations

PP-01 Innovative strategies with sustainable fungicides of new composition towards mycotoxin fusarium control in cereals

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Several Fusarium species, including the cereal pathogens *F. culmorum* and *F. graminearum*, are the predominant incitants of Fusarium head blight (FHB) and crown and foot rot (CFR) worldwide. Yield and quality losses are caused by FHB, which develops from infection that occurs at anthesis and spreads until grain harvest, causing the grain to be contaminated with mycotoxins. The mycotoxins type B trichothecenes deoxinivalenol (DON), their acetylated derivatives (3-ADON and 15-ADON), and nivalenol (NIV), produced by *Fusarium* spp., are predominant both in Europe and Argentina.

Natural inhibitory compounds and fungicides are mostly extracted from plants and are involved in host resistance response. The CNR-CONICET bilateral project, started on August 2017, aims at identifying, designing, formulating, and characterizing a series of molecules based on the structure of natural/natural-like inhibitors, able to counteract the pathogenic and mycotoxigenic potential of natural populations of Fusarium, or capable to stimulating natural resistance responses by the host plant. The activity has be supported by a preliminary in silico study based on the best interaction between trichodiene synthase TRI5 (enzyme which catalyzes the synthesis of trichodiene, precursor of trichothecenes) and a collection of molecules, mainly phenols and biphenols. This activity has be performed by the CNR UNIT as well as the study and synthesis of natural occurring compounds [1, 2]. In vivo testing has be carried out with selected molecules in order to evaluate their efficacy in reducing FHB symptoms in wheat as well as trichodiene accumulation in the spikes, which has be evaluated using a straightforward method developed by the CONICET UNIT [3]. In vitro evaluation of sustainable carriers to improve bioavailability of molecules with fungicide and/or inhibitory activities has be also investigated.

Preliminary results of the project activity will be presented in the poster communication.

Acknowledgements: *CNR-CONICET Bilateral project 2017-2018* SAC.AD002.001.014. SIPCAM ITALIA S.p.A. for *PhD* Research *Fellowship* (SO).

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PP-02 Isolation and identification of an unusual, modified, D-L-α-cyclic hexapeptide from the filamentous fungus *Fusarium graminearum*

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Filamentous fungi produce many secondary metabolites with diverse biological activities interesting for medical applications. Among these are polyketides and non-ribosomal peptides. The fungus *Fusarium graminearum* holds 15 genes coding for polyketide production and 19 genes coding for non-ribosomal peptide production. So far, only 8 polyketides and 4 non-ribosomal peptides have been assigned to the genes responsible for their production. The yet unknown secondary metabolite products hold the potential to provide new candidates for the drug discovery pipeline serving the increasing demand for novel drugs, including antibiotics and anti-cancer agents.

In this poster I present the detection, isolation and identification of a new fungal secondary metabolite applying RP-HPLC-MS-SPE-NMR and Marfeys method for absolute configuration.

PP-03 Assessment of the effects of the Fusarium mycotoxin Deoxynivalenol in Wistar rats: monitoring of fecal samples by LC-MS/MS and metagenomic analysis of gut microbiota by next-generation sequencing

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The negative influence of mycotoxin consumption in human and animal health has been widely investigated, but their impact in the microbial diversity is more uncertain. In the present research, a metagenomic research that evaluates the response of gut microbial composition to deoxynivalenol (DON) administration at levels easily consumed in the Mediterranean diet is carried out. This evaluation was performed administrating two different DON concentrations to male Wistar rats. Thus, three groups were evaluated: the control group where no mycotoxin was administered, the group P60 and P120 which was administrated with 60 µg kg-1 body weight (bw)/day and 120 µg kg-1 bw/day of DON doses, respectively. Fecal samples were individually collected and the presence of mycotoxins was monitored by solid-liquid extraction (SLE) followed by ultra-high performance liquid chromatography coupled with tandem mass spectrometry detection (UHPLC-(ESI)MS/MS). Two mycotoxins were quantified in fecal samples which were DON and its metabolite deepoxy-deoxynivalenol (DOM-1). Along the seven weeks of treatment, the concentration of DOM-1 present in fecal samples increased gradually. The hypothesis was to relate this increase of DOM-1 concentration with the increasing detoxification capability acquired along the daily DON administration, as it was observed previously in pigs by Eriksen et al [1]. Once these adaptions were observed, the microbial diversity of gut samples analysed by high-throughput sequencing was performed. The microbial relative abundance between groups was compared, and some significant differences were found in the genus taxonomic level. The relative abundance of the Coprococcus genus was slightly higher in the P60 and in the P120 groups as compared with the control group. These differences between the control group and the two groups with DON treatment suggests that the Coprococcus genus could be a factor in the DON detoxification. Further experimentation is needed to evaluate the incidence of DON in long-term exposure and to identify specific bacterial genus or species with specific detoxification capability in order to use them as probiotics.

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PP-04 New compounds isolated from *Fusarium avenaceum* grown on exotic media

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Fusarium avenaceum is a filamentous fungus found to infect a wide range of host plants, including potato tubes, mung beans and apples. Apart from the economic damage that such infections incur, the fungus also produces a wide range of potentially poisonous secondary metabolites. Therefore, it is important to gain more knowledge about the biosynthetic potential of the fungus. After growing *F.avenaceum* on unusual media, we identified a group of new compounds produced by a yet unknown polyketide synthase. Compounds were purified and their structures were elucidated by NMR. Initial screening for bioactivity was performed.

PP-05 Accuracy of genomic selection models for predicting Fusarium head blight resistance in bread wheat

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Canada has been a major force internationally in wheat (*Triticum aestivum* L.*)* production and export representing a value of over $4.5 billion every year. Fusarium head blight (FHB) is one of the most devastating diseases of wheat, reducing both seed yield and end-use quality (due to mycotoxin accumulation), while increasing production costs through the use of fungicides. The disease is caused by a complex of *Fusarium* species of which *F. graminearum* is the most dominant species in Canada. Although multiple approaches have been used to improve genetic resistance to FHB in recent years, the progress has been slow due to the complex inheritance and significant effect of G x E interactions. The major limitation with improving genetic resistance is access to FHB nurseries. Genomic selection (GS) offers an opportunity to improve phenotyping efficiency by selecting individuals of higher breeding value before being tested in field or nurseries, thus speeding up the breeding cycle and increasing the rate of genetic gain (ΔG) per cycle. GS predicts genomic estimated breeding values (GEBVs) of individuals based on genome-wide markers, and one of the factors that affects the prediction is the accuracy of GS models. We optimised parametric and semi-parametric statistical genomics-based GS models for predicting FHB resistance in bread wheat. The models ridge regression-best linear unbiased predictor (rrBLUP), genomic-BLUP (gBLUP), least absolute shrinkage and operator selection (LASSO), Bayesian B (Bayes B), reproducing kernel Hilbert space (RKHS) were trained empirically using 3,581 predictor (single nucleotide polymorphism) variables and a training population of 175 advanced breeding lines of bread wheat. We then assessed the ability of models to predict FHB resistance for a validation set (n= 69 lines), for which genotypic and phenotypic data (FHB index) were also available. LASSO ($\mathbf{r}_{g\hat{g}}$ = 0.462; explained h^2 = 0.456) out-performed all other models in predicting FHB resistance with cross-validation. No discernable difference was observed between the prediction accuracies of gBLUP ($\mathbf{r}_{g\hat{g}}$ = 0.197; explained h^2= 0.292) and rrBLUP ($\mathbf{r}_{g\hat{g}}$ = 0.197; h^2 explained= 0.292). Taken together, results obtained in the study suggest that LASSO captured the linkage disequilibrium between SNP markers and FHB resistance effectively, leading to higher accuracy than RKHS ($\mathbf{r}_{g\hat{g}}$ = 0.222; explained h^2= 0.330), Bayes B ($\mathbf{r}_{g\hat{g}}$ = 0.210; explained h^2= 0.456), rrBLUP and gBLUP.

PP-06 Factors influencing the accumulation of mycotoxins in grain of cereals

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Fusarium head blight (FHB) poses a serious threat to small grain cereal. Infection by *Fusarium* spp. is influenced by factors such as moisture and temperature, cultivar susceptibility and cultivation practice. The best way to prevent or reduce Fusarium infection is to grow cultivars with high levels of disease resistance.

Grain colors are of adaptive importance for wheat and are caused by antioxidant substances. It appears that pigment accumulation could be associated with better resistance to FHB infection. Based on preliminary results of wheat evaluation, it was found that varieties and lines with colored grain showed on average a lower DON content in comparison with conventional grain. Based on the two-year results of the oats evaluation, it was found that after artificial infection, there was a statistically significantly higher accumulation of mycotoxins (DON, T2 and HT2 toxin) in the grain of hulled oats than in naked grains. On the contrary, wheat was repeatedly found to have a lower content of DON in the grain of hulled wheat. More frequent occurrence of lower DON content in grain of the hulled wheat species could be connected with peeling off the hulls before grain processing.

PP-07 NX-toxins: toxicity, stability and detoxification

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Deoxynivalenol (DON) produced by *Fusarium graminearum* is a well-known virulence factor on wheat and most likely on other hosts. A new type A trichothecene mycotoxin, NX-2, was previously reported to be produced by North American isolates of *F. graminearum* (NX-strains; Varga *et al*. (2015) Environ Microbiol, 17: 2588–2600). NX-2 has a similar structure as 3-acetyl-DON, but lacks the keto group at C-8. While the NX-strains produce mainly NX-2 on rice or synthetic media, NX-2 is de-acetylated to NX-3 *in planta*. The toxicity of NX-3 towards eukaryotic ribosomes is comparable to that of DON.

The new NX-chemotype is caused by a different *TRI1* allele. Despite the fact that *TRI1* is unlinked to the core cluster, all natural isolates so far were genotyped as 3-acetyl type. Introduction of the NX-allele of *TRI1* into the 15-ADON producer PH-1 led to formation of NX-4, the C-15 acetylated analoga of NX-2. Like other trichothecenes NX-4 is a potent inhibitor of eukaryotic translation with a similar inhibitory activity as DON. Nevertheless, in wheat infection assays with cultivar Apogee, NX-4 producing strains seem to be less virulent than the isogenic DON producer PH-1, indicating that the plant response might be different. NX3-glucoside (and also NX4-glucoside) can be formed by recombinant UDP-glucosyltransferases *in vitro* and presumably *in planta*.

Recently up to 20% NX-producers were detected in an area of New York State, a much higher frequency than previously reported (Lofgren *et al*. *in press*, Eur J Plant Pathol DOI 10.1007/s10658-017-1314-6). A selective advantage could be that NX-type toxins can escape a second type of detoxification inactivation by Michael-adduct formation with glutathione.

Conversion of NX-3 into a non-toxic rearrangement product (NX3-M1) lacking the epoxide, has been observed during alkaline treatment, conditions mimicking food processing (baking), and even at temperature conditions that might occur in the field and during extended grain storage.

PP-08 Diversity in the tomato wilt fungus, *Fusarium oxysporum* f. sp. *lycopersici*: variation of mutations in *AVR* genes in the field isolates

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Race-cultivar interactions between the tomato wilt fungus, *Fusarium oxysporum* f. sp. *lycopersici* (*Fol*) and tomato (*Solanum lycopersicum*) are determined by the interactions between the avirulence genes (*AVR1*, *AVR2* and *AVR3*) carried by *Fol* and the resistance genes (*I*, *I2*, and *I3*) carried by tomato. The avirulence genes often exist on small conditionally dispensable (CD) chromosomes, which are not necessary for growth but harbor pathogenicity. Mutation in the avirulence genes determines races (race 1, 2 and 3). A collection of isolates of races 2 and 3 of *Fol* was made from the fields in Japan from 2000 to 2017. In the nine isolates analyzed, we found 5 kinds of mutations (*avr1*^{null}, *avr1*^{th380}, *avr1*^{th685}, *avr1*$^{tf\text{-}380}$, and *avr1*$^{5'\text{-}ND}$) in *AVR1*, and 4 kinds (*avr2*^{null}, *avr2*^{G121A}, *avr2*^{T122A}, and *avr2*^{C146T}) in *AVR2*. None of the nine isolates had the same combination of mutations in *AVR1* and *AVR2*, even some of them are phylogenetically very close. These suggest that race differentiation of *Fol* in the field is more diversified than expected. This possibly relates to that the avirulence genes exist on CD chromosomes, and we will discuss the karyotypes of the field isolates, too.

PP-09 Enzymatic production of 3-lactyl-deoxynivalenol and 3-propionyl-deoxynivalenol

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In 1982 a then new mycotoxin “responsible for vomiting in humans and swine” was isolated from naturally contaminated barley in China. Structural characterization of the substance initially called CDB2 indicated that this compound was 3-lactyloxy-7,15-dihydroxy-12,13-epoxy-trichothec-9-en-8-one or 3-lactyl-deoxynivalenol (3-lactyl-DON) [1]. This proposed structure was confirmed by NMR spectroscopy [2]. Later a group from Brazil seemingly isolated this compound either from inoculated wheat heads or from Fusarium culture material (no details given), and reported the GC-MS fragmentation pattern of the trimethylsilyl-ether derivative [3]. Treatment of contaminated grain with lactic acid bacteria or even lactic acid is an increasingly suggested treatment method to reduce the DON content. We wondered how 3-lactyl-DON compound might be generated. It is still unclear whether 3-lactyl-DON is a plant-derived metabolite or produced by Fusarium. We started testing the obvious and simple hypothesis, that the Tri101 acetyltransferase of *Fusarium graminearum* can also utilize lactyl-CoA instead of acetyl-CoA to acylate the C3-OH of DON. Lactate is normally utilized via lactate dehydrogenase and pyruvate-dehydrogenase and no lactyl-CoA is formed. Yet, it has been reported [3] that *Escherichia coli* grown on propionic acid as sole carbon source induces a propionyl-CoA synthase (*prpE*) that can also act as lactyl-CoA synthase (ATP + CoA + lactate = lactyl-CoA + AMP + PPi). We have therefore prepared and combined recombinant (6xHis-tagged) *prpE* from *E. coli* and FgTri101 protein *in vitro* with the substrates. Enzymatic synthesis of both 3-lactyl-DON and 3-propionyl-DON was possible, which are useful analytical standards to address the question under which environmental conditions Fusarium produces these derivatives, and which gene(s) are required to provide the CoA-derivatives.

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4. Megraw RE, Reeves HC, Ajl SJ. (1965) Formation of lactyl-coenzyme A and pyruvyl-coenzyme A from lactic acid by Escherichia coli. J Bacteriol. 90(4):984-8.

PP-10 Incidence of Fumonisin B_1 in durum wheat grains in Serbia

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A wide variety of commodities in the world have been analysed for fumonisins contamination. However, they have mostly been reported in maize and maize-based foods and feeds. Little research has been carried out on the natural contamination of wheat with these mycotoxins, although *F. verticillioides* (Sacc.) Nirenberg isolated from wheat has been reported to produce high concentrations of fumonisins, and fumonisin-producing isolates of genus *Fusarium* were reported to be as well-adapted to grow on wheat as on maize. Durum wheat (*Triticum turgidum* L.) is an important small grain cereal, used for human consumption and it’s mainly used for the elaboration of pasta in Serbia.

Survey was carried out to determine Fumonisin B_1 contamination in 46 samples of durum wheat (*Triticum turgidum* L.) samples collected during three harvest seasons (2015, 2016 and 2017) from 12 different locations in Serbia. The primary samples were homogenised and quartered to obtain a 1 kg sample for laboratory analyses. Concentration of FB_1 were analysed with the Enzyme-Linked Immunosorbent Assay (ELISA). Positive results were found in 50.7%, 31.6% and 10.1% samples in 2015, 2016 and 2017, respectively. FB_1 concentration varied from 750 to 1300 µg kg^{-1}, and the mean levels recorded were: 1305.7 µg kg^{-1}in 2015; 962.7 µg kg^{-1} in 2016 and 768.2 µg kg^{-1} in 2017.

PP-11 *Fusarium graminearum* might be able to downregulate plant ethylene signalling and to deplete L-cysteine of the host plant with cysteine-racemase

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Fusarium graminearum is a prominent mycotoxin producing fungus with a broad range of hosts. While it was proposed that *Fusarium graminearum* is able to exploit ethylene signalling to increase host susceptibility [1], most publications propose a role of ethylene production in plant defense and increased plant resistance. The bioinformatic analysis of the *F. graminearum* genome revealed two genes annotated as ACC deaminases (ACD). ACDs degrade the immediate precursor of ethylene, ACC (1-aminocyclopropane-1-carboxylic acid), releasing NH_3 and 2-oxobutyrate. One of these candidate genes indeed showed ACD activity with a K_m value of 3.3 mM, which is well in the range of bacterial enzymes reported to decrease “stress ethylene” [2]. The other candidate gene turned out to be a D-cysteine desulfhydrase specifically converting D-cysteine into pyruvate and H_2S. The K_m value of the D-cysteine desulfhydrase was determined to be 18.1 mM and V_{max} 5.5 µmol mg^{-1} min^{-1}. The gaseous signalling molecule H_2S has recently been reported to antagonize the effect of ethylene [3]. Yet, since D-cysteine is not normally present in relevant amounts *in planta*, *F. graminearum* should be able to convert L- into D-cysteine. Evidence for a still unknown D/L-cysteine racemase activity in protein extracts was obtained. Potentially *Fusarium* might therefore be able to deplete cysteine and affect glutathione levels in the host besides affecting ethylene signalling. However, testing knockout strains (single ACD or cys-desulfhydrase and double mutants) on the highly susceptible ‘Apogee’ wheat revealed no significant changes in *Fusarium* virulence.

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PP-12 Killing or modulating. Selection strategies of Streptomycetes limiting trichothecene type B production in cereals affected by Fusarium.

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Streptomycetes are an important group of soil inhabiting bacteria, major producers of secondary metabolites that find application in medicine and agriculture. Streptomycetes can also interact with plants behaving as endophytes. Their diversity represents an interesting source for developing new biological control agents. Selection of antagonists able to limit toxin production can focus on the ability to modulate the toxin pathway or on the ability to limit fungal growth by impairing fungal development and consequently toxin production.

Two phenotyping methods for selection based on fungal growth and trichothecene type B inhibition in the wheat-fusarium pathosystem have been employed in order to identify effective antagonistic strains within a 1500 streptomycetes historical collection obtained from plants and soil worldwide.

From our preliminary screening it is evident that control of toxins production can be achieved more effectively and consistently by direct inhibition of fungal growth. Strains employed as potential modulators of toxin production when fungal growth is already established were not able to provide consistent and significant protection against toxin accumulation.

By integrating the data from the two previously mentioned screening methods, strains of *Streptomyces* which showed promising results both *in vitro* and *in vivo* (grain storage simulation) were selected for deciphering their mode of action against fusaria.

PP-13 Cropping factors which increase the risk of *Fusarium* infection and mycotoxin contamination in Swiss silage and grain maize

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Maize is frequently infected by a complex of *Fusarium* species, leading to mycotoxins, which represent a severe threat to animal health. To assess the risk of infection and mycotoxin contamination in silage (SM) and grain maize (GM), multi-year surveys were conducted (SM: 2010-14; GM: 2008-10). Harvest samples from commercial SM fields (n=169) of two cantons and GM fields (n=288) of 16 cantons, together with information on the cropping history, were collected. Despite a high *Fusarium* species variability in SM and GM as well as a strong year effect on their occurrence, deoxynivalenol (DON) was the most prevalent mycotoxin. An alarmingly high proportion of SM (68%) and GM (51%) samples exceeded the European guidance value for DON in swine feed (0.9 mg kg^{-1}). The mean content of DON was equal for SM (2.2 mg kg^{-1}) and GM (2.5 mg kg^{-1}), however, the mean zearalenone level was higher in SM (0.5 mg kg^{-1}) than in GM (0.3 mg kg^{-1}). In both surveys, the average concentrations of fumonisins were substantially lower (SM: 0.6; GM: 0.3 mg kg^{-1}) and mainly found in samples from the South. As DON was the dominant mycotoxin in SM and GM samples and *F. graminearum* (FG) the predominant species in GM, we focussed on identifying cropping factors influencing these two parameters. In SM and GM, samples from fields with reduced tillage showed a significantly higher mean DON content (SM: 2.9; GM: 3.2 mg kg^{-1}) compared with samples from ploughed fields (SM: 1.5; GM: 2.1 mg kg^{-1}), irrespective of the previous crop. In addition, late harvest dates resulted in higher levels of FG and DON compared with those from earlier harvest dates. In the GM survey, maize grains from mid-late maturing hybrids had higher mean DON contents (3.8 mg kg^{-1}) than grains from early (2.3 mg kg^{-1}) or mid-early (1.8 mg kg^{-1}) maturing hybrids. Cereals or maize as pre-previous crops significantly increased the DON content in samples from ploughed fields, compared with samples from fields with other pre-previous crops. Since effects of hybrids and pre-crops might have been masked by the highly variable cropping conditions in our first dataset, we examined GM samples from Agroscope variety trials with eight different hybrids (2011-2013). The analyses demonstrated that samples from fields with previous crop maize resulted in increased FG infection and DON contents. In addition, we revealed substantial differences in hybrids irrespective of the maturity class, with DON mean values ranging from 0.6 to 1.9 mg kg^{-1}.

