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### Report on the 34th Annual Meeting of the Working Group "Beneficial Arthropods and Entomopathogenic Nematodes"

The 34<sup>th</sup> Annual Meeting of the Working Group "Beneficial Arthropods and Entomopathogenic Nematodes" of DPG and DGaaE was held on 30<sup>th</sup> November till 1<sup>st</sup> of December at. Thanks to Dr. Rainer Meyhöfer and his team of the, Gottfried Wilhelm Leibniz Universität, Institute of Horticultural Production Systems, we could enjoy a very comfortable and well-organized get-together in the historical "Leibnizhaus" and also later in the evening in restaurant nearby in the Old City of Hannover.We were happy to welcome about 30 participants from universities, research institutes, plant protection extension services and biocontrol companies and we would like to thank all contributors.

The next meeting will be organized probably in the year 2017. Please expect our invitation in spring 2017.

Dr. Annette Herz & Prof. Dr. Ralf-Udo Ehlers

## Identification and selection process of potential antagonistic pathogens against the insect pests *Drosophila suzukii* and *Cydia pomonella*

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The spotted wing drosophila (SWD, Drosophila suzukii Matsumura) occurs in Germany as an invasive pest since 2011 and caused urgent research activities to improve biological control systems. D. suzukii 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 2008. Nowadays, it has emerged to one of the most harmful pests on commercially grown fruit plants. It prefers ripe and overripe fruits of stone fruits and nearly all kind of berries in Germany. 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 including microsporidia, bacteria, viruses, and protista) in drosophilids from different geographical origins, isolate putative entomopathogenic microorganisms and reinfect lab populations of *D. suzukii* to investigate the antagonistic potential. We already found microsporidia from related species that are currently evaluated on their pathogenic potential on D. suzukii in larval bioassays. Furthermore, we will integrate the pomaceous fruit pest codling moth (Cydia pomonella), which is an ongoing problem in apple orchards also because the pest develops resistance against commercially available insecticides and biological control agents. The long-term aim of this study is to establish a stable system for pathogen detection that can be used for rapid identification of microorganisms. Moreover, this detection tool provides a miscellaneous application range e.g. as integration in standard diagnostic research for random screening of arthropod breeding systems. Furthermore it can be discussed as an application in risk and safety assessment for beneficials and the detection of pathogenic antagonists in natural pest populations.

#### Trichogramma and Cydalima perspectalis - a still unsolved mystery

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The Box tree pyralid Cydalima perspectalis (Walker, 1859) is an invasive species in Europe, which is not yet regulated by natural enemies. Larval feeding is highly destructive to ornamental and native Buxus plants, due to defoliation. Trichogramma wasps are well known egg parasitoids of many Lepidopteran pests and successful host egg parasitization prevents larval hatching and defoliation. The potential to control C. perspectalis with Trichogramma wasps was investigated in the laboratory. The ability of eight Trichogramma species (T. brassicae Bezdenko, T. bourarachae Pintureau & Babault, T. cacoeciae Marchal, T. cordubensis Vargas & Cabello, T. dendrolimi Matsumura, T. evanescens Westwood, T. nerudai Pintureau & Gerding and T. pintoi Voegele) to accept and parasitize eggs of this new host was determined in direct observation tests and subsequent cage tests on potted Buxus plants. C. perspectalis eggs are attractive to various Trichogramma species, which are able to oviposit, to locate this host on the plant and produce successfully in it but the resulting parasitism was unexpectedly low. Therefore, the female parasitism behavior and the embryonic development in parasitized eggs were surveyed in further observation tests. They were conducted with T. dendrolimi, T. brassicae and T. achaeae females on eggs of C. perspectalis and on another alternative host, the European Corn borer Ostrinia nubilalis. The examination included photo and video documentation of parasitism behavior and parasitoid development inside the host egg. Again, all tested Trichogramma species showed high host encounter and active female rates but only T. dendrolimi was successful in parasitizing the eggs of C. perspectalis. Results were comparable to the T. brassicae - O. nubilalis system but T. dendrolimi spent more time for the examination and parasitization of C. perspectalis eggs. The photo documentation brought no new information to what is happening in the egg after the oviposition process. There is still the unsolved mystery, what leads to the ineffective parasitization although C. perspectalis eggs are attractive to various Trichogramma species and what enables T. dendrolimi for superiority to the other species?

