Nachwuchswissenschaftlerforum
Young Scientists Meeting

Achtes Nachwuchswissenschaftlerforum 2015

19. - 21. Oktober
in Quedlinburg

- Abstracts -

Berichte aus dem Julius Kühn-Institut

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Bundesforschungsinstitut für Kulturpflanzen

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Kontaktadresse
Anja Hühnlein
Julius Kühn-Institut (JKI)
Bundesforschungsinstitut für Kulturpflanzen
Informationszentrum und Bibliothek
Erwin-Baur-Straße 27
06484 Quedlinburg

Telefon +49 (0)3946 47-123
Telefax +49 (0)3946 47-255

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Telefax +49 (0)5374 6577

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Greetings from the President

Dear Young Scientists,

Welcome to the 8th Young Scientists Meeting of the Julius Kühn-Institute at the JKI headquarters in Quedlinburg.

The meeting has seen several innovations over the recent years, such as the get together with a tasting of the JKI's own wine creations. Another novelty are short, high speed poster presentations, where participants let their creativity run free and even put their talk in verses or brought a jar of candy smell. This year's rerun of the elevator talks is highly anticipated.

Like the meeting on a small scale, the entire JKI is constantly undergoing changes. Construction works at Braunschweig and Dossenheim update the research facilities to modern standards with state-of-the-art equipment. The recently perceived endangerment of pollinators by agrochemicals led to the foundation of an Institute for Bee Protection which is going to be established in 2016.

To fulfill the various and ever-changing responsibilities in future-oriented basic research, translation of research results into practical applications and counseling the German government on agricultural, horticultural and forestry issues, top level scientific expertise, dedication and networking are essential. With half of the institute's scientific staff facing retirement within the next fifteen years, it is a major objective to preserve the acquired knowledge and skills, and keep young scientists working at the JKI.

To this end, we welcome Kathrin Lehmann, head of Human Resources Unit, to this year's meeting and look forward to her key note talk on the perspective of young scientists at our institute. In addition to this 'infrastructure talk', which has become a tradition lately, Cornel Adler will give the second key note on the topic of stored product protection.

I am convinced that all participants will personally benefit from the NWF by expanding their knowledge and by increasing their presentation skills. In addition, you get the chance to get to know each other during the joint evening events and expand your personal network, an important base for your future professional life.

I hope you all enjoy your stay at Quedlinburg and return home enriched by the experience and inspired both scientifically and socially.

Quedlinburg, October 2015

Dr. Georg F. Backhaus
President of the JKI

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Keynote 1

Organic Stored Product Protection – Why, how, and for whom?

Cornel Adler, Agnès Ndomo-Moualeu, Christina Müller-Blenkle
Julius Kühn-Institut, Institute for Ecological Chemistry, Plant Analysis and Stored Product Protection, Berlin
Email of corresponding author: cornel.adler@jki.bund.de

All we produce in agriculture will be harvested at one point and used as food, feed, or for other purposes. It is a principle in nature that all plants and plant parts are broken down by organisms. Durable parts of plants or plant products after harvest are called stored products. If stored products are attacked we call the attackers stored product pests. However, we should perhaps change our perspective and call them indicators for poor stored product protection.

According to the FAO (2011), man looses about one third of stored products between harvest and consumption. Projected onto grain and 2014, losses in Germany alone amount to billions, while research is done by three scientists alone. In industrialized countries, a lot of these losses occur due to the waste of food, in tropical climates there is often less waste but no protective winter and insects are developing all year round.

In Germany, stored product protection became a topic for science after World War I, because the devastation had caused hunger. After World War II, chemical plant protection promised to solve all problems, even in stored product protection. But today the consumer does not want any residues in food or animal feed. The last new stored product protection chemicals, pyrethroids were invented in the 1970s. Is organic protection a future for all plant protection? Perhaps not completely, but the tendency is clear: less hazardous, more sustainable techniques are wanted.

In stored product protection, solutions are customized to the particular situation. A good start is to avoid the establishment of pests by an optimized structural design. Hermetic storage, cooling, drying, sanitation, and sufficiently sealed packages are important preventive measures. If prevention failed, an infestation needs to be detected as early as possible. At present we run a project on acoustic detection that may detect single insects in a grain mass, perhaps even give the species and stage. This would help to use biological control, like parasitoids early on. We also test the orientation of insects by smell and try to develop lures that copy dried fruits, nuts and grain. Other modern devices for pest detection are cameras scanning surfaces and attached computers comparing shapes to identify pests. We could even use automated laser guns to control pests (and make Lara Croft unemployed).

For control we use extreme temperatures, milling, carbon dioxide at ambient or high pressure, anoxic atmospheres at ambient or high temperatures, biological control or other techniques. There is a lot of high-tech available, but little has been implemented yet because stored products were cheap, - too cheap. This changes now, and we better be prepared!

Session 1

Viticulture

New insights in the molecular structure and function of the resistance locus *Ren3* against powdery mildew (*Erysiphe necator*) from the grapevine cultivar 'Regent'

Pierre Schneider, Daniel Zendler, Reinhard Töpfer, Eva Zyprian
Julius Kühn-Institut, Institute for Grapevine Breeding Geilweilerhof, Siebeldingen
Email of corresponding author: pierre.schneider@jki.bund.de

Powdery mildew is one of the most devastating diseases of grapevine (*Vitis vinifera* L). The disease is caused by *Erysiphe necator* Schw. (syn. *Uncinula necator* (Schw.) Burr, anamorph *Oidium tuckeri* Berk.), an ascomycete fungus, which was introduced from North America to Europe in 1845. The traditional European cultivars are highly susceptible to the fungus and still today huge amounts of fungicides are necessary to defeat the pathogen.

Many North American *Vitis* species developed resistance against *Erysiphe necator* due to co-evolution of host and pathogen. This process promoted the development of a genetic locus called *Ren3*, which was characterized in the cultivar 'Regent'. There are several genes located within this region, which show great similarity to genes known to mediate resistances in other plants.

Until this day, the resistance locus *Ren3*, located on LG15 of the grapevine cultivar 'Regent', spans an approximate interval of 4 Mb, which contains three different clusters of resistance genes analogs (RGAs). To determine which one of the RGA clusters harbors the gene which causes resistance against *Erysiphe necator* we performed a fine-mapping of LG15 in the crossing populations 'Regent' x 'Lemberger' and 'Regent' x 'Cabernet Sauvignon'.

The results of recent studies suggest an involvement of more than one RGA cluster in the recognition of *Erysiphe necator* over the course of a year. This finding leads to the assumption that we are confronted with more than one *Erysiphe necator* strain which causes Powdery Mildew infections in Germany. Furthermore the sequencing of one of the three RGA clusters could be completed via BAC-clone sequencing.

The project is funded by DFG (Deutsche Forschungsgemeinschaft) Zy 11/9-1.

Analysis of bunch architecture in grapevine

Robert Richter[1], Susanne Rossmann[2], Eva Zyprian[1], Klaus Theres[2], Reinhard Töpfer[1]
[1] Julius Kühn-Institut, Institute for Grapevine Breeding, Siebeldingen
[2] Max Planck Institute for Plant Breeding, Research Dept. of Plant Breeding and Genetics, Köln
Email of corresponding author: robert.richter@jki.bund.de

A lose grape cluster is a desirable trait in grapevine breeding, since it reduces the abundance and severity of fungal infections. This is partly due to a better coverage of the grape bunch with antifungal spraying agents but nonetheless due to a higher air exchange within the grape cluster. The reduced exposure to high humidity acts as a physical barrier against pathogens which are in need of high moisture to proliferate, e.g. *Botrytis cinerea*. The aim of this study is to identify bunch architecture influencing genes and to introduce molecular markers to accelerate the selection process in grapevine breeding.

To calculate the compactness factor for the bunch architecture a phenotyping procedure was established. The experimental trails are spread over three wine growing regions in Germany and located in the climate areas A and B. The plant range contains a mapping population (GF.GA-47-42 x 'Villard blanc'), a set of 'Pinot Noir' clones with lose as well as compact clusters and additionally extremely lose clustered table grapes of the 'Cardinal' family.

A phenotype-DNA based approach uses the 150 F1 individuals of the mapping population for QTL analysis. The F1 progeny segregates in terms of cluster size and compactness. QTL calculations performed in 2012 suggest QTLs on one chromosome related to rachis length. The calculation of two consecutive years (2012, 2013) confirmed a QTL related to pedicel length. However the QTLs still cover wide genomic regions and candidate gene suggestion is therefore hampered. Using the phenological data of the upcoming two years in association experiments should result in verification and definition, as well as revelation of QTLs.

For model plants, the literature provides information about genes involved in the regulation of floral meristem formation. The expression of these genes is conserved over genetic distances and displays a great impact on the inflorescence and bunch architecture. Based on the grapevine reference genome (PN40024) orthologues of these genes should be detected. Furthermore molecular markers linked to these traits need to be established.

In a transcriptional profiling approach two lose and three compactly clustered clones of 'Pinot Noir' were compared. In a first step RNA from dormant winter buds and compound buds harvested during the growing period were used in a differential gene expression experiment. The RNA sequencing was performed at the Max-Planck-Institute for Plant Breeding. First candidate genes have to be verified with chip-based micro fluidic PCR (Fluidigm) and quantitative PCR.

Phaeomoniella chlamydospora - the Esca pathogen in grapevine nursery production

Nicolai Haag, Michael Fischer
Julius Kühn-Institut, Institute for Plant Protection in Fruit Crops and Viticulture, Siebeldingen
Email of corresponding author: nicolai.haag@jki.bund.de

Esca is a grapevine trunk disease (GTD) that can be found in wine-growing regions throughout the world. In Europe the wood-inhabiting fungi *Phaeomoniella chlamydospora* (*Pch*), *Phaeoacremonium aleophilum* (*Pal*) as well as *Fomitiporia mediterranea* (*Fmed*) are considered the main causal agents of this disease. Not only does Esca occur in older vineyards, but also young vineyards and planting material may be affected by the so called Petri disease, a precursor disease of Esca. Considerable economic losses caused by these diseases could be observed over the last two decades. To date only limited data are available about biology, incidence, infection paths and spreading behaviour of the involved fungi. Direct and effective control measures are not available up to now.

Information on occurrence and epidemiological aspects of *Pch*, probably the most important Esca pathogen in plant material production, are required in order to build up the basis for development of effective control strategies in the nurseries. Consequently, this project investigates the occurrence of *Pch* during the grapevine production process of three different grapevine nurseries over a period of three years. The investigated substrates comprise grapevine wood, callusing media, different dipping baths, air and soil.

In 2014 and 2015 wood samples from grafting material (different rootstock and scion cultivars), recently grafted vines as well as grafted vines in the nursery were collected and investigated for the presence of *Pch*. In addition, samples from dipping baths and callusing media were investigated prior to grafting and planting in the nursery, respectively. During the nursery stage, i.e. between May and November, also soil and air (spore traps) were checked for *Pch*. Wood samples were investigated through visual evaluation of *Pch*-characteristic wood symptoms, cultivation measures on potato dextrose agar (PDA) and a specific nested PCR. Likewise water samples and callusing media were investigated by cultivation measures and nested PCR. Spore traps and soil samples were checked by nested PCR only.

In general an increase of characteristic wood symptoms in the course of the production process was observed. *Pch* was frequently detected in wood samples whereat detection rates were higher after planting of the grafted vines in the fields; it also could be frequently found in dipping baths and spore traps. Sporadic detection exists in callusing media. To date no occurrence in soil was determined. Our findings show the presence of *Pch* in different stages of the plant material production process and that potential infection sources do exist. In order to better estimate the risk of infection emanating from these various substrates further investigations will be conducted in 2015 and 2016.

A new wound closure for vines made of electrospun polymer fibers as a protection against the Esca disease

Melanie Molnar, Michael Fischer
Julius Kühn-Institut, Institute for Plant protection in Fruit Crops and Viticulture, Siebeldingen
Email of corresponding author: melanie.molnar@jki.bund.de

Due to climatic changes and the international trade with vines, the Esca disease, originally seated in the Mediterranean area, has spread worldwide during the last decades. Esca causes high losses in yield every year by symptoms on leaves, black measles and leather berries. The disease is considered to be a complex of three wood living fungi *Phaeomoniella chlamydospora (Pch)*, *Phaeoacre-monium aleophilum* (*Pal*) and *Fomitiporia mediterranea (Fmed)*. In addition symptoms can also be found in the trunk like gummosis, caused by *Pch* and *Pal*, and white rot, caused by *Fmed.* Wounds in the bark of the vines are considered as the main entrance for spores of these fungi.

Therefore particular attention has to be paid to the winter pruning of vines where spores can easily invade the plant through numerous pruning wounds close to the stem head. For that reason the wounds have to be protected. Conventional ways of protection like waxes, resins or the application of fungicides have not led to an improvement of the situation so far. As a result a prototype of a new wound closure based on electrospun polymer fibers has been developed and tested in a three year project.

The process of electrospinning provides elastic and physically stable non woven fiber mats with a defined pore size providing a physical barrier for the pathogens of the Esca disease, especially *Pch* and *Pal* as they infect the plant in an early age. Additionally, the material provides important properties like air- and water permeability promoting the healing process of the plant and prevent rotting.

At the JKI the tightness of different poymers was tested against spores and germination tubes of *Pch*. Also the degradability of the materials was tested in field trials and in aging tests. Furthermore different methods of applications were evaluated as the fibermats should be applied directly after the winter pruning.

Session 2

Plant Pathogens

Fine mapping and identification of candidate genes for a BaYMV/BaYMV-2 resistance gene located on chromosome 5H of barley

Katja Perner [1,2], Dragan Perovic [2], Antje Habekuß [2], Andreas Graner[1], Frank Ordon[2]
[1] Leibniz-Institute of Plant Genetics and Crop Plant Research, Gatersleben
[2]Julius Kühn-Institute, Institute for Resistance Research and Stress Tolerance, Quedlinburg
Email of corresponding author: katja.perner@jki.bund.de

One of the most important diseases of winter barley in Europe and East Asia is barley yellow mosaic virus disease caused by different strains of *Barley yellow mosaic virus* (BaYMV) and *Barley mild mosaic virus* (BaMMV) leading to yield losses up to 50% in susceptible winter barley varieties. Due to the transmission by the soil-borne plasmodiophorid *Polymyxa graminis*, chemical measures to prevent high yield losses are neither effective nor ecologically sound. Thus, breeding for resistance is of prime importance in order to ensure winter barley production in the growing area of infested fields. Up to now, nine different loci conferring resistance to the different strains of BaMMV and BaYMV are known. In order to get detailed information on the structure and function of a resistance gene being only effective against BaYMV and BaYMV-2 located in the centromeric region of chromosome 5H a map based cloning approach was conducted. For marker saturation of the target interval all available sequence information in barley and the synteny to rice, *Sorghum*, *Brachypodium* and sequence information included in the genome zipper was used. Phenotyping for BaYMV/BaYMV-2 resistance of respective segmental RILs derived from a high resolution mapping population comprising 5000 F_2-plants was carried out in field trials followed by DAS-ELISA. Based on marker saturation and phenotyping of 691 RILs the resistance gene was mapped in an interval of 0.22% recombination. By an additional exome capture sequencing approach of the parental lines, 249 morex contigs containing 256 genes were located in this interval. Out of these, two candidate genes were identified of which one is co-segregating with the resistance locus. Sequence analysis of this gene revealed 3 functional SNPs and a 6 bp deletion in the resistant parent.

Further analyses are in progress to get information whether this gene confers resistance to BaYMV/BaYMV-2. This shows that combination of different barley genomic resources and new generation sequencing technologies, applied in map based cloning procedures, is a powerful tool to accelerate soil-borne virus resistance gene isolation in barley.

Different symptomatologies of infected tobacco plants from two full-length cDNA clones of *Apple chlorotic leaf spot virus*

Lei Zhang, Wilhelm Jelkmann
Julius Kuehn-Institut, Institute for Plant Protection in Fruit Crops and Viticulture, Dossenheim
Email of corresponding author: lei.zhang@jki.bund.de

Apple chlorotic leaf spot virus (ACLSV) is the type species of genus *Trichovirus* within the family *Betaflexiviridae*. It has a filamentous particle containing positive single-strand RNA genome of about 7.5 kb. In nature ACLSV infects pome and stone fruit and induces obvious symptoms or hidden disorders on them.