PP-14 Dynamic of the genome in *Fusarium graminearum*: insights on evolution of pathogenicity related traits

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The ascomycete *Fusarium graminearum* is a devastating pathogen of cereal crops, and can contaminate food and feed with harmful mycotoxins. Previous studies suggest a high-adaptive potential of this pathogen, illustrated by an increase of pathogenicity and resistance to fungicides in some populations. The foundation of this ability to evolve remains largely unknown. The pathosystem *F. graminearum*/wheat does not fit to the gene-for-gene relationship and is a model to study the quantitative host-pathogen interaction. Deciphering the genetic architecture of the pathogenicity in *F. graminearum*, in terms of number of loci/genes, heredity, and evolvability, is a key prerequisite to understand how the fungus adapts to its environment. Through a combination of classical quantitative genetic and genomic approaches, we provide new insights on the evolutive potential of this pathogen. The construction of a high-density genetic linkage map gave a detailed picture of the recombination landscape. The recombination rate had a strong positive correlation with nucleotide diversity, and recombinant active regions were enriched for genes with a putative role in host-pathogen interaction. The quantitative trait locus (QTLs) involved in pathogenicity related traits, such as disease severity or toxin production, obtained in our studies or described in literature, were mainly found in the most dynamic part of the genome. Consequences for the evolutive potential of this major pathogen will be discussed.

PP-15 Evaluation of fusarium head blight resistance by digital pictures analysis for triticale populations with *Fhb1* introgression

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Fusarium Head Blight (FHB) is considered worldwide as a cereal disease of economic importance. Though genetic resistance is now well understood for bread wheat, relatively little research has been done for triticale. We propose here to characterize the FHB genetic resistance of triticale, and to evaluate the efficiency of resistance QTL from wheat in a triticale background. To this end a FHB resistant triticale line G8.06, has been produced after a back-cross of the highly FHB-resistant wheat line CM-82036 and the triticale cultivar 'Santop'. This line carries the two FHB resistance QTL from wheat: *Fhb1* and *Qfhs.ifa-5A*. It has been used as resistant parent in three crosses: 1) x 'Tulus'; 2) x (F1: 'Agostino'x'Grenado'); 3) x 'El Paso'. The three populations were advanced to the F4 generation. RILs were evaluated in field experiments for FHB resistance in four seasons using spray inoculation and genotyped with SSR as well as genotyping-by-sequencing markers (DArTseq). Phenotyping was performed on two phases. First, we visually evaluated the percentage of infected spikelet to determine the area under the disease progress curve (AUDPC). Secondly, we digitally evaluated the whitened kernel surface (WKS) using a newly developed image analysis program. Four QTL with major effect on the resistance were identified on chromosomes 2B, 3B, 5R and 7A. The QTL on 3B and on 5R were detected at the *Fhb1* and *Ddw1* intervals, respectively, no QTL was detected in the *Qfhs.ifa-5A* interval. The four major effects QTL were detectable with both variables, AUDPC and WKS. So far, it is the first time that an efficient introgression of *Fhb1* is documented in triticale. It is a significant step forward for enhancing FHB resistance in this crop. The impact of *Ddw1* on plant height and on FHB resistance is confirmed. These QTL, and the other we detected, are well characterized on our maps with SNP markers, and could be easily used through a marker assisted selection program. The new notation criterion WKS, based on picture analysis, is a promising tool for breeders and researchers. Fast and easy to use, it presents good correlation with mycotoxin content (DON). A very economical way to evaluate mycotoxin content and to enable the large scale scoring and ranking needed to select resistant cereal varieties for the future.

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PP-16 ADEPIDYN^{TM}: A new broad spectrum foliar fungicide for multiple crops

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ADEPIDYN^{TM} is the new carboxamide fungicide discovered by Syngenta, which is the first member of a new chemical group among the succinate dehydrogenase inhibitor (SDHI) fungicides, the N-methoxy-(phenyl-ethyl)-pyrazole-carboxamides. The ISO common name for ADEPIDYN^{TM} is pydiflumetofen. ADEPIDYN^{TM} was selected based on its particular strength against *Fusarium* species, especially Fusarium Head Blight of cereal crops. ADEPIDYN^{TM} possess high binding properties to the complex II enzyme. It sets a new performance standard against many leaf spots (such *as Septoria tritici*, *Cercospora arachidicola*, *Alternaria solani* and *Venturia inaequalis*) in various crops (such as wheat, peanuts, potatoes and apples). Further, it provides excellent control of powdery mildews across multiple crops. In addition, ADEPIDYN^{TM} is highly active on difficult to control diseases such as *Botrytis cinerea, Sclerotinia sclerotiorum, Corynespora cassiicola* and *Fusarium species* that cause severe damage on important crops. This spectrum makes ADEPIDYN^{TM} the ideal fungicide to complement the Syngenta fungicide portfolio and to introduce a new mode of action for *Fusarium* control. An ADEPIDYN ^{TM} based product has successfully been introduced in Argentina end of 2016. A range of products based on ADEPIDYN ^{TM} will be offered to address the most pressing needs for disease control across a range of crops around the world. In the poster, we will provide an introduction to the molecule and provide selected examples from *Fusarium* related assays in the lab, greenhouse and field.

PP-17 *Fusarium* spp. associated with 'bud rot' of oil palm in San Lorenzo, Esmeraldas province, Ecuador

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African oil palm (*Elaeis guineensis* Jacq.) was introduced in Ecuador in 1953. Ecuador is the second largest exporter country worldwide. The Provinces with major production of oil palms are: Esmeraldas, Los Ríos, Santo Domingo de los Tsáchilas, Pichincha, Sucumbíos and Francisco de Orellana. The most important disease of oil palm in Ecuador is 'bud rot' also called *pudrición del cogollo*" (PC) in Spanish. So far 23 000 ha of oil palm planted areas are affected by this disease. Several fungi are associated with 'bud rot' worldwide, but none has been confirmed as the causal agent. To better understand the etiology of the disease, several samples were collected from four private enterprises (Palesema, Palmeras De Los Andes, Palpailón, Energy&Palm) that are representative for San Lorenzo Canton. Different portions of five to eight-year-old plants were selected and analyzed. At the same time, samples from nursery plants of two private oil palm enterprises (Alespalma and Palmeras de Los Andes) were collected. *Fusarium* species of all isolates were determined by morphological and molecular characterization. A total of 70 isolates belonging to *F. oxysporum*, *F. proliferatum*, *F. sacchari*, *F. solani*, *FIESC*, *F. mangiferae*, were obtained from field and nursery samples. Twenty-five strains (36%) were isolated from the roots and 45 strains (64%) from the spear and meristem. Only two species were isolated from the basal portion, *F. solani* (52%) and *F. oxysporum* (48%), while the main species isolated from spear and meristem were *F. oxysporum* (33%), *F. sacchari* (31%), and *F. proliferatum* (24%). Pathogenicity tests were conducted in the open field on four-month-old oil palm plants. One isolate of each identified *Fusarium* species was tested by single or combined inoculation on 4 month-old nursery plants. In the pathogenicity test series complete spear rot was caused only by *F. oxysporum* and *F. proliferatum*, but no symptoms of wilt were observed in any of the inoculated plants. *F. sacchari* caused general chlorosis of young leaves and necrotic lesions in spears, whereas *F. solani* caused disperse necrotic points in spears. Our study suggests that the main species contributing to cause the 'bud rot' on oil palm are *F. proliferatum*, *F. oxysporum*, and *F. sacchari*.

PP-18 Identification of candidate genes present on 7EL chromosome of *Thinopyrum elongatum* and responsible for FHB resistance in *Triticum aestivum*

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Thinopyrum elongatum, a close wild relative of wheat, carries genetic resistance to fusarium head blight (FHB) on the long arm of its chromosome 7E (7EL). By cross referencing genomic sequence databases for wheat chromosome 7 and *Th. elongatum* 7EL, a series of 7EL- and 7DL- specific markers have previously been designed. Using the current wheat 7D pseudomolecule information, a genetic order for the 7EL markers is proposed. In parallel to the primer design, a Chinese Spring (CS) ph1b line was crossed with a CS-7E(7D) substitution line to facilitate introgression of 7E fragments from *Thinopyrum* into the 7D chromosome of wheat. Progeny from BC_1F_5 and BC_1F_7 families have been genotyped and phenotyped to determine the 7EL region of introgression and the FHB response of the plant material. Genotyping was performed through PCR assays using the 7EL- and 7DL- specific markers. Phenotyping was performed by inoculating wheat heads with *F. graminearum* and measuring disease progression at 7 and 14 days post inoculation (dpi). In the progeny, FHB resistance is carried by the 7EL chromosome, in a small region of approximately 41-42 million base pairs, based on synteny with 7DL. Further work is being done to identify and test potential candidate genes present in this introgressed region.

PP-19 Phenamacril: a potent, but reversible, inhibitor of *Fusarium* class I myosin

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Fungicide application remains a vital component in the management of *Fusarium*-induced plant infections, but environmental concerns and resistance toward some of the commonly used fungicides has spurred efforts into developing new specific fungicides. Phenamacril, a cyanoacrylate fungicide, represents a novel class of seemingly *Fusarium*-specific and environmentally benign fungicides that could hold great potential in our future efforts toward minimizing the occurrence and severity of *Fusarium*-induced plant infections. Phenamacril targets the motor domain of *Fusarium* class I myosin [1-2], an ubiquitous molecular motor which facilitates numerous ATP-driven and actin-associated processes within the cell. The myosin superfamiliy is divided into approximately 20 different classes (classes I, II, V and XVII are present in fungi), each potentially having several isoforms. While the task-specific C-terminal tail domains are highly variable in size and function, the N-terminal motor domains remain highly conserved, both structurally and functionally. Interestingly, Phenamacril-resistance has been shown to correlate with single amino acid mutations in a highly conserved region of the motor domain [1], a region involved in the allosteric communication between the actin- and nucleotide binding-site [3].

To gain further insight into the specificity, the inhibitory potential and nature of the Phenamacril-mediated inhibition of *Fusarium* myosin, we overexpressed *Fusarium* class I myosins in Sf9 insect cells. Through a combination of NADH-coupled and *in vitro* motility assays, we demonstrated that Phenamacril is a very potent but functionally reversible inhibitor of *Fusarium* class I myosin. Similarly, we demonstrated that Phenamacril had no or only minimal inhibitory effect on human myosin 1c, *Dictyostelium discoideum* class I and II myosins, but that the motor domain of the Phenamacril-resistant *F. solani* f. sp. *pisi* was highly inhibited by Phenamacril *in vitro*. This suggests that despite of the high degree of structural conservation in the myosin super-family, subtle differences render Phenamacril a highly specific and potent reversible inhibitor of *Fusarium* class I myosin.

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PP-20 Ethylene biosynthesis by *Fusarium graminearum* in vitro.

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Certain plant pathogens possess the ability to exploit plant hormones to aid in their infection strategies. Different *Fusarium* species have the ability to synthesise a range of plant hormones. Previous work has identified that the plant hormone Ethylene (ET) is a susceptibility factor in *Arabidopsis thaliana* and wheat (*Triticum aestivum*) towards *Fusarium graminearum*. Here we attempt to identify whether *F. graminearum* has the ability to synthesise its own ET and whether ET acts as a virulence factor.

The *F. graminearum* isolate PH1 was grown in defined liquid media with a range of prerequisite substrates necessary for ethylene production in fungi and/or microorganisms. The headspace from the sealed cultures was extracted and analysed using gas chromatography with a flame ionisation detector. Results showed that *F. graminearum* PH1 has the biosynthetic capacity to produce ET in-vitro with both methionine and α-Keto-γ-methylthiobutyricacid (KMBA) as primary and secondary precursors respectively. Furthermore, we showed that ET production is conserved between multiple *F. graminearum* and *Fusarium culmorum* isolates regardless of chemotype. Expression of candidate genes in this pathway were compared using RT-qPCR. We found genetic redundancy for the conversion of methionine to KMBA, and one candidate gene for the conversion of KMBA to ET. RNA-seq analysis will be used to identify any other genes highly upregulated in cultures producing ET. In future work, we will knockout key ET producing genes and determine whether ET production acts as a virulence factor for *F. graminearum* on *Brachypodium distachyon* and *Triticum aestivum*.

PP-21 Effect of Fusarium Head Blight pathogens on gluten quality in wheat

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The proportion of Norwegian wheat used for food has varied significantly during the recent decade, mainly because of the instability of factors that are essential to baking quality (i.e. protein content and gluten functionality). During the same period, serious contamination of *Fusarium* spp. and mycotoxins was observed in some grain lots [1, 2].

A project was established to generate greater knowledge of the interface between gluten functionality and effects of *Fusarium* species and other microorganisms on Norwegian wheat quality. Instances of severe degradation of gluten proteins that resulted in an almost complete loss of gluten functionality were observed in some lots of Norwegian wheat. The degradation of the gluten appeared to be caused by exogenous proteases. Metabarcoding of fungi and bacteria in these grain lots identified fungi within the Fusarium Head Blight complex, as well as one bacterial species, as candidate species for influencing gluten functionality. Some of these candidates were inoculated on wheat during flowering [3]. Analysis of baking quality of the flour from this experiment revealed a reduced proportion of un-extractable polymeric proteins (%UPP) and severe reductions in the gluten's resistance to stretching (R_{MAX}) in wheat flour from plants inoculated with *Fusarium graminearum*. Flour from wheat inoculated with *Fusarium avenaceum* was generally less infested, and showed minimal or no reduction in gluten functionality and %UPP compared to flour from the *F. graminearum* infested samples. Flour from wheat inoculated with *Michrodochium majus* is yet to be analysed.

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PP-22 Functional analyses of RecQ helicase and SMS-2 protein genes in *Fusarium oxysporum* f. sp. *lycopersici*

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RNA silencing is one of the post transcriptional gene regulation systems in eukaryotes, and has been extensively studied in many model organisms. Each eukaryote has own indigenous components of RNA silencing pathway and each carries different functions.

In *Neurospora crassa*, one (ReqQ) of DNA helicases has roles not only in repairing DNA lesions, but also in quelling, a mechanism of RNA silencing. Additionally, SMS-2 (suppressor of meiotic silencing 2) is a main component of the MSUD (meiotic silencing by unpaired DNA) RNA silencing pathway and plays an important role in repair of chromosomes during sexual reproduction in *N. crassa*.

Fusarium oxysporum is a filamentous ascomycete fungus which includes important plant pathogenic strains of which f. sp. *lycopersici* (Fol) causes wilt of tomato. According to the recent studies, the RNA silencing of fungus is known to play a role in their growth, pathogenicity and life cycle. Through this clue, we are analyzing the function of RNA silencing pathways in Fol.

We identified both RecQ and SMS-2 homologues from Fol by BLAST. A RecQ gene was identified in Fol by BLAST in comparison to the amino acid sequence of RecQ in *N. crassa*. We generated RecQ-disruptants in Fol which presented an abnormal aerial hyphal growth on PDA (Potato Dextrose agar) compared with the wild-type. SMS-2 disruptants in Fol also exhibited an abnormal aerial hyphal growth on PDA (Potato Dextrose agar). This suggests that both of the two genes are involved in vegetative growth in Fol. We're now checking other phenotypes and pathogenicity in the disruptants.

PP-23 Combinatory effects of glutathione-modulation on the toxicity of the *Fusarium* mycotoxins deoxynivalenol (DON), NX-3 and butenolide (BUT)

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Various *Fusarium* mycotoxins, such as the trichothecenes DON and NX-3 or BUT, have been reported in previous studies to trigger ROS production and oxidative damage in mammalian cells (Mishra et al, 2014; Wang et al, 2006). Glutathione (GSH) plays on one hand a central role in the maintenance of the intracellular redox homeostasis and on the other hand represents a crucial substrate to detoxify thiol-reactive xenobiotics. Previous reports argue for an involvement of GSH in cellular detoxification processes of BUT and DON (Wang et al, 2006; Stanic et al, 2016).

In the present study, we addressed the question whether the intracellular GSH level affects the toxicity of DON and NX-3. We furthermore investigated whether the co-occurrence of BUT, reported to decrease the intracellular GSH-level, might enhance the toxicity of DON and NX-3. Pre-treatment of human liver cells (HepG2) with L-buthionine-[*S,R*]-sulfoximine, a well-known inhibitor of GSH synthesis, aggravated substantially BUT-induced cytotoxicity (≥50 µM, 24 h) but only marginally affected the cytotoxicity of DON and NX-3. In line with literature, BUT itself (≥100 µM) diminished the intracellular GSH level after 3 h of incubation. However, after 24 h incubation a substantial increase of the GSH levels was observed for DON (≥5 µM), NX-3 (≥5 µM) and, even more pronounced, for BUT (≥50 µM). Using a reporter gene approach, these concentrations were found to activate the redox-sensitive Nrf2/ARE-pathway, triggering among others the expression of γ-glutamyl cysteinyl ligase, the key enzyme in GSH biosynthesis. Thus, for combinatory experiments, the experimental design with respect to time and concentration ratios of pre- or co-incubation was found to be critical and 3 h BUT pre-treatment followed by 24 h incubation with DON showed increased cytotoxic effects. Taken together, in HepG2 liver cells, only limited impact of the intracellular GSH on the cytotoxicity of DON and NX-3 was observed. However, the GSH-level was found to be critical for the toxicity of BUT. The observed combinatory effects underline the potential contribution of non-regulated secondary metabolites like BUT to the toxicity of DON or structurally related trichothecenes, arguing for further studies on the toxicological relevance of these metabolites in naturally occurring mixtures.

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PP-24 Automated multispectral phenotyping for better understanding the plant-Fusarium interaction.

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In the past, virulence- and other bio-assays are predominantly evaluated by the human eye. However, in the omics era high-throughput automated multispectral analysis of a biological trait is a keystone for downstream omics analyses and dissecting plant-pathogen interactions.

The Laboratory of Applied Mycology and Phenomics (LAMP) combines the phenomics technology with fungal genome-editing approaches to better understand the drivers of a pathogen's virulence. LAMP recently installed a state-of-the-art high-throughput sensor-to-plant phenotyping and micro-dispenser platform: the **PathoViewer**. The platform allows to visualize diverse physiological traits in real time, based on specific absorption, reflection and emission patterns. This includes anthocyanin levels, chlorophyll (fluorescence), NIR, and GFP- and RFP-tagged organisms. The central part of the platform comprises a 3CCD camera mounted on a Cartesian coordinate robot housed in an acclimatized environment, including hard- and software pipelines to fully automate the image acquisition process. This platform is furthermore equipped with a dispenser which can be fitted with a nozzle to treat the plants with agrochemicals in a standardized manner. Growing plants under optimal conditions of light is one of the key-factors which is often underestimated when plants are grown in growth chambers. Therefore, the housing of the robot is a growth chamber equipped with Sunlight Led Modules (SLMs), which can be set to match natural light environments, with programmable day profiles up to a maximum of 1000 µmol m^{-2} s^{-1} PAR.

Here, we will present examples on the use of multispectral imaging in different plant-*Fusarium* interactions including wheat and maize.

PP-25 Exploring the role of ethylene signalling and wheat resistance to *Fusarium graminearum*

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Ethylene is a gaseous plant hormone involved in both plant defence and development. Often ethylene-mediated plant defence responses to necrotrophic fungi involve synergistic interactions with the jasmonate signalling pathway. On the other hand, ethylene is also an inducer of senescence and cell death, which could be beneficial for some invading necrotrophic pathogens. *Fusarium graminearum* is a hemibiotrophic pathogen, with both biotrophic and necrotrophic phases, that can infect wheat seedlings and inflorescence to cause fusarium seedling blight (FSB) and head blight (FHB), respectively. Interestingly, the role of ethylene signalling in the host-response to *Fusarium* species is unclear: some studies indicate that ethylene mediates resistance, while others have shown that it is associated with susceptibility. In an effort to understand the discrepancies in the literature, a series of FHB and FSB experiments involving exogenous hormone applications were carried out. The effects of ethylene inhibitor or enhancer treatments on the FHB response were compared in a detached head assay. Ethylene inhibition broke down resistance to initial infection and disease spread in three resistant wheat genotypes, whereas ethylene enhancer treatments resulted in reduced susceptibility in three susceptible genotypes. A similar trend was observed in FSB assays when comparing one resistant and one susceptible line. These results demonstrate that ethylene can mediate resistance to *Fusarium* in wheat, and that this pathway may not be up-regulated in susceptible genotypes. While these observations were consistent across multiple genotypes, and conserved in two different assays, they do not resolve the confusion in the literature regarding the role of ethylene in the host-response to *Fusarium*. There may be more to the story, and some important considerations should be made. For example, it is not known whether interactions with other signalling pathways affect the observed response. In addition, the timing of ethylene signalling and it's interaction with the pathogens lifestyle has not yet been evaluated. In both sets of experiments presented herein, the treatments were applied 2 h prior to inoculation. Additional work is underway to determine whether this effect is conserved when the ethylene signalling pathway is affected after the fungus has already established infection and entered into the nectrophic phase of its lifecycle.

PP-26 Charaterization of the interaction between cotton and *Fusamium oxysporum* isolates causing seedling disease.

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The fungus Fusarium oxysporum f. sp. vasinfectum (FOV) is an important wilt pathogen of cotton. Within the United States, FOV race 4 currently is geographically limited, and was first identified in the state of California in 2001 and in Texas in 2017. In FOV race 4 infested fields the fungus has also been known to cause early damping off and seedling mortality, often before the 4-leaf stage. To better assess disease impacts under California field conditions, more needs to be known of the relative pathogenicity and phenotypic characteristics of FOV races as seedling pathogens. Therefore, the goal of this study was to assess the reaction of cotton to seedling infection by different isolates of FOV. During the summer of 2017, isolates of FOV were isolated from infected cotton seedlings from three FOV race 4-infested commercial fields across the San Joaquin Valley of California. To test seedling pathogenicity a rolled towel assay was used to evaluate the pathogenicity of 16 FOV isolates. Seeds from the FOV race 4-moderately resistant Upland (Gossypium hirsutum L.) cultivar FM-2334 and susceptible Pima (G. barbadense L.) cultivar PHY-830 were individually inoculated with 100 µl of a 1×10 6 conidia/ml suspension. At ten days, seedlings were rated using an ordinal rating scale (1= no disease, 5= dead). There was a significant difference for isolate and variety (P<0.0001). Ordinal rating averages for isolates ranged from 2.1-3.4 and 3.7-4.3 for the FM-2334 and PHY-830, respectively. Based on these results, the use of the rolled towel assay should be examined as an early screening method for resistant cotton genotypes for FOV races.