## Use and establishment of predatory mites for sustainable control of two-spotted spider mite (*Tetranychus urticae*) in hop

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The aim of the project (term: May 2013-April 2016), funded by the German Federal Ministry for Food and Agriculture (2812NA014), was to establish predatory mites, especially Typhlodromus pyri, in hop yards during several growing seasons by ground cover management. For this purpose tall fescue Festuca arundinaceae, gallant soldier Galinsoga parviflora and stinging nettle Urtica dioica were initially sown between the rows in randomly positioned plots. As it became evident during the first project year, that neither gallant soldier nor stinging nettles were practicable for ground cover management, a mix of grasses including field meadow foxtail Alopecurus pratensis and annual meadow grass Poa annua was established instead. In addition, some plots were equipped with strawberries Fragaria x ananassa (cv. Asia). A second part of the study focused on the determination of practicable release methods and the efficiency of different species of predatory mites. The trials were conducted in five different hop yards with following predatory mites: (a) overwintering T. pyri on felt strips, (b) overwintering T. pyri on grape vine cuttings from a vineyard in Rhineland-Palatinate, (c) Amblyseius andersoni in vermiculite and (d) a mix of allochthonous Phytoseiulus persimilis and Neoseiulus californicus on bean leaves. As T. pyri for both variants were delivered already in February, they had to be stored in a cooling chamber by at 5-8°C till an application was possible in May. In the year 2015, a hot dry summer offered ideal conditions for a strong population growth of spider mites. The best results of spider mite control were achieved with T. pyri applied on grape vine cuttings. At the end of May, the cuttings were distributed on every third crown, thus releasing approximately 30,000 predatory mites per ha before a noteworthy spider mite infestation. During the season, in this hop yard spider mite numbers did not increase; at harvest with 7 mites per leaf the numbers of the T. pyri plots were below the damage threshold and significantly lower than in the untreated control (20 mites per leaf). On the other hand, in another hop yard with T. pyri released on felt strips spider mites increased more than 500 per leaf, causing a loss of yield of 50 % and visible damage on all cones. The short project duration however does not allow yet a statement about the success of a long-term establishment of predatory mites in hops with the aid of different ground cover management.

#### Microbial antagonists of various Otiorhynchus species in Germany

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As part of the project on soil snout beetles, numerous harmful or potentially harmful Otiorhynchus species were bred for different purposes in breeding boxes. Occasionally, it came to losses because the beetles had reached their natural age, were attacked by pathogens or rarely died early due to stressors. These beetles were examined for their infection by microorganisms. 178 dead beetles of nine Otiorhynchus species were investigated. Entomopathogenic organisms were found in 48 specimens (27%); in five beetles each two pathogens and in 43 beetles each one pathogen were diagnosed. Most commonly, the Ascomycetes Lecanicillium sp. (17 beetles, 9.5% and 35.4% respectively), Beauveria bassiana (15 beetles; or 8.4% and 31.3% respectively) and Gliocladium sp. (12 beetles; 6.7% and 25% respectively) occurred. In two specimens undetermined microsporidia, nematodes and *Mattesia* sp. were found and once each *Arthrobotrys* sp., Isaria sp., and one species of the Entomorphthorales. B. bassiana and Lecanicillium sp. are known insect pathogenic fungi, *Gliocladium* sp. is primarily also insect pathogenic, but in this genus, there are also species that are used against soil-borne phytopathogens (e.g. G. catenulatum). Among the rare species Isaria sp. (syn. Paecilomyces) was found. Mattesia is a parasitic protozoan that infects the fat body of various insect orders. Arthrobotrys sp. is a nematophagous predatory fungus. Among the remaining organisms were mainly unspecified bacteria (17.4%) and various fungi. Among these, especially the saprophytic fungi Aspergillus and Penicillium (together at 10.6% of beetles) and as a single evidence also a weakness parasite (*Mucor*), phytopathogenic fungi (Fusarium, Diplosporium Cylindrocarpon) and nematodes. The role of entomopathogenic microorganisms in the field. and how their pathogenicity is assessed, is not known. After a clarification of their identity, it would also be obvious to carry out investigations on the pathogenicity of the isolates. Data on identified pathogens of this study have already been included into the database of the Institute for Biological Control (JKI, Darmstadt) about arthropod diseases:

http://arthropodenkrankheiten.julius-kuehn.de

#### First insights into heat shock responses of Harmonia axyridis PALLAS

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Invasive coccinellid Harmonia axyridis PALLAS has rapidly spread over Germany during the last 15 years. Harmonia axyridis is assumed to have a high potential to compete to or even eliminate native species such as Coccinella septempunctata L.. Many scientists all over the world try to quantify their potential under the local environmental conditions in their new conquered distribution areas. Own comparative investigations in climate chambers showed different responses of multivoltine H. axyridis and univoltine C. septempunctata to 3 K and 6 K elevated temperatures. For both species increased feeding rates could be measured but probably due to their different ecological strategies food conversion efficiencies were higher for C. septempunctata. These results lead to the question how these species respond to environmental stress, in particular heat stress periods. Therefore a set of biochemical and ecological studies was designed. Proteomics should be established to investigate potential responses to heat stress in the coccinellid proteome. Based on these findings targeted ecological studies shall prove the results within ecological and behavioral studies in order to quantify potential changes in live table processes and ecological parameters. Meanwhile 2Dgelelectrophoreses could be established for H. axyridis. A first study on heat stress responses in H. axyridis resulted in significant changes of altogether 37 proteins. Nine of them were up or down regulated for at least 50%. The identification of these proteins via mass spectroscopy and homology modeling with Tribolium castaneum HERBST could be realized for 6 protein spots. Currently additional ecological investigations are conducted. Further studies will compare the responses of the two coccinellids and prove the influence of for instance acclimatization, length of stress induction and developmental stage where heat stress is induced.

#### Evaluation of banker plant systems against the cabbage whitefly Aleyrodes proletella

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The cabbage whitefly Aleyrodes proletella has become a major pest on several cabbage crops in recent decades. Conventional control methods based on insecticides has shown to be insufficient mainly due to the whiteflies' cryptic behaviour on the underside of leaves and insecticide resistances. Although offering promising alternative or supplementary tools, conservational and augmentative biological control methods in field crops have often been neglected. For instance, the release and promotion of natural enemies with the help of banker plants is one way to increase biological control services not only in greenhouses, but also in the open field. This field study evaluated the potential of two banker plant systems against A. proletella: (1) the greenhouse whitefly Trialeurodes vaporariorum on pumpkin Cucurbita maxima 'Uchiki Kuri' (TVP) and (2) the honeysuckle whitefly Aleyrodes Ionicerae on European columbine Aquilegia vulgaris (ALC) both inoculated with the whitefly parasitoid Encarsia tricolor. We determined the potential of both systems in terms of parasitoid production and their effects on parasitism rates and whitefly infestation on Brussels sprouts Brassica oleracea var. gemmifera in two different distances from the banker plants (1m and 3.5m). Over the entire experimental period TVP produced 3.1 times more parasitized nymphs than ALC (1,715 per m<sup>2</sup> and 558 per m<sup>2</sup>, respectively). The TVP system enhanced A. proletella parasitism on cabbage in 1m and 3.5m distance, whereas ALC only in 1m distance. Average parasitism rates on cabbage plants increased by 53% and 51% in TVP and ALC, respectively, compared to the control treatment without banker plants. Furthermore, the number of parasitized whitefly nymphs on banker plants correlated with the parasitism rate on cabbage 14d later (TVP: r= 0.69 and ALC: r= 0.44) underlining the direct impact of *E. tricolor* from banker plants. However, neither of the banker plant systems decreased A. proletella populations significantly. In conclusion, especially the TVP system showed promising potential as banker plant system against A. proletella. It produced not only high amounts of parasitoids continuously, but also marketable pumpkins with a yield (32 t/ha) comparable with not infested pumpkin plants. Nevertheless, further improvements are still needed to increase the impact on A. proletella population. These may include pumpkin plants initially inoculated with a higher number of *E. tricolor*, a combined inoculation with *E. tricolor* and a compatible whitefly predator like the ladybeetle *Clitostethus arcuatus* or a combination of TVP with flowering plants to further promote natural enemies like hoverflies and ladybeetles.

# Ecoorchard: Innovative design and management to boost functional biodiversity of organic orchards

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Researchers of nine EU member states are involved in the project ECOORCHARD which is funded by the Core Organic Plus programm for a period of three years (2015-2017, http://coreorganicplus.org/research-projects/ecoorchard/).The aim is to develop approriate strategies and actions to promote functional agro-biodiversity in organic pome fruit production, with an emphasis on apple cultivation. One common action is the performance of a field trial aiming on the establishment of flowering strips within the orchard to improve conservation biocological control of the main apple pests like Rosy apple aphid (Dysaphis plantaginea) and Codling moth (Cydia pomonella). In Germany, the entomological field trial is conducted at a fruit orchard (Obstplantage Latz) in Saarwellingen. In this orchard organic, pome fruit has been produced for more than 15 years on about 17 ha. In spring 2015, flowerstrips were sown into the inter-rows at two different varieties (Fuji and Braeburn). The seed mixture contained about 30 perennial herbaceous plant and grass species, that can be mulched occasionally. Natural enemies like Syrphidae, Coccinelidae and parasitoids of codling moth are supposed to be promoted with these additional floral ressources. To optimise the seed mixture it is necessary to study the effect of these flowering plants on the main pests and potential beneficials. To monitor the pest pressure and the state of biodiversity, different methods are being used: visual control, beating sampling, corrugated cardboard bands, sentinel prey and fruit damage assessment. The requirement and availability of food resources, like nectar and pollen has to be investigated. Therefore, further experiments on feeding ecology and nutrition physiology of beneficials and pest species need to be carried out. Besides faunal and floristic studies on the suitability of sown wildflower-strips in the inter rows, an adjusted habitat- and cultivation management will be developed. Furthermore, practical methods and indicators will be worked out to monitor functional biodiversity on the farm level. In addition to all these various studies a web-based thematic portal (EBIO-Network = European Biodiversity Orchard Network) has been created, where the achieved results and recommendations for practical use are continously documented.

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## Biological control as an ecosystem service in integrated and ecological pome fruit production – the project "Demoapfel"

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The project evaluates the role of natural antagonists of the codling moth (Cydia pomonella) in integrated and organic pome fruit production regarding their pest controlling function. The aims on the development of strategies for a reasonable and sustainable research conservation, promotion and use of these elements of the agricultural system. For this purpose, field samples are taken in three main apple growing regions in Germany which are located in the North (Altes Land, an area near Hamburg), Center ("Kraichgau" near Karlsruhe) and the Lake Constance region several times per year. In all regions, samples were taken in orchards with integrated or organic management and also in cider orchards without any plant protection. Different sampling methods were applied (e.g. sweeping net, beating sampling, collecting attacked apples, installing and again collecting corrugated cardboard, window traps, soil samples). The samples were further processed in the laboratory to detect natural enemies and pathogens of all developmental stages of codling moth. The objective of this sampling is to determine species diversity and abundance of selected natural enemies in the different apple growing regions, especially the parasitic hymenoptera. Specimen collected by sweep netting and beating are currently processed for further identification to genus or species level. A part of the samples was incubated under outdoor conditions in order to monitor the natural emergence of parasitoid species in relation to the codling moth. Furthermore, rearing cultures of selected parasitoid species will be established to investigate their biology and their interactions. Potential side-effects of plant protection products used in integrated and organic apple production will also be tested on these organisms. These investigations will allow to inform the fruit grower how to apply compatible plant protection measures more precisely in order to preserve the natural enemies and their ecosystem service.

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#### Promotion of natural enemies by tailored flower strips in cabbage

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In German agriculture, flower strips are established as a part of landscape conservation and are subsidized by states. Their primary functions are promotion and conservation of biodiversity, wildlife and pollinators. Also the function for plant protection is of interest. Studies showed positive effects on biological pest management in agricultural crops. But in horticultural crops these flower strip types may be also highly attractive to pests (Lepidoptera) and therefore need to be adapted. In this project a tailored flower strip was developed with the aim to promote natural enemies of common cabbage pests (esp. Aleyrodidae, Aphididae, Pieridae) but not the cabbage pests. Other demands were high flower density, long flowering period and high competitiveness to weeds. The adapted flower strip consists of 11 annual flowering plant species which were carefully selected. The flower strip "Hannover Mix" was compared with the conventional "Tübinger Mix" in their efficiency to promote natural enemies. In 2015 both types of flower strips were sown along Brussels sprouts  $(3q/m^2, 2.2 \times 6m, n=9)$ . Insect visits and flowers density were determined in the flower strip as well as densities of pests and natural enemies in Brussels sprouts. The results show the potential of tailored flower strips as a strategic part of biological pest management. "Hannover Mix" outperformed the conventional flower strip in attractiveness to natural enemies while being less attractive to butterflies. Numbers of pest insects in Brussels sprouts were significant lower along the tailored flower strip. Therefore, higher abundances of natural enemies can be achieved by careful selection of attractive plant species. Optimization of the composition, validation by large-scaled field trials in cooperation with local farmers and investigations on the effective range in field will follow.

# Ecological relationship between hoverflies (Diptera: Syrphidae) and *Salvia bogotensis* in an urban environment

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Urban agriculture is an important topic for food security program. The populations of biological control agents and pollinators in urban environments are low. Hence, it is important to identify the richness of these beneficials and study plants that favor their fitness. Salvia bogotensis (Lamiales: Lamiaceae) is an ornamental plant found in both urban and natural areas of the Cordillera Oriental in Colombia and their flowers are frequently visited by hoverflies. These insects can be pollinators, natural enemies of aphids and saprophagous organisms among others. The main goal of this study was to describe the ecological relationship between hoverflies and S. bogotensis in order to know if this plant offers benefits to these insects. To get this objective, first, we recognized the high number of hoverflies species attracted to *S. bogotensis* and then the following aspects were studied: morphology, syndrome and type of pollination of S. bogotensis, as well as hoverflies' behavior, pollen load and accessibility to the S. bogotensis nectar and pollen. Salvia bogotensis attracted adults of nine species belonging to the family Syrphidae. Eight species of the subfamily Syrphinae (Allograpta neotroica, Allograpta exotica, Allograpta anulipes, Allograpta aenea, Platycheirus ecuadoriensis, Platycheirus fenestrata, Toxomerus s.p. and Syrphus shoree) and one species of the subfamily Eristalinae (Lejops sp.) could be identified. The pollination syndrome of S. bogotensis is entomophily. This plant has morphological features that favour crosspollination such as: staminal levers, nectar guides, purple colour that attracts insects, hercogamy, anthers covered by the corolla, and the upper part of the corolla with numerous hairs where pollen grains that insects bring from other plants are attached. Hoverflies were not able to take nectar from S. bogotensis flowers but their pollen could be identified in the hoverflies' guts in high quantity. Additionally, these insects are neither pollinators nor do they cause plant damage thus suggesting a commensalism relationship with the plant. These species of hoverflies has low pollen load and few hairs which mean they cannot be pollinators in plants with cross-pollination but they could optimize the self-pollination in plants with autogamy. In conclusion, S. bogotensis attracts and provides pollen to hoverflies which could play a relevant role in crops sowed in the city as pollinators or biological control agents.

## The role of flowering plants in improving the fitness and efficiency of parasitoids used as biological control agents of cabbage whiteflies and aphids

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In agroecosystems, the lack of food resource is a factor that affects the fitness and efficiency of parasitoids. Parasitoids are omnivorous, therefore if hosts are absent they have to rely on alternative food sources, i.e. nectar of flowers. Hence, the aim of this research was to explore the effect of flowering plants on the attraction, longevity, fecundity and efficiency of the microhymenoptera Diaeretiella rapae M'Intosh (Hymenoptera: Aphidiidae) and Encarsia tricolor Förster (Hymenoptera: Aphelinidae). In our findings, both adult parasitoids had access to the floral nectar of alyssum and buckwheat but not to the faba beans. In the attractiveness experiment, it was observed that i.) alyssum had higher attractiveness for *D. rapae* compared to all other flowering plants, ii.) D. rapae parasitoids showed a high preference for the HPC (Host Plant Complex) compared to flowering plants, iii.) alyssum had higher attractiveness for E. tricolor compared to buckwheat, iv.) E. tricolor showed similar response for HPC and flowering plants. The climate chamber experiments showed that fitness of both parasitoid species was increased substantially: i.) the longevity of D. rapae females was 166 % longer in buckwheat treatment as compared to alyssum and of E. tricolor 38 %, ii.) the longevity of *D. rapae* females was 433 % longer in the buckwheat treatment as compared to host-honeydew and of E. tricolor 78 %, iii.) the number of D. rapae mummies was 334 % larger in the flowering plants treatment (mix of alyssum and buckwheat) as compared to the control, iv.) number of mummies produced by E. tricolor was similar in all treatments. Finally, field experiments showed that the selected flowering plants increased the parasitism rate by 112 % for D rapae and by 46 % for E. tricolor. In conclusion, flowering plants are able to attract parasitoids and play an important role in optimizing the fitness of parasitoids. In D. rapae females, flowering plants increased survival, fecundity and parasitism rate. In E. tricolor females, flowering plants enhanced the survival and parasitism rate. It means that flowering plants could contribute in the cabbage integrated pest management to reduce the use of agrochemicals.