In lab experiments *Chenopodium quinoa* and *Nicotiana occiddentalis* are useful host plants of ACLSV. In the present work different symptomatologies were observed on *N. occiddentalis* 37b which were infected with constructed full-length cDNA clones of two different ACLSV isolates of apple and pear, respectively. ACLSV-infected samples of apple and pear leaves were collected from the Virus Resource Center at the JKI Dossenheim. The In-Fusion™ cloning system was used for constructing cDNA clones. The cDNA genome of ACLSV was ligated to a binary vector (a pBin vector, E. Maiss, Leibniz University, Hannover) under control of a 35S promoter.

The efficiency of the constructs was tested on *N. occiddentalis* 37b by Agrobacterium-mediated transformation. Symptoms of yellow/chlorotic spots developed between 8 to 12 dpi on infected plants. The infectious cDNA clone of the apple isolate showed considerably stronger symptoms compared to the one of pear. Over time systemic symptoms were observed on infected plants.

Development of Fire Blight antagonists after application on apple flowers

Christine Hübert[1], Helmut Junge[2], Kristin Dietel[2], Annette Wensing[1], Wilhelm Jelkmann[1].
[1] Julius Kuehn-Institute, Institute for Plant Protection in Fruit Crops and Viticulture, Dossenheim
[2] ABiTEP GmbH, Berlin
Email of corresponding author: christine.huebert@jki.bund.de

Fire Blight is a devastating bacterial disease which can cause great economic losses for fruit growers. Control mechanisms are limited to a few compounds with adequate efficiency, but the need for alternatives still remains.

Two main problems have to be considered in the development of control agents against the Fire Blight pathogen *Erwinia amylovora*: First, the ability of exponential growth leads to high cell densities in a short amount of time. Second, the most critical phase of Fire Blight infection occurs during blossoming, when the pathogen is transported to open flowers by various insects. To prevent infection, it is important to avoid invasion inside the plants tissue by interfering with growth of *E. amylovora* cells.

We are testing two different bacterial antagonists in their ability to inhibit the Fire Blight pathogen: The Gram negative bacterium *Erwinia tasmaniensis* and a Gram positive representative of *Bacillus amyloliquefaciens*.

E. tasmaniensis is closely related to the pathogen and is well adapted to the Fire Blight habitat. On the other hand, *Bacillus amyloliquefaciens* is commonly known as a classical soil habitant, its ability to adapt to the flower might be restricted.

Both antagonists are able to establish themselves in the field. However, efficiency against *E. amylora* differs between *E. tasmaniensis* and *B. amyloliquefaciens*.

Bacillus is fairly simple stored as spore formulations. Nevertheless, before growth of vegetative cells and inhibition against a pathogen can occur, spore germination has to take place. This might be a factor that can cause a delay in development on the flower.
Therefore, we investigated and compared the development of our antagonists after application onto apple flowers in a detached-flower assay in the lab to determine the growth rate and maximum cell densities.

Even though *E. tasmaniensis* as well as *B. amyloliquefaciens* start to grow 12 to 15 hours after application on the flower, the former reveals a steeper rise. Moreover, end cell densities differ and *B. amyloliquefaciens* reaches total cell numbers 10 to 100 fold below *E. tasmaniensis*.

A lower cell density might be a reason for a weaker efficiency in *E. amylovora* inhibition. To overcome this discrepancy an additional nutrient which is only favorable for the antagonist might be a useful additive to increase efficiency.

Session 3

Entomology

Establishing of the entomopathogenic fungus *Isaria fumosorosea* as an endophyte in *Triticum aestivum* and molecular detection of strain JKI-BI-1496

Manuel Werner, Katharina Saar, Dietrich Stephan
Julius Kühn-Institut, Institute for Biological Control, Darmstadt
Email of corresponding author: katharina.saar@jki.bund.de

Entomopathogenic fungi gain more and more influence in biocontrol and protection of plants. Many species of entomopathogenic fungi in various genera are known and some of them show promising effects as biocontrol agents (BCA).

The entomopathogenic fungus *Isaria fumosorosea,* formerly known as *Paecilomyces fumosoroseus,* has got a relatively wide host range. Within the scope of the EU project BIOCOMES investigations were done to validate the use of *I. fumosorosea* as a BCA against several pest insects. Under laboratory conditions it could be shown that *I. fumosorosea* seems to be a suitable BCA against *Bemisia tabaci* (silverleaf whitefly) and *Spodoptera exigua* (beet armyworm).

Based on the previous investigations we tried to establish three different strains of *I. fumosorosea* (JKI-BI-1496, JKI-BI-1497 and JKI-BI-1508) as endophytes in *Triticum aestivum* cv. 'Apogee' to protect plants from pest insects and phytopathogenic fungi like *Fusarium proliferatum, Verticillium dahliae* and *Fusarium solani.* Moreover, the potential as an antagonist against the mentioned phytopathogenic fungi as well as positive or negative influences on plant biomass production were analyzed. Therefore, different parts of the plant were surface sterilized and incubated with the objective to re-isolate the three strains.

Furthermore, DNA was extracted from these *I. fumosorosea* strains to amplify partial sequences of different genes like beta-tubulin and the ITS-region of the ribosomal RNA operon. In addition, after sequencing, phylograms were created.

To determine, if the re-isolated probes from dead insects and plant tissue are the given strains, molecular investigations were done. To begin with and set as a priority the aim is to design a specific primer for *I. fumosorosea* strain JKI-BI-1496 to determine its presence when used in experiments.

Characterization of the Tunisian PhopGV isolate TU1.11; molecular identification and biological activity on *Phthorimaea operculella* larvae

Saoussen Ben Tiba[1,2,3], Andreas Larem[1], Eva Fritsch[1], Karin Undorf-Spahn[1], Asma Laarif[2], Sami Fattouch[3], Johannes A. Jehle[1]

[1] Julius Kühn-Institut, Institute for Biological Control, Darmstadt, Germany

[2] Regional Center for Agriculture and Biological Agriculture, Chott Meriem, Tunisia

[3] National Institute for applied technological Science Tunis (INSAT), Carthage University, Tunisia

Email of corresponding author: saoussen.ben.tiba@jki.bund.de

The *Gelechiidae Phthorimaea operculella* (Zeller) is a Lepidopteran pest, which causes serious damages in potato crops in the fields and stored tubers in sub-tropical and tropical regions. One of the infectious enthomopathogenic agents against this pest is *Phthorimaea operculella* granulovirus (PhopGV) of the family *Baculoviridae.* Baculoviruses are considered as potent biological control agents for different insect orders. The genome of the Tunisian isolate PhopGV 1346 is fully sequenced. It is used as a reference to characterize the PhopGV isolate TU1.11, which is in the focus of this study. For that, different fragments of the TU1.11 genome are sequenced, such as the granulin gene and the ecdysteroid UDP-glucosyltransferase (*egt*) gene. The *egt* gene of the reference isolate PhopGV-1346 is 1353 bp in length unlike the size of *egt* gene identified in PhopGV TU1.11, which was 1086 bp, placing this isolate in *egt* group III.

TU1.11 was tested against *P. operculella* larvae for its biological activity. First bioassay results showed that the larval development was retarded and that larvae, which were still alive, did not emerge into pupae, even 15 days post infection. Exposed to PhopGV, *P. operculella* do not develop to adults, thus, the population size of the next generation may decrease because of PhopGV infection.

Investigation on pathogenic antagonists of selected insect pests – an overview

Sarah Biganski, Regina G. Kleespies, Johannes A. Jehle
Julius Kühn-Institut, Institute for Biological Control, Darmstadt
Email of corresponding author: sarah.biganski@jki.bund.de

One recently occurred invasive insect pest has caught the attention for investigating biological control mechanisms and systems: The spotted wing drosophila (SWD, *Drosophila suzukii* Matsumura) is endemic in East China and Japan but has been introduced to the western hemisphere about 10 years ago and has been found in Europe since 2009. Nowadays, it has emerged to one of the most harmful pests to commercially grown fruit plants like stone fruits and nearly all kind of berries while it prefers ripe and overripe fruits. Our intention is to investigate the possible usage of natural antagonists for biological control.

Therefore, we examine the natural load of parasites and pathogens (i.e. fungi, bacteria, viruses, microsporidia and protista) in fruit flies, isolate them and re-infect lab populations of *D. suzukii* for investigating the antagonistic potential. Furthermore, we will integrate the fruit pest codling moth (*Cydia pomonella*), which is an ongoing problem in apple orchards also because the pest develops resistance against commercially available insecticides. The long-term aim is to establish a stable system for pathogen detection that can be used for rapid identification of microbial antagonists in natural populations.

Determination of the potential of the entomopathogenic fungi *Isaria fumosorosea* as a BCA against pest insects

Katharina Saar[1], Jasmin Philippi[2], Edgar Schliephake[2], Andreas Leclerque[3], Manuel Werner[1], Johannes A. Jehle[1], Dietrich Stephan[1]

[1] Institute for Biological Control, Julius Kühn Institute, Darmstadt

[2] Institute for Resistance Research and Stress Tolerance; Julius Kühn Institute, Quedlinburg

[3] Institute for Microbiology and Biochemistry, Geisenheim University, Geisenheim

Email of corresponding author: katharina.saar@jki.bund.de

Entomopathogenic fungi like *Isaria* sp., *Metarhizium* sp. and *Beauveria* sp. are important biological control agents due to their lethal effects against several insect pests. The aim of the EU-funded project BIOCOMES is therefore the use of the natural ubiquitous entomopathogenic fungus *Isaria fumosorosea* (Ascomycota: Hypocreales: Cordycipitaceae) in an integrated pest management strategy to control pest insects in greenhouses.

The selection of highly virulent and effective strains is one of the necessary prerequisites. Therefore, several strains of *Isaria* sp., obtained from different geographical origins and hosts, were compared for their efficacy and virulence against various pest insect species from the orders Lepidoptera and Hemiptera. By means of various pathogenicity assays, the efficacy and median lethal concentration (LC_{50}) of different *Isaria* strains against *Spodoptora exigua* (beet armyworm) and *Bemisia tabaci* (whitefly) were investigated. Depending on the *Isaria* strain, the mortality rates differed from 30 % to 76 %.

The feeding of whiteflies on plants may cause direct damage to the leaves as well as virus transmission by their sucking behaviour. In addition to the mortality effects, insects showed changes within their behaviour when they were infected with *I. fumosorosea*. In order to evaluate whether adults of *B. tabaci,* which had been infected and non-infected with *Isaria*, showed differences in their feeding (sucking) behaviour, the electrical penetration graph method (EPG) was used to observe behavioural changes, e.g. penetration frequencies. First results of the comparisons concerning the changes within the waveform and durations will be shown.

Presently, different molecular methods like RFLP are used for the characterization of virulence factors and differences between the strains of *I. fumosorosea* from different geographical origins and hosts and its influence on different activity patterns as mentioned above.

Session 4

Climate Change and Ecological Aspects

Development and field test of a pneumatic seeder for a sustainable reduction of risk to the ecosystem and the user by dust of seed dressings

Christoph Kämpfer, Jan-Philip Pohl, Dieter von Hörsten, Dirk Rautmann
Institute for Application Techniques in Plant Protection, Julius Kühn-Institut, Braunschweig
Email of corresponding author: christoph.kaempfer@jki.bund.de

The project is dealing with the improvement of a universal pneumatic seeder which is suitable for sowing a wide range of different seeds in order to decrease particulate emissions of seed dressings. Aim of the project is to identify leakages in the pneumatic system in order to develop opportunities for the subsequent improvement of existing equipment to fulfill the high demands of user and environmental protection. Pneumatic seeders are characterized by a central, funnel-shaped hopper. The batch feeder being an airlock and dosing feeder is located in the outlet of the seeder. The metered seed is fed into the air stream and transported to the sowing distributor by a conveying air stream.

In 2008 around 12,000 bee colonies where eliminated in the south-east of Germany because of particulate emissions of Clothianidin being discharged in the process of sowing maize. The following evaluation of this incident led to the conclusion that the quality of the seed dressing as well as the seeding technology used was not sufficient in order to suppress particulate emissions of pesticides in a satisfactory manner with regard to human health and the environment. Nevertheless, there is a strong need for seed dressing in agriculture being a highly effective method of plant protection in practice. Therefore, it is necessary to develop a seeder, which reduces the drift of particulate emissions from seeding to a minimum being acceptable with regard to human health and the environment.

The demand for large pneumatic seeders rises because of structural change in agriculture. The average field sizes are increasing and large seeders are a basis for high performance. But this development also leads to higher air volume output on seeding machines which will increases the drift problem, if further technical developments reducing the risk of drift will not be done.

The focus of the project is the development of an efficient and ecofriendly pneumatic seeder, which fulfills the high requirements in terms of minimizing the risk of drift. The industry has made some efforts and is interested to adopt the technical requirements to their pneumatic seeders in order to develop innovative solutions for future markets.

A literature-based approach to estimate the effect of climate change on plant protection: The example winter wheat

Johanna Hemetzberger, Bernd Freier
Julius Kühn-Institut, Institute for Strategies and Technology Assessment, Kleinmachnow
Email of corresponding author: johanna.hemetzberger@jki.bund.de

Climate change not only affects crops directly, but also changes conditions for plant diseases, pests and weeds as well as their antagonists.

This contribution is part of the project "Konsequenzen des Klimawandels für die Nachhaltigkeitsziele beim Pflanzenschutzmitteleinsatz" and aims to collect the current knowledge about these effects on the occurrence of pests, diseases and weeds of winter wheat as well as the resulting changes in plant protection strategies.

The diseases Septoria leaf spot, powdery mildew, brown and yellow rust, eyespot and Fusarium head blight as well as insect pests such as cereal aphids, wheat midges and cereal leaf beetles were covered in this study. In addition, the most common weeds of winter wheat were included. The focus of the literature survey was the effect of the expected warming, increased frequency of spring and early summer drought and heavy precipitation events.

In the first step, predictions for future occurrence of the above-named pests were compared. The information was summarized in climate sheets for each pest including their climatic requirements and scenarios for their response to climate change.

In the next step, the expected changes in occurrence of diseases, pests and weeds were each classified into three categories (minor, medium and high increase/decrease) based on the respective climate sheets.

In the third step, treatment frequency indices (TFI) were used to describe the intensity of the plant protection strategies. The TFI for the scenario 2050 were based on values for the reference period 2007 - 2013 and adjusted according to the findings from the literature survey. An expected minor, medium or high increase/decrease in disease, pest or weed occurrence was translated into a change of the TFI by 25%, 50% or >50% for fungicides, insecticides or herbicides.

Results of this literature-based analysis imply that especially rusts will benefit from future warmer conditions, while the occurrence of other diseases like eyespot and powdery mildew is likely to remain unchanged. Insect pests in general are expected to gain importance in winter wheat mainly due to warming. Weeds are diverse in their response to climatic change depending on their individual requirements.

Consequently the derived scenarios for future plant protection strategies imply that the intensity of fungicide and herbicide use will remain mostly unchanged, while a medium increase in insecticide use is expected.

The gained information on possible trends in plant protection is useful for the evaluation of ecological impacts of plant protection in the future.

Session 5

Molecular Insights

Analysis of SPO11 protein interaction during meiosis

Thorben Sprink, Frank Hartung and Joachim Schiemann
Julius Kühn-Institut, Institute for Biosafety in Plant Biotechnology, Quedlinburg
Email of corresponding author: thorben.sprink@jki.bund.de

Meiosis as the specialized cell division of sexual reproduction plays a crucial role in the exchange and reorganization of genetic material between two individuals by dividing the chromosome set in half and forming gametes. Even though in the last years major findings in the field of meiosis have been achieved, especially in plants, some key questions remain concealed. For a proper meiosis the initiation of double strand breaks (DSBs) during early prophase I is essential. Without DSBs no physical connection can occur between homologous chromosomes and recombination, pairing, and crossing over are excluded. So far in all analyzed eukaryotes SPO11, a meiosis specific transesterase, is the key enzyme inducing DSBs. But other than in animals and fungi where a single SPO11 is sufficient, plants need at least two different SPO11, referred to as SPO11-1 and SPO11-2, for proper meiosis. In *Arabidopsis thaliana* both have crucial functions and are essential in a functional form for the induction of meiotic DSBs as single knock out mutants are leading to near sterility by random chromosome distribution. Despite the same function of the homologs SPO11-1 and -2, the identity between both proteins is quite low. Homology of the orthologous SPO11 from different organisms is much higher. By exchanging SPO11-1 and -2 in Arabidopsis by their orthologs from various organisms we could demonstrate a species specific function of each SPO11, as a functional complementation of sterility could only be achieved with SPO11 from closely related species from the Brassicaceae. By exchanging non conserved regions between SPO11-1 and -2 of Arabidopsis we additionally could show a sequence specific function for each SPO11, as a functional rescue could not be achieved with all chosen regions. Interestingly, we could reveal a specific pattern of aberrant spliced isoforms for each SPO11 which are also sequence as well as species specific. By producing antibodies against AthSPO11-1 and -2 we were able to analyze for the first time the binding of SPO11-2 onto the DNA and perform co-immunolocalization studies with SPO11-1 and -2.

With this novel knowledge we further want to analyze possible interactions of SPO11-1 and -2 with each other and/or with other proteins involved in meiosis. To date nothing is known about such interactions in plants. By using our self developed antibodies as well as SPO11 fused to tags, we want to perform pull down assays as well as interaction studies, aiming to identify possible interaction partners of SPO11-1 and -2. Additionally, we will mutate and exchange various regions between SPO11-1 and -2 in Arabidopsis using CRISPR/Cas9 to deepen and confirm the results gained from the previous studies.

Identification of transcriptome-based molecular markers linked to stem-rust resistance in perennial ryegrass

Jens Bojahr[1], Björn Rotter[3], Nicolas Krezdorn[3], Ottilia Nhengiwa[4], Bernhard Saal[4], Brigitte Ruge-Wehling[1], Christine Struck[2], Peter Winter[3], Peter Wehling[1].

[1] Julius Kühn-Institut, Institute for Breeding Research on Agricultural Crops, Rudolf-Schick-Platz 3a, 18190 Groß Lüsewitz

[2] University of Rostock, Faculty of Agricultural & Environmental Sciences, Group Crop Health, D-18059 Rostock

[3] GenXPro GmbH, Altenhöferallee 3, D-60438 Frankfurt

[4] Saatzucht Steinach GmbH & Co KG, Wittelsbacherstrasse 15, 94377 Steinach

Email of corresponding author: jens.bojahr@jki.bund.de

Perennial ryegrass (*Lolium perenne*) is one of the most important cool-season grass species in temperate zones worldwide and used in forage production and as turf grass. Seed production of ryegrass is affected by stem rust caused by the obligate biotrophic pathogen *Puccinia graminis* f.sp. *graminicola* and causes yield loss up to 98 %. A perennial ryegrass mapping population segregating for stem-rust resistance was screened with three stem rust field isolates in a leaf-segment test. Leaf segments of inoculated and non-inoculated resistant and susceptible individuals were bulked, respectively, at three time points (before inoculation, 4–8 and 18–24 hpi). Bulked segregant analysis of differential gene expression was accomplished via massive analysis of cDNA ends (MACE) and RNAseq. MACE detected genome-wide, quantitative expression profiles of 57 million transcripts along with their allelic diversity. Transcripts were assembled into 144,000 contigs and functionally annotated to the SwissProt database using BLASTX and the NCBI *Viridaeplantae* database using BLASTN. In total, 401 transcripts exclusively expressed in the "resistant" bulks (ERTs) were identified, including eight transcripts with homology to disease resistance genes. In addition, SNPs of RNAseq and MACE which occurred exclusively in the “resistant” bulks were filtered. ERTs and SNPs were annotated to the genome of the model grass species *Brachypodium distachyon*. Most of the ERTs and SNPs mapped on *Brachypodium* chromosome 1 (Bd1), with a peak falling into the physical region of 25.5 – 34.5 Mbp. To predict the genomic location of the stem-rust resistance gene in *L. perenne*, the perennial ryegrass GenomeZipper based on the conserved synteny between the grass species including *B. distachyon* was used. The peak of ERTs and SNPs on Bd1 showed macro-synteny to *L. perenne* chromosome 7. ERTs and SNPs annotated to Bd1 were used for PCR primer design. In total, 87 primer pairs were designed, of which 27 showed genetic linkage to stem-rust resistance.

Identification of genomic regions involved in tolerance to drought stress and drought stress induced leaf senescence in juvenile barley

Gwendolin Wehner[1,2], Christiane Balko[1], Klaus Humbeck[3], Frank Ordon[1,2]
[1] Julius Kühn-Institut (JKI), Institute for Resistance Research and Stress Tolerance Quedlinburg/ Groß Lüsewitz
[2] Interdisciplinary Center for Crop Plant Research (IZN), Halle (Saale)
[3] Martin-Luther-University Halle-Wittenberg, Institute of Biology, Halle (Saale)
Email of corresponding author: gwendolin.wehner@jki.bund.de

Premature leaf senescence induced by external stress conditions, e.g. drought stress, is a main factor for yield losses in barley (*Hordeum vulgare* L.). Research in drought stress tolerance has become more important as due to climate change the number of drought periods will increase and tolerance to drought stress has become an important goal in barley breeding.

Therefore, the aim of this subproject was to identify genomic regions involved in drought tolerance and leaf senescence in early developmental stages of barley. For this purpose phenotyping, genotyping and expression analyses were conducted on 156 genotypes and based on these data genome wide association studies were performed. After a four weeks stress period (BBCH 33) six physiological parameters for drought stress and leaf senescence were determined in the control and stress variant in greenhouse pot experiments.

Significant phenotypic variation was observed for all traits analysed and significant effects of genotype, treatment and genotype x treatment were estimated for most traits analysed. Based on these phenotypic data and 3,212 polymorphic SNPs with a minor allele frequency >5 % derived from the Illumina 9k iSelect SNP Chip, 181 quantitative trait loci (QTL) were detected for all traits analysed. Major QTL for drought stress and leaf senescence were located on chromosome 2H and 5H. Expression analyses of a set of 14 genes involved in drought stress and early leaf senescence on these 156 genotypes resulted in the identification of 13 expression QTL. One of those is located in the same region of chromosome 5H as the QTL for biomass yield and leaf colour under drought stress. Respective markers may be used in future barley breeding programmes for improving tolerance to drought stress and leaf senescence.

Identification of candidate genes for the prehaustorial resistance of *Triticum monococcum* against *Puccinia triticina*

Mathieu Deblieck[1], Albrecht Serfling[1], Frank Ordon[1]
Julius Kühn-Institut, Institute for Resistance Research and Stress Tolerance, Quedlinburg
Email of corresponding author: mathieu.deblieck@jki.bund.de

Leaf rust caused by *Puccinia triticina* can cause yield losses up to 60 % and is the most common rust disease of wheat in the world. Vertical leaf rust resistance genes (*Lr*-genes) have been introduced in cultivars. Many of these resistances are broken down by virulent pathotypes. Horizontal resistances which are independent from races of a pathogen are known but show a quantitative characteristic which is carried by a few cultivars.

In Einkorn (*Triticum monococcum)* a horizontal resistance against *P. triticina* was identified. Microscopic analysis revealed that this type of resistance is accompanied by an early hypersensitive response that prevents the fungus to form haustoria 8-24 hours after infection. Furthermore expression studies using the massive analysis of cDNA ends (MACE) pointed out that prehaustorial resistant plants increase the expression of peroxidase and chitinase genes after an inoculation. To elucidate the genetic base of this quantitative resistance, a resistant and susceptible Einkorn accession was crossed, F2 plants were phenotyped and the most diverging plants were used for differential array technology analyses (DarTs). As a result QTL-regions on Chromosome 2A and 5A which were related to the prehaustorial resistance could be identified.

In the scope of this project 41 *T. monococcum* contigs were anchored to these QTL-regions. 13 genes within three of four QTL-regions could be assigned to MACE-tags. Two genes seem to be involved in defense responses of the plant. Only one of the 13 genes was differentially expressed between the resistant *Einkorn* accession and the susceptible one. This gene encodes a beta carotene hydroxylase and was downregulated in the resistant Einkorn after infection with *P. triticina*. The enzyme beta carotene hydroxylase is involved in the carotenoid synthesis which affects the generation of abscisic acid (ABA). An experimental treatment of leaves with ABA led to partially inhibited defense of the prehaustorial resistant Einkorn. This confirms that the beta carotene hydroxylase encoding gene is one of the candidate genes involved in the prehaustorial resistance.

Poster Session

PRUNI-REPEL: Developing an innovative push-and-pull strategy

Jannicke Gallinger, Jürgen Gross
Julius Kühn-Institut, Institute for Plant Protection in Fruit Crops and Viticulture, Dossenheim
Email of corresponding author: jannicke.gallinger@jki.bund.de

European Stone Fruit Yellows (ESFY) is one of the most serious diseases in European fruit production. Infected *Prunus* cultivars yield poorly and lead to high economic losses. ESFY is caused by a specialized bacterium located in the phloem tissue of *Prunus* ssp., the Phytoplasma '*Candidatus* Phytoplasma prunorum'. It is spread by the phloem-feeding plum psyllid (*Cacopsylla pruni*) which acquires the bacterium by feeding on infected plants and is able to transmit it to healthy plants.

C. pruni is a homometabolic, univoltine jumping plant lice. Within one generation it is changing its host plant two times. After development of nymphal stages on *Prunus* ssp., the young adults, called emigrants, migrate to overwinter on conifers (e.g. spruce). In early spring they come back (remigrants) to reproduce on *Prunus*.

Many insects use allelochemicals for localisation of their hosts. To take advantage of the olfactory orientation of *C. pruni*, the volatile organic compounds released by the different host plants are collected by headspace technique and analysed via GC-MS. Previous investigations showed that *C. pruni* is more attracted by *Prunus* rootstocks than by cultivars. Due to differences in the emission of plant volatiles from *Prunus* rootstocks, *Prunus* cultivars and spruce potential attractants and repellents were identified. The effects of single compounds and mixtures on the behavior of *C. pruni* emigrants and remigrants were proven by bioassays, in a Y-shaped dynamic olfactometer.

In spring 2015 first field experiments were carried out to elaborate a push-and-pull strategy against *C. pruni*. A trap developed by the JKI Dossenheim was established as monitoring tool for psyllids.

To offend the vector from very attractive *Prunus* rootstocks, dispensers filled with a mixture of repellent compounds, were applied in the field. This application reduced the number of captured individuals of *C. pruni* emigrants in monitoring traps. Next step for developing an efficient push-and-pull system is to improve the formulation of the repellents and to find a highly attractive substance, to lure the plum psyllids into traps.

Genetic Background Selection in Grapevine Breeding exemplified by the Cross `Blaufraenkisch´ x `Catawba´

Franziska Huber, Florian Schwander, Rudolf Eibach, Reinhard Töpfer
Julius Kühn-Institut, Institute for Grapevine Breeding, Siebeldingen
Email of corresponding author: franziska.huber@jki.bund.de

Wild vines originally from North America, like the species *Vitis labrusca*, are an important resistance source in grapevine breeding. But a common problem of interspecific crosses is to meet the wine quality requirements due to possible off-flavor inheritance. In order to face this problem more efficiently, it is proposed to use the marker-assisted selection (MAS) strategy. In this study it is shown how this can be accomplished.

The natural methyl anthranilate (MA) is assumed to contribute to the Labrusca-typical "foxy" flavor and called to account mothball taste. MA does not exist in perceivable concentrations in the high-quality European *Vitis vinifera*. For the localization of the chromosomal region responsible for MA synthesis, MA was quantified in 226 descendants of the cross population `Blaufraenkisch´ x `Catawba´ by gas chromatography.

The triannual studies revealed one main quantitative trait locus (QTL) on the linkage group (LG) 4. In this QTL, a DNA sequence similar to the *aamt1* gene can be found on the *Vinifera* `Pinot noir PN40024´ reference genome. AAMT1 is a key enzyme in the MA synthesis in maize where it is produced in case of leaf damage and therefore is probably involved in plant defense attracting the herbivores' predators.

The second step was to perform ancestry investigations: `Catawba´ is one of the oldest commercialized North American cultivars. In former times it was speculated to originate from *V. vinifera* and a native wild species. Its genetic fingerprint was compared systematically with the data of the Geilweilerhof grapevine repository and revealed *V. vinifera* `Sémillon´ as its ancestor. The genetic map of `Blaufraenkisch´ x `Catawba´ consists of 337 genome-wide SSR markers. Using this marker data it was possible to identify the *Vinifera* alleles of `Blaufraenkisch´ and `Sémillon´ in the progeny for background selection in a marker-assisted backcrossing (MABC) approach. As expected, the *Vinifera* portion was 73.54 % in average with a maximum value of 82.95 % and a minimum of 62.90 % among the offspring. The trait-independent *Vinifera* markers plus the knowledge about the MA QTL can now promote breeding programs with the `Catawba´ progeny: A screening can be done for the genotypes with the highest *Vinifera* portion and having no MA QTL on LG 4 to use them for crossing the next generations. For the breeders, the information about the chromosome where the allele for the undesired quality trait is located is already sufficient without enclosing the region more precisely.

NoViSys*:* Novel Viticulture Systems for sustainable production and products

Anna Kicherer[1], Florian Schwander[1], Katja Herzog[1], Reinhard Töpfer[1], Bernd Weisshaar[2]
[1] Julius Kühn-Institut, Institute for Grapevine Breeding, Siebeldingen
[2] Bielefeld University, Faculty of Biology, Center for Biotechnology, Bielefeld
Email of corresponding author: anna.kicherer@jki.bund.de

The aim of the NoViSys project is the investigation of innovative new growing systems in viticulture.

Within the last 100 years high quality, fungus resistant grapevine cultivars so-called PIWIs (for "Pilzwiderstandsfähige Rebsorten") are the biggest innovation in viticulture. These varieties are the approach to reduce the environmental concerns of the public regarding the extraordinary high input of fungicides in both conventional and organic viticulture. The novel cultivation method of the "minimal pruning of trellis trained grapevines" (MPTS) enables a high reduction of manual work by exclusive mechanical pruning and yield reduction. Combining the use of resistant cultivars with the MPTS cultivation system, a grapevine production which is environmental friendly, economically beneficial and adapted to the on-going climatic changes can be achieved.

To evaluate new resistant grapevine cultivars in such an advanced production system an interdisciplinary consortium was formed comprising a high level of expertise for all important fields of the wine sector. This includes grapevine breeding, plant protection, visual and sensor based screening, application engineering, physiological and molecular analyses, analytical and sensorial evaluation of grape must and wine, business management, wine marketing, and social economy.

The goal of NoViSys is to evaluate the behavior of different grapevine cultivars in the most common vertical shoot positioning in trellis-system (TS) and compare it to MPTS. In addition the typical reaction of those cultivars to the mechanical thinning which is needed in MPTS cultivation to reduce yield, is validated. Therefore sensor-based approaches using cameras and 3D point cloud reconstruction is used.

Another focus is laid on the validation of grape cluster architecture, the biodiversity in the vineyards and the quality of resulting wines will be evaluated and compared. We intend to unravel the cause of ripening delay upon viticultural treatments, and to develop the technological basis for a broad introduction of the new cultivation system into viticultural practice.

Our comprehensive investigation will empirically and functionally address field studies where new cultivars are raised in the new cultivation concept. We will generate validated information for the wine growers to demonstrate benefits and risks. In addition the economical advantages as well as supporting marketing to improve consumer acceptance will be investigated.

Biotechnological investigations on the natural rubber producer *Taraxacum koksaghyz* concerning the herbicide target AHAS

Regina Kölzsch, Katja Thiele, Thomas Berner, Frank Hartung, Joachim Schiemann
Julius Kühn Institute, Institute for Biosafety in Plant Biotechnology, Quedlinburg
Email of corresponding author: regina.koelzsch@jki.bund.de

The Russian dandelion *Taraxacum koksaghyz* (Tks) is one of the most promising candidates as an alternative for the rubber tree *Hevea brasiliensis*. This crop is currently the sole commercial source for natural rubber, saving the worldwide needs for producing tires and other goods which cannot be replaced by synthetic analogs. Presently, the global population of this tree is threatened by the fungus *Microcyclus ulei* that leads to dieback of trees. Tks is excellently appropriate to build up new resources concerning the physical and chemical characteristics of the rubber. Until now the cultivation and harvest of Tks is elaborative and the yield is not yet comparable to the one of *H. brasiliensis*. Further biotechnological and agricultural optimization is needed to achieve a crop that can substantially contribute to the world rubber market.

Tks grows under sparse conditions in the region of the Tian Shan Mountains and is therefore very eligible for being grown on marginal acreages in the European climate zone. As Tks has a very slow early growth phase, weed is overgrowing the young plant resulting in reduced size and rubber yield. In addition to other projects focusing on improvement of field performance by e. g. breeding or fertilization, our part of the "EVITA" project aims to develop an imidazolinone resistance for a sustainable weed control during Tks seed production. By screening the gene sequence of the herbicide target (acetohydroxyacid synthase, AHAS) of wild type and EMS-mutagenized plants which are imidazolinone tolerant, we get information about the natural sequence variability and induced mutations. So far we could not find herbicide resistance endowing SNPs – suggesting that the investigated AHAS1 gene might not play the prominent role in this herbicide resistance mechanism. By using degenerated primer as well as the RACE method we are searching for other AHAS genes in the Tks genome which are relevant for imidazolinone resistance. Further we want to use the recently developed but well known CRISPR/Cas9 nuclease system for introducing specific mutations in a targeted manner to provoke imidazolinone resistance. As a proof of concept this will be done first with the already known AHAS1 gene.

As this work is generating a lot of sequencing data, it is necessary to have an easy, fast and reliable tool for their analysis. In addition to manual evaluation with the CLC Main Workbench, usage of the web-based workflow management system Galaxy proved to be efficient for detecting SNPs in hundreds of reads. By defining workflows with parameters and error rates, analysis is simplified and less afflicted to subjective errors.

Establishment of a fingerprinting method for analysis of fungal communities on grapevine

Christian Kraus[1], Michael Fischer[1], Ralf Vögele[2]
[1] Julius Kühn-Institut, Institute for Plant protection in Fruit Crops and Viticulture, Siebeldingen
[2] University of Hohenheim, Department of Phytopathology, Hohenheim
Email of corresponding author: christian.kraus@jki.bund.de

Since the last two decades a number of ´fingerprinting´ methods have been developed to analyze microbial communities and their dynamics, including Terminal Restriction Fragment Length Polymorphism (T-RFLP), Length Heterogeneity-Polymerase Chain Reaction (LH-PCR) and Automated Ribosomal Intergenic Spacer Analysis (ARISA). Because the latter provides a quick and cheap way together with high accuracy, we have chosen this method to investigate the fungal communities on grapevine, wood, leaves and berries.

As a first step in ARISA, a PCR approach amplifies the internal transcribed spacer (ITS) region of fungal DNA samples. As the ITS region represents non-coding DNA, it is extremely variable in nucleotide sequence and length. One of the primers used in the PCR is labeled with a fluorescent dye, which can be detected as a peak in an electropherogram. Because of the length variability of the ITS region, each fungal species ideally forms an individual peak in the electropherogram. Furthermore the height of the peak gives a hint about the abundance of the fungus within the community.

To assign the peaks to the corresponding fungus, a database has to be created, containing the peak position (correlating with the size of the amplicon) for each fungal species.

Taken together the ARISA provides a valuable tool which gives important information not only about the fungal diversity within a given sample but also about the quantity of each fungus.
This technique shall help us to study the composition of fungal communities on grapevine and how they change compared to different environmental conditions in relation to cultivars as well as training and plant protection regimes.

Optimized gap detection for precise application of PPP in orchards

Verena Overbeck[1], Jonas Huhs[2], Jens-Peter Ralfs[2], Tanja Pelzer[1]
[1] Institute for Application Techniques in Plant Protection, Julius Kühn-Institut Braunschweig
[2] Research and Extension Centre for Fruit Growing (OVA), Chamber of Agriculture Lower Saxony, Jork
Email of corresponding author: verena.overbeck@jki.bund.de

The use of plant protection products (PPP) in orchards is indispensable to maintain crop health and to secure yield. To reduce potential inputs into the environment, the reduction of the amount of PPP and the minimization of drift are important social and political objectives. Therefore, the use of sensor technology for gap detection and precise application offers a technical solution to comply with these aspects.

In a prior project (LADUS), a master plan for an optimized tree gap detection based on an optimal harmonization of sensors and nozzles was developed. For this purpose, a sprayer with a radial fan (NH 63) was equipped up to twenty optical infrared sensors on both sides of the sprayer. The technical request for the gap detection was to achieve a high compliance between the scanning field of a sensor and the spatial extend of the spraying area of a single nozzle.

Field experiments in Jork ("Altes Land") have shown, that in different fruit orchards a precise application can reduce the output quantity of PPP, especially in young orchards with a saving potential of 70 %. At the same time, the evaluation of the coverage in the gap showed significant lower values compared to a conventional application without gap detection.

In general, the acquired knowledge out of the different trials enables the construction of a cross-flow fan sprayer and the retrofitting of sprayers in use. One focus of the new project OLSVA will be the evaluation of the biological efficiency of the gap detection system with the different sprayer types. However, for precise recommendations there are several years of investigations necessary.

The aim of the project is to develop a product with a market maturity for new sprayers just as for retrofitting sprayers in use. Therefore, the equipment has to be extremely reliable and robust as well as affordable. In the same way, the product has to fulfill the requirements for a high biological efficiency.

Analysis of color-related mutations in bud sports of *Vitis vinifera* L.

Franco Röckel, Ludger Hausmann, Erika Maul, Reinhard Töpfer
Julius Kühn-Institut, Institute for Grapevine Breeding Geilweilerhof, Siebeldingen
Email of corresponding author: franco.roeckel@jki.bund.de

Grapevines are one of the oldest agricultural crops and produce table fruits, dried fruits, juice and wine. The number of different varieties worldwide is estimated at 8000 to 10 000 and most of these cultivars belong to the domesticated European species *Vitis vinifera ssp. vinifera*. The increased breeding effort of the last centuries produced a lot of new varieties; however, most arose due to steady crosses between domesticated and wild grapes or growing seedlings from open pollination.

All known wild grapes have a blue/black berry color, whereas the cultivated grapes show a broad color spectrum ranging from green/white to blue/black. Responsible for the coloration of the berries is mainly the composition and concentration of five anthocyanins exclusively in the grape skin. The anthocyanin biosynthesis is controlled by a cluster of four MYB-related transcription factors located on chromosome 2 from which only two adjacent genes are considered as functional (*VvmybA1* and *VvmybA2*). At least one of these two genes needs to be functional for color expression. An insertion of a retrotransposon (*Gret1*) in the promoter region of *VvmybA1* and two amino acid-changing mutations in the coding sequence of *VvmybA2* lead in combination to a non-functional "white" allele. It could be shown that white grapes are almost all homozygous for the non-functional allele variant.

Due to the fact that grapevine is vegetatively propagated, a lot of different somatic mutations can occur during planting from which only a few affect the phenotype. Because berry color mutations can easily be observed in the vineyard just by visual inspection, a lot of different color mutants have been selected since the rise of viticulture. These mutants can easily be vegetatively propagated by cuttings. The new resulting cultivars might differ not only in the color of the ripe fruits, but also in the coloration of autumn leaf or the prostrate hair of the young shoot. The most mutations found are from white to red berry color, but mutations from black to white can also rarely be found.

This study focuses on the molecular analysis and sequencing of color-related allele variants in berry color mutants of grapevine cultivars found in the German speaking area. Novel *myb*-related gene recombinations at the color locus were found and will be discussed.

Arthropod biodiversity and natural pest suppression in vineyards under innovative management.

Theresa Thiele[1,2], Christoph Hoffmann[1], Martin Entling[2].
[1] Julius Kühn-Institut, Institute for Plant Protection in Fruit Crops and Viticulture, Siebeldingen
[2] University of Koblenz – Landau, Institute for Environmental Sciences, Landau
Email of corresponding author: theresa.thiele@jki.bund.de

Current growing practices in viticulture are lacking sustainability regarding environmental, economic and social aspects.

One promising approach to a more environmentally friendly viticulture is growing fungus resistant cultivars with the novel cultivation method of the minimal pruning of trellis trained grapevines (MPTS). This practice reduces the tremendous amounts of fungicides needed to protect traditional cultivars and is expected to increase biodiversity compared to vineyards with traditional trellis trained grapevines (TS).

In MPTS vineyards, plant architecture is very different from TS trained systems. The higher canopy volume might lead to a changed microclimate and to a higher structural complexity of the grapevine, offering more habitat and more options for overwintering. In vineyards with fungus resistant cultivars, the input of plant protection chemicals is reduced and disturbances from applying these products are decreased. These factors could support a richer biodiversity which is an important ecosystem service to viticulture. A shift in the arthropod community structure could potentially improve natural pest control. The main pests in focus are grape berry moths, pest mites and botrytis vectors such as earwigs and drosophilids.

Arthropods are sampled throughout the vegetation period using a variety of sampling methods to obtain a comprehensive view of their general and functional diversity. To compare predation pressure between MPTS and TS systems, grape berry moth eggs and pupae are exposed in the vegetation and removal rates as well as parasitization will be examined. In addition, grapes are infected with grape berry moth eggs and the resulting damage is compared between traditional and novel pruning systems, and different plant protection intensities.

New plant production systems with autonomous agricultural machinery

Lisa-Marie Urso[1], Till-Fabian Minßen[2], Cord-Christian Gaus[3], Dieter von Hörsten[1], Jens Karl Wegener[1]

[1] Institute for Application Techniques in Plant Protection, Julius Kühn-Institut, Braunschweig
[2] Institute of Mobile Machines and Commercial Vehicles, Technische Universität Braunschweig
[3] Institute of Farm Economics, Thünen-Institute, Braunschweig
Email of corresponding author: lisa-marie.urso@jki.bund.de

Over the last centuries, production systems in arable farming have been adopted constantly to changing demands, like the world´s increasing population, advancements in genetics or breeding and also to steadily improving technological possibilities. This development is driven by the aim to produce food and feed in the most efficient way and to optimize the relation between input and yield.

Processes have been optimized and as a consequence machinery has become more productive which also increased their weight and size in general. Furthermore, improved technologies like assistance and automation systems contributed their part in increasing the productivity. Simultaneously, structural change in agriculture is a main driver for the demand of larger machinery. Nevertheless, modern conditions raise the question of whether today's ways of cultivation are still the ones to be followed in the long term view. The potential of autonomous machinery to change farming processes is shown by highly diverse ideas, approaches and solutions from industry and academia.

The aim of this project is to re-think the ways in which plants are cultivated nowadays and consequently build scenarios on how plant production could be using the chances of autonomous machinery in future.

In order to achieve this aim, today's plant production systems are analysed under consideration of agronomic, economic and technological factors aiming to identify favourable paths for the introduction of autonomous machinery into newly created cultivation systems. Based on the evaluation results, scenarios of production systems will be developed for two kinds of machinery. In a first scenario, the future machines will be about the same size as today's but completely autonomous. The second scenario is about small machines known as autonomous field robots. In a third scenario the combination of both is evaluated.

The final result of the project will not be a single precise scenario on how to include autonomous machines into new plant production systems. Instead, the aim of project is to point up the most favorable path for the utilization of autonomous agricultural machinery by showing the connection between plant production, technology and farm economics. Further needs for research and development for an economic and sustainable new plant production system will be identified.

Genetics of flowering time in grapevine

Anna Werner[1], Iris Ochßner[1], Ludger Hausmann[1], Nadia Kamal[2], Daniela Holtgräwe[2], Thomas Rosleff Sörensen[2], Bernd Weisshaar[2], Reinhard Töpfer[1]

[1] Julius Kühn-Institut, Institute for Grapevine Breeding Geilweilerhof, Siebeldingen

[2] Bielefeld University, Faculty of Biology, Center for Biotechnology

Email of corresponding author: anna.werner@jki.bund.de

Due to climatic change, phenology traits are becoming increasingly important in grapevine breeding, since a premature flowering and ripening time could be observed for grapevine in the last decades. However, knowledge about these traits is still limited as they are genetically very complex and highly influenced by environmental factors. The analysis of the genetic basis of flowering time therefore will enable the development of tightly linked molecular markers useful for marker-assisted selection of especially late flowering breeding lines.

A QTL analysis approach was followed to detect major genetic factors within a biparental mapping population derived from a cross of the early-flowering breeding line GF.GA-47-42 and late-flowering cultivar 'Villard blanc'. Phenotyping of flowering time was carried out for the 150 F1 individuals in seven years. First investigations indicate QTLs on six chromosomes, some of which could also be confirmed in a second unrelated mapping population. Analysis of the QTLs indicates that certain combinations of marker alleles from the different QTL regions lead to a very early, medium or late flowering phenotype, respectively, and suggests a dominant effect for early flowering. However, the revealed QTLs still cover large regions of the respective chromosome which hampers the detection of credible candidate genes.

Future aims of the project are: (1) Fine mapping of the QTL regions by developing new SSR markers and also with genotyping-by-sequencing (GBS) of a subset of F1 individuals. Additionally, an extended mapping population of approximately 900 F1 plants is available for phenotyping, in order to refine the QTL regions, hence limit the number of candidate genes and determine more reliable ones. (2) A deeper investigation of allelic variation at the QTL regions and the impact of their combination on the flowering time phenotype. (3) Flowering time homologues described in model plants and located within a QTL are examined for their expression at various developmental time points in both parents to find differences between the early and late genotype.

The resistance locus *Ren3* and its powdery mildew isolate-specificity

Daniel Zendler, Pierre Schneider, Reinhard Töpfer, Eva Zyprian
Julius Kühn-Institut, Institute for Grapevine Breeding, Siebeldingen
Email of corresponding author: daniel.zendler@jki.bund.de

The obligate biotrophic pathogen *Erisyphe necator* causing powdery mildew on grapevine which belongs to the order of *Ascomycetes* was first introduced to Europe from America around 1845. Since then it has spread and is now present in every winegrowing region worldwide. It is thought to be one of the most devastating fungal diseases known to *Vitis vinifera* L. and is responsible for a considerable annual yield loss if no counteractions such as the application of fungicides are taken.

To this date several natural genetic resistances are known to confer resistance to *E. necator.* Nearly all of them originate from wild Amercican *Vitis* species which have obtained the aforementioned by their co-evolution with the pathogen. These genetic resources are sources for breeding resistant grapevine cultivars with high vine quality and therefore reducing the application of fungicides. The grapevine cultivar 'Regent' is one example of this approach trying to combine resistance with high vine quality. In 2004 Fischer *et al*. detected two resistance loci conferring resistance to downy and powdery mildew in 'Regent' by QTL-analysis in the cross population of 'Regent' x 'Lemberger'. The latter locus was named *Ren3* (resistance to *E. necator*) and was found to be located on chromosome 15.

To this date only few genes have been characterized to be responsible for resistance to *E. necator.* One of the best described resistance gene analogs (RGA) is *MrRun1* which belongs to the family of TIR-NBS-LRR (Toll/Interleukin-Receptor-Nucleotide Binding Site-Leucin Rich Repeats) genes.

So far the resistance locus *Ren3* spans an approximate interval of 4 Mb and covers a total of three clusters of these so-called RGAs. Recently we observed a shift of the maximum QTL over these RGA clusters. Therefore we analyzed plants with recombination in *Ren3* to discriminate the involvement in resistance to *E. necator* of the different RGA clusters.

However, due to a lack of recombination in *Ren3* in the F1 progeny of the 'Regent' x 'Lemberger' cross population, we had to screen other *Ren3* carrying cultivars originated from the repository at the JKI Geilweilerhof for recombination. This approach identified two useful recombinants which delimit the identified RGA clusters.

By controlled infection of detached leafs of the identified *Ren3* recombinant cultivars and the *Ren3* recombinant F1 progeny we were able to detect hypersensitive reactions (HR) associated with appresoria. Two of the RGA clusters seem to be able to trigger HR due to the fact that plants carrying either one of the two clusters showed the same HR response.

These results suggest the involvement of both RGA clusters in the recognition of the same or probably different *E. necator* isolates.

Keynote 2

Beschäftigungsmöglichkeiten für junge Wissenschaftlerinnen und Wissenschaftler im JKI

Kathrin Lehmann
Julius Kühn-Institut, Central administration - human resources department, Quedlinburg
Email of corresponding author: kathrin.lehmann@jki.bund.de

Hand-out

„Berichte aus der Biologischen Bundesanstalt für Land- und Forstwirtschaft“
erscheinen seit 1995 in zwangloser Folge

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