Contributed paper. Tuesday, 11:45. **104**

A role of polychaetes in transmission of white spot syndrome virus in shrimp ponds?

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White spot disease (WSD) is caused by white spot syndrome virus (WSSV) (*Nimaviridae*). WSSV emerged in the early-to-mid 1990s in Southeast Asia and became panzoonotic since. The disease can be mitigated by introducing rigorous sanitation protocols, proper pond management, use of specific pathogen-free shrimp and by early diagnosis followed by eradication. The virus is transmitted horizontally by healthy individuals predating on diseased ones, via feeding on detritus or by intake of WSSV-contaminated water. WSSV can also be transmitted vertically via broodstock. The virus infects a wide range of crustaceans beyond the penaeids such as crabs and crayfish, and these co-inhabitants of ponds form a reservoir of WSSV for disease transmission to penaeids. Much less knowledge is there on the potential of resident benthic organisms as vectors for WSSV. A literature survey indicates that WSSV is present in a number of non-crustacean invertebrates, which sometimes vector the disease to penaeid shrimp. *Dendronereis* spp. is a most ubiquitous resident annelid in shrimp ponds and used as food source for shrimp. We showed that WSSV replicates in *Dendronereis* spp. and can be transmitted from this polychaete to penaeid shrimp. Furthermore there appears to be a positive correlation between the past incidence of WSD in ponds and the occurrence of WSSV in resident *Dendronereis* spp, whereas there is no correlation with other pond parameters. We hypothesize that *Dendronereis* spp., as a replicative host for WSSV, may serve as a reservoir for WSSV and may be associated with the persistence of this virus in pond systems.

Contributed paper. Tuesday, 12:00. **105**

Novel Pattern Recognition Receptor Protects Shrimp from *Vibrio* Infection by Binding Flagellin and LPS through Different Recognition Modules

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Pattern recognition receptors (PRRs) recognize pathogens thorough the pattern recognition modules. For example, Toll like receptors recognize the ligands through leucine-rich repeats (LRRs), and C-type lectins bind to glycans on the surface of pathogens by the C-type carbohydrate recognition domain (CRD, also called C-type lectin like domain CTLD). Many PRRs contain more than one kind of modules. In the present study, we identified a novel PRR, named *Leulectin*, which contains several LRRs and a CTLD. Such unique arrangement has not been found in any other organisms. Recombinant Leulectin and the modules (LRRs and CTLD) were found to protect shrimp from *Vibrio* infection. An ELISA-based screen was performed to identify the potential ligands the two modules may recognize. Results showed that LRRs could recognize the *Vibrio* flagellins, and CTLD could recognize lipopolysacchraides (LPS). The Leulectin-flagellin interaction was determined by the third LRR of Leulectin and the N-terminus of flagellin, and the Leulectin-LPS interaction was dependent on the long loop region of CTLD in a calcium-independent manner. The ligand-recognizing activity of LRRs and CTLD was critical for Leulectin to bind to bacteria, and the binding was the basis for Leulectin to protect shrimp from bacterial infection. This study clearly showed the interesting synergy between distinct modules of a PRR.

Contributed paper. Tuesday, 12:15. **106**

Observations on *Agmasoma penaei* and *Perezia nelsoni* in White shrimp *Litopenaeus setiferus* from the Gulf of Mexico

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In June 2012 a few shrimp from Plaquemines parish LA with the symptoms of microsporidiosis were delivered to the Louisiana Aquatic Diagnostic Laboratory for identification. Light microscopy including examination of Luna-stained paraffin sections, and electron microscopy showed the infection was limited to ovaries and was caused by a microsporidium producing roundish pansporoblasts with 8 spores (3.6 x 2.1µm) and anisofilar (2+6) polar filaments, the features corresponding to the diagnosis of *Agmasoma penaei* (=*Thelohania penaei* Sprague 1950, n.comb Hazrad and Oldacre, 1973). Comparison of the SSUrDNA sequence of the novel isolate to *A.penaei* from Thailand revealed 95% similarity, which suggests these geographical isolates, may be two different species, a conclusion supported by several ultrastructural dissimilarities and different tissue tropism. Phylogenetic analyses places this species as a divergent taxa within the clade IV (microsporidia of terrestrial origin) sensu Vosbrinck, Debruner-Vossbrinck, 2005. In two shrimps infection of ovaries with *A. penaei* was accompanied by heavy infestation of muscles with another microsporidium *Perezia nelsoni. P.nelsoni* produces individual spores (2.0 x 1.1µm). Structurally and genetically (SSUrDNA sequence similarity >99%) LA isolate was identical to *Perezia nelsoni* from the Mississipi coast of the Gulf (Canning et al., 2002). Previously reported infection of muscles with *A.penaei* may be due to overlooked double infection with *P.nelsoni.* Supported by Louisiana Department of Wildlife and Fisheries.

CONTRIBUTED PAPERS Tuesday, 10:30-12:30

FUNGI 3

Contributed paper. Tuesday, 10:30. **107**

Comparison of ecological traits of co-existing *Metarhizium*: What does it take to dominate an agricultural field?

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It is expected that particular adaptive ecological traits influence species abundance and distribution within an ecosystem. We evaluated selected traits of different co-existing species and genotypes of the entomopathogenic fungi *Metarhizium* isolated from an agroecosystem in Denmark. Fifteen fungal isolates representing 11 genotypes were tested for: UVB tolerance, *in vitro* growth at 12.5°C and 21.5°C, mycelial growth from the insect cadaver into the surrounding soil, virulence and conidia production on cadavers. The results showed that the relative performance of the most abundant *Metarhizium* genotype was intermediate for mycelial growth in soil and in vitro growth at 12.5°C / 21.5°C while it showed high UVB tolerance and conidia production compared to other genotypes. We discuss whether the two latter traits are most important to dominate the *Metarhizium* community in agricultural habitat or whether the "Jack of all trades" performance could be the key to understand the dominance of a particular genotype.

Contributed paper. Tuesday, 10:45. **108-STU**

Effect of entomopathogenic fungal strains on non-target arthropods in sour cherry orchard

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Efficacy of *Metarhizium* and *Beauveria* entomopathogenic fungal strains for the control of cockchafer grubs was evaluated in sour cherry orchards. Safety like possible effect of the inoculum on natural soil microbiota as well as efficacy and fate of these fungi need to be investigated. The applied fungal strains have wide host range, thus we have to determine the risks of their use during repeated long-term applications. Different inoculation methods were compared and the persistence of inoculum was monitored in the soil and on target and non-target organisms. The treatments were applied 2 times (May and July) in the space rows and we used pitfall traps as sampling method. Samples were collected 8 times during the summer of 2013. The samples were processed in laboratory and the numbers of different arthropods (collembolans, mites, thrips, flies, ants, spiders, centipedes, crickets, rove beetles, ground beetles) were recorded in each sample. The comparison of un-treated and treated areas, and the microscopical examination showed no significant differences in the frequency of species. As a conclusion, the effect of these entomopathogens on non-target arthropods is minimal and as such they do not impose any environmental risk.

Contributed paper. Tuesday, 11:00. **109-STU**

Potential of endophytic *Beauveria bassiana* in grapevine against insects

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Fungal entomopathogens are important antagonists of arthropod pests and have attracted increased attention as biocontrol agents in integrated pest management programs. In addition, evidence has accumulated that some entomopathogenic fungi like *Beauveria bassiana* (Bals.) Vuill. can endophytically colonize plants and provide a systemic protection against insect pests. Currently, it is unknown whether *B. bassiana* can exist as an endophyte in grapevine, *Vitis vinifera* (L.) and still maintains its antagonistic potential against insect pests. In the present study, the antagonistic activity of *B. bassiana* (strain ATCC 74040) after plant inoculation and endophytic establishment in grapevine against the vine mealybug *Planococcus ficus* was assessed using surface sterilized leaves for a bioassay. Possible effects of endophytic *B. bassiana* on the feeding preference of black vine weevil *Otiorhynchus sulcatus* choosing between control and inoculated plants was examined through choice assays. A significant effect of endophytic *B. bassiana* on growth during the whole observation period and on mortality of mealybugs one week after initial settlement was evident. Adult *O. sulcatus* chose significantly more often the control plants as a host plant compared to grapevine plants with endophytic *B. bassiana.* In addition, a microarray analysis was performed to get insights into genetic mechanisms behind the plant-fungus-interaction. The results indicate an up-regulation of diverse defense related genes in grapevine due to the endophytic establishment of *B. bassiana*. In conclusion, endophytic establishment of the entomopathogenic fungus *B. bassiana* in grapevine might represent an alternative and sustainable plant protection strategy, with the potential for reducing pesticide applications in viticulture.

Contributed paper. Tuesday, 11:30. **111**

Horizontal transmission of entomopathogenic fungi by ectoparasitoid *Habrobracon hebetor*

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Horizontal transmission of entomopathogens by parasitoids is well known for viruses but did not registered for fungi. Our experiments were carried out on the laboratory system *Galleria mellonella* (Lepidoptera, Piraliidae), – *Habrobracon hebetor* (Hymenopera, Braconidae) – *Beauveria bassiana* (Hypocreales, Cordycipitaceae). We found out that contamination of *H. hebetor* ovipositor with low titers of conidia *B. bassiana* and following envenomation of *G. mellonella* larvae led to mycoses followed by host colonization and conidiaformation. In addition *H. hebetor* females transmitted fungal conidia from infected (6 hours post inoculation with conidia) to native *G. mellonella* larvae, and this transmission led to successful mycosis of native host larvae. The decreasing of cellular and humoral immune reactions, significant increasing of adhesion and germination of fungus on cuticle of envenomated larvae were registered. As a result susceptibility of envenomated *G. mellonella* larvae to fungal infection was increased in thousands times compared with native control. Thus the paralyzation and strong inhibition of immune reactions of larvae by venom of *H. hebetor* allows to minimize quantity of transmitting with parasitoid fungal inoculum. We assumed that «paralyzing» parasitoids can take part in transmission of entomopathogenic fungi particularly in out-of-the-way places (shelters) as well as disperse of fungal infection under low density of hosts.

Contributed paper. Tuesday, 11:45. **112**

Fast spread of the parasitic *Laboulbenia formicarum* in a supercolony of the invasive garden ant *Lasius neglectus*

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Many ant species are highly successful invaders and can dominate vast areas by forming dense networks of connected nests in contrast to the smaller and discrete, spatially dispersed colonies of most social insects. However, it was recently proposed that such supercolonies are more vulnerable to infection by parasites and diseases as they would serve as large targets with high rates of transmission from nest to nest. Here we studied the invasive garden ant *Lasius neglectus*, a new pest species which is currently spreading throughout Europe where several populations are infected with the ectoparasitic fungus *Laboulbenia formicarum*. In one population (supercolony) we followed the prevalence and intensity of the infection over 10 years, revealing an epizootic spread of the ectoparasite with the mean annual prevalence increasing from 0.126 to 0.997. Distinct body parts of the ants had markedly different infection intensities, and at low intensities antennae and thorax were free from signs of infection. There were no seasonal differences in infection intensity and no other *Lasius* species in the area was found to be infected. These results give the first direct support to the hypothesis that supercolonies of invasive ants potentially face a significantly higher threat from parasites and diseases compared to ants with normal colonies, implying interesting perspectives for biological control of these pest species.

Contributed paper. Tuesday, 12:00. **113**

The dietary preference of a beneficial predator in apple orchards reveals an undocumented spore dispersal mechanism for entomopathogenic fungi.

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In the course of a floristic and ecological study of the entomopathogenic fungi found in apple orchards and strawberry fields (part of the IMBICONT biological control project), we observed resting spores in the frass of the earwig *Forficula auricularia*, a beneficial predator in apple orchards. The presence of resting spores in earwig frass suggests that in addition to being a beneficial predator, earwigs may play a role in the dispersal of Entomophthoromycota—a spore dispersal mechanism not previously documented for this group of fungi. In the lab, we observed that earwigs avidly consumed entomophthoromycotan-infected insects even while the fungus was actively ejecting conidia. We hypothesize that this fungus-insect meal might confer a nutritional benefit but that earwigs avoid foraging on insects infected by generalist entomo-pathogenic fungi (e.g. *Metarhizium*, *Beauveria*) because these generalist entomopathogens pose a risk that would potentially outweigh any nutritional benefit. We present the preliminary results from a series of choice-experiments to test these hypotheses.

Contributed paper. Tuesday, 12:15. **114**

Effects of entomopathogenic fungi on the "*Trialeurodes vaporariorum – Encarsia formosa*" system: preliminary results

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The effects of a selected group of six entomopathogenic fungal isolates (including the mycoinsecticide Naturalis and the *Beauveria bassiana* ATCC74040 strain contained into the commercial product) on the system "*T.vaporariorum* - *E.formosa*" were evaluated, considering the direct effect on the parasitoid development but also on the *E. formosa* activity and behaviour. The effect of fungal treatments on the parasitoid development was evaluated submitting infested tomato plants to the fungal treatments at different times from the parasitization and recording the parasitization rate and the parasitoids emergence. Then, the effect of fungal isolates on *E. formosa* behaviour and activity was examined in "free-multichoice" and "no-choice condition". Finally, the role of *E. formosa* in trasmitting the mycoses from infected to uninfected host population was estimated. Results showed that fungal treatments can affect the *E. formosa* development, particularly when applied before the parasitoids introduction and using the mycoinsecticide Naturalis. *E. formosa* showed no differential tropism in "free - multichoice" conditions and it was not able to locate and select the uninfected hosts "at distance" but it was able to detect and avoid infected hosts by direct exploration. Furtheremore, *E. formosa* was able in vectoring the fungal propagules from contaminated to uncontaminated hosts trough its activity. Results of these laboratory experiments provided important information about the possibility to integrate the entomopathogenic fungal treatments and the *Encarsia formosa* releases and clarified some biological and behavioural aspects of the "host–pathogen–parasitoid" system.

WEDNESDAY - 6 August

SYMPOSIUM 5 (Microbial Control) Wednesday, 8:00–10:00

Developments/Issues in the Regulation of Microbial Products: Harmonization across Jurisdictions

Symposium. Wednesday, 8:00 **115**

The authorisation and regulation of microbial biopesticides: why bother?

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The use of microbial biopesticides and other minimal-risk products is starting to become more widespread as a result of new government legislation that aims to reduce the excessive use of conventional chemical pesticides and increase the use of "alternative" control methods. In the European Union, a paradigm shift in pesticide policy has occurred recently with the enactment of the Sustainable Use Directive on pesticides. This legislation makes IPM mandatory for farmers and growers and gives specific emphasis to biologically based controls including microbial biopesticides. There has been significant recent activity in the biopesticides industrial sector, with multinational agchem / agri-business companies buying up biopesticide