PP-27 *Fusarium* species isolated from *Pennisetum clandestinum* collected during outbreaks of kikuyu poisoning in cattle in South Africa

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Kikuyu poisoning occurs sporadically in South Africa, but it is of major economic importance as valuable dairy cows are often poisoned and once affected the mortality rate is high. Kikuyu poisoning is not only a local problem and has also been reported from New-Zealand, Australia and eastern Africa[1]. Australian researchers have reported that *Fusarium torulosum* was consistently isolated from kikuyu grass collected during an outbreak. They hypothesized that this fungus might be the causal agent of kikuyu poisoning in Australia[2]. On the other hand, Brazilian researchers reported a similar syndrome in ruminants in the state of Rio Grande de Sul. However, this intoxication is caused by another plant, namely two *Baccharis* species, a member of the Asteraceae family. In this case, soil fungi synthesize a range of trichothecenes which are then absorbed by the roots of the plant[3]. As *Fusarium* species are known to synthesize trichothecenes the objective of this study was to submit kikuyu grass samples specifically for the isolation and molecular identification of *Fusarium* species.

Pennisetum clandestinum samples were collected during eight outbreaks of kikuyu poisoning in cattle in the Eastern Cape Province of South Africa. Ninety-four *Fusarium* isolates were retrieved from the grass samples, of which 72 were members of the *F. incarnatum/F. equiseti* species complex (FIESC) based on morphology and phylogenetic analyses of the translation elongation factor 1α sequence data. The South African isolates from kikuyu identified as members of the FIESC grouped together in six separate clades. The other isolates were *F. culmorum* (n = 3), *F. redolens* (n = 4) and *F. oxysporum* (n = 15). Contrary to findings in Australia, *F. torulosum* could not be isolated from *P. clandestinum* collected from toxic pastures in South Africa. However, the mycotoxicosis theory is still highly plausible and should be pursued.

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PP-28 *Fusarium* species diversity and deoxinivalenol level in grain of different spring wheat and spring barley varieties in Lithuania

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Barley and wheat are the main spring cereal crops in Lithuania. Spring wheat is the largest spring crop with approximately 200.000-265.000 ha sown in recent years. There are approximately 140.000-200.000 hectares of spring barley sown annually. Fusarium head blight (FHB) is a key fungal disease of cereals in Lithuania in recent years. Earlier studies showed that spring cereals are more susceptible to this disease than winter cereals and risk of mycotoxins is also higher in spring crop. FHB control and management strategies rely heavily on the host plant resistance. The aim of study was to determine the differences in the susceptibility of Lithuania-grown spring wheat and barley varieties to FHB severity, *Fusarium* species diversity and deoxinivalenol (DON) concentration in harvested grains. Field experiment was carried out at the Institute of Agriculture of the Lithuanian Research Centre for Agriculture and Forestry over 2 growing seasons (2016 and 2017).

High variability of FHB severity between different spring barley and wheat cultivars was established. The disease severity in spring wheat varieties was ranged from 1.40% to 11.05% and from 3.53% to 16.86% in 2016 and 2017, respectively. Spring barley was less susceptible to FHB severity than wheat and FHB severity was ranged in the interval of 3.35-7.65% in both years. In most cases, the level of DON in grains increased due to increase the severity of FHB. *Fusarium* species diversity varied within crop species and genotypes. The most prevalent species occurring on spring barley and wheat were *Fusarium graminearum*, *F. avenaceum*, *F. poae* and *F. culmorum*.

PP-29 A field study on the reduction of *Fusarium* toxins in wheat by the biological control agent *Clonostachys rosea* and the evaluation of a novel formulation strategy

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Fusarium Head Blight (FHB) of wheat is one of the major cereal diseases worldwide, caused by a complex of toxigenic fungi. The dominant species worldwide is *Fusarium graminearum* (teleomorph *Gibberella zeae*) which produces the mycotoxin deoxynivalenol (DON) and the mycoestrogen zearalenone (ZEA), leading to high economic losses and representing a threat to food and feed safety. High-risk farming systems that rely on cereal intensive crop rotations and the overall aim to reduce chemical inputs are in need of innovative solutions to minimize the disease pressure. The application of microbial antagonists to protect wheat at the time of susceptibility is integral in sustainable mycotoxin management.

Within the scope of the Horizon 2020 project MycoKey, our two year study examines the ability of the fungal biological control agent (BCA) *Clonostachys rosea* to reduce the contamination of wheat grains with *Fusarium* toxins under field conditions in Switzerland and in the growth chamber. In the field, maize residues deliberately infected with *F. graminearum*, provide semi-natural disease pressure throughout the season. We hypothesize that a novel oil-in-water formulation strategy that includes protection against desiccation and harmful UV-B radiation enhances the biological control efficacy over commonly used suspension of conidia in water. The comparison between locally isolated BCA strains and strains from abroad might underline the importance of sourcing antagonists in proximity to the application area.

First year results from a growth chamber experiment demonstrate that application of *C. rosea* conidia suspensions significantly reduced the mean disease severity and the mean DON content up to 100% and 96% relative to the mock treated control, respectively. Under controlled environmental conditions, the addition of sunflower oil and a UV-B absorber revealed no or even a negative effect on the ability of the BCA to protect the host. In contrast, *C. rosea* in combination with this formulation applied under field conditions resulted in highest disease control compared with the other treatments. Currently, harvested grains from field and growth chamber experiments are analysed for the content of *Fusarium* toxins, the *Fusarium* species incidence as well as the amount of *F. graminearum* DNA by qPCR. A protocol for molecular quantification (qPCR) of BCA biomass is under development. First year results and the latest findings of the study will be presented and discussed.

PP-30 Mycotoxins in two size fractions in Norwegian oat grains

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Fusarium infections and mycotoxin occurrences have increased in Norwegian cereals over the recent 15 years. The mycotoxin deoxynivalenol (DON), produced by *Fusarium graminearum*, has become a threat for the cereal grain quality, especially in oats, which covers approximately 25 % of the cereal cultivation acreage in Norway. During the last five years, we have seen a decline in the DON levels, however, at the same time, high concentrations of the mycotoxins HT-2 and T-2 (HT2+T2), primarily produced by *Fusarium langsethiae*, have been detected in some grain lots Norwegian oats. Moreover, a number of other *Fusarium* toxins, including modified DON (3-acetyl-DON (3-ADON), DON-3-glucoside (DON-3G)), nivalenol (NIV), zearalenon (ZEA), beauvericin (BEA) and enniatins (ENN) have been recorded in Norwegian cereal grain surveys.

The quality of a grain lot may be improved if the most toxin-contaminated kernels can be removed. Sorting and aspiration techniques have shown that DON and HT2+T2 contaminated kernels can be separated from healthy grain. In one of the work packages in the Norwegian SafeOats project we explore the possibilities for reducing the content of the most frequent mycotoxins in oat grain lots by removing small kernels. So far, samples from 15 oat grain lots have been sorted into two fractions (< 2,2 mm >). The mycotoxin content in these two fractions and a sample of un-sorted grain from each lot have been analysed by LC-MS/MS. On average, the levels of both HT2+T2 and EnnB were reduced by more than 50 % after removal of the small fractions. The smaller grain fractions had almost four times higher levels of T2+HT2 and approximately two times higher EnnB than the larger grains. The levels of the mycotoxins DON, 3-ADON, NIV, BEA, EnnB1, EnnA and EnnA1 were also reduced after removal of the small fractions, however, the levels of these toxins were in general low.

PP-31 *Fusarium* landscape of asparagus decline syndrome in Spain and influence of soil biodisinfection and *Trichoderma* spp. on crop establishment.

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Asparagus decline is a complex syndrome that involves weakening of commercial asparagus plant and limits the productive period of the crop preventing replantation. It is mainly caused by *Fusarium oxysporum* f. sp. *asparagi* (FOA) (1), although the composition of the consortium varies according to the regions (2). Affected fields have been surveyed in the main asparagus growing areas in Spain. More than 200 *Fusarium* isolates obtained from tissues of roots, crowns, and stems were identified to species level by morphological approach (3) and sequencing of a portion of the translation the elongation factor 1 alpha gene (4). This work also aims to evaluate the efficacy of *Trichoderma* spp. and organic amendments combined with soil solarization as control methods. A randomized block design has been implemented on a farm with seven years history of asparagus culture. The effects of treatments on asparagus replantation have been evaluated by measuring the soil inoculum density of *Fusarium* spp. In addition, parameters such as the interception of photosynthetically active radiation or the total biomass production have been studied. The results show significant differences between the different production areas with the presence of different *Fusarium* species involved in the disease complex. Regarding the field trial, results showed higher number of *Fusarium* colonies in the control treatment than in all soil disinfection treatments evaluated, being highly significant the reduction of *Fusarium* propagules of the soil treated with Dazomet, which also corresponds to the highest growth parameters analyzed. Although to a lesser extent, biosolarization with brassica and hen manure pellets have been shown to reduce *Fusarium* infection and increase the dry weight and radiation interception of asparagus plants. Treatments with *Trichoderma* spp. reduced FOA populations, but did not enhance plant growth. Both chemical fumigation and biosolarization with organic amendments can be considered as crop strategies before replanting of asparagus plants.

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PP-32 Towards a CRISPR-Cas9 system in *Fusarium poae*

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CRISPR-Cas technology is an efficient genome editing tool, which has been successfully used in several fungal genera such as *Penicillium*, *Aspergillus* and *Neurospora*. This system consists of two elements: a Cas9 endonuclease and a single guide RNA (sgRNA) containing a 20-nt guide sequence matching the target DNA. The sgRNA can recruit Cas9 to create specific DNA double strand breaks (DSBs) in the target region. Two common mechanisms for repairing DSBs in DNA exist: One of them leads to deletions, insertions or substitutions through error-prone non-homologous end-joining (NHEJ). A second system involves homologous recombination-directed repair (HDR) in the presence of a gene, which can then be inserted at the target site.

Our work aims at establishing a CRISPR-Cas9 platform in *Fusarium poae*, an omnipresent member in the Fusarium head blight complex of wheat in Europe. We used a dual expression system of Cas9 and sgRNA from a single vector. First, we codon-optimized the Cas9 coding sequence for *F. poae*. This codon-optimized Cas9 was equipped with a *F. poae* gpdA- or tef1 promoter. The sgRNA fragment was put under the control of a U6 *F. poae* promoter. After the protoplast-mediated transformation, positive transformants were selected for sequencing to assess the gene-editing features.

In the future, we will use this CRISPR-Cas technology combined with the high-quality PacBio genome of *F. poae*, to investigate the role of specific effectors and secondary metabolite gene clusters. This will shed new light on genes involved in the specific cryptic disease symptoms of *F. poae*.

PP-33 INFLUENCE of agronomic and climatic factors on the epidemiology of *Microdochium* species and assessment of wheat cultivars susceptibility to these pathogens in France.

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Fusarium Head blight (FHB) is an important disease of small grain cereals caused by a number of toxigenic (*Fusarium* spp.) and non-toxigenic (*Microdochium*) species. In France, many studies focused on the survey of occurrence of *Fusarium species*, on forecasting models specific to *F. graminearum*, mycotoxins production or cultivar resistance to *Fusarium*. However, *Microdochium nivale* and *M. majus* are also frequently identified in small grain cereals and can cause alone significant yield losses. This study investigated the influence of agronomics and climatic factors on the epidemiology of *Microdochium* species and evaluated wheat cultivars susceptibility to *Microdochium* in field trials.

More than 500 seed samples of wheat were collected from 2007 to 2017 and analysed by quantitative PCR (qPCR) for *M. nivale, M. majus* and *F. graminearum*. Agronomic practices and several climatic variables were tested to explain DNA quantity of these pathogens. To evaluate susceptibility of wheat, in 2016 and 2017, 50 cultivars separated in 2 subpanels of 25 cultivars (sharing 5 cultivars used as control) were both inoculated by *M. nivale* and *M. majus* singly. 3 trials per subpanel were done each year as the notations of symptoms on leaves and ears, and were harvested. Efficiency of artificial inoculations was measured by qPCR analysis for all 5 control cultivars. Each year, qPCR analyses were also done for all varieties of 2 field trials.

Results of the survey indicate that climatic factors are more explicative than agronomic factors for the abundance of *Microdochium* species conversely to *F. graminearum*. Moreover, proportion between *M. nivale* and *M. majus* seems also dependant of climatic factors. Efficiency of artificial inoculation in field trials was very aleatory and interactions with *F. graminearum* complicate notations. However, qPCR analyses allow us to bring out different susceptibilities between cultivars for *Microdochium*, both for leaves and spikes. We also compare these results with cultivar resistance to *F. graminearum*. All these results will help us to develop a predictive model for wheat contamination by *Microdochium* and improve wheat resistance to *Microdochium*.

PP-34 Control of Fusarium head blight in wheat by using *Bacillus velezensis* RC 218 and chitosan in Argentina

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Both bread wheat (*Triticum aestivum* L) and durum wheat (*Triticum turgidum* L. var. *durum*) can be affected by *Fusarium* head blight (FHB) being *Fusarium graminearum* sensu stricto the main pathogen in Argentina. In the management of FHB, crop rotation, tillage practices, fungicide application and planting less susceptible cultivars are the main options to counteract the disease. Among the environmentally-friendly strategies, the use of biocontrol and chitosan have been proposed. Chitosan is considered as GRAS (generally recognized as safe), easily biodegradable, and has antioxidant and antifungal activity. *Bacillus velezensis* RC 218 showed effectiveness in controlling FHB severity at under field conditions and deoxynivalenol accumulation in harvested grains. The aim of the present study was to evaluate the effectivity of the co-application of *B. velezensis* RC 218 (Bvel) and chitosan (Chi) to control FHB on both bread and durum wheat cultivars. At the anthesis period, the bacteria (1×10^6 ufc/ml) and chitosan (0.1 % v/v) were applied over spikes prior the inoculation of an inoculum mixture of *F. graminearum* strains. *Bacillus velezensis* RC 218 and Bvel+Chi treatment were able to reduce FHB incidence on both types of wheat cultivars. Disease severity was significantly reduced by Bvel, Chi and the combination Bvel+Chi treatments on both bread and durum wheat ($p \leq 0.005$). FHB severity reduction levels averaged 53 and 58.6 % for Bvel treatment on bread and durum wheat, respectively; 47.8 and 49.7 % for Chi treatment on bread and durum wheat, respectively; and finally the combination Bvel+Chi reduced FHB severity by 38.7 and 54.2 % on bread and durum wheat, respectively. The effect of the treatments on other parameters such as deoxynivalenol accumulation, *Fusarium*-damaged kernel and *F. graminearum* DNA levels were also evaluated.

PP-35 Exploring Fungal endophytes as new biocontrol agents against *Fusarium graminearum* in maize

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Fusarium Head Blight (FHB) is a devastating fungal disease which affects small grain cereals such as wheat and maize. Although FHB is caused by a species complex, *Fusarium graminearum* (Fg) is the most important member involved. Beside the economic losses due to the decrease in yield, the fungus has an impact on grain quality due to the production of mycotoxins. Driven by the awareness that reduced tillage systems result in soil structure improvement, conservation tillage practices are often implemented leaving more stubble/straw residues on the field. This organic material can serve as the primary inoculum of Fg. Over the last decade, different strategies for FHB management have been proposed. Among them, biological control using non-pathogenic bacteria and fungi is encouraged as safe and sustainable long-term solution in comparison with chemical control. However, survival of these biocontrol agents is often a serious impediment for their use in practice. Therefore, we explored whether endophytic fungi can be used as biocontrol agents. Their close association with the plant might be an interesting trait to guarantee a better survival in the rhizosphere. To isolate these endophytes, we started from crop residues. Despite its role as source for primary Fg inoculum, we hypothesized that crop residues also harbor valuable antagonistic fungi. In the current project, we isolated several novel fungal endophytesfrom crop residues, and tested them for their ability to control the growth of Fg and of mycotoxin production both *in vitro* and *in planta*.

The endophytic fungi were tested for their effects against Fg. *In vitro* plating assays (contact and volatile) and *in vivo*, maize pot experiments, were performed for each endophyte to assess its biocontrol capacity against Fg. The obtained results show that the selected biocontrol endophytes have a promising effect on Fg infection. Furthermore, measuring the mycotoxin levels (deoxynivalenol, 15-acetyldeoxynivalenol, deoxynivalenol-3-glucoside and zearalenone) through a validated multi-mycotoxin LC-MS/MS method, the selected biocontrol endophytes had an inhibitory effect on mycotoxins production. Using a non-targeted metabolomics approach, with Q-TOF LC/MS, the mode of action is being investigated. The project will contribute to a great extend to reduction of *fusarium* mycotoxins level in grain cereals especially wheat and maize.

PP-36 Response of oat genotypes to Fusarium head blight in western Canada.

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Oat (*Avena sativa* L.) is one of the most important cereal crops in western Canada and has become desirable for human consumptions due its high nutritional value. In recent years, Fusarium head blight (FHB) emerged to be one of the most serious diseases on oat in the Canadian Prairies and mycotoxin contamination in Fusarium damaged kernels has become a potential problem for oat production. To minimize damage and mycotoxin contamination, suitable sources of FHB resistance need to be identified and used in oat breeding programs to develop cultivars suitable for production in FHB-affected environments. In this study, we investigated FHB resistance in advanced oat breeding lines (Western Cooperative Oat Registration Trial -WCORT) and oat cultivars already being grown commercially by screening oat lines in a mist-irrigated artificially-inoculated FHB disease nursery in 2016 and 2017. Deoxynivalenol was detected in all tested genotypes, and up to 59 µg/g of DON was observed in the oat grain. Genotype ranking in DON content was relatively consistent across years, planting dates, and experiments. It is concluded that Fusarium mycotoxin could be a potential problem for oat production under high FHB pressures in western Canada; however, the severity of this problem needs to be assessed by extensive monitoring of DON level under natural conditions. Additionally, genotypes with consistently low and high DON levels are identified and could be used in the future genetic analysis of FHB resistance.

PP-37 Quantification of fumonisins and hydrolysed fumonisins in pig serum with LC-MS/MS

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Fumonisins are mycotoxins produced by *Fusarium* strains. The predominant form is Fumonisin B_1 (FB1), followed by FB2 and FB3. The most affected commodity is maize. FB1 was shown to have several negative effects on animal and human health.

Chemically, all fumonisins are long-chain alkylamines with two tricarballylic acid (TCA) groups. The hydrolyzed FB1, after cleavage of both TCA groups, has been shown to have a highly reduced toxicity compared to FB1 *in vivo*. Therefore, a novel feed additive, containing fumonisin esterase (FUMzyme®), which can enzymatically hydrolyse fumonisins in the gastrointestinal tract of animals, is a way to counteract the negative effects of these mycotoxins on livestock health and productivity.

The quantitative determination of biomarkers in different biological matrices is a prerequisite for detailed analysis of animal feeding trials. Due to the low bioavailability of fumonisins and their hydrolysis products, the concentration of the analytes is very low in matrices like serum, urine or bile. Therefore, the sample preparation has to be optimized for each matrix, prior to analysis by HPLC-ESI-MS/MS.

We describe the validation of an analytical method for the quantification of twelve analytes (FB1, FB2, FB3 and all their fully and partially hydrolysed forms) in pig serum. Serum samples were extracted with acidified methanol-acetonitrile solvents. As pre-trials have shown that the use of internal standards is a promising means to achieve a sufficient overall recovery, ^{13}C-labeled internal standards were added before sample preparation to compensate for losses during protein precipitation and matrix effects in ESI-MS/MS analysis. The recoveries of fumonisins in serum with internal standard correction were between 85 and 110 percent for all twelve analytes. The stability of the analytes in serum samples was also tested under different storage conditions. Serum samples spiked at three concentration levels were analysed immediately or frozen at -20 °C and worked up after one week, one month, three months, and after one and three freeze-thaw cycles. The recoveries were mostly between 75 and 115 percent. The sample stability was also tested in ready-to-inject samples at room temperature and at 4 °C for 24 and 48 hours. Again, no losses were observed.

The validation and stability data demonstrate the usability of the developed analytical procedure and measurement method for accurate quantification of fumonisins in pig serum.

PP-38 An investigation into *Fusarium langsethiae* resistance in UK oat breeding lines

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In 2004 *Fusarium langsethiae* was described as a new species, since then it has been shown to be a weak pathogen in cereals, producing mostly symptomless infections if present. However *F. langsethiae* has a preference for oats causing symptomless infections resulting in combined concentrations of the mycotoxins HT2 and T2 (HT2+T2) in unprocessed grains as high as 9990 µg kg^{-1}. The tolerable combined daily intake for which is 0.02 µg/kg body weight. Genetic resistance exists within current UK oat breeding lines and has been examined through the measurement of HT2+T2 concentrations found in oat cultivars grown in the Agricultural and Horticultural Development Board Recommended List trials. To further investigate varietal differences a mapping population of winter oat recombinant inbred lines derived from crossing Buffalo and Tardis parents were used to identify quantitative trait loci (QTL) that could infer resistance to HT2+T2 accumulation. Height and flowering time were identified as traits with the potential to infer resistance, and therefore QTL for those traits were introgressed into the background of the opposing parent to generate near isogenic lines (NIL) that could be assessed for mycotoxin accumulation. The expression of two of the flowering time QTL are different in spring drilled plants as opposed to winter drilled. As such two NIL experiments from the 2017 season are discussed; one drilled in the Autumn and one in the Spring. This will allow the examination of the impact of flowering time itself against any genetic linkage associated with the QTL in question. Experiments were laid out as randomised complete block designs and grown in a field which maintains a winter oat and wheat rotation. Grain was sampled at harvest, milled and assessed for HT2+T2 concentration using an ELISA. Analysis of the HT2+T2 content of the NIL and the parent varieties will be presented.

