European PhD Network **"Insect Science"** XIII Annual Meeting

16-18 November 2022

SCIENTIFIC PROGRAM
&
BOOK OF ABSTRACTS

CREA
Via di Lanciola, 12,

50125 Comune di Impruneta (FI)

PROGRAMME

Wednesday 16 November 2022

14:00 **Registration**

14:25 **Welcome address**

14:30 - 15:00 **Senior scientist lecture** Roberto Romani – University of Perugia
Functional morphology of sensory structures in spittlebugs

15:00 - 16:30 **Oral Presentations** (regular & short talks)

Chair: ROBERT CALVERT

15:00 - 15:15 R Andrea Arpellino – University of Turin
Investigations on the vine-feeding species complex: insect-microorganism interactions and their implications for vineyard management

15:15 - 15:30 R Daniele Bruno - University of Insubria
Insect-mediated bioconversion: from organic waste to biobased materials. Conversion efficiency and protein valorization

15:30 - 15:45 R Marco Bonelli - University of Milan
Insect-mediated bioconversion: from organic waste to biobased materials. Digestion capability and lipid valorization

15:45 - 15:50 S Aurora Bozzini - University of Padua
Development of innovative methods for the early-detection of the European spruce bark beetle outbreaks

15:50 - 15:55 S Mathieu Bussy - IRBI CNRS / Université de Tours
Role of thermal heterogeneity in driving co-existence of competing species

15:55 - 16:10 R-O Maja Fluch - Free University of Bozen-Bolzano
Reconstruction of the feeding behaviour of the brown marmorated stink bug *Halyomorpha halys* via molecular gut analysis

16:10 - 16:25 R Andree Cappellari - University of Padua
Effects of seasonality and landscape composition on pollen collected by honeybees

16:25 - 16:30 S Sara Caramella - University of Insubria
Characterization of RNASET2 in *Hermetia illucens*

16:30 - 16:35 S Ivana Carofano - University of Padua
Symbiotic control of harmful insects as an innovative sustainable method: the case study of *Bactrocera oleae* (Diptera, Tephritidae)

16:35 - 17:05 **Coffee Break**

17:05 - 17:45 **Oral Presentations** (regular & short talks)

Chair: ELIA RUSSO

17:05 - 17:20 R Elena Chierici - University of Perugia
Effects of the rearing host on *Trissolcus japonicus* olfactory responses

17:20 - 17:25 S Corentin Clavé - University of Naples "Federico II"
Plant-mediated effects of the entomopathogenic fungus *Beauveria bassiana* on *Spodoptera littoralis*

17:25 - 17:30 S Giovanna De Leva - University of Naples "Federico II"
Impact of Deformed Wing Virus infection on honey bee gut microbiota

Thursday 17 November 2022

09:30 - 10:00 **Senior scientist lecture** Myron Zalucki – University of Queensland
Eruditione luce canticum

10:00 - 11:05 **Oral Presentations** (regular & short talks)

Chair: MARCO BONELLI

10:00 - 10:15 R Sara D'Arco – University of Modena and Reggio Emilia
Evaluation of imidacloprid detection and behavioral resistance of *Musca domestica* L. using proboscis extension response to sucrose containing imidacloprid

10:15 - 10:20 S Maria Giovanna De Luca – University of Naples "Federico II"
Beneficial fungi in the genus *Trichoderma* are effective control agents of noctuid moth larvae

10:20 - 10:35 R Luca Deganutti – University of Padua
Effect of biological control agents at field conditions to control the spruce bark beetle *Ips typographus*

10:35 - 10:50 R Matteo Dho – University of Turin
Advancements in symbiotic control of stink bug pests of hazelnut from the local to the global scale

10:50 -11:05 R Nicolò Di Sora – University of Tuscia
Biological control of *Toumeyella parvicornis* with *Exochomus quadripustulatus* and *Cryptolaemus montrouzieri*

11:05 - 11:30 **Coffee Break**

11:30 - 13:00 **Oral Presentations** (regular & short talks)

Chair: ANDREE CAPPELLARI

11:30 - 11:35 S Juan Sebastian Enciso – Free University of Bolzano-Bozen
Microbiome composition and distribution of major endosymbionts in *Scaphoideus titanus* (Hemiptera: Deltocephalinae)

11:35 – 11:50 R Jay Darryl Ermio – University of Palermo
Nectar-dwelling filamentous fungi affect nectar attraction and survival of the stink bug egg parasitoids *Trissolcus basalis* and *Ooencyrtus telenomicida*

11:50 - 11:55 S Elena Eustacchio – University of Milan
Plant-insect interactions in mountain areas: insights for an analysis of pollen loads comparing light microscopy and ITS2 metabarcoding

11:55 - 12:10 R Robert Calvert – Newcastle University
The development of novel biopesticides derived from the venom of insect antagonists

12:10 - 12:25 R Elena Gazzea – University of Padua
Recovery of plant and pollinator communities after severe wind disturbance in Alpine forests

12:25 - 12:40 R Francesco Lami – University of Bologna
The potential of ground-dwelling arthropods as biodiversity indicators in maize agroecosystems of Northern Italy

12:40 - 12:55 R Serena Malabusini – University of Milan
Studies on the bioethology of *Sclerodermus brevicornis* to improve biological control strategies

12:55 - 13:00 S Frédéric Manas – Institut de recherche sur la biologie de l'insecte - Université de Tours
Reproduction of the black soldier fly, an insect for bioconversion – study of a trade-off between immunity and reproduction

13:00 - 14:30 **Lunch**

14:30 - 15:15 **Senior scientist lecture** Angharad M. Gatehouse – University of Newcastle
Bioinspired pest control technologies: a new frontier in biological control