PP-39 *Fusarium chlamydosporum* species complex associated with Brazilian rice: species diversity and toxigenic potential

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Amongst several species of the genus *Fusarium,* strains of the *Fusarium chlamydosporum* species complex FCSC were isolated from samples of rice grains collected in Brazil. Morphological markers of the FCSC species such as the presence of chlamydospores and formation of conidiophores in the aerial mycelium with a typical ramification allowed the identification of isolates as members of the FCSC. The main aim of this study was to characterize isolates of the FCSC by molecular phylogenetic analyses of partial sequences of the genes *Tef-1α, Rpb2* and *Cal,* and to evaluate its potential to produce nivalenol (NIV), deoxynivalenol(DON) and acetylated forms 3-acetildeoxynivalenol (3-ADON) and 15-acetildeoxynivalenol (15-ADON), which are members of the harmful group of mycotoxins called trichothecenes. Analyses of *Tef-1α* sequences identified three of the known phylogenetic lineages belonging to FCSC. Sequences of the regions *Cal* e *Rpb2* were generated for a subset, representative for the three distinct lineages. Sixty-one isolates grouped together within lineage 1, together with reference material and the *F. chlamydosporum* type CBS 125.45. Lineage 2 and lineage 3 were represented by 7 and 12 isolates, respectively. Selected isolates of all three lineages produced detectable levels of NIV and DON *in vitro*. Five out of eight isolates analyzed for the occurrence of *tri5* gene, involved in trichothecenes production, showed to possess this gene. In conclusion, we report here, for the first time, the occurrence of *F. chlamydosporum* and the two still unnamed phylogenetic sister lineages, in association with rice in Brazil. Moreover, the potential of these species to contribute to the contamination of rice with trichothecenes needs to be better investigated, in order to elucidate the potential capability of the FCSC species to produce trichothecenes on rice grains.

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PP-40 Catechization of a wheat leucine rich receptor like kinase for *Fusarium graminearum* resistance.

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Receptor-like kinases form the largest family of receptors in plants and play an important role in recognizing pathogen-associated molecular patterns and modulating the plant immune responses to invasive fungi, including cereal defences against fungal diseases. But hitherto, none have been shown to module the wheat response to the economically important Fusarium head blight (FHB) disease of small-grain cereals. Homologous genes were identified on barley chromosome 6H, (*HvLRRK-6H*) and wheat chromosome 6DL(*TaLRRK-6D*), which encode the characteristic domains of surface-localized receptor like kinases. Gene expression studies validated that the wheat *TaLRRK-6D* is highly induced in heads as an early response to both the causal pathogen of FHB disease, *Fusarium graminearum*, and its' mycotoxic virulence factor deoxynivalenol. The transcription of other wheat homeologs of this gene located on chromosomes 6A and 6B was also up-regulated in response to *F. graminearum*. Virus-induced gene silencing (VIGS) of the barley *HvLRRK-6H* compromised leaf defense against *F. graminearum*. VIGS of TaLRRK-6D in two wheat cultivars, CM82036 (resistant to FHB disease) and cv. Remus (susceptible to FHB), confirmed that TaLRRK-6D contributes basal resistance to FHB disease in both genotypes. Although the effect of VIGS did not generally reduce grain losses due to FHB, this experiment did reveal that *TaLRRK-6D* positively contributes to grain development. Further gene expression studies in cv. Remus indicated that VIGS of *TaLRRK-6D* suppressed SA signaling, which is a key hormonal pathway involved in the initial defence against FHB disease. Thus, this study provides the first evidence of receptor like kinases as important component of cereal defence again Fusarium and highlights this gene as a target for enhancing cereal resistance to FHB disease.

PP-41 Hunting for orphan genes involved in wheat defence against Fusarium head blight disease.

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Fusarium head blight (FHB) disease, caused by several rapidly evolving *Fusarium* species reduces grain yield and contaminates grain with harmful mycotoxins. Such pathogenic stress may lead to species-specific adaptive processes and the emergence of mysterious, yet biologically significant, orphan genes in the host. The focus of this study is the identification and characterisation of genes that have specifically evolved for defence and/or disease mitigation in wheat. The mining of next generation sequencing (NGS) RNAseq data is ongoing, to identify wheat-specific genes involved in defence against FHB disease. This involves a deductive process of differential expression analysis, homology searches, motif detection, domain archiving and functional profiling. Three RNAseq data sets (Gou et al., 2016; Hofstad et al., 2016; Schweiger et al., 2016) have been extracted from the NCBI SRA database, each consisting of sequence data from studies investigating *Fusarium* infection in wheat. Analysis of the first dataset is underway, wherein the treatments were mock and *Fusarium* and the genotypes used were the FHB-susceptible cv. Chinese Spring and the resistant combination of cv. Chinese Spring containing a long arm of the chromosome 7E from *Thinopyrum elongatum* (CS-7EL).

The differentially expressed transcripts were screened using limma / voom (Law et al., 2014) after *de novo* assembly of RNA seq reads using Trinity (Haas et al., 2013). In the *Fusarium*-inoculated susceptible vs mock-inoculated susceptible comparison, 9563 putative genes were up regulated and 17515 genes down regulated in response to *Fusarium*. In the resistant genotype, 1878 putative genes were up regulated and 13622 down regulated in response to *Fusarium.*

A new automated pipeline is being established for the screening and expression profiling of such novel putative interactors in the ongoing wheat/FHB conflict. The orphan genes were hunted from *Pooideae* family. In susceptible genotype 2738 orphan genes were found to be up-regulated yet, 1458 orphan genes were downregulated in response to *Fusarium*. 473 orphan genes were found to be up regulated while 249 orphan genes were found to be down regulated in response to *Fusarium* in resistant genotype. The role of putative orphans in disease resistance will be assessed using virus induced gene silencing (VIGS) and qPCR.

PP-42 Kdm5, a jumonji histone H3 demethylase is a chromatin regulator of secondary metabolite gene expression and virulence in *Fusarium graminearum*

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Fusarium graminearum is a fungal plant pathogen causing fusarium head blight (FHB), a devastating disease of several important cereal crops. It is now well established that chromatin-based epigenetic mechanisms are key determinants of the coordinated gene expression programs underlying pathogenic processes. Of particular interest are chromatin-based mechanisms which regulate the expression of effector gene expression and secondary metabolite gene clusters. These genomic regions seem to feature a very special chromatin landscape that either mediates silencing or activation of underlying genes by a unique combination of particular histone marks.

Here we show that Kdm5, a multi-domain histone demethylase responsible for the removal of activating histone H3-lysine 4 trimethylation (H3K4me3) marks, not only mediates gene repression, as expected, but Kdm5 is also an important activator, particularly of genes involved in secondary metabolism. To determine whether the histone demethylase domain of Kdm5 is required for SM gene regulation, we constructed in addition to the Kdm5 deletion mutant a Kdm5 variant carrying point mutations in the highly conserved catalytic domain. This region of the protein is directly responsible for the removal of the activating H3K4me3 mark. Similar to the recently reported work in *Aspergillus nidulans*, first results in F. graminearum indicate that Kdm5-mediated regulation is not restricted to its catalytic JmjC domain. Furthermore, cross-complementation experiments show that the A. nidulans KdmB protein is only partially able to replace the function of Kdm5 and thus only in part complements the deletion phenotype. Pathogenicity assays with the kdm5 mutants on Apogee wheat showed a significant hypovirulence of the mutants although biosynthesis of deoxynivalenol (DON) is not strongly deviating from the WT in axenic cultures. This would mean that additional virulence-determining genes are regulated by Kdm5.

PP-43 Diallel analysis of Fusarium head blight resistance caused by *Fusarium graminearum* in winter wheat

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Fusarium head blight (FHB) caused by *Fusarium* spp. reduces grain yield and quality in wheat contaminating the infected grain with mycotoxins. Development of resistant wheat cultivars is believed to be the most efficient and economical mean of reducing damage caused by FHB. The FHB resistance in wheat is a quantitative trait, which can be evaluated using different measures based on visual disease symptoms or by measuring mycotoxin contamination of grain. In the present study we present the results of an F1 diallel analysis of FHB resistance involving eight European winter wheat genotypes and their F1 crosses. Parents and F1s excluding reciprocals were grown at location Zagreb over three consecutive years in a field trial arranged as a RCBD with two replications. Artificial inoculations with the isolate of *F. graminearum* Schwabe using spray method were performed at the beginning of anthesis and repeated on the same plots two days later. The resistance was scored visually 18, 22, 26 and 30 days after inoculation and expressed as % of wheat head area infected with FHB (VRI). After ripening 10 random spikes from each plot were manually harvested and % of fusarium damaged kernels (FDK) was assessed on threshed grains. In addition, milled samples of grain from each plot were used for quantitative analysis of mycotoxins deoxynivalenol (DON) and zearalenone (ZON) using an HPLC-MS/MS based-multianalyte method. For all traits both general combining ability (GCA) and specific combining ability (SCA) effects were statistically significant, whereas the interaction with year was significant only for GCA. Additive genetic variance accounted for 85, 76, 89 and 65% of the total genetic variance for VRI, FDK, DON and ZON, respectively. A high broad sense heritability was observed for analyzed traits ranging from 0.84 (ZON) to 0.92 (VRI) allowing for considerable progress by selection. A significant mid parent heterosis was observed in a number of F1 combinations for all traits, whereas a significant better parent heterosis was observed in several F1 crosses only for FDK and ZON content. Correlations between content of mycotoxins (DON and ZON) and disease ratings (VRI and FDK) were very strong ranging from 0.90 to 0.96 indicating that mycotoxin content in infected grain can be accurately predicted based on visual disease ratings.

PP-44 Using Brachypodium as a tool for screening resistance against wheat root diseases

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Brachypodium distachyon (Bd) is a model species for the study of the more complex cereal genomes, having not only a great gene synteny to most of the small grain cereal species but also the advantage of its small size and short life cycle. The purpose of this project is to use Bd to identify potential resistance / susceptibility genes that play a significant role in response to Fusarium root rot (FRR, caused by *Fusarium graminearum* and *Fusarium culmorum*) and Take-all (caused by *Gaeumannomyces graminis* var. *tritici*). Exploiting the recent availability of the genomic sequence of a number of different Brachypodium accessions by the Vogel group in the Joint Genome Institute in the United States Department of Energy, a set of accessions was tested in order to obtain a resistance profile against the two diseases. A special focus was made in the characterization of accessions that are parents to populations that have already been developed.

Two populations were demonstrated to have a moderate resistance differential to FRR: Luc1xFoz1 and Bd21XABR6, with Bd21xABR6 having a genetic map already generated for further analysis. The Luc1xJer1 population also showed a resistance contrast to Take-all, where Luc1, like in FRR, is the most susceptible parent and Jer1 the resistant parent. Since there are no available genetic maps for the Luc1xFoz1 and Luc1xJer1 populations, these will be developed to the F5 generation and genotyped using KASP markers. A QTL analysis will be performed to identify the most promising region(s) of the genome with potential effects on plant defence.

Even though resistance to the two diseases seem to work through different defence pathways within the plant, an effort will be made to pinpoint genomic regions involved in the resistance response to both diseases. These preliminary investigations reveal that the use of Brachypodium as a model can be a powerful tool for identifying the genetic bases of disease resistance mechanisms allowing a potential transfer of this knowledge onto crop species.

PP-45 Response of some durum and bread wheat genotypes to fusarium foot and root rot disease under dry conditions in Tunisia

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Fusarium foot and root rot (FRR) is an important cereal disease in dry land conditions. It is mainly caused by *Fusarium culmorum* and/or *F. pseudograminearum* but *F. culmorum* remains the most frequently isolated species in Tunisia. Reaction to this disease varies among cereal species. Durum wheat (*Triticum turgidum* ssp. durum) (DW), the principal grown cereal in Tunisia (60% of grown area), seem to be susceptible. In parallel, significant damages are observed on bread wheat (*Triticum aestivum*) (BW), but fortunately some finding identified partial resistances in this species. The behavior of these two wheats to FRR, under Tunisian climatic conditions, is not well documented. Thus, response of 24 DW advanced lines and five commercial BW were studied. DW genotypes were selected from the national breeding program for their tolerance to Septoria and rust leaf diseases and their high productivity. BW was included as tolerant-check. The trial was carried out in a field located in a dry region of Tunisia during the growing season 2016/2017. Plant material was inoculated by *F. culmorum* at stage three leafs. Disease severity was estimated using a scale ranging from (0-5) of stem bases discoloration and percentage of whiteheads senescing prematurely. Grain yield (GY), Thousand Kernel Weight (TKW), Heading (HD) and the height (H) of plants were also evaluated. Results revealed that inoculation has significantly increased both disease severity and whiteheads, decreased GY and TKW, delayed HD but had no effect on H. In parallel, disease severity and whiteheads respectively, were significantly correlated with GY (r= -0.60, $P < 0.01$; r= -0.63, $P < 0.01$), TKW (r-0.34, $P < 0.01$) and HD (r=0.56, $P < 0.01$; r=0.62, $P < 0.01$). In conditions of this trial, DW was more affected by the disease than BW with a significant ($P < 0.0001$) difference observed between DW genotypes. One group of three DW lines, characterized by their tolerance to water stress, seems to be more tolerant and one cultivar, adapted to wet conditions, was more susceptible than the others. These results present the work of one year, thus, more investigation are needed to understand and reinforce these findings.

PP-46 Global mycotoxin survey 2017 for animal feed

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For more than ten years, BIOMIN has been conducting an annual mycotoxin survey, monitoring the incidences of different mycotoxins in several agricultural commodities intended for use in animal feed. The samples are collected on a worldwide basis and are analyzed for the following mycotoxins: DON (deoxynivalenol), T-2 toxin, ZEN (zearalenone), FUM (fumonisins B_1 and B_2), Afla (aflatoxins B_1, B_2, G_1, G_2) and OTA (ochratoxin A). Analysis is carried out in different laboratories, using HPLC, LC-MS/MS and ELISA methods.

In the first three quarters of 2017, more than 13,000 samples, originating from 69 countries were analyzed. As expected, regional differences can be observed. For example, the most prevalent mycotoxin in Europe was DON: 72% of 2556 tested samples were positive, i.e. showed values above the limit of quantitation (depending on the analysis method between 1 and 200 ppb). The average level of DON in the positive samples was 448 ppb, 50% of all tested samples were above the risk threshold of 150 ppb DON for potential effect on sensitive species. A higher risk was reported in South and Central America, where even 82% of 3990 samples tested for DON had contamination above this risk threshold, with an average level of 898 ppb in all positive samples. FUM also showed variation between regions, and was found above the risk threshold (500 ppb) in 15% of 1556 European samples, in 40 % of 1,754 Asian samples and in 66% of 4,611 samples from South and Central America.

In general, 75% of all samples analyzed for at least three mycotoxins were positive for more than one of the tested mycotoxins, 20% were contaminated by a single detected mycotoxin and only 6% of all samples tested negative.

From these data it is obvious that mycotoxins are not only a worldwide threat, but also that co-contamination proves to be the rule, not the exception. Therefore, further research on the effects of co-occurrence of several mycotoxins is warranted.

PP-47 Breeding for high resistance to Fusarium head blight (FHB) in cultivar selection programs with and without known FHB resistance sources aided by artificial inoculation

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The last decades increased our knowledge on resistance of wheat to FHB, but less progress was made in cultivar breeding. The screening for resistance should be introduced into running breeding programs. Further, we should test a breeding program focused on FHB that considers other traits necessary to breed marketable cultivars as a competitive procedure. The C and D lines contained also the corresponding lines from the FHB program. Visual symptoms, Fusarium damaged kernel (FDK) and deoxynivalenol (DON) were measured.

In the FHB breeding program (2011-2015) 897 F_3-F_5 lines) from more resistant adapted winter wheat and exotic sources were used. The exotics gave slightly better FDK performance (58% R), than adapted genotypes (55% R) between 0 and 10% (R), and 20.8% and 8.8% were between 40 and 80% (S).

The 1281 B lines (343 local and 554 exotic lines) indicate that both systems can produce resistant and highly resistant breeding material. In the B lines (514 local, 767 exotic) the rate of exotic lines is higher and within local 41.4% R, 21.4% S, and within exotic 48.2% R and 11.3% S rate shows the advantage of the exotic model.

From the non FHB program 1370 C lines were tested (replicated yield trials) with 1283 locals, 87 exotics). From the local program 41.3 % yielded 0-10 and 14.7% gave between 40 and 250 mg/kg DON. For the exotic FHB program 58.7% yielded 0-10 mg/kg DON and only 2.3% was between 40-50 mg/kg. In the 636 advanced lines and variety candidates (612 local and 24 exotic) 32% of the local program contained DON between 0-10 mg/kg and 8.2% gave higher DON content than 45 mg/kg (maximum: 256 mg/kg). From the exotic lines 66.7% belonged to the 0-10 mg/kg group, but all were lower than 5 mg/kg and no one hade DON above 25 mg/kg. that is a significant difference.

The conventional breeding programs contain a large variation for FHB resistance that can be utilized by consequent screening. In the FHB program we have created well adapted winter wheat lines. As FHB resistance itself is not enough to breed cultivars, the high FHB resistance was combined with leaf rust, yellow rust, powdery mildew and leaf spot resistance. Many lines of them exhibit high protein content (15-18%) and baking quality (up to alveograph W=390).

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PP-48 Enzymatic deactivation of *Fusarium* mycotoxins in corn cob mix silage

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Infection of crops with mycotoxin producing fungi in the field are very common. Field fungi, such as *Fusarium* species, produce mycotoxins that are capable of inducing both acute and chronic toxic effects in livestock. Fumonisin B_1 (FB_1) and zearalenone (ZEN) are two of the most toxicologically important *Fusarium* mycotoxins.

To counteract undesirable effects of mycotoxins in animal feed we developed enzymes that can be mixed into contaminated feed for detoxification of *Fusarium* mycotoxins in the gastrointestinal tract. FUM*zyme*®, a fumonisin esterase, which is registered in the European Union as a feed additive, is an example. The enzyme hydrolyses the two tricarballylic acid side chains of fumonisins, leading to the formation of the non-toxic hydrolysed fumonisins. Another mycotoxin-degrading enzyme is the zearaleonone hydrolyase (ZEN*zyme*®), which exhibits lactonase activity against ZEN, forming the non-estrogenic hydrolysed zearalenone (HZEN) as the primary reaction product.

Application of mycotoxin degrading enzymes in silage is a new approach for the degradation of *Fusarium* mycotoxins during the ensiling process. Here, we tested the FB_1- and ZEN-degrading ability of FUM*zyme*® and ZEN*zyme*® in naturally contaminated corn cob mix silage in two independent trials. In case of FUM*zyme*® the influence of an acid-based feed preservative on the enzyme activity was evaluated additionally. For this purpose, we sprayed the enzymes and the preservative onto the shredded maize kernels. Silos were prepared by putting treated maize kernels (~ 700 g) into plastic bags, applying pressure to the bags to compact the material and subsequently closing the bags airtight with a cable binder. The silos were stored at room temperature for 90 days.

First results indicated that FUM*zyme*® was highly active during silage and FB_1 was fully degraded within 90 days of ensiling, even in silos containing the acid-based preservative.

PP-49 MAPK overexpression in *Fusarium graminearum*—towards characterization of the Mgv1 signalling pathway

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Mitogen-activated protein kinases (MAPKs) are key regulatory enzymes involved in numerous cell processes. They function in a cascade, where upstream a MAP kinase kinase kinase phosphorylates a MAP kinase kinase, which in turn phosphorylates a MAP kinase. The MAP kinase has a myriad of targets that upon phosphorylation have altered activity and/or cellular localization. Three MAPK pathways have been identified in *Fusarium graminearum*, one of the main *Fusarium* species responsible for fusarium head blight (FHB) disease of cereals. *F. graminearum* Mgv1 (MAP kinase for growth and virulence 1) is thought to act in a Bck1-Mkk2-Mgv1 cascade. *Mgv1* knockout mutants in *F. graminearum* have altered trichothecene mycotoxin profile, cell wall integrity, reduced fitness and virulence, and are female-sterile. While it is clear that Mgv1 affects various traits, little is known about the biochemical pathway of this MAP kinase. With the aim of identifying downstream targets of Mgv1, four fungal colonies overexpressing the *Mgv1* gene *in locus* were generated. Characterization of these transformants is presented here. Mycelial growth on potato dextrose agar were compared to the wild type (WT) strain GZ3639. FHB disease assays were carried out in *Brachypodium distachyon*, a model plant for cereal crops, and no difference in pathogenicity was observed between the WT and transformants. Quantitative RT-PCR analysis confirmed up-regulation of the *Mgv1* transcripts, and also revealed down-regulation of the *TRI5* gene involved in trichothecene synthesis. These results may explain the low abundance of the deoxynivalenol mycotoxin detected following HPLC analysis of one of the transformants compared to the WT. No changes were detected in the expression of the *RLM1* gene, a transcription regulator previously shown to interact with Mgv1—although altered expression of *RLM1* is not required for these gene products to interact. The *MGV1* overexpression transformants will be used to further characterize the role of Mgv1 in various pathways, including cell wall formation, and to identify the downstream proteomic and genetic elements of the Mgv1 signalling pathway.

PP-50 *Bacillus velezensis* RC 218 as biocontrol agent of Fusarium head blight: effect on *Fusarium graminearum* penetration and growth on wheat spike

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Fusarium head blight (FHB) causes important yield and quality losses on wheat worldwide. *Fusarium graminearum sensu stricto* is the main pathogen in Argentina and the disease occurs when high humidity conditions prevail during anthesis. Among management strategies to control the disease, biocontrol offers an environmentally friendly tool to be used in the frame of an integrated pest management. In previous studies we have demostrated the effectiveness of *B. velezensis* RC 218 to reduce both FHB severity and DON accumulation. The aim of the present study was to evaluate the effect of *B. velezensis* RC 218 on *F. graminearum* spike infection, penetration and deoxynivalenol (DON) production. For this purpose, wheat plants were grown under greenhouse conditions and inoculated by spray at anthesis stage (DC65) with: i) *B. velezensis* RC 218 and *F. graminearum TRI5prom::GFP* conidial suspension (this strain produces a completely functional TRI5 protein, synthetizes GFP protein under the control of TRI5 gene promoter and shows constitutive expression of *dsRed* gene) and ii) *F. graminearum TRI5prom::GFP*. Spikes were sampled at 14 days post inoculation and freehand cuts were carried out in different symptomatic spike portions (grain, upper and lower rachis internodes and spikelet glumes). GFP and dsRed production was characterized through FV1000 Olympus confocal microscope and images were captured. Results showed that *B. velezensis* RC 218 reduced *F. graminearum* hyphae growth, and DON accumulation was more evident in the grains and its corresponding upper and lower rachis internode. The effect was not observed in spikelet glumes. According to these results, *B. velezensis* RC 218 effectiveness over the pathogen seems to be higher in the spike inner structures, preventing pathogen growth and movement through grains and into the rachis.

PP-51 *Fusarium langsethiae* artificial oat infection under field conditions

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Fusarium langsethiae is a recently described species belonging to the complex of fungi causing *Fusarium* Head Blight in small grain cereals. The epidemiology of this species is poorly understood compared with the more extensively studied species of the *Fusarium* genus such as *F. graminearum*. However, it is known to be a potent producer of T-2 and HT-2 trichothecene mycotoxins and has been implicated in the rising levels of these toxins found in European cereals, particularly in oats for which *F. langsethiae* shows host preference. Infection under experimental conditions has mostly relied on natural infection in the field or artificial inoculation in glasshouse conditions. Here, we describe a method for field inoculation of *F. langsethiae* in oats under the Irish environment. The field trial consisted of 190 oat varieties with 10 checks planted in 1.5m rows in a randomised complete block design with 2 replications. Each variety received three separate inoculations at Zadok's growth stages 55, 60 and 65, using a spore suspension consisting of a mix of three isolates of *F. langsethiae* at 1 x 10^5 conidia/ml in Tween 20 (0.02% v/v in sterile distilled water). Each inoculation consisted of 100ml of spore suspension distributed across each variety row using a pressurised hand-sprayer. Checks received the same volume of Tween 20 solution. After each inoculation, a misting irrigation system suspended above the trial area was used to maintain a high humidity conducive to fungal growth. Mist was produced in the plot area at 20-minute intervals for up to 4 hours after inoculation, depending on prevailing weather conditions. At harvest, 30 main tillers of each variety were hand harvested, cleaned, dried and milled. T-2 and HT-2 contamination and *F. langsethiae* infection analyses will be performed by LC/MS Q-TOF and qPCR, respectively. Preliminary data have successfully shown *F. langsethiae* infection.

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PP-52 The epidemiology of *Fusarium langsethiae* in oats

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The plant pathogenic fungus *Fusarium langsethiae* produces the highly potent mycotoxins HT-2 and T-2. Since these toxins are frequently detected at high levels in oat grain lots, they pose a considerable risk for food and feed safety in Norway, as well as in other north European countries. To reduce the risk of HT-2/T- 2-contaminated grain lots to enter the food and feed chain, it is important to identify factors that influence *F. langsethiae* infection and mycotoxin development in oats. However, the epidemiology of *F. langsethiae* is unclear.

A three-year survey was performed to reveal more of the life cycle of *F. langsethiae* and its interactions with oats, other *Fusarium* species, as well as insects, mites and weeds. We searched for inoculum sources by quantifying the amount of *F. langsethiae* DNA in crop residues, weeds, and soil sampled from a selection of oat-fields. To be able to define the onset of infection, we analysed the amount of *F. langsethiae* DNA in oat plant material sampled at selected growth stages (between booting and maturation), as well as the amount of *F. langsethiae* DNA and HT-2 and T-2 toxins in the mature grain. We also studied the presence of possible insect- and mite vectors sampled at the selected growth stages using Berlese funnel traps. The different types of materials were also analysed for the presence *F. graminearum* DNA, the most important deoxynivalenol producer observed in Norwegian cereals, and which presence has shown a striking lack of correlation with the presence of *F. langsethiae* in oat. Results show that *F. langsethiae* DNA may occur in the oat plant already before heading and flowering. Some *F. langsethiae* DNA was observed in crop residues and weeds, though at relatively low levels. No *Fusarium* DNA was detected in soil samples. Of the arthropods that were associated with the collected oat plants, aphids and thrips species were dominating.

PP-53 Genome wide association studies to identify markers, QTL and genes associated with Fusarium kernel rot resistance in two maize populations

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Fusarium verticillioides infects maize, inciting several diseases including seedling blight, root rot, stalk rot, kernel rot, and ear rot. Interest in *F. verticillioides* has been renewed by the evidence that the fungus can produce the secondary metabolite fumonisins. In this work, a rolled towel bioassay (RTA) was applied for the first time to screen the maize Goodman association panel and the MAGIC population for Fusarium kernel rot (FKR) resistance. For the Goodman association population, phenotypic data were used to perform a Genome Wide Association Study (GWAS) using a set of 227K SNP markers that revealed 164 SNPs associated with the disease severity trait. Furthermore, a set of genes were found associated to the SNPs and many of these having a role in disease response, such as transcription factors, chitinase, cytochrome P450, and ubiquitination proteins. Co-localization between genes found in the analysis and QTL reported for Fusarium ear rot (FER) or fumonisin accumulation resistance revealed that the majority of the genes, associated to the SNPs, localize inside QTL for FER and/or fumonisin accumulation resistance, previously described. Similar genomic/genetic analysis were run for the MAGIC population in order to increase the number of DNA markers and candidate genes for FKR resistance, and further 8 QTL and 3 genes associated to resistance were identified. Additional phenotyping and genotyping on recombinant intercrosses (RIXs) that were produced by crossing MAGIC RILs will shed new light on *F. verticillioides* resistance mechanisms in hybrid backgrounds.

PP-54 A Genome-Wide Association Analysis Study (GWAS) for Fumonisin Content in Maize Kernel

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In current study, GWAS has been performed for the first time for detecting high-resolution QTL for resistance to fumonisin accumulation in maize kernel. A subset of 270 inbred lines from a maize diversity panel (composed of 302 inbred lines) that represents much of the diversity available in public breeding sector around the world was evaluated under inoculation with *Fusarium verticillioides.* Significant associations between polymorphisms at SNPs in bins 1.07, 1.09, 2.08, 3.02, 3.04, 3.05, 3.06, 3.08, 3.09, 4.02, 4.05, 5.02, 6.07, 7.05, 8.07, 9.03 and fumonisin accumulation (determined by ELISA tests) in maize kernel were detected. At least, four associations should be behind novel QTL because QTL were not found in that bin or in adjacent bins in previous studies (Robertson-Hoyt et al. 2006; Maschietto et al. 2017). These novel QTLs for fumonisin content would be at bins 3.02, 5.02, 7.05, and 8.03. Genes likely involved in ROS production, ROS-scavenging and ROS-detoxification, in signaling and/or regulation of plant hormone-mediated responses and lipid metabolism were proposed as candidate genes behind significant associations.

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PP-55 Enzymatic detoxification of fumonisin and zearalenone in the bioethanol process

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The U.S. ethanol industry surpassed the 1 million barrels of ethanol produced per day mark at the beginning of 2017 (U.S. Energy Information Administration). The production of one barrel of ethanol leads to 98 kg of Distiller Dried Grains with Solubles (DDGS) (Bothast & Schlicher, 2005), which is an interesting by-product for the feed industry. Mycotoxins present in the raw materials for the bioethanol production are not degraded but even concentrated in the process. According to the latest results of the BIOMIN Mycotoxin Survey, all 212 corn DDGS samples tested were at least positive for one of the major mycotoxins (aflatoxin, zearalenone, deoxynivalenol, T2-toxin, fumonisin, ochratoxin). The use of DDGS in the feed industry therefore increases the likelihood of exposing animals to higher mycotoxin levels. In the animal, mycotoxin exposure can lead to adverse health effects, decreased animal performance and economic losses.

Counter measures against high mycotoxin concentrations in DDGS comprise rejection of highly contaminated raw materials or dilution by blending high and low contaminated materials. Actual degradation of mycotoxins in the bioethanol production process by application of mycotoxin-degrading enzymes offers a new possibility to tackle mycotoxin contaminations.

Lab-scale tests with mycotoxin degrading feed additives showed the degradation of fumonisin B_1 (FB_1) and zearalenone (ZEN) and the formation of the degradation product hydrolyzed FB_1 and hydrolyzed ZEN proving the detoxification of these two important mycotoxins during the process. FUM*zyme*® added either before liquefaction or before fermentation (60 U/kg corn) during the bioethanol production process with naturally contaminated corn (2324 ppb FB_1) led to a ≥97% reduction of FB_1 in the mash. Likewise, addition of ZEN*zyme*® addition (40 U/kg corn) during fermentation with naturally contaminated corn (1486 ppb ZEN) led to a 89% reduction of ZEN. Mycotoxin degrading additives can be used to produce high quality DDGS with low mycotoxin levels thereby benefitting livestock producers and boosting bioethanol industry's revenues.

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PP-56 Use of *Fusarium* mycotoxin degrading enzymes in corn cob mix silage

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Animal feed contains a wide range of contaminants and toxins. One major factor contributing to feed spoilage is the growth of mycotoxigenic fungi. It is well known that contamination of animal feed with mycotoxins compromises animal health and may lead to carry-over into eggs, meat, or milk. *Fusarium* mycotoxins e.g. fumonisins and zearalenone (ZEN) are among the most common mycotoxins found in feed and food.

Our group developed enzymes that catalyse the degradation of *Fusarium* mycotoxins in feed in the gastrointestinal tract, i.e. the fumonisin esterase FumD (FUM*zyme*®, registered in the European Union as a feed additive) and the zearalenone hydrolase ZenA (ZEN*zyme*). FUM*zyme*® hydrolyses the two tricarballylic acid side chains of fumonisins, resulting in the formation of the non-toxic hydrolysed fumonisins. ZEN*zyme*, on the other hand, exhibits lactonase activity against ZEN, forming the non-estrogenic hydrolysed zearalenone.

The application of enzymes for the detoxification of mycotoxins in feed during the ensiling process seems to be a promising approach. In this study, we investigated the influence of FUM*zyme*® and ZEN*zyme* on certain indicators of silage quality (pH, volatile fatty acid levels and water soluble sugars levels). The trial was set up with naturally contaminated corn cob mix (CCM) in two long-term silage trials, performed in presence and absence of a silage inoculant *(Lactobacillus plantarum, L. brevis, L. kefiri)* and an acid-based preservative.

First results showed that FUM*zyme*® completely degraded fumonisin B_1 during the experiment (not a part of this presentation), without any negative impact on the fermentation of CCM. Acidity of silage and production of desired lactic acid in FUM*zyme*® treated samples was comparable to those of a FUM*zyme*® free control treatment. Addition of silage inoculant stimulated the fermentation and induced a rapid pH drop in presence or absence of FUM*zyme*®.

PP-57 Screening a wheat mutant population for deoxynivalenol sensitive lines

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Fusarium head blight (FHB) is a devastating fungal disease affecting bread wheat and other small grain cereals. It is mainly caused by *Fusarium graminearum* and leads to severe losses in both yield and quality. The fungi can produce mycotoxins such as deoxynivalenol (DON) and other trichothecenes. Mycotoxin contamination of grain poses a global hazard to food and feed safety. FHB resistance is a quantitative trait, the most prominent QTL *Fhb1* is located on wheat chromosome 3BS and confers resistance to fungal spread and to DON. A pore-forming toxin-like gene was identified to confer resistance to fungal spread (Rawat et al. 2016). DON resistance is linked to the formation of the nontoxic DON-3-O-glycoside (Lemmens et al. 2005); however, the gene mediating DON resistance is still unrevealed. But this knowledge is crucial for breeders as the development of DON and FHB resistant cultivars is the most effective way to reduce mycotoxin contamination. In previous studies the *Fhb1* locus could be fine-mapped to a 860 kb sequence comprising 28 genes. To identify DON resistance genes we worked on a mutant wheat population in a forward genetics approach. 1500 EMS mutated lines of the resistant CM-82036 (carrying *Fhb1*) were each infiltrated with DON solution at anthesis. We evaluated the DON resistance level 26 days after the application. Mycotoxin treatment induced typical symptoms such as straw-colored heads in DON sensitive lines, as expected DON resistant lines did not develop any symptoms. In our field experiment we could select 16 mutant lines showing DON induced bleaching of the heads. We sequenced these DON sensitive mutants for the most promising *Fhb1* candidate genes to reveal common mutations. In addition, we confirmed the phenotypic selection of the sensitive mutant lines by Fusarium inoculation and DON infiltration in the greenhouse.

Acknowledgements

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PP-58 Reliable method for *Gibberella fujikuroi* species complex differentiation by matrix-assisted laser desorption/ionization time-of-flight mass spectrometry

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The *Fusarium* species within the *Gibberella fujikuroi* complex are responsible for big economic losses in many market segments due to destructive diseases on cereals and maize and reduction in staple food crop quality. Many of the members are known for the production of mycotoxins in food/feed worldwide, which result in serious human and animal health problems and even death if prolonged exposure occurs. Differentiation of the species is a major challenge considering their similarity and overlapping morphological traits that underestimate the true diversity of the complex. The multi-locus sequencing technology (MLST) can be used to clearly characterize species. However, the technique is time consuming and cost intensive. Matrix-assisted laser desorption/ionization time-of-flight mass spectrometry (MALDI–TOF MS) analysis of subproteome patterns of these fungi can be applied as a promising alternative tool for the differentiation of closely related species. The purpose of this study was to discriminate species in the *Gibberella fujikuroi* species complex (GFSC) using MALDI–TOF MS and to evaluate our database by identifying unknown isolates. Analysis included pre-cultivation on Synthetic Nutrient Agar (SNA) and subsequent cultivation in Sabouraud Dextrose Broth for 48 hours before acetonitrile/formic acid based protein extraction to obtain distinct reference mass spectra for species identification. We established a supplementary database including reference spectra of 50 GFSC species. To evaluate the discriminative potential of the database, more than 200 fungal isolates were investigated with MALDI-TOF MS and identified to species level using transcription elongation factor 1 α (TEF1-α) sequences as reference method. Results showed that MALDI-TOF MS is as suitable and accurate technique for the identification and species-level differentiation of the species within the *Gibberella fujikuroi* species complex as well as an innovative, time efficient alternative to MLST.

PP-59 Dissecting a wheat chromosome in search of a Fusarium head blight susceptibility factor

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In the effort to protect wheat from fungal diseases, such as Fusarium head blight (FHB), genes promoting resistance are generally sought. However, there is increasing evidence to suggest that the fungus can exploit plant physiological processes to aid its infection. The removal of such genes promoting susceptibility offers an alternative means to enhance FHB resistance.

We have identified a susceptibility factor on the short arm of wheat chromosome 4D that compromises resistance to spread of the fungus in the head. We conducted both point inoculation and DON application experiments on Chinese Spring and ditelosomic lines (missing 4DS or 4DL) to establish whether the factor is conferring susceptibility to the fungus, or to DON mycotoxin, a known virulence factor. When 4DS is missing, there is increased resistance to the fungus, but an increase in susceptibility to DON (observed both as bleaching severity and its effect on grain weight). This suggests the presence of a DON resistance factor, in addition to the fungal susceptibility factor, on 4DS. We also carried out a spray inoculation experiment on these lines, to identify whether the susceptibility factor has a type I (initial infection) or type II (infection spread) effect. The loss of 4DL resulted in much higher type I susceptibility, suggesting there is a type I resistance factor situated on 4DL. The line missing 4DS showed a similar phenotype to the wild type, suggesting that the susceptibility factor does not influence type I resistance.

To refine the positions of DON resistance and fungal susceptibility factors, we used Chinese Spring material possessing terminal deletions of 4DS varying in size. From these data we identified a ~33 Mbp region on 4DS containing the susceptibility factor; distinct from the DON resistance factor which is positioned on the distal third of 4DS. We have obtained 4DL terminal deletion lines to refine the position of the 4DL type I resistance factor.

We designed markers, capable of detecting deletions across 4DS and used a subset to screen a gamma-irradiated Paragon population for deletions within the susceptibility factor region. From this screen we identified lines containing both homozygous and heterozygous deletions, varying in size and position, within the susceptibility factor region. These lines provide the means of further refining the position of the susceptibility factor.

Our findings to date reiterate the complex polygenic situation underlying FHB resistance.

PP-60 Diversity and incidence of *Fusarium* species on maize under different pesticide treatments

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Fusarium ear rot (*Fusarium* spp.) and the European corn borer (*Ostrinia nubilalis*) are two major yield reducing factors in maize production. It has been well documented that *O. nubilalis* can enhance Fusarium ear secondary infections (Blandino et al, 2015). The aim of this study was to monitor the influence of different pesticide treatments (a.i. Chlorantraniliprole, Indoxacarb, and Chlorantraniliprole + Lambda-Cyhalothrin) on *O. nubilalis* and *Fusarium* disease severity in the field in a four year trial (2013-2016). The diversity of *Fusarium* species and their incidences on infected kernels were also estimated. Fusariosis in the field was assessed on a total of 160 plants per treatment, according to Reid et al. (1996) scale, while disease severity was expressed as McKinney's Index. For further analyses, 40 ears with fusarium ear rot symptoms were randomly chosen per treatment. The diversity of *Fusarium* species was analysed on 400 kernels per treatment, according to standard laboratory procedure, and morphologically identified according to Leslie & Summerell (2006).

In total 11 *Fusarium* species were identified on maize kernels in a four year period: *F. equiseti, F. graminearum, F. oxysporum, F. proliferatum, F. pseudograminearum, F. semitectum, F. solani, F. sporotrichioides, F. subglutinans, F. verticillioides*, and *Fusarium* sp. As expected, the dominant species were: *F. proliferatum, F. verticillioides* and *F. graminearum*.

Low abundance of *O. nubilalis* and fewer injuries on kernels were registered in 2014 and 2015, which led to low differences in disease severity between treatments (15.88-17.24 and 10.72-13.04, respectively), and no significant effects of insecticides comparing to the control treatment. However, the years 2013 and 2016 were more favorable for the pests development and disease severity varied from 7.74-14.09 in 2013, and 8.18-11.55 in 2016. Significant differences in the efficacy of the insecticides compared to the control were registered in all treatments, but the most efficient a.i. was Chlorantraniliprole.

PP-61 Genetic resources for maize breeding programmes to improve ear rot resistance in Poland

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In Poland, during the last years the maize growing area has significantly increased because of the high demand from fodder industry - total area is about 1200K ha. Maize grain, being similar feed value to wheat, is much cheaper. Red and pink ear rots caused by *Fusarium* spp. are important factors affecting the yield and it's quality, mainly because of it's contamination with mycotoxins produced by fungi. It was observed, that during the last years contamination of these toxins increase also in Poland.

Based on our study conducted in 2015 and 2016, when 102 samples of commercially grown maize cultivars were collected in 14 localities throughout Poland, it was possible to determine that *F. verticillioides* was the most abundant species, followed by *F. proliferatum* and *F. graminearum* (60%, 11% and 12% of all *Fusarium* isolates, respectively). A number of minor species were also present: *F. temperatum*(5,0%), *F. poae*(2%), *F. subglutinans*(8%), *F. sporotrichioides*(1,0%). DON contamination in 22% of evaluated grain samples exceeds 1750 µg/kg, FUM in 5% of evaluated grain samples exceeds 1000 µg/kg and ZEA contamination in 7% samples was higher than 350 µg/kg. Differences between localities and varieties were significant.

Because of this, the major objective of this study was to determine effectiveness of the recurrent selection to develop maize flint and dent genotypes resistant to the ear rot based on genetic resources used in Polish breeding programmes. Two separate gene pools were evaluated: SH and KOB. The earliest flowering $S_{0:1}$ were self-pollinated and kernel inoculated with *F. gramninearum*. Disease development was visually assessed at harvesting time using 1-7 scale. Next, all genotypes scored within the range 2-3 were selected to continue the selfing procedure to obtain $S_{1:2}$, $S_{2:3}$ and $S_{3:4}$ lines. Additionally, anthocyanin content in corncobs, silks and pollen was described. Selection effectiveness was much higher within the gene pool SH ($S_{1:2}$: 51%, $S_{2:3}$: 58,9%, $S_{3:4}$: 87,2%) than within KOB pool ($S_{1:2}$: 56%, $S_{2:3}$: 57,9%, $S_{3:4}$: 59,6%). Ear rot resistance of selected S_4 lines was confirmed under field conditions after inoculation and based on DON contamination determined by RIDA QUICK SCAN. It was possible to conclude that DON content positive correlate with ear rot. Disease severity and DON contamination, negatively correlated with anthocyanin content in silks.

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PP-62 *Fusarium* head blight in German oats - genetic, toxigenic and phenotypic diversity of *Fusarium langsethiae, F. sporotrichioides* and *F. poae*

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Information on *Fusarium* head blight in German oats is still scarce regarding symptom development, the predominant causal agents, and mycotoxin contamination Therefore, we analysed the occurrence and diversity of *Fusarium* spp. and its mycotoxins in oat kernels from different locations in Germany in a 3-year monitoring. *Fusarium* species were identified based on spore morphology and the analysis of the elongation factor alpha region after sequencing. Sequences were further used to analyse phylogenetic relation among the obtained species. Mycotoxins in oat kernels were determined with HPLC-MS/MS. Although the frequency of *Fusarium* spp. varied between years, the predominant species was always *F. poae*, followed by *F. sporotrichioides,* and *F. langsethiae*. Further, we found that nivalenol (NIV) and T-2/HT-2 were among the most dominant mycotoxins in oat kernels. In greenhouse trials we tested the aggressiveness and mycotoxin production of the 3 predominant *Fusarium* species. According to our data, *F. sporotrichioides* was more aggressive than *F. langsethiae* under greenhouse and field conditions, which suggests that *F. sporotrichioides* is the main producer of T-2/HT-2 in German oats. Further, the results of this study indicate that *F. poae* might be responsible for high NIV levels in German oats. Over 40% of the analysed oat kernels were colonized with *F. poae* leading to frequently high levels of NIV with more than 900 µg/kg. Interestingly, the mycotoxin content was reduced by up to 90% when kernels were dehulled. However, the results of this study highlight that *Fusarium* head blight in oats is a problem of significant importance in Germany, which needs more attention in future research.

PP-63 Positive-negative selectable markers based on fusion genes allow efficient marker-free gene editing in *F. graminearum* as well as *Cre*-loxP aided recycling of transformation markers for consecutive knockouts

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Marker-free genetic engineering of filamentous fungi, such as F. graminearum, has proven to be a laborious task in the past, mainly due to the inability to efficiently recycle previously integrated markers. To amend this shortcoming, a series of bifunctional marker genes was generated by N-terminal fusion of the Herpes simplex virus thymidine kinase (*HSVTK*) to the resistance markers *HPH* (hygromycin B), *NPTII* (G418) and *NAT1* (nourseothricin). These fusion genes can be selected for using the respective antibiotics in the fungus, and also in *E. coli*, while additionally allowing forced excision of the marker via 5-fluoro-2-deoxyuridine (FdU) counterselection. Here, we demonstrate the applicability of HSVTK fusion genes by constructing a *TRI8* allele-swapped *F. graminearum* PH-1 transformant. *TRI8* encodes an esterase that uses 3,15-diacetyl-deoxynivalenol as a substrate, and cleaves off an acetyl group from either position 3 or position 15, depending on the allele. PH-1 possesses the *TRI8* allele responsible for producing 15-acetyl-deoxynivalenol (15-ADON), however, the 3-acetyl-deoxynivalenol (3-ADON) chemotype is also frequently encountered in the wild.

An isogenic PH-1-derived strain, carrying the 3-acetyl *TRI8* allele, was generated in two steps. First, *TRI8* was deleted by integration of the *HSVTK-NAT1* marker, after which the marker was replaced with the 3-acetyl *TRI8* allele via homologous recombination. The resulting strain no longer possessed the nourseothricin resistance of its precursor Δ*tri8:HSVTK-NAT1* strain, and was PCR-verified on both flanks for correct integration. As expected, the chemotype was switched from 15-ADON to 3-ADON production. Furthermore, we constructed a *F. graminearum* PH-1-derived mutant that carries seven consecutive gene deletions using the *Cre*-loxP system for marker recycling. Treatment of protoplasts with recombinant *Cre* recombinase removes loxP-flanked cassettes very efficiently, reducing the screening effort required to obtain marker-free deletion strains.

PP-64 Developing FHB resistance and lowered DON content in two-row barley at the Agriculture and Agri-Food Canada, Brandon Research and Development Centre

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Fusarium head blight (FHB) epidemics, caused primarily by *Fusarium graminearum* Schwabe, occurred on barley (*Hordeum vulgare* L.) in the western Canadian province of Manitoba in the mid-90's. Since then the pathogen has progressively spread westward to occupy all western Canadian, barley-growing regions. A long-term effort has been undertaken at the Agriculture and Agri-Food Canada, Brandon Research and Development Centre to breed resistant barley varieties for lower deoxynivalenol (DON) content. Over the years, many exotic genotypes have been used in crosses to introgress resistance. Large efforts have also been conducted to expose genetic variation through application of *in vitro* selection. A large FHB nursery at Brandon, MB has been essential to phenotype elite and developmental germplasm. Progress has been limited for fact that many of the current resistant sources in barley only confer partial resistance and are linked with undesirable agronomic and quality characteristics. While many difficulties exist, several varieties have been released that represent a balance of improved resistance with advanced agronomics and maintenance of high quality. New breeding tools are being investigated for application within the breeding program to continue progress on developing resistance to this economically important disease, such as use of genome-wide molecular markers (SNP) and next generation sequencing (RNA-Seq). Developing FHB resistant varieties with lowered DON content continues as an extremely high priority for the breeding program, and it is hoped that new breeding tools may hasten this activity.

PP-65
Monitoring of the content of mycotoxins in the feedstuffs of the central zone of the Russian Federation for 2015-2016

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Analysis of various feedstuffs for mycotoxins maintenance is conducted in the laboratory of the All-Russian Scientific and Technological Institute of Poultry with the support of the Russian branch of the company "Biomin". The purpose of this work was to monitor the content of mycotoxins in feeds entering the laboratory from farms and enterprises in the Russian Midlands. Screening of feed samples for the content of mycotoxins was performed by ELISE, using test - systems AgraQuant® ELISA (Romer Labs, Austria).To confirm the results of the ELISA screening, as well as to conduct routine tests for mycotoxin content in feed, a method the HPLC-MS / MS system-tandem liquid chromatography-mass spectrometry was used (Agilent Infinity LC Systems 1290 and AB SCIEX Triple Quad ™ 5500).The method used for the analysis "dilute and shoot". During the period 2015-2016 about 600 samples were analyzed.Cereals were analyzed: wheat - 17%, corn - 12%, barley - 6%. The majority of samples were compound feeds: for poultry - 27%, for pigs - 18%, for cattle-4%. The rest 16% are meals, oilcakes, haylage and silage. The analyses revealed the presence of trichothecene mycotoxins type A: HT2 toxin in 79% of the samples, T2 toxin in 76%, Diacetoxyscirpenol in 4% of the samples; trichothecene mycotoxins type B: Deoxynivalenol in 70% of the samples, Nivalenol in 24%, Deoxynivalenol-3 glycoside in 22% of the samples. Ochratoxin A was also found in feed in 51% of samples, Fumonisin B1 in 37%, Fumonisin B2 in 19% and Fumonisin B3 in 15% of samples. A significant part of the feed was contaminated with zearalenone(69% of all samples). Aflatoxin B1 and Aflatoxin G1 were detected only in trace amounts in 4% of the samples. Maize became the leader in the diversity and quantity of mycotoxins. The greatest concentration of T2 toxin in maize was 424 mkg / kg, HT2 toxin- 689 mkg / kg, Fumonisin B1- 6821 mkg / kg, Fumonisin B2-1808 mkg / kg, Fumonisin B3-681 mkg / kg, Zearalenone 277 mkg / kg. High concentrations of B-type trchotecene mycotoxins were found in samples of wheat. The greatest amount found for Deoxynivalenol was 1940 µg / kg, and practically all samples contained Nivalenol. The highest concentration of Zearalenone in wheat was 220 µg / kg. However, the highest concentration of Deoxynivalenol was found in barley (8985 µg / kg). This amount was found only in one sample, all other barley samples differed relative well-being in the content of mycotoxins.

PP-66 Nutrient preferences in *F. langsethiae* revealing competitive skills

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Fusarium langsethiae is one of the Fusarium head blight fungi and the main causal agent for T-2/ HT-2 contamination in small grain cereals in the Nordic region and the UK. Despite its slow growth and low aggressiveness, *F. langsethiae* infections persist in the field, some years causing fatal levels of mycotoxin contamination in oats.

Overall, available data indicate that *F. langsethiae* benefits from molecular strategies and/or pathological and epidemiological niches different from that of *F. graminearum*. Epidemiological data show that *F. langsethiae* has a strong preference for oats, and the occurrence of T-2/HT-2 and DON in oats, more often than not, is mutually exclusive, indicating that strong co-infection of both fungi is not likely to occur in the field. It has also been suggested that *F. langsethiae* predominates over *F. graminearum* in dry summers, suggesting that it might tolerate a dryer climate.

In order to investigate possible nutritional preferences for *F. langsethiae* compared to other Fusarium species (especially *F. graminearum*), we conducted a pilot experiment investigating *F. langsethiae* growth over a range of different carbon and nitrogen sources under a constant temperature of 18°C. A minimal medium was used as a base, and selected carbon and nitrogen sources were added to a final concentration of 5 mM for amino acids and 20 g/L for carbon compounds. Care was taken to choose components particularly available in plants, and enriched in oat compared to wheat. We also tested drought tolerance by incubation of *F. langsethiae* and other *Fusarium* species on potato dextrose agar (PDA) with decreasing water activities, adding salt (NaCl).

Our results show that *F. langsethiae* is equally (or more) sensitive to reduced water activity/increased salt concentration as are other *Fusarium* species, and is therefore not likely to be more drought resistant. Furthermore, our data indicate that *F. langsethiae* may have advantages at low temperatures given certain nutritional preferences, and that, under favorable conditions, it may be able to compete with *F. graminearum*. Some of these favorable nutritional needs may be met in oats.

PP-67 Oats for the future

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Fusarium langsethiae is an increasingly important pathogen of oats in Nordic countries, Central Europe, United Kingdom (UK) and Ireland. *F. langsethiae* can produce T-2/HT-2, which are type A trichothecenes; they can inhibit protein synthesis, cell proliferation and cause acute or chronic intoxication of humans and animals. In this study, we screened oats for resistance to *F. langsethiae;* we screened 12 UK cultivars, 8 of which are currently recommended by the UK Agriculture and Horticulture Development Board (AHDB, 2018), and 26 uncharadcterised heritage Irish oat varieties. These genotypes were grown under glasshouse conditions and at flowering heads were inoculated with *F. langsethiae*. At harvest time, the grains were analysed for yield, fungal DNA and toxin levels. Yield analysis showed there is no effect of *F. langsethiae* on seed quantity or weight. Fungal DNA quantification using real-time PCR (qRT-PCR) revealed potential resistant and susceptible oat cultivars, with fungal DNA values ranging between 0 to 0.025 pg fungal DNA/ng plant DNA. Fungal DNA was detected in 11 out of 12 UK cultivars (0 – 0.016 pg fungal DNA/ng plant DNA), with the highest fungal DNA quantity found in the cultivar Yukon, which is in the recommended list 2018. In the Irish heritage cultivars, fungal DNA was detected in 24 out of 26 cultivars, with values ranging 0.0008 – 0.025 pg fungal DNA/ng plant DNA. Subsequently, using LC/MS Q-TOF, T-2 concentration was quantified in these cultivars, which ranged between 0 to 108 ppb, with the highest levels in cultivar Harmony. Results showed weak correlation (P<0.05, Spearman's Rho = 0.35) between *F. langsethiae* DNA and T-2 concentration in the whole population. This experiment is being repeated several times in order select material for further breeding work.

PP-68 Species specific impact of *Fusarium* ssp. on *Asparagus officinalis* crown and root rot

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Asparagus is an economically important vegetable in many countries, including Germany. Plant production can be severely limited by soil borne pathogens, resulting in large yield losses and decreased quality of asparagus spears over the typical growing period of 10 years. One major cause is attributed to infections of roots with *Fusarium* species, the causal agents of Fusarium wilt and the crown and root rot disease. The most important *Fusarium* ssp. in this regard are *F. oxysporum* f.sp. *asparagi*, *F. proliferatum* and *F. redolens*. Infections result in necrotic lesions on root and stem surfaces, rot of roots, rhizomes and stems, reduced plant size, and wilt of the ferns. Chemical control of the disease is difficult once plants are infected with *Fusarium*. Thus either preventive measures need to be developed to inhibit infection, or breeding of resistant cultivars should be targeted. In order to get deeper insight into the interaction between the pathogen and asparagus cultivars on the molecular level, we have established a host pathogen system. Asparagus seedlings of two different cultivars were grown six weeks under controlled conditions. Roots were then inoculated with isolates of *F. oxysporum* f.sp. *asparagi*, *F. proliferatum* and *F. redolens*, subsequently plants were potted in a soil/quarz-sand mixture and cultivated for eight more weeks. Disease development and pathogenicity of the isolates were evaluated eight weeks after inoculation. While no clear disease symptoms became visible and measurable in the above ground parts of both plant cultivars, a significant impact of the fungal infections on the roots was already evident. Reduced root fresh and dry mass and necrotic lesions were detectable for each isolate tested. Based on available sequence information qPCR protocols were developed to clearly identify and quantify species specific fungal colonisation. In addition, an automated analysis pipeline of root images was designed to determine root morphology alterations in response to fungal infection. Using such systems we will gain better knowledge about the diversity in the interaction process of asparagus with different species of *Fusarium*.

PP-69 Two approaches for the detection and quantification of Fusarium Head Blight on common wheat: hyperspectral imaging on ears and multispectral imaging on kernels

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The aim of Ecophyto IRIGAM program was to compare different methods for phenotyping Fusarium Head Blight (FHB) on different samples: plant, seed and flour. Spectral imaging has been used to improve the evaluation of varietal resistance to Fusarium Head Blight of common wheat.

On ears, the objective was to identify the most relevant wavelengths to discriminate healthy ears from scabbed ears, using a hyperspectral camera (400nm-1000nm). Subsets of between two and five discriminating wavelengths were identified using three chemometric methods. Four subsets of wavelengths were validated on 20 cultivars in two inoculated trials by *F. graminearum* for 2 years. Significant correlation was observed between scabbed area rate obtained by spectral analysis and scabbed ears observed in field: R^2>0.91 in 2016 and R^2>0.87 in 2017**.** The study of phenological stages has shown that these wavelengths are accurate between 300°C/d and 500°C/d after inoculation. This method of classification by hyperspectral imaging will be transferable to other hosts/bioagressors. The next step will be to use these selected wavelengths to quantify the FHB on ears directly in the field.

On kernels, the algorithm "Fusaspectral soft wheat", was developed by GEVES to quantify the Fusarium Damaged Kernels (FDK) by using multispectral imaging. The aim is to replace in-field scorings and mycotoxin measurements for DON content on kernels, measured by LCMS-MS. This tool showed good results in case of *F. graminearum* prevalence*:* FDK was strongly correlated with Fusarium spikelet levels and with the fungal mass of *F. graminearum,* FDK was also a satisfactory predictor of DON. In the frame of IRIGAM project, 3 replicates of 100 common wheat cultivars of breedwheat collection were assessed by VideometerLab3^R and studies of correlations are ongoing.

PP-70 Distribution of secondary metabolite biosynthetic gene clusters in 343 *Fusarium* genomes

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Fusarium consists of over 200 phylogenetically distinct species, many of which cause important crop diseases and/or produce mycotoxins and other secondary metabolites (SMs). Some fusaria also cause opportunistic infections in humans and other animals. To investigate the distribution of biosynthetic gene clusters (clusters) responsible for synthesis of mycotoxins and other SMs, we conducted phylogenomic analyses of 343 genome sequences that represent 26 single and multi-species lineages of *Fusarium*. Using antiSMASH and OrthoFinder, we identified 15,647 putative clusters, and observed tremendous variation in presence and absence of homologous clusters among the genomes. We noted three patterns of cluster distribution: limited, wide and sporadic. Among the clusters identified in the genomes were 192 that are likely to be responsible for the synthesis of sphinganine-analog metabolites (SAMs), a class of metabolites that disrupt sphingolipid metabolism and that include fumonisin mycotoxins. We based the identification of the SAM clusters on the presence within a putative cluster of genes encoding three enzymes that we predict are required for synthesis of all SAMs: a reducing polyketide synthase, an amino transferase, and a dehydrogenase. Phylogenetic analyses resolved the putative SAM clusters into six homolog groups: the fumonisin cluster and clusters SAM1 – SAM5. We propose that each of these six clusters confers the ability to synthesize a family of structurally distinct SAMs. Our analyses also suggest that some SAM clusters have undergone horizontal transfer among *Fusarium* species, and indicate that two or more SAM clusters occur in some *Fusarium* genomes. In addition, SAM clusters tend to have limited and/or sporadic distributions among *Fusarium*. The SM products of clusters SAM1 – SAM5 are not known, but we predict that SAM5 is responsible for synthesis of 2-Amino-14, 16-dimethyloctadecan-3-ol, a SAM previously reported in *F. avenaceum*. The somewhat common occurrence of SAM clusters among the 343 *Fusarium* genomes suggests that the ability to disrupt sphingolipid metabolism provides an ecological advantage to many but not all fusaria.

PP-71 Exploiting natural and induced variation to increase Fusarium head blight and brusone resistance in wheat

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Fusarium head blight (FHB) and wheat blast (brusone) are two major causes of wheat yield loss in Brazil. *Fusarium graminearum* is the main causal species of FHB in Brazil and brusone is caused by *Magnaporthe oryzae* (*Triticum* pathotype MoT). Identifying sources of resistance to both diseases is critical for continued wheat production in Brazil. Two F_6 populations of Brazilian varieties (Anahuac × BR18 and BR18 × BRS 179), were developed at the John Innes Centre (JIC). Anahuac is susceptible to brusone, whilst BR18 displays moderate resistance yet is susceptible to FHB. Contrastingly, BRS 179 is resistant to FHB and susceptible to brusone. The populations were phenotyped for FHB at JIC in 2016 and 2017 and for brusone resistance using a detached leaf assay in 2018. Genetic maps were constructed for QTL analysis.

An FHB QTL explaining up to 11.8% of the phenotypic variance was identified on 5B in the Anahuac × BR18 population across trial years, with Anahuac conferring susceptibility. QTL associated with DON accumulation were identified on 3B, 4D, 5B and 7A but were not consistent across trials. Detached leaf assays for brusone identified a QTL on 2B with susceptibility associated with Anahuac, suggesting that within this population FHB adult plant resistance and brusone seedling resistance do not co-locate. In the BR18 × BRS 179 population two FHB QTL were identified on 2A and 7A, with susceptibility being associated with the BR18 allele. A single QTL for DON accumulation was detected in both trial years on 4B. The 4B QTL explained up to 14.9% of the phenotypic variance and was not co-incident with QTL for disease symptoms. Detached seedling assays are currently being undertaken to determine genomic regions associated with brusone resistance in this population.

Further FHB and brusone field phenotyping will be carried out in Brazil. This will allow QTL for both diseases to be identified across several environments and will identify whether QTL associated with adult plant resistance for both FHB and brusone are co-incident. Determining if increased resistance to FHB increases susceptibility to brusone, or vice versa, is critical to ensure there is no potential for trade-off when introgressing resistance to these diseases into elite Brazilian cultivars.

PP-72 Identification, phylogeny and fumonisin potential capability of *Fusarium fujikuroi* species complex strains isolated from Brazilian rice grains

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Members of the *Fusarium fujikuroi* species complex (FFSC) are causal agents of bakanae disease in rice and, among them, many species can produce fumonisins, a harmful group of mycotoxins. In Brazil, bakanae was reported in 1970 and since then symptoms of this disease were no longer observed in the field. However, in 2013, low levels of fumonisin B1 were reported in samples of commercial rice in the State of São Paulo. These reports supported the hypothesis that species of the FFSC are associated with rice grains in Brazil. The study aimed to identify the FFSC species associated to bakanae symptomized plants, establish phylogenetic relationships among the species identified and verify in each Fusarium strain, the presence of the *FUM1* and *FUM8* genes that are essential for synthesis of the 20-carbon-long chain that forms the backbone of fumonisin. A collection of 109 isolates of FFSC was obtained from rice grains from the major producing regions in Brazil. Based on morphological traits and phylogenetic analysis of *TEF*, *CAL* and *TUB* gene fragments we identify eight members belonging to FFSC: *F. andiyazi*, *F. anthophilum*, *F. begoniae*, *F. fujikuroi*, *F. proliferatum*, *F. pseudocircinatum*, *F. sterilihyphosum* and *F. verticillioides*. Also, strains that could not be assigned to any known species were analyzed. Finally, *FUM1* and *FUM8* genes were detected in a significant amount of strains. These preliminary results emphasize the toxigenic potential of from Brazilian *Fusarium* isolates occurring on rice and suggest of both monitoring rice at a wider level and developing control strategies and prevention in the field. Studies for assessing the mycotoxin profile of these species, with special attention to fumonisins, are in progress as well as studies aimed to evaluate the conditions leading to fumonisin production by the Brazilian FFSC species will be carried out.

PP-73 Towards elucidating the structure and role of the *in planta* produced *Fusarium graminearum PKS15*-metabolite

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The genome of *the* plant pathogen *Fusarium graminearum* contains a large number of predicted secondary metabolite biosynthesis gene clusters, for most of which still no corresponding metabolites are known. Particularly genes expressed only during plant infection may be involved in synthesis of compounds with a role in virulence, but obviously such compounds escaped classical identification procedures based on analysis of mutants grown on axenic media or on autoclaved rice cultures. The *PKS15* gene encodes a non-reducing polyketide synthase solely expressed *in planta*. To test the impact on virulence, the *PKS15* gene (FGSG_04588) was disrupted in the *F. graminearum* strain PH-1 and virulence tests on the wheat cultivar Apogee were performed. In agreement with [1] no reduced virulence was found, but according to our preliminary results, *pks15* deletion mutants displayed even slightly increased virulence. Connolly et al. reported that *PKS15* (called *PKS29* in their study, [2]) and several other secondary metabolite clusters were constitutively expressed in mutants with a deletion of a histone K27 methyltransferase gene (*KMT6*). Our constructed *kmt6* mutant, despite overexpressing multiple secondary metabolites, was non-pathogenic (avirulent) on Apogee wheat, and likewise the *kmt6 pks15* double mutant was not able to spread from the infection site. The constructed strains are now utilized to search for metabolites present in the extracts of *kmt6* cultures but absent in extracts of *kmt6 pks15* double mutants. Results of the metabolome analysis will be presented.

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PP-74 Response of a collection of Kenyan and Ethiopian wheat lines to Fusarium head blight

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Fusarium head blight (FHB) has emerged as a threat in wheat growing regions worldwide, including in Africa. Kenya and Ethiopia have reported increased epidemics of Fusarium head blight in the past decade. The increase of Fusarium head blight in African countries appears to have been driven by similar factors to those that have contributed to an increase in FHB in other parts of the world, including the adoption of conservation tillage practices and the increased production of alternative hosts, including maize. Little work has been done to identify the response of African wheat germplasm to FHB.

A collection of 215 lines, representing commercial wheat varieties and advanced lines in Kenyan and Ethiopian breeding programs, was screened for response to *Fusarium graminearum* in the United States. The lines were screened in replicated, inoculated, and mist-irrigated field nurseries at two locations in Minnesota USA over two years, providing three location years of data. The field nurseries were established as randomized complete blocks with three replicates using single row plots (ca. 1.5 meters long). Plots were inoculated twice, first at anthesis and then three days later, with a suspension of *Fusarium graminearum* macroconidia (100,000 macroconidia/ml; 50 ml/plot). FHB incidence and FHB index were visually assessed 21 days after inoculation. Approximately 30 heads were hand-harvested from each row at maturity and threshed. The harvested grain was used to estimate the percentage of visually scabby kernels (VSK) and then the grain was ground and submitted for mycotoxin analysis.

Significant differences were observed among the germplasm tested for all variables examined though, as has been in other FHB screening nurseries, maturity class was observed to impact disease development with both early and late lines escaping the disease. Despite the impact of heading date on disease development, a range of FHB responses were observed within the main maturity groups. Significant correlations were recorded between FHB severity, VSK and DON.

Our results suggest that there is native resistance within the germplasm screened in this study and that a number of resistant lines may be of value to wheat breeding programs in East Africa.

PP-75 *Evaluation of integrated control for managing Fusarium Head Blight and DON content in spring wheat in Lithuania*

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Fusarium head blight (FHB) is one of the important fungal diseases of grain crops in Lithuania at recent years. The disease periodically causes significant yield loss and reduced grain quality because FHB species complex produces mycotoxins. The timing of wheat anthesis greatly influences the severity of wheat FHB, because the wheat is susceptible to infection only for a short period around anthesis, when weather is warm and wet. Earlier studies in Lithuania have been shown that FHB especially affecting spring wheat and the highest deoxynivalenol (DON) quantities were identified in the grain of this crop. DON contamination of crops may cause economic losses at all levels of the food and feed production chain. Spring wheat is the largest spring cereal in the country and the quality of grains plays a significant role in market of grains, therefore great attention to grain quality management during season must be paid.

Anthesis growth stage in spring wheat is usually at beginning of July when warm and rainy weather prevail in Lithuania, therefore favourable weather conditions may favour infection and high disease pressure in this crop. No single disease management measure will provide effective control of the disease, especially if environmental conditions favour disease development - an integrated approach needed. The experiment in Institute of Agriculture, Lithuanian Research Centre for Agriculture and Forestry (LAMMC) was set up in to evaluate the efficacy of chemical control against FHB and influence on (DON) content in spring wheat, grown with the presence of wheat and maize crop residue on soil surface. The highest severity of FHB in spring wheat, grown with maize crop residue on soil surface, was established. No any crop residue on soil surface (ploughing) showed 11.6% and 24.3% lower DON content than the with wheat and maize residue respectively. Treatments at the anthesis with fungicide prothioconazole significantly reduced both FHB disease severity and DON content in the grains as compared to the untreated control with the wheat and maize crop residue on soil surface. Diseases severity and DON content in sprayed with fungicide crop with maize residue were higher compared to crop with wheat residues. Consequently, chemical FHB control and DON content reduction in spring wheat is more complicated when previous crop residues on soil surface is maize than that wheat.

PP-76 QTL for Fusarium Ear Rot in a Multi-parent Advanced Generation Intercross (MAGIC) population of maize

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Multi-parent Advanced Generation Intercross (MAGIC) populations offer intermediate characteristics between bi-parental and association mapping populations. MAGIC populations enclose higher polymorphism diversity than bi-parental populations, but keeps detection power at higher levels than in association mapping populations. On the other hand, resolution is greatly improved compared to bi-parental populations due to inter-mating of multiple founders for several generations before self-crossing. A MAGIC population of approximately 700 inbred lines was generated after inter-mating for six generations a synthetic variety composed of eight inbred lines with contrasting susceptibility to Fusarium ear rot (FER). A subset of 339 inbred lines of this MAGIC population have been genotyped by genotyping by sequencing (GBS) and evaluated for resistance to FER; polymorphisms at 9 SNPs were significantly associated with resistance to FER. Significant SNPs are mapped at 53103370 and 55250804 bp in chromosome 2; at 216876071 bp (bin 3.09) in chromosome 3; at 164651229, 164651230, and 164651232 bp (bin 5.04) in chromosome 5; at 156669384bp (bin7.04) in chromosome 7; and at 45294620 and 45294624 bp in chromosome 9 (RefGen_v2). The region 160-175 Mbp in chromosome 5, where three significant SNPs were located in the current study, deserves special attention because it seems to contain allelic variability relevant for differences for resistance to FER in many backgrounds.

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PP-77 Occurrence of *Fusarium* species associated with the Mediterranean perennial alfa grass (*Stipa tenacissima*) in Tunisia

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In this work, we investigated the diversity of *Fusarium* species associated with the indigenous alfa grass (*Stipa tenacissima*), which is considered one of the most interesting plants for fiber production in papermaking. A total of 324 isolates was collected from roots and stems of alfa grass in six different sites in central Tunisia in April 2011 and 2012. Based on morphological characteristics, isolates were classified into eleven species; *F. equiseti, F. polyphialidicum*, *F. compactum*, *F. solani*, *F. oxysporum*, *F. scirpi*, *F. acuminatum*, *F. proliferatum*, *F. nygamai*, *F. chlamydosporum* and *F. burgessii*. Portions of the translation elongation factor 1-alpha (*TEF* 1-α) gene was sequenced for 58 isolates including, at least one representative isolates of each species. Sequence data were compared against both the Fusarium multilocus sequence typing (MLST) database and the Fusarium-ID database. Except for some isolates, BLAST results revealed generally a linear consensus between morphological identification and molecular identification. Two isolates of *F. oxysporum* were classified as *F. redolens* (99.8 % homology) and *F. flocciferum* (97.8 % similarity) when compared to both databases. The TEF1 region of one isolate of *F. burgessii* showed 90.4 % homology with *F. hostae* when compared to Fusarium-ID database whereas it showed a similarity of 98.7 % with *F. burgessii* when compared to the MLST database. Moreover, the sequences of two isolates that were morphologically identified as *F. acuminatum* and *F. compactum* showed high homology with *F. tricinctum* species complex (95.9 %) and *F. brachygibbosum* (99.9 %), respectively.

These results demonstrate a great biodiversity of the community of *Fusarium* spp. population associated with alfa grass in central Tunisia.

PP-78 Identification of the first steps in phenalenone pigment biosynthesis in *Fusarium solani*

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Most *Fusarium* species produce bikaverin and aurofusarin for mycelium pigmentation and fusarubins for perithecial pigmentation [1]. However, *Fusarium solani* produces fusarubins during mycelial growth and another unknown pigment during sexual reproduction. This unknown pigment is predicted to be biosynthesized by a non-reducing polyketide synthase (PKS35 = PKSN [2]). *PKS35* encodes a 2106 aa protein and share 52% identity to PhnA from *P. herquei*, which catalyzes the first step of the herqueinone biosynthetic pathway [3]. Genetic analyses of the *PKS35* gene cluster suggests that it contains eleven genes, of which four are present in the herqueinone cluster.

Herqueinone contains tricyclic phenalenone core ring structure cyclized by a FAD-dependent monooxygenase. To unravel the biosynthetic products in the *PKS35* pathway the intron stripped *PKS* was cloned and put under control of a galactose inducible promoter in a 2µ vector, which was transformed into a *Saccharomyces cerevisiae* strain co-expressing a Sfp-Type 4′-Phosphopantetheinyl Transferase (Ppt1). The transformed yeast strain was cultivated under induced conditions in liquid cultures for five days and before production of secondary metabolites was analyzed by high-resolution mass spectrometry (HRMS). The results showed that the yeast strain was able to produce prephenalenone, which is also the first step of the herqueinone pathway. We also detected dehydroxyprephenalenone, which is formed through spontaneous dehydration [3]. Through heterologous expression of the other genes in the cluster, we will follow the biosynthetic route and ultimately unravel the entire pathway of the perithecial pigment of *F. solani*.

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PP-79 Characterisation of *Fusarium* infection rate in wild oats

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The wild species of *Avena* genus can be considered as a potential source for breeding resistant cultivars of oats. The 57 genotypes belonging to *A. atlantica*, *A. canariensis*, *A. clauda*, *A. damascena*, *A. hirtula*, *A. longiglumis*, *A. wiestii*, *A. agadiriana*, *A. barbata*, *A. vaviloviana*, *A. insularis*, *A. magna*, *A. murphyi*, *A. fatua*, *A. ludoviciana*, *A. occidentalis*, *A. sterilis* from the collection of the N.I. Vavilov Institute of Plant Genetic Resources were tested for infection of grain by trichothecene-producing *Fusarium* fungi (Tri-*Fusarium*). The plants were grown in the field and inoculated by *Fusarium culmorum*. Real-time PCR was used to quantify fungal DNA, and the deoxynivalenol (DON) was analysed by ELISA.

The DNA of Tri-*Fusarium* was detected in all genotypes in the range 0.0065–1.2 pg/ng of total DNA. The three genotypes belonging to *A. canariensis* (Canary Islands), *A. magna* (Morocco) and *A. ludoviciana* (Ethiopia) were the most infected by Tri-*Fusarium* while *A. ludoviciana* (Syria), *A. fatua* (Armenia), and *A. barbata* (Syria) genotypes were the least infected. The amounts of DON varied between 52–3862 ppb. The genotypes most contaminated by DON (>1500 ppb) were *A. barbata* (Ethiopia), *A. canariensis* (Canary Islands), three genotypes of *A. fatua* (Bulgaria, Ethiopia, China), *A. insularis* (Italy), two genotypes of *A. magna* (Morocco), and two genotypes of *A. sterilis* (Israel, Morocco). The low amounts of DON (A. byzantina (China), *A. sterilis* (Turkey, Israel), *A. fatua* (Russia), and *A. wiestii* (Egypt).

The content of Tri-*Fusarium* DNA and DON in the groups of tetraploid oats were higher in compared with the groups of di- and hexaploid species. A strong correlation was observed between the presence of Tri-*Fusarium* DNA and DON content ($r=+0.66$, $p<0.01$). The connection of the morphological traits of the *Avena* species and grain infection by Tri-*Fusarium* was also analysed. The amounts of Tri-*Fusarium* DNA had a significant connection with the weight of 1000 grains ($r=+0.38$, $p<0.05$) and husk hardness ($r=+0.53$, $p<0.01$). The husk hardness also had a significant correlation with DON amounts ($r=+0.37$, $p<0.05$). A negative correlation was detected between plant height and amounts of DNA ($r=–0.26$, $p<0.05$).

The investigation was supported by the Russian Science Foundation (No. 14-26-00067).

PP-80 Spraying rice flowers with a non-pathogenic *Fusarium oxysporum* W5 effectively controls “Bakanae” by competition with the pathogen *in planta*

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“Bakanae”, or foolish seedling, caused by *Fusarium fujikuroi* is one of the most destructive seedborne rice diseases worldwide. At present, application of chemical fungicides (e.g. ipconazole) to seeds shows the effective control of the disease. However, such application often leads to the occurrence of fungicide resistant strains of the pathogen. Thus, establishing a new method using a non-pathogenic *F. oxysporum* as biocontrol agent could be beneficial.

Seventy isolates identified as non-pathogenic *Fusarium* were obtained from rice tissues in “Bakanae” infested fields. Then, we screened them for biocontrol activity to “Bakanae” by seed treatment with their bud cell suspension, and obtained two effective isolates (*F. oxysporum* W3 and W5). The two isolates also exhibited significant “Bakanae” control activity by rice flower treatment.

To reveal the mode of action of biocontrol agent *in planta*, we observed the dynamics of *F. fujikuroi* expressing red fluorescent protein (RFP) and the most effective control agent *F. oxysporum* W5 expressing enhanced green fluorescent protein (eGFP) on the rice tissues. The observed rice flower was sprayed with W5 and ten minutes later, sprayed with *F. fujikuroi*. Not only the flower but the next generation plants developed from seeds obtained from treated flower were observed. In the single inoculation with *F. fujikuroi*, the fluorescence of *F. fujikuroi* was frequently and extensively observed in the sprayed flowers and tissues of next generation plants especially roots and basal part of leaf sheathes. In contrast, the frequency of *F. fujikuroi* fluorescence was remarkably reduced under the presence of W5. Interestingly, the fluorescence of W5 was not significantly affected by the presence of *F. fujikuroi*. In Addition, W5 did not inhibit the growth of *F. fujikuroi* on dual culture assay on potato dextrose agar (PDA), suggesting that W5 has no antagonistic activities.

The non-pathogenic *F. oxysporum* W5 may protect the sprayed flowers and the rice plants, generated from the seeds obtained from the sprayed flowers, from the infection of *F. fujikuroi* by competing with *F. fujikuroi* for nutrition and/or niche.

PP-81 Profiling of tryptophan derived compounds in *Fusarium graminearum* infected *Triticum durum* lines with different resistance levels

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Fusarium Head Blight (FHB) of wheat and other small grain cereals, a disease caused by *F. graminearum*, leads to reduced quality and yield and most importantly to mycotoxin contamination of the harvested grains. In plants, tryptophan (Trp) related metabolic pathways are found to be affected by pathogen infection. In addition, Trp-derived indolic secondary metabolites are known to play an important role in the defense against phytopathogens. The aim of this study is to shed light on the behavior of Trp-derived metabolites of T. durum by evaluating the Trp-derived submetabolome in T. durum differing in FHB resistance under infection conditions. To this end, different *T. durum* genotypes, which differ in the presence/absence of the FHB resistance QTL Fhb1 were chosen, and cultivated to anthesis in the glass house under controlled conditions. After the inoculation with *F. graminearum* spores, deoxynivalenol and water as control, the treated ears were sampled after six distinct time points between 0h and full ripening and immediately shock frozen. The spikelets were milled, extracted and measured with LC-HR(MS/)MS according to our latest isotope-assisted metabolomics workflow. Additionally, the ^{13}C-Trp tracer-based labeling approach was used to extract the target tryptophan derived metabolites in the *T. durum* cultivars.

This study revealed approximately 70 Trp-derived metabolites in wheat under the tested conditions. Closer insights into the Trp-derived submetabolome and comparative quantification of the Trp-related metabolic profile will be reported and discussed in this presentation.

PP-82 Mycotoxin analysis of Bt and non-Bt maize grain infested with lepidopteran insect larvae and inoculated with *Fusarium subglutinans* and *Fusarium temperatum*.

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Fusarium subglutinans (Fs) is one of the most common *Fusarium* spp. associated with Fusarium ear rot in North America. However, there is limited information on the role of the closely related species *F. temperatum* (Ft), formally Fs group I, in Fusarium ear rot and mycotoxin contamination of grain. In 2015 and 2016, randomized complete block design field experiments were conducted in Iowa, USA with Fs or Ft silk channel inoculations and with corn earworm (*Helicoverpa zea*) or European corn borer (*Ostrinia nubilalis*) infestations on Bt and non-Bt maize. Ear rot severity and insect injury were estimated visually and a multi-mycotoxin analysis was performed on milled grain. In both years, Fusarium ear rot severity and insect injury was lower in the Bt hybrid, and Ft inoculation resulted in higher levels of moniliformin compared to Fs (P<0.0001). In 2016, *H. zeae* infestation increased ear rot severity (P=0.0452) compared to the non-infested control and both insects caused increased moniliformin levels in Ft-inoculated treatments ($P\leq$0.0055). In 2015, grain from Fs-inoculated treatments had higher levels of fusaric acid than Ft treatments (P<0.0001). In 2016, Fs-inoculated treatments had the highest levels of fusaproliferin which were increased by insect infestations, whereas Ft-inoculated treatments had the highest levels of beauvericin, also increased by insect infestations (P<0.0001). There were large differences between Ft strains regarding moniliformin, fusaproliferin, beauvericin, and gibberellic acid levels. The highest levels of fumonisins in 2015 were detected in non-inoculated, non-Bt, insect-infested treatments. However in 2016, insect infested, non-inoculated treatments of both hybrids had the highest fumonisin levels ($P\leq$0.0001).

PP-83 Changes in expression of stress-responsive genes in *Fusarium proliferatum* exposed to host plant extracts

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Stress factors (biotic or abiotic) are critical players in functional characteristics of plant-pathogenic fungi. Our previous studies have shown that *Fusarium proliferatum*, a casual fungal pathogen, besides increased fumonisin biosynthesis, activates the expression and accumulation of numerous proteins to minimize the effect of potential stressing factors, such as host plant extracts. Some of them are related with stress response, *e.g.* the heat-shock proteins (HSPs) which often show protective and stimulating biological functions. Therefore, here we tried to understand the role of HSPs in response to stress conditions exerted by the presence of plant metabolites from host species. The main objective of present study was to examine the changes in the expression of two *Hsp* genes after the addition of aqueous extracts from four potential host species in *Fusarium proliferatum*.

Asparagus-derived *F. proliferatum* strains was cultured *in vitro* using a liquid medium, which at the 5th day of cultivation was supplemented with plant extracts made from asparagus, maize, garlic and pineapple tissues. Mycelia were collected every two days from liquid cultures before and after extract added and subjected to the total mRNA extraction. To validate the expression profiles of *Hsp70* and *Hsp88* genes, specific primers were designed based on previously obtained partial genomic sequences and quantitative RT-PCR technique was used and normalized against the constitutively expressed *tub2* gene.

The expression levels of *Hsp70* and *Hsp88* genes varied relating to host extract used. The *Hsp88* gene was expressed in all samples whereas the expression of the *Hsp70* gene was observed 24 h after the extract supplementation. The asparagus extract induced the highest increase of *Hsp70* and *Hsp88* transcript levels and garlic extract was the second most effective inducer. In the *Hsp88* expression profile we observed two expression peaks: after 24 h of extract supplementation and after 12 days of cultivation (7 days after extract application). We have demonstrated that dissimilar nutritive environments have significant impact on stress-related gene expression profiles in *F. proliferatum*.

This research was supported by the Polish National Science Centre Project 2015/17/B/NZ9/03577.

PP-84 Cadmium and Deoxynivalenol in durum wheat grains: physiological and biological basis of the co-contamination.

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Cadmium (Cd) and mycotoxins are among the most worrying contaminants that threaten the safety of food products derived from cereal kernels. Indeed, as recently supported by the last French total diet survey (Anses, 2011), deoxynivalenol (DON) and Cd human exposure through food mainly results from the consumption of cereal-derived products. Durum wheat is the most sensitive cereal culture for both DON and Cd accumulation in kernels, leading to a high frequency of co-contaminated harvests. This frequent co-occurrence (even though each toxic substance is in concentration within the EU regulatory limits) combined with the fact that Cd and DON are likely to be distributed in the same milling fractions raises the concern of consumer exposure to the cocktail Cd+DON.

To address the issue of DON+Cd co-contamination, the CaDON initiative (funded by the French National Agency for Research, 2015-2019) investigates the relations between Cd and DON occurrence in durum wheat, from crops co-contamination in the field to the milling end products, as well as the toxicity of Cd and DON mixtures upon ingestion. One of the objectives pursued by the CaDON project aims to elucidate the physiological bases of Cd+DON contamination of durum wheat kernels. The effect of soil Cd contamination on *Fusarium graminearum* infection and DON accumulation in kernels and conversely of *F. graminearum* infection on Cd accumulation are investigated through the implementation of *in vitro* and greenhouse experiments. As a first step, the way *F. graminearum* exposition to Cd affects the production of DON is analysed and the mechanisms underlying this modulation investigated with a strong focus on the relation between Cd/oxidative stress/DON biosynthesis. These insights will be the subject of the present communication together with the first data delivered by greenhouse experiments.

Anses, 2011. https://www.anses.fr/fr/content/les-etudes-de-lalimentation-totale-eat

PP-85 Do botanical extracts and potentially antifungal mulch treatments suppress *Fusarium graminearum* inoculum?

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Fusarium Head Blight (FHB) is one of the most important cereal diseases worldwide causing not only significant reductions in grain yield but also severe contaminations of the harvested products with mycotoxins that jeopardise food and feed safety. The predominant species of the FHB disease complex is *Fusarium graminearum* (FG; teleomorph *Gibberella zeae*). In maize-wheat rotations with reduced- or no-till systems, the remaining maize residues on the soil surface represent an important inoculum source for infection of the subsequent cereal crop. Hence, agronomic practices such as suitable crop rotations and management of crop residues with ploughing are commonly used to control FHB in wheat cropping systems. Nevertheless, continuous ploughing has also several drawbacks, such as increased soil erosion risks and decreased soil fertility in the upper soil layers. Within the framework of the Horizon 2020 project MycoKey, the main objective of this study is to develop prevention strategies to suppress FHB and thus decrease the risk of mycotoxin accumulation in wheat. One approach consisted of applying various botanical extracts and freshly mulched material from crops with potential antifungal activity onto FG infected maize residues in the field. The botanicals included two mustard extracts and suspensions of milled Chinese galls, while the mulch treatments were harvested from mustard and clover crops. Preliminary results revealed that the mulch treatments decreased the deposition of discharged *G. zeae* ascospores and reduced FHB disease symptoms on wheat heads. Currently, the analyses of the mycotoxin content (deoxynivalenol and zearalenone) in the harvested grains is in progress and results will be discussed. Furthermore, *in vitro* studies showed that the botanical extracts have the ability to suppress not only the mycelium growth but also the germination of conidia and ascospores as well as their discharge from mature perithecia. The preliminary data of this study suggest that there is a great potential to suppress FG inoculum using non-conventional agricultural methods using botanical extracts and mulch layers from crops with antifungal properties.

PP-86 A model to predict DON content in French winter wheat and Norwegian spring oats

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Fusarium head blight is a major disease of cereals all over the world. *Fusarium graminearum* is an important deoxynivalenol (DON)-producing species in wheat and oats. In addition to climatic parameters such as rain at flowering (Hjelkrem et al. 2017), previous crop, tillage and varietal susceptibility significantly influence the DON content in cereals. To reduce the risk of elevated mycotoxin levels in grains at harvest, but simultaneously minimize the use of pesticides, ARVALIS and NIBIO have developed a forecasting model that aims to optimize fungicide treatment against *F. graminearum*. The model was built in two step: first, a model to predict ascospore discharge was developed based on spore trap data, thereafter a second model including output of the first one, was developed based on DON content in grains collected from farmers' fields.

The ascospore model consists of a logistic equation, including the sum of temperatures on rainy days, fitted from two years observations in wheat fields with maize as previous crop and no-till. Output of this model and various weather variables were calculated for 1400 fields of wheat in France and 250 of oats in Norway. Daily weather data were recorded from the nearest weather station provided by Agrometeorlogy Norway or from spatially explicit climatic data for the geographic coordinates of each plot in France (Deudon et al., 2017). Finally, logistic regressions were calculated for elevated DON levels in harvested grains with a backward selection on weather parameters, ascospores prediction and agronomic variables, using the ROC curve (receiver operating characteristic) method in a cross year validation. For both countries, the best model was selected according to the area under the curve (AUC) and achieved a good prediction performance.

Deudon, O, *et al.*, 2017. Interest and implementation of a spatialization method of meteorological data used for in Agricultural Decision Support Tools. Proc. 2017 EFITA CONGRESS – Montpellier, France.

Hjelkrem, A-G.R., *et al.,* 2017. DON content in oat grains in Norway related to weather conditions at different growth stages. European Journal of Plant Pathology. 148: 577-594

PP-87 *FUSARIUM culmorum* monitoring in cereal residues in Tunisia

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Fusarium foot and root rot, caused mainly by *F. culmorum,* is an important soil-borne disease affecting several cereal crops in Tunisia. This fungus survives as chlamydospores and mycelium in the soil, or infecting plant residues. The aim of this study was to monitor the *F. culmorum* population in the residues of four cereal crops (durum wheat, soft wheat, barley and oat) stored at different depth in the soil. A total of 75 stems inoculated with a *F. culmorum* strain were enclosed in three nylon bags (25 each) for each cereal species and disposed on the soil surface and at 10 cm depth, in December 2014. The study was carried out in three different sites: Mateur, Bou Sallem and Kef, located in the sub-humid, humid and semi-arid regions, respectively.The inoculum rate of *F. culmorum* in crop residues was estimated two times: at the beginning of the experiment and 8 months after the trial setup. Frequency of recovery of *F. culmorum* from the residues was estimated on ¼ PDA, by isolating the fungus from the all samples collected from the three sites. In addition, DNA of *F. culmorum* was quantified using quantitative real-time polymerase chain reaction (qPCR). At the beginning of the trial, residues were highly infested with *F. culmorum* and the rates were 85%, 75%, 70% and 65% for oat, soft wheat, barley and durum wheat, respectively. Statistical analysis showed a significant decrease of *F. culmorum* recovery rate and amount of DNA after 8 months. Overall, there was a strong correlation (r^2=0.78) between DNA levels and the rate of *F. culmorum*. In addition, there was no significant difference of both the incidence and the amount of DNA of *F. culmorum* between the different sites and the different cereal species. This work also showed that *F. culmorum* was significantly higher ($P < 0.001$) in residues kept on the soil surface than the ones placed at 10 cm depth. These results suggest that the recent adoption of no-till farming system by Tunisian farmers could contribute to increase the inoculum level of *F. culmorum* and, consequently the incidence of foot and root rot on cereals, compared to conventional cropping system.

Keys words: soil-borne disease, Cereal, Foot and root rot, *F. culmorum*, no-till, survival, residues.

PP-88 Mycotoxin contamination in maize parcels in relation to agronomic practices in Flanders

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Mycotoxins are toxic secondary metabolites produced by a variety of fungal species, such as *Fusarium*, *Penicillium* or *Aspergillus*, among others. Contamination of feed with mycotoxins can cause severe health problems in dairy cattle. Especially high yielding dairy cows with a high feed uptake and rapid ruminal flow are susceptible to gastroenteritis, reduced reproduction and reduced milk production, as a result of mycotoxin contamination. Maize silage is one of the main components of dairy feed in the region of Flanders, Belgium, and is therefore one of the main sources for mycotoxin uptake in dairy cows.

This research aims towards providing dairy farmers in Flanders with a user-friendly prediction model, able to foresee mycotoxin contamination based on weather, cultivation, harvest and silage conditions. This model will be constructed based on mycotoxin analyses of freshly harvested maize and maize silages across Flanders, and on research focusing on methods to prevent mycotoxin contamination. During the first sampling in 2016, 91 samples were taken of Flemish maize parcels during harvest. These samples were analyzed for 23 different mycotoxins using LC-MS/MS.

The results of these first 91 field samples indicate that nearly every maize parcel in Flanders contains at least one of the 23 mycotoxins analyzed. NIV was the most prevalent one, being present in all but one sample. DON and ZEA were found in 84 and 59 samples, resp.. Concentrations went up to 2368ppb for NIV and 2777ppb for DON. No aflatoxins or fumonisins (except FB1 in 2 samples) were found. These results were then used to search for correlations in the concentrations of different mycotoxins, and to detect certain agronomic practices that are correlated with a higher or lower mycotoxin concentration. DON, ZEN and ENN B were positively correlated with later harvest dates, meaning that the longer the maize plants stay on the field, the more mycotoxins will be present. Significantly more DON was found on loam and sandy loam soils compared to sandy soils. No significant influence was found for sowing density, fertilization, tillage and dry matter content, among other factors. This however does not mean that these factors have no influence on mycotoxins at all. Further sampling is needed to confirm these results.

PP-89 *Fusarium*-resistance of climate-adapted barley plants

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Barley is an important crop and mainly used for animal feed and malt production. The Fusarium-complex is an increasingly important fungal pathogen in barley. Its occurrence is favoured by specific climatic factors and causes losses in yield and quality[1]. Natural resistance against several *Fusarium* species is genetically complex and imperfect. Furthermore, chemical control is deficient and its sustainability questioned. Within the project BayKlimaFit, spring barley cultivars that adapted to climate conditions are tested as well for resistance against fungal pathogens to develop tools to optimise plant breeding and to screen for resistant genotypes. Therefore, a range of barley genotypes is used, which is relevant for Bavarian plant breeders. This assortment was pretested for resistance against abiotic stress factors such as extreme climate/whether conditions. In 2016 and 2017, field trials for artificial inoculation experiments at TUM Weihenstephan (Freising, Germany) and under controlled drought stress, in cooperation with the Bavarian State Institute for Agriculture (Freising, Germany), were conducted. Molecular genetic studies, visual assessments and microclimatic measurements examined a differentiated *Fusarium*-infestation. By artificial single and mixed inoculation with *Fusarium culmorum* and *Fusarium avenaceum*, a differentiation in infestations was achieved. In 2017, weak respectively good resistance of some candidates was again validated. The results show a high and dynamic range in Fusarium disease severity of different barley cultivars. This can work as a basis for classifying disease resistance (patho-phenotyping) under combined abiotic and biotic stress. Based on results provided by the other project partners, the range of barley genotypes was narrowed down to a set of interesting candidates for further research in the upcoming season. The comparison of resistant and susceptible plants should provide insights into mechanisms of disease resistance of climate-adapted barley cultivars. In addition it is questionable to what extent the resistance to local climate conditions of barley is coupled to a resistance against *Fusarium* species. The results of this project can be applied directly for the practical plant breeding in Bavaria in form of selective physiological and molecular markers.

1 Hückelhoven et al (2017) Fusarium infection of malting barley has to be managed over the entire value chain. J. Plant Dis. Prot. DOI: 10.1007/s41348-017-0101-0

PP-90 The infection progress of *Fusarium oxysporum* f. sp. *niveum* on susceptible and resistant watermelons using green fluorescent protein-transformed isolates

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Fusarium wilt of watermelon is caused by the soil-borne fungal pathogen, *Fusarium oxysporum* f. sp. *niveum* (E. F. Smith) Snyder & Hansen (Fon). Fon invades the host root or stem tissues via the contaminated seeds or soil residues, and leads the host plant to death. The infected host shows vascular browning. The chlamydospores of Fon could survive in the soil for more than a decade, thus causes tremendous loss on watermelon production. In this study, the green fluorescent protein gene (GFP) was introduced to Fon H0103 isolate via the *Agrobacterium tumefaciens*-mediated transformation (ATMT) method. The GFP-integrated transformants, emitting the green fluorescence, were used to inoculate the Fon-susceptible watermelon cultivar (Grand Baby, GB). Watermelon seedlings showed no, mild, or severe symptoms when challenged with different Fon transformants. The infection progress of the very low-virulent isolate (Fon-Y1) and Fon-X3 isolate, showing similar virulence as the wild-type Fon H0103, was subjected to confocal laser scanning microscopic observation on the Fon-susceptible watermelon line (Sugar Baby, SB) and Fon-resistant watermelon line (JSB). The results suggested that at the beginning stage of infection, there were no much difference for Fon-Y1 and Fon-X3 invading SB or JSB watermelon seedlings. The spore germination ability and mycelium invasion for both transformants were similar. But at the later stage, the colonization, mycelial growth, and chlamydospore formation of Fon-X3 was faster and more abundant than Fon-Y1 transformant in Fon-susceptible SB host. The infection progress of Fon-X3 and Fon-Y1 transformants was similar in both Fon-susceptible SB and Fon-resistant JSB watermelons. The T-DNA insertion site was analyzed using thermal asymmetric interlaced PCR (TAIL-PCR). The function of the T-DNA interrupted Fon gene was studied for the involvement in pathogenicity or virulence.

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PP-91 Development of SNP markers for *Fusarium* head blight (FHB) resistance breeding in wheat.

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Fusarium head blight (FHB), primarily caused by the fungus *Fusarium graminearum,* is an economically important disease of wheat resulting in reduced yields and grain contaminated with mycotoxins, rendering unfit for consumption by humans and animals. Many FHB resistance QTLs have been genetically mapped and in our lab, several genes responsive to FHB and mycotoxin deoxynivalenol (DON) have been functionally characterized. These include a cytochrome P450*,* a receptor kinase, the *Pooideae*-specific gene *TaFROG*, and *Ta-FROG*-interacting proteins such as *TaNAC5.*

Wheat consists of three sub genomes (A, B and D) and the objective of this project is to determine whether varietal-specific differences within these FHB/DON responsive genes and/or their promoter region are associated with FHB resistance in the field. Gene expression studies were conducted using homoeolog-specific primers, which confirmed that the homoeologs were differentially expressed in response to FHB infection and the toxigenic *Fusarium* virulence factor (DON). Sequence analysis of the coding and promoter regions has revealed sequence differences that could, potentially, underlie these observed expression differences.

On-going studies are identifying homoeolog and genotype–specific single nucleotide polymorphisms (SNPs) in conserved domains and the promoters of these FHB/DON-responsive genes. These SNPs will be converted to high-throughput KASP assays and used to genotype the Remus x CM82036 doubled haploid population, which has been scored for FHB infection over several site and seasons. QTL analysis will then be conducted to determine which SNPs are associated with FHB resistance in the field. Significantly associated SNP markers will then be used for marker–assisted selection (MAS) for FHB resistance breeding.

PP-92 Development of a forecasting system for *Fusarium graminearum* and deoxynivalenol in barley

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An early prediction of mycotoxin levels is important for farmers to minimise the risk of contaminated food and feed entering the cereal supply chain. The vast majority of studies investigating Fusarium head blight focus on wheat, however, since cereals differ in terms of the main occurring *Fusarium* species, a barley monitoring was conducted throughout Switzerland in 2013 and 2014 by collecting harvest samples from farmers along with information on respective cropping factors. The analysis of these samples using a seed health test and LC-MS/MS measurements revealed *F. graminearum* (FG) and deoxynivalenol (DON) as the most prominent species and mycotoxin, respectively. The evaluation of the impact of cropping factors demonstrated that barley samples from fields with maize as a previous crop had a substantially higher FG incidence and elevated DON content compared with other previous crops. In addition, the use of reduced tillage resulted in a higher disease incidence and toxin content compared with samples from ploughed fields.

In addition, climate chamber experiments with artificial FG infection during anthesis were performed to investigate the influence of different temperatures (10°C, 15°C, 20°C) and durations (4h, 8h, 12h) at 99% relative humidity on toxin accumulation in barley. For the fodder variety Ascona, an up to three times higher DON contamination in samples from the 15°C treatments was detected compared with those from the 10°C and 20°C treatments. For the malting variety Concerto, the prolonged humidity durations had a stronger effect under all tested temperatures and resulted in up to two times higher DON contaminations. Currently, we are investigating whether barley could also be susceptible to FG infections in growing stages before and after anthesis.

The combination of these findings together with results from resistance experiments with various barley varieties will be used to extend the forecasting model FusaProg for DON in wheat towards barley. This system will contribute to a targeted, effective and reduced application of fungicides and will also provide a tool for farmers to decrease the infection risk.

PP-93 Screening of mycoviruses infecting *Fusarium culmorum populations*

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Fusarium head blight (FHB) is a fungal disease that affects cereals, in particular wheat (*Triticum aestivum* L.). This disease causes a severe loss grain yield, but the most worrying concerns the release of secondary metabolites, also known as micotoxins. The presence of toxic substances to humans and animals makes this problem common to many economically important processes well as to the health of consumers all over the world. Unfortunately the efficiency fungicides to eradicate fungal pathogens could be variable depending on field environmental conditions. For this reason, additional defense measures could be adopted, like biocontrol, based on the possibility to reduce fungal virulence. Mycoviruses are viruses that infect fungi and have the potential to control fungal diseases of crops when associated with hypovirulence (Yu *et al*. 2010). Mycovirus-mediated hypovirulence is a phenomenon in which the virulence of fungal pathogens is reduced or even completely lost as a consequence of virus infection. The successful utilization of hypoviruses for biological control of chestnut blight (caused by *Cryphonectria parasitica*) in Europe has attracted much interest and led to the discovery of hypovirulent strains in other fungi (Yu *et al*., 2010). The potential of new sequencing technologies such as MinION can support the identification of new mycovirus species. MinION consists of a chip with incorporated nanopores that measure the changes in electrical conductivity generated by the passage of DNA strands through a biological pore. Italian, Syrian, Tunisian and Iranian populations of *Fusarium culmorum* (W.G. Smith) Sacc. strains, one of the main FHB agent, were analyzed for mycoviruses presence. Bioinformatic analysis showed that most of the micoviruses belong to *Partitiviridae*, *Endornaviridae*, *Narnaviridae*, *Tombusviridae*, *Totiviridae* and *Metaviridae* families. Some of these mycovirus families have been studied as inducers of hypovirulence in some phytopathogenic fungi, therefore further studies are necessary to investigate their role as possible biocontrol agents.

Yu X, et al. A geminivirus-related DNA mycovirus that confers hypovirulence to a plant pathogenic fungus. Proc Natl Acad Sci USA. 2010;107(18):8387–8392

PP-94 Stable isotope labeling guided metabolomics of Fusarium Head Blight in wheat: Investigation of the phenylalanine derived submetabolome

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Fusarium Head Blight is a fungal disease of small grain cereals such as wheat. It leads to yield losses of the crop and its contamination with mycotoxins. Although breeding of resistant wheat cultivars is considered as one of the most sustainable strategies to counteract the disease, this approach is not straight forward as resistance in wheat is multigenic and thus mediated by a large number of different quantitative trait loci (QTLs). In the presented study wheat lines with different levels of resistance are compared with respect to their metabolic response upon treatment with F. graminearum and its major mycotoxin deoxynivalenol (DON). Besides a resistant and a susceptible parent line, two near isogenic lines only differing in the presence or absence of the QTL Fhb1 (located on chromosome 3B), were investigated. A combination of LC-HRMS and stable isotopic labeling of whole wheat and the tracer $^{13}C_9$ phenylalanine (Phe) were chosen to allow the evaluation of both the global and Phe-derived submetabolome under infection conditions.

Flowering wheat ears of four different wheat genotypes were treated with Fusarium graminearum (Fg), the mycotoxin DON or water as a control. At five different time points after treatment, samples were harvested, milled, extracted and measured with LC-HRMS. Automated data processing allowed the detection of about 1000 plant metabolites including 175 metabolites belonging to the Phe-submetabolome. The presentation will illustrate our isotope-assisted metabolomics approach and focus on the very diverse Phe-derived submetabolome and the abundance of those metabolites over time. Phe related metabolic differences between the two parent and near isogenic lines and the putative association of significantly differing metabolites with Fhb1 mediated resistance or susceptibility to FHB will be discussed in detail.

PP-95 Microbial detoxification of deoxynivalenol

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Mycotoxins are secondary metabolites produced by fungi. When these fungi are present on agricultural commodities, they can pose a high risk to animal and human health. As the European Commission imposes strict regulations on maximum levels of several mycotoxins, it is a prerequisite for farmers to meet these regulations to avoid complete loss of a contaminated batch. In an agro-ecosystem, farmers try to avoid fungal infection and the occurrence of mycotoxins in their commodities via prevention and intervention approaches, but often fail in completely eliminating the risk. Therefore, detoxification strategies are needed such as the application of binders or the application of microorganisms able to detoxify these mycotoxins.

Our project focuses on the microbial detoxification of deoxynivalenol (DON), a worldwide prevalent mycotoxin. Microorganisms were screened for their biotransformation capacity of DON through non-targeted LC-MS/MS. Furthermore, a bioassay using the aquatic plant *Lemna minor* was developed to assess the residual toxicity of DON metabolites after biotransformation. Pursuing this approach, two mixed cultures were obtained, originating from soil and activated sludge, capable of biotransforming DON into the epimer of DON and the epimer of DOM-1, both conveying no residual toxicity for the *Lemna minor* plant.

PP-96 Quantitative analysis of *Fusarium graminearum* and *Microdochium* fungi in winter wheat tissues

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Fusarium and *Microdochium* fungi can produce identical symptoms such as head blight on winter wheat. Recently, numerous head blights and necrotic lesions on flag leaves of wheat have been observed in the South-European region of Russia.

The aim of the study was to reveal the relationship between *F. graminearum* and *Microdochium* fungi in wheat tissues.

The whole plants of three commercial cultivars of wheat (100 plants per cultivar) were collected at the early dough growth stage and then divided into four parts (head, peduncle, flag leaf, and stem). This was followed by the homogenisation of each part and extractions of DNA and mycotoxins.

Seven samples of winter wheat were incubated on PSA for one week, and then the percentage of grain infection in those samples was scored.

Quantitative PCR was used to analyse the DNA content of *Microdochium nivale*, *M. majus* (Nielsen et al., 2013), and *F. graminearum* (Yli-Mattila et al., 2008) in the wheat tissues. The amount of deoxynivalenol (DON) was determined by ELISA.

The content of *M. majus* DNA was significantly higher (from 0.85 to 120 pg/ng of total DNA) than that of *M. nivale* (from 0.08 to 3.6 pg/ng) in all parts of the plant. The maximum DNA of both the *Microdochium* species was detected in the flag leaves. The highest content of *F. graminearum* DNA was observed in the heads (3.0×10^{-3} pg/ng) compared with the other parts of the plant ($3.7\text{-}5.4)\times10^{-4}$ pg/ng. The maximum DON value observed was 779 ppb in the heads. In addition, a significant positive correlation was found between *F. graminearum* DNA and DON in the plant tissue ($r = +0.84$).

The percentage of *Microdochium* infection varied from 0 to 31% and that of *F. graminearum* varied between 0 and 3%. The average DNA content of *M. majus* in the grain samples was 3.8 pg/ng, *M. nivale* was 6.6×10^{-2} pg/ng and *F. graminearum* was 4.4×10^{-3} pg/ng of total DNA.

Significant negative correlations between the percentage of *F. graminearum* infected grains and content of *M. nivale* DNA, and between DNA of *F. graminearum* and DNA of *M. nivale*.

The study showed that the head blight symptoms on winter wheat were associated mainly with *M. majus* than with *F. graminearum* and *M. nivale*. However, the symptoms alone are inadequate for identifying the disease in the fields.

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