15:15 - 16:00 **Oral Presentations** (regular & short talks)

Chair: NICOLO DI SORA

15:15 - 15:20 S Federico Marangoni – University of Verona
Evaluation of the efficacy of two different "Mass trapping" devices for integrated *Euzophera pinguis* management

15:20 - 15:25 S Giuseppe Morgante – University of Padua
Investigation on Ips typographus (Coleoptera, Scolytidae) vision and integration of visual cues in trapping systems for plant pests

15:25 - 15:40 R Aurora Montali – University of Insubria
An alternative use of silkworm larvae for screening antimicrobial molecules against staphylococcal infections

15:40 - 15:55 R Davide Nardi – University of Padua
Effects of wind disturbance on forest arthropod communities

15:55 - 16:10 R Ahmed Oraby - University of Turin
Survey for *Candidatus Liberibacter* spp. in North-Western Italy

16:10 – 16:40 **Coffee Break**

16:40 - 17:30 **Oral Presentations** (regular & short talks)

Chair: VALERIO SAITTA

16:40 - 16:45 S Silvia Parenzan – University of Udine
First steps towards an artificial rearing of *Varroa destructor*

16:45 - 17:00 R Bianca Orrù – University of Turin
New insights on symbiotic control of *Halyomorpha halys* and other Pentatomoidea

17:15 - 17:30 R Cristina Pogolotti – University of Turin
Variation of *Torymus sinensis*'s phenology and impact on the effectiveness of biological control

17:30 – 17:35 S-O Simone Pitton – University of Milan
Effective and eco-friendly delivery of a *Bacillus thuringiensis*-based insecticide in *Aedes albopictus* larvae

17:35 - 17:50 R Sofia Victoria Prieto – University of Turin
Study of the gut microbiota in *Nezara viridula* and other pentatomids for symbiotic control

17:50 - 17:55 S Valeria Rossi – University of Perugia
Lethal effects of natural products and dusts on *Trissolcus japonicus*

Friday 18 November 2022

09:30 - 10:00 **Senior scientist lecture** Matteo Montagna – University of Naples Federico I*/*
Insect molecular systematics: species delimitation and identification

10:00 - 11:00 **Oral Presentations** (regular & short talks)

Chair: GENNARO VOLPE

10:00 - 10:15 R Alessandro Roman – Wageningen University & Research
Effects of *Asaia* bacteria on mosquito fitness for improvement of SIT programs

10:15 - 10:30 R Elia Russo – University of Naples "Federico II"
***Aphidius ervi* venom modulates the host-parasitoid developmental interactions**

10:30 - 10:45 R Valerio Saitta – University of Perugia
Attachment ability of the melon ladybird beetle *Chnootriba elaterii* and the effect of trichomes in different species of Cucurbitaceae

10:45 - 11:00 R Giacomo Santoiemma – University of Padua
Improving trapping strategies for *Agrilus* beetles at international scale

11:00 - 11:30 **Coffee Break**

11:30 - 13:00 **Oral Presentations** (regular & short talks)

Chair: DANIELE BRUNO

11:30 - 11:45 R Elisa Seffin – University of Udine
An unexpected interaction between pollen and nicotine

11:45 – 12:00 R Maria Carmen Valoroso – University of Naples "Federico II"
Fighting the mosquitoes spread: targeting immune genes of *Aedes albopictus* larvae to improve microbial biopesticides performance

12:00 - 12:15 R Gennaro Volpe – University of Naples "Federico II"
RNAi-mediated silencing of an immune gene in *Spodoptera littoralis* (Lepidoptera, Noctuidae) alters its embryonic development

12:15 - 13:00 **Discussion on PhD education and future careers and meeting closure**

REGULAR PRESENTATIONS

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ABSTRACTS

Investigations on the vine-feeding species complex: insect-microorganism interactions and their implications for vineyard management

A. Arpellino

University of Torino - Department of Agricultural, Forest and Food Sciences, Italy

Pest management in vineyard is changing to establish new eco-friendly control methods. Symbiotic control takes advantage of insect-microorganism interactions, since manipulating these interactions has a significant impact on insect life cycle. In North-West Italy, worrying grapevine pests are Scaphoideus titanus, the main vector of the phytoplasmal agents of Flavescence dorée, and Drosophila suzukii. The aim of this project is to provide a contribution to knowledge regarding the microbial interactions in the two pest species, to support the development of sustainable control strategies. First, to assess the fitness of S. titanus in relation to gut microbiota variation related to the host cultivar, the longevity of nymphs was tested on populations reared on Barbera and Erbaluce vines. The high throughput sequencing of 16S rRNA gene will reveal if some specific components of the gut microbiota are related to feeding on different grape cultivars; however, statistical analysis of survival curves did not show differences between the groups. To further evaluate the occurrence of differences in the gut microbiota associated with S. titanus according to the feeding source, adult leafhoppers were collected in two areas of Piedmont characterized by a single prevalent cultivar or on Vitis spp.; high throughput sequencing of 16S rRNA gene is currently ongoing on these samples. To study microbial attraction in D. suzukii, different preference bioassays were performed using different yeast and bacterial strains; Stamarella bacillaris was the most attractive strain, revealing the potential for implementing new mass capture and/or attract and kill techniques against this pest.

Insect-mediated bioconversion: from organic waste to biobased materials. Conversion efficiency and protein valorization

D. Bruno[1], M. Bonelli[2], M. Orlando[3], G. Molla[1], L. Pollegioni[1], E. Testa[4], E. Fasoli[4], M.S. Galimberti[4], V. Torretta[1], A. Vezzulli[1], U. Giese[5], M. Casartelli[2,6], G. Tettamanti[1,6]

[1]University of Insubria, Department of Biotechnology and Life Sciences; [2]University of Milano, Department of Biosciences; [3]University of Milano-Bicocca, Department of Biotechnology and Biosciences; [4]Politecnico of Milano, Department of Chemistry, Materials and Chemical Engineering "G. Natta"; [5]Institut für Kautschuktechnologie, Germany; [6]Interuniversity Center for Studies on Bioinspired Agro-environmental Technology (BAT Center), Italy

One of the most important challenges that must be addressed in coming years is the transition from linear to circular economy models. The valorization of waste plays a key role in this scenario. In particular, the reduction and bioconversion of organic waste by using insects can mitigate problems related to waste management and, at the same time, generate valuable biobased products.
To this aim, the goal of the RICH (Turning Rubbish Into biobased materials: a sustainable CHain for the full valorization of organic waste) project is to develop an innovative circular supply chain to bioconvert the organic fraction of municipal solid waste (OFMSW) into biobased materials as bioplastics, characterized by high technological value and environmentally friendly, by using the larvae of the black soldier fly (BSF), *Hermetia illucens*.
The results obtained so far demonstrate the efficacy of BSF larvae to reduce two different OFMSW, which differ in nutritional composition, as indicated by the bioconversion indexes. In particular, although differences in the efficiency of waste reduction and conversion of the ingested food were identified, no difference was recorded in the larval growth rate. In addition, the comparison of the nutritional composition of larvae and pupae reared on the two substrates revealed a similar amount of proteins and lipids. Protein extracts obtained from both developmental stages were biochemically characterized and used to produce free-standing and flexible biofilms, featured by thermal stability and high quality.

The study was funded by Fondazione Cariplo (2020-0900).

Insect-mediated bioconversion: from organic waste to biobased materials. Digestion capability and lipid valorization

M. Bonelli[1], D. Bruno[2], M.G. Pellegrino[1], M.C. Valoroso[1], A. Aldrigo[1], E. Gussoni[1], D. Roma[1], G. Molla[2], L. Pollegioni[2], M.S. Galimberti[3], S. Caccia[1], G. Tettamanti[2,4], M. Casartelli[1,4]

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The use of insects as agents for the bioconversion of organic waste and by-products could represent an innovative strategy to valorize low-value materials and obtain sustainable biobased products. To this purpose, one of the most promising insects is Black Soldier Fly (BSF) (*Hermetia illucens*; Diptera: Stratiomyidae) whose larvae can grow on a wide variety of low-quality biomass. In the framework of a project funded by Fondazione Cariplo, we are evaluating the biotransformation of Organic Fraction of Municipal Solid Waste (OFMSW) by BSF larvae to obtain sustainable biobased materials with high technological value, such as bioplastic films and bionanocomposites (from insect proteins), and biodiesel (from insect lipids).
As the midgut plays a fundamental role in bioconversion processes, first we evaluated the digestion capability of BSF larvae reared on two substrates that mimic the composition of OFMSW. Our results show that the larvae can set in motion post-ingestion responses to compensate variations in nutrient composition of the substrate through the regulation of expression and activity of digestive enzymes. Moreover, we assessed if and how the different nutritional content of two rearing substrates affected the insect composition. In particular, we performed lipidomics analysis on larvae and pupae to evaluate the impact of the rearing substrates and development on lipids profile. The results indicate that changes occur in lipid composition during insect development from both a quantitative and qualitative point of view and the rearing substrate affects lipid composition both in the larvae and pupae, but the total relative amount is not altered.
The study was funded by Fondazione Cariplo (2020-0900).

Reconstruction of the feeding behaviour of the brown marmorated stink bug *Halyomorpha halys* via molecular gut analysis

M. Fluch[1], E. Corretto[1], S. Fischnaller[2], L. Borruso[1], H. Schuler[1,3]

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Invasive species are an increasing problem and threat to ecosystems and agricultural production worldwide. A recent example is the brown marmorated stink bug *Halyomorpha halys*, which is native in Northeast Asia and has invaded North America and Europe in the last decades. This insect is extremely polyphagous and has been described on more than 300 host plants, including a variety of agriculturally important crops. Host plants are usually identified by visual inspections, meaning that any plant on which adults or nymphs of this species are found is considered a host. But the individuals are highly mobile and can change the host plants according to their needs, so a molecular analysis of the gut content is a useful tool to get further knowledge about which plants they feed on. Here we describe a molecular gut analysis of naturally occurring individuals of *H. halys*, in and around apple orchards in South Tyrol, Italy. DNA of dissected guts was amplified by PCR using plant-specific primers targeting the ITS2 region of plants. The amplicons were subsequently sequenced on a Nanopore Flongle device, which resulted in up to 220,000 sequences per individual and allows to assess the feeding behaviour of *H. halys*. Our study demonstrates the potential of using this tool which helps to gain new insights into the ecology of insect pests.

Effects of seasonality and landscape composition on pollen collected by honeybees

A. Cappellari[1], V. Malagnini[2], L. Marini[1], L. Zanotelli[2], L. Tonidandel[2], G. Angeli[2], C. Ioriatti[2], P. Fontana[2]

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The honeybee is the most important managed pollinator. Its diet is based on nectar and pollen, and since the quality of pollen varies among plant species, bees must have access to diverse pollen sources to assure colony health. Only landscapes with high floristic diversity, e.g., related to the presence of semi-natural areas, can therefore guarantee adequate resources for honeybees. The amount of agricultural area can also have a strong effect on bee health due to pesticide contamination in pollen.

This work aimed to explore how the composition and pesticide residues of pollen collected by honeybees were modulated by seasonality and landscape heterogeneity in Northern Italy. We selected 13 locations from which we collected pollen samples every month during the flowering season over two years. For each pollen sample, we determined pollen type composition and diversity and the Pollen Hazard Quotient (PHQ), which provides a measure of pollen toxicity. Finally, we determined the cover of the main habitat types in 3-km radius buffers around the sampling locations.

Landscape composition did not affect pollen diversity from April to August, however, in September the diversity increased with increasing semi-natural areas. Also, PHQ decreased with increasing semi-natural areas. Our research highlighted the scarcity of floral resources in late summer and the resulting importance of semi-natural habitats for honeybees. Since pollen quality can affect the response of bees to pesticides, beekeepers should evaluate landscape composition before placing beehives, to maximise floral resources, while minimising the likelihood of pollen contamination by pesticides.

Effects of the rearing host on *Trissolcus japonicus* olfactory responses

E. Chierici[1], G. Sabbatini Peverieri[2], P. F. Roversi[2], G. Rondoni[1], E. Conti[1]

[1]University of Perugia - Department of Agricultural, Food and Environmental Sciences, Italy; [2]CREA - Research Centre for Plant Protection and Certification, Florence, Italy

Female parasitoids must optimize their ability to find a suitable host for reproducing because foraging time is limited and the host often inconspicuous and hard to detect. Host induced plant volatiles play an essential role in orienting these females to the host. Moreover, host-derived chemicals represent an additional cue that can possibly be exploited by the parasitoid. The egg parasitoid *Trissolcus japonicus* (Ashmead) (Hymenoptera: Scelionidae) is an effective classical biological control agent of the invasive *Halyomorpha halys* Stål (Hemiptera Pentatomidae). Adventive populations of the parasitoid were detected in several countries, and field releases have been allowed in some Italian regions. Laboratory no-choice and choice tests showed *T. japonicus* potential to develop in native non-target Pentatomidae. The aim of this study was to evaluate the olfactory responses of *T. japonicus* towards plant and host-derived volatiles associated with *H. halys* and non-targets. Furthermore, we wanted to assess how the rearing host species affected host-location behaviour of the parasitoid. Our results showed that parasitoids reared on eggs of the coevolved host, *H. halys*, positively responded only to plants with *H. halys* feeding damage and a deposited egg mass. Noteworthy, *T. japonicus* reared on the native species did not exhibit attraction to any of the tested stimuli. Results seem to confirm that, when a parasitoid is reared on an uncommon host, this affects its preference for the target host.

Evaluation of imidacloprid detection and behavioral resistance of *Musca domestica* L. using proboscis extension response to sucrose containing imidacloprid

S. D'Arco[1], A.C. Gerry[3], L. Maistrello[2]

[1,2] *University of Modena and Reggio Emilia - Department of Life Sciences, Italy*
[3] *University of California, Riverside-Department of Entomology, California*

The cosmopolitan housefly *Musca domestica* L. (Diptera, Muscidae) is a major pest of livestock, also known for the appearance of many insecticide-resistant populations. This study examined differences in proboscis extension response (PER) of imidacloprid susceptible and resistant fly strains and determined whether imidacloprid detection occurs at the tarsi or proboscis sensory system. Flies were glued to wooden toothpicks using clear nail polish applied to the dorsal thorax of each fly allowing for easy manipulation of the flies. In the tarsal contact assay, 150 female flies from each fly strain that responded correctly to a negative (water) and positive (sucrose 30%) test solution were subsequently exposed to 30% sucrose containing imidacloprid at two concentrations (high: 4000 µg/ml or low: 10 µg/ml) with contact by only the tarsi. PER was recorded for each fly at 0, 2 and 10 s following continuous tarsal contact. In the proboscis contact assay, 150 females were tested similarly to the tarsal contact assay except that flies were allowed to contact the test solutions with their proboscis, and flies were allowed to feed on the 30% sucrose test solution for 2 s to confirm a positive response. Results show that in the tarsal contact assay, there was no difference in PER for flies of either fly strain or to either imidacloprid concentration, suggesting that imidacloprid was not detected by the tarsal sensilla. In the proboscis contact assay, the PER was different between fly strains and varied between imidacloprid concentration only for the resistant fly strain, suggesting that imidacloprid detection occurred at the proboscis.

Effect of biological control agents at field conditions to control the spruce bark beetle *Ips typographus*.

L. Deganutti

Padua University – Department of Agronomy, Food, Natural resources, Animals and Environment, Italy

During spring and summer 2022 a field an experiment to test biological control for *I. typographus* was carried out in south-eastern Alps. The effects of different biological control agents were assessed in field on Norway spruce bolts infested by the spruce bark beetle. The treatments were applied in 45 cm long log bolts with 20 cm diameter, baited with a specific aggregation pheromone to promote colonization. We tested the entomopathogenic fungi *Beauveria bassiana* (Naturalis) and *Metarhizium anisopliae* (with *Beauveria bassiana*) (Bomet); the bacteria *Bacillus thuringiensis* var. *tenebrionis* (Novodor FC), and the predator *Thanasimus formicarius* attracted on the logs by a specific pheromone (ThanasiWit Witasek). The breeding performances of *I. typographus* assessed in term of emerging adults of the new generation were compared among treatments and the control (baited but untreated bolts). Bolts were treated and placed in the field on May 17th, during the emergence and dispersal of the overwintering beetles to ensure full bark colonization. The bolts were then removed from the sites two weeks after positioning and placed in breeding plastic pipes closed with metal mesh. Pipes were checked every week collecting bark beetles and other insects emerging from the bolts. Results showed ineffective reduction in bark beetle production in *Metarhizium anisopliae* and *Bacillus thuringiensis* var. *tenebrionis* treatments (in average 177.1 and 169.1 beetles per bolt respectively) compared to control (in average 177 beetles per bolt). *Beauveria bassiana* reduced offspring by 14.6%, while *Thanasimus formicarius* by 23.1% (in average 151.1 and 136.1 beetles per bolt, respectively). Other non-target species such as *Thanasimus formicarius*, *Acanthocinus* sp. and *Tomicobia seitneri* were also observed in the bolts.

Advancements in symbiotic control of stink bug pests of hazelnut from the local to the global scale

M. Dho

University of Torino – Department of Agricultural, Forest and Food Sciences, Italy

Halyomorpha halys is a phytophagous insect native to Eastern-Asia which causes severe damages to hazelnuts. So far, the genetic variability of *H. halys* and the primary endosymbiont *Candidatus* Pantoea carbekii has been studied by sequencing few targets. A deeper study may highlight the occurrence of still underexplored genetic variants of the insect-symbiont system, which may also affect the pest susceptibility to the recently proposed symbiotic control strategy. This approach allows nymph suppression after symbiont elimination; however, the insect-symbiont dynamics after anti-symbiont treatments have been poorly studied under field conditions. The main aims of this research are: 1) identify novel genetic markers to thoroughly study *H. halys* and *P. carbekii* genetic variability; 2) study symbiotic control efficacy in Piedmontese hazelnut orchards. A Multi Locus Sequence Typing approach was initiated as a first step to expand the knowledge on *H. halys* genetic variability: five genetic markers showed increased variability when tested on the presently known insect haplotypes. Moreover, the outcome of symbiotic control was studied in a hazelnut grove by applying sentinel egg masses before symbiotic control field applications. The egg mass position on plants was correlated to the wetting percentage resulting from treatment, to mortality of first instar nymphs emerged from treated eggs, and to elimination/reduction of symbionts in emerged nymphs. No correlation was seen between wetting percentage and nymphal mortality, whereas the position significantly affected the wetting percentage. The nymphal acquisition of *P. carbekii* was not totally prevented by treatments, but a reduced symbiont concentration was often observed.

Biological control of *Toumeyella parvicornis* with *Exochomus quadripustulatus* and *Cryptolaemus montrouzieri*

N. Di Sora[1], L, Rossini[1], M, Contarini[1], S, Speranza[1]

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The sap-sucking insect *Toumeyella parvicornis* (Cockerell, 1897), commonly known as pine tortoise scale, is an alien pest for the European territories infesting the genus *Pinus*. In the recent years the species is rapidly spreading, causing several diebacks of stone pines (*Pinus pinea* L.), its main host plant. The infestations are widespread, since the stone pine is largely distributed in the Italian Peninsula especially in urban areas where plants are considered as a landscape symbol. Despite local authorities are controlling the pest through endotherapic treatments, it is necessary to investigate an alternative control strategy which may also be applied in natural systems such as the pinewood forest, a forest type widely distributed along the Italian coastal areas. We tested the activity of two predators belonging to the Coccinellidae family, in controlling *T. parvicornis*. More precisely, we focused on two ladybugs' species: *Exochomus quadripustulatus* (Linnaeus, 1758) and *Cryptolaemus montrouzieri* Mulsant, 1850. Parallel tests were conducted both in laboratory and in semi-field conditions. Under laboratory conditions we investigated the: *i)* longevity, and *ii)* attraction response of the ladybugs' species fed with the pest. In an overtime semi-field experimentation, instead, we applied net sleeves on infested stone pine twigs, focusing on the predation activity of ladybugs, forced to stay inside. Preliminary results showed a positive response of the two ladybugs in controlling the prey and encourage us to better explore this potential strategy, hence more studies on the use of these two coccinellids for biological control of *T. parvicornis* should be considered.

Nectar-dwelling filamentous fungi affect nectar attraction and survival of the stink bug egg parasitoids *Trissolcus basalis* and *Ooencyrtus telenomicida*

J.D. Ermio[1], A. Cusumano[1], P. Bella[1], E. Peri[1], M. Rostas[2] and S. Colazza[1]

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[2]*University of Goettingen – Division of Agricultural Entomology, Department of Crop Sciences, Germany*

Adult parasitoids utilize floral odors to forage and locate suitable food resources such as floral nectar. Recent studies have highlighted the role of volatiles from nectar-dwelling microbes in parasitoid foraging behavior. Bacteria and yeasts are thus far the reported microbes found to elicit attraction in parasitoids. Although these microbes may coexist with filamentous fungi in floral nectar, the latter has received little to no attention, especially in plant-parasitoid interactions. Here, we have isolated and identified 6 culturable nectar-dwelling filamentous fungi (3 *Cladosporium* spp., 2 *Aspergillus* spp., and 1 *Stachybotrys* sp.) from buckwheat - a flowering plant largely used in conservation biological control. We further investigated how fungal fermentation of synthetic nectar affects the preference and performance of the stink bug egg parasitoids *Trissolcus basalis* and *Ooencyrtus telenomicida*. Both parasitoids were attracted to nectar fermented by *Cladosporium* sp. SAAF 22.2.11, while *O. telenomicida* was also attracted to nectar fermented by *Cladosporium* sp. SAAF 22.3.29. Parasitoid olfactory responses were neutral toward other test fungi. Interestingly, nectar fermented by *Cladosporium* sp. (SAAF 22.2.11) improved the performance in terms of survival of *T. basalis* as compared with the unfermented synthetic nectar, suggesting that it has a direct influence on insect fitness. For *O. telenomicida*, no significant effects on its survival were observed after feeding with fungus-fermented synthetic nectars. These results are important in understanding the complex interactions between parasitoids and flowering plants that are mediated by nectar-dwelling microbes, and can help to unravel strategies on how to improve conservation biological control.

The Development of Novel Biopesticides Derived from the Venom of Insect Antagonists

R. Calvert, M. Edwards, A. M. R. Gatehouse

Newcastle University – School of Natural and Environmental Science – United Kingdom

Climate change and pesticide resistance continue to challenge global food security, largely by contributing to the rise of resistant insect pest populations. Therefore, it remains essential to develop novel and sustainable methods of insect pest control. Arthropods such as spiders and parasitic wasps remain a largely untapped resource of insecticidal compounds with novel modes of action. These could be highly effective if developed for use in agriculture. This work involves the expression of known insecticidal proteins derived from the venom of two parasitoid wasps in the microbial expression system *Pichia pastoris*.

Recovery of plant and pollinator communities after severe wind disturbance in Alpine forests

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Natural disturbances are fundamental drivers of forest dynamics. By changing the structure and the composition of forests, disturbances reallocate available resources and promote stand successional development. Amongst abiotic disturbances, windthrows are recognised as those affecting European forests more severely. While windthrow impacts on ecosystem services are generally negative, the enhancement of the forest structural heterogeneity seems to benefit biodiversity. However, the mechanisms influencing the recolonization dynamics of biotic communities still remain unclear. Here, we focused on exploring the recovery of plants and of pollinators after a large-scale windthrow event in Alpine forests. To study the response of target communities, we sampled 6 forest sites and 35 salvaged areas 3 years after the windthrow event. Preliminary results suggest that topographical and large-scale variables, such as climate and landscape, play a key role in determining the recovery of the herbaceous layer, and subsequently the presence and abundance of pollinators. Understanding the recovery mechanisms of biodiversity after natural disturbances can provide important insights on the forest management strategies targeted to the provision of multiple ecosystem services.

The potential of ground-dwelling arthropods as biodiversity indicators in maize agroecosystems of Northern Italy

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Reliable monitoring of arthropod diversity is pivotal for the efficient conservation of related ecosystem services such as biological control. The complexities of arthropod collection and identification, however, highlight the need for surrogate taxa that can be easily sampled and be representative of other taxa in term of diversity, general community features and specific composition.
In this study, we used pitfall traps to sample three ground-dwelling arthropod taxa important as biocontrol agents (ground beetles, rove beetles and spiders) in 9 maize agroecosystems of Northern Italy over the course of two years, with the goal of characterizing their assemblages and evaluate their reciprocal potential as indicators of general community features and specific species cooccurrence.
Although dominated by few generalist species, sampled arthropod communities were relatively species-rich, with an interesting finding being represented by the first Italian records of the spider *Zelotes metellus* (Roewer). Ground beetles were confirmed as promising indicators for the species richness and community turnover of the other two groups. We also showed that, while the cooccurrence of individual arthropod species is limited for the studied taxa, a few species such as the ground beetle *Parophonus maculicornis* (Duftschmid) do show promise as species-level bioindicators. Our results could be useful in improving the monitoring and management of these important natural enemies in maize-rich regions.

Studies on the bioethology of *Sclerodermus brevicornis* to improve biological control strategies

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Parasitoids have a key role in the control of xylophagous species that spend most of their life cycle inside wood, and thus are difficult to reach by chemicals. *Psacothea hilaris hilaris* (Pascoe) (Coleoptera, Cerambycidae, Lamiinae, Lamiini), is an Asiatic beetles detected in Italy in 2006 that causes damages to plants belonging to Moraceae, in particular mulberry and fig plants. The Hymenoptera *Sclerodermus brevicornis* Kieffler (1906) (Hymenoptera: Bethylidae) appears as a good candidate for its biological control. In order to optimize its release, it is crucial to screen in detail its bioethology, in particular life cycle, reproduction and behavioural strategies. *S. brevicornis* is a quasi-social parasitoid, and it is known that usually behavioural studies must take into account that the interactions become more intense as the degree of sociality increases. In addition, it is very difficult to observe *S. brevicornis* in nature as its lifecycle is spent mostly inside wood galleries. For these reasons, studies of bioethology in laboratory were aimed at balance the number of parasitoids with the hosts (intraspecific competition), analyze the interaction with other parasitoids (intraguild parasitization) and the possibility to find an alternative host for mass rearing.

An alternative use of silkworm larvae for screening antimicrobial molecules against staphylococcal infections

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The spread of antibiotic-resistant bacteria has highlighted the need to develop and commercialize new antimicrobial molecules. The standard protocols for screening new antibiotics include preclinical trials performed on mammals but, due to ethical problems and high costs, the identification of alternative infection models among invertebrates is an aspect of primary importance. Lepidoptera, in particular the silkworm, *Bombyx mori*, proved to be valuable models that can partially reduce the use of mammals in this context.
In this study, a silkworm infection model was used to test the efficacy of three glycopeptide antibiotics (vancomycin, teicoplanin, and dalbavancin) against two Gram-positive nosocomial pathogens (*Staphylococcus aureus* and *Staphylococcus epidermidis*). The larvae were reared at 37 °C to reproduce human physiological conditions, and the infection was monitored by analyzing the survival of the larvae, the bacterial load in the hemolymph, and immunological markers. In particular, the activation of the cellular and humoral response was assessed through the viability of hemocytes, the activity of the prophenoloxidase system and lysozyme, and the expression of antimicrobial peptides. The results showed that all the three antibiotics were effective in curing the infection elicited by both bacteria, reducing insect mortality, and inhibiting the activation of the immune system. The present study demonstrates that the silkworm can be introduced into preclinical trials and used as an alternative infection model for a rapid and cheap screening of novel antimicrobial compounds against staphylococcal infections.

Effects of wind disturbance on forest arthropod communities

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Natural disturbances, such as windstorms, can strongly affect forest ecosystems and are expected to increase in the coming years, due to the climate change. Therefore, understanding the multi-facet impacts of forest disturbances on communities is an urgent objective for targeting appropriate management strategies. I focused on understanding the effects of a large-scale wind disturbance on forest biodiversity, and evaluating the ecological role of post-disturbance sites to protect biodiversity at the landscape scale.

First, I focused on the effects of wind disturbance on three groups of ground-dwelling arthropods: spiders, ground beetles, and harvestmen. Our results highlighted that wind disturbance had different consequences depending on the taxonomic and the functional traits. Moreover, underlying ecological gradients, such as topography and climate, modified the impact of windstorm.

Second, I investigated the abandoned patches as proxy for the long-term response of post-disturbance sites. Using spiders as target group, we found that temporary abandoned patches played a pivotal role in enhancing the spider diversity at the landscape scale. These transition habitats created by disturbance or abandonment enhanced the landscape environmental heterogeneity providing novel ecological niches compared to homogenous forest landscapes.

In conclusion, forest disturbances are extremely complex phenomena. The high complexity of such phenomena and the variety of their outcomes should be considered when designing conservation and management actions, and when planning future forest landscapes.

Survey for '*Candidatus* Liberibacter' spp. in north-western Italy

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The gram-negative bacteria '*Candidatus* Liberibacter' spp. are phloem-limited plant pathogens causing diseases in several crops' families. In Europe, some haplotypes of '*Ca.* Liberibacter solanacearum' (CLSol) pose a threat causing damage to carrots beside being hosted by other commercial crops and wild plants. They are carried by numerous species of psyllids, among them *Bactericera trigonica* Hodkinson, *Trioza apicalis* Förster and *T. urticae* L. On the contrary, '*Ca.* Liberibacter europaeus' (CLeu) which is transmitted by *Cacopsylla pyri* to pear trees and is present in different rosaceous plants, apparently do not behave as a pathogen but as an endophyte, but not in New Zealand where it causes serious damage to *Cytisus scoparius* (L.). In order to define the presence and spreading of the pathogens in north-western Italy, a wide screening for "*Ca.* Liberibacter" species in insects and in cultivated and wild plants has been implemented. Insect samplings were done in 21 different locations, on 18 plant species resulting in the collection of around 1,500 individual psyllids. After species identification, total DNA extraction was done from more than 700 psyllids. Different PCR-based detection protocols were tested. Currently, special attention is posed on CLeu that was found in four different psyllid species [*Cacopsylla pyri* L., *C. pyricola* (Förster), *Arytainilla spartiophila* Förster and *Arytaina genistae* (Latreille)] with two associations new to Italy. Further search for '*Ca.* Liberibacter' species/haplotypes is ongoing.

New insights on symbiotic control of *Halyomorpha halys* and other Pentatomoidea

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Halyomorpha halys is a polyphagous insect, native to eastern Asia and recently introduced in the Americas and Europe, which severely hampers many agricultural productions. To control its populations, an intensive use of insecticides is applied; however, the use of many active substances is currently raising important environmental and health concerns. For this reason, major efforts are being conducted to implement biological control using egg parasitoids. Another insecticide-free control tool is symbiotic control, i.e. the interruption of vertical transmission of the obligate bacterial symbiont '*Candidatus* Pantoea carbekii'. The potential related to symbiotic control application to counteract *H. halys* was assessed under laboratory conditions, exposing fresh egg masses to different commercially available products with antibacterial activity. Products application results in high nymphal mortality, providing a considerable contribution to the reduction of stink bug populations. Moreover, the impact of symbiotic control on parasitism by native and exotic egg parasitoids was evaluated. Treatment of the egg masses did not affect emergence rates of the tested parasitoid species. A field assessment of the effect of symbiotic control on the populations of *H. halys* and *Eurygaster maura* was performed in different agroecosystem areas; a significant reduction of the percentage of stink bug-damaged fruits was observed in plots used for anti-symbiont treatments, compared with untreated control plots. Moreover, no reduction of parasitism activity and no natural enemies' biodiversity change was reported in treated groves, suggesting that symbiotic control may represent a valid sustainable tool supporting biological control in the containment of productive losses caused by *H. halys*.

Variation of *Torymus sinensis*'s phenology and impact on the effectiveness of biological control

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The biocontrol agent *Torymus sinensis* Kamijo represents one of the most successful examples of classical biological control programs, able to obtain long-term control of the Asian chestnut gall wasp (ACGW), *Dryocosmus kuriphilus* Yasumatsu. Recently, an unusual presence of galls was recorded in a few chestnut orchards of Northern Italy, although *T. sinensis* was previously released. Investigations were performed in the five-year period 2018–2022 in six chestnut orchards, selected according to the ACGW infestation level, and divided in "non-critical sites" (negligible presence of galls) and "critical sites" (high presence of galls). We evaluated *T. sinensis*'s parasitism rate and phenology by dissecting chestnut galls. Temperatures were monitored in all the orchards with data loggers. In non-critical sites the parasitism rate by *T. sinensis* was stable and/or growing in all years, accounting from 77% to 99%, whereas in critical sites it experienced a severe decline, reaching values under 50%. The dissection of the galls recorded in non-critical sites in February highlighted that most *T. sinensis* were larvae and immature pupae (70%). Conversely, in critical sites most of the individuals were mature pupae (76%), with presence of newly formed adults (12%), and the mean temperature recorded was 2.72°C and 2.34°C higher in January and February respectively. Moreover, in critical sites the early emergence of *T. sinensis* (late February-early March) was recorded when current-year ACGW fresh galls were not available, suggesting that the asynchrony between *T. sinensis* and the ACGW heavily affected its role in the suppression of the pest's outbreaks.

Study of the gut microbiota in *Nezara viridula* and other pentatomids for symbiotic control

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Insects of the Pentatomidae family harbour an obligated symbiont in a gut ventricle, vertically transmitted via egg surface contamination by the females. Since stink bugs are high impact pests, alternative management strategies are under development. The symbiotic control consists on disrupting the symbiont acquisition by treating the egg masses with low-impact anti-symbiont products. This leads to high mortality in species such as *Halyomorpha halys*. We tested the efficacy of a micronutrient biocomplex to eliminate the symbiont of another pentatomid, *Nezara viridula,* and we assessed mortality on nymphs. Mortality was higher on the treated group, although many samples showed strong survivorship. The genetic background and the interaction with other bacteria could be involved in this response. Through total microbiota 16s rRNA gene sequencing, we discovered six strains of *N. viridula*'s symbiont; isolation efforts of these strains revealed their uncultivability. We also detected the presence of *Pantoea* strains – diverging from the primary symbionts – in samples where the symbiont had been eliminated; the role of replacing bacteria for the host fitness is under investigation. Regarding other pentatomids, egg masses of four species present in North Italy where treated with the anti-symbiont biocomplex and preliminary results indicate an increase in mortality in the treated group. These tests have been contrasted with field trials, which indicated a reduction of production losses where symbiotic control is applied.

Effects of *Asaia* bacteria on mosquito fitness for improvement of SIT programs

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Arboviruses are responsible for many diseases in both humans and animals. *Aedes aegypti* is the primary vector of yellow fever, dengue, chikungunya and Zika virus. Disease-control strategies strongly rely on mosquito population management through insecticides and larvicides. However, the threat of insecticide resistance is rising, resulting in less effective vector control techniques. Over the last decades, new methods for population management have been developed, including the Sterile Insect Technique (SIT). To successfully apply SIT, healthy and competitive mosquito colonies are required, allowing sterilized males to compete with their wild counterparts. Recently, it has been shown that mosquito microbiota could play an essential role in larval development and fitness. Bacteria of the genus *Asaia* were identified as potential actors in paratransgenesis techniques and as probiotic. However, little is known about the effects of *Asaia* spp. on adult fitness, in *Ae. aegypti* and in other mosquito species of public health interest. In this study, we inoculated first instar larvae of *Ae. aegypti*, *Ae. albopictus*, *Culex pipiens* biotype *molestus* and *Anopheles coluzzii* with different species of *Asaia* and for different exposure times. Fitness parameters were analysed as well as the microbiome composition following the exposure to *Asaia* spp. Ongoing efforts include exploring potential microbiome manipulations on male attractiveness and competitiveness in laboratory and semi-field contexts. Our findings underline the importance of understanding host-microbe interactions. In addition, improving mosquito fitness can be beneficial for large-scale sterile male production.

Aphidius ervi venom modulates the host-parasitoid developmental interactions

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The endophagous parasitoid *Aphidius ervi* (Hymenoptera, Braconidae) modulates the physiology and reproduction of the pea aphid *Acyrthosiphon pisum* (Homoptera, Aphididae), to enhance its nutritional suitability for the developing offspring, using host regulation factors both of maternal (i.e., venom) and embryonic (i.e., teratocytes) origin. Here we investigated the functional role of the main component of *A. ervi* venom, *Ae*-γ-glutamyl transpeptidase (*Ae*-γ-GT), using the RNA interference (RNAi) technique. The suppression of *Ae*-γ-GT was obtained through microinjection of dsRNA solution in *A. ervi* female pupae, and its relative expression was checked by qRT-PCR. Parasitism by *Ae-γ-GT* knocked down females induced a significant increase of aphid size and bacterial load of the primary aphid's symbiont *Buchnera aphidicola*. Similarly, both *A. ervi* larvae and teratocytes showed a significant increase in size. The silencing of *Ae-γ-GT* significantly reduced the host castration induced by control wasps, which resulted similar to that induced by wild type females. Parasitoid adults were of larger size, but this trait was associated with a reduced survival and fecundity, suggesting a trade-off of the body size increase. These results shed light on the role of venom in the intricate network of interactions among the parasitoid, the host aphid and its symbiont, which finely orchestrate the development of parasitoid's progeny.

Attachment ability of the melon ladybird beetle *Chnootriba elaterii* and the effect of trichomes in different species of Cucurbitaceae.

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The attachment ability of the oligophagous melon ladybird beetle *Chnootriba elaterii* was investigated in relation to the leaves' trichomes coverage of several Cucurbitaceae species. We described adult and larva tarsal attachment devices and leaf surface structures using the Cryo-SEM (glandular and non-glandular trichomes) in *Citrullus lanatus*, *Cucumis melo*, *Cucumis sativus*, *Cucurbita moschata*, *Cucurbita pepo, Ecballium elaterium*, *Lagenaria siceraria,* and *Luffa aegyptiaca*. We measured the friction force exerted by females and larvae on plant leaves using traction force experiments and centrifugal force tests. We observed that Cucurbitaceae glandular trichomes do not affect insect attachment ability at both developmental stages; this fact suggests an adaptation of *C. elaterii* to its host plants. The non-glandular trichomes instead, when are dense, short, and flexible, heavily reduce the attachment ability of both insect stages. When trichomes are dense but stiff, only the larval force was reduced probably because the larva has a single claw, in contrast to the adult having paired bifid dentate claws. The data on the mechanical interaction of *C. elaterii* at different developmental stages with different Cucurbitaceae species, combined with data on the chemical cues involved in the host plant selection, can help to unravel the complex factors driving the coevolution between an oligophagous insect and its host plant species.

Improving trapping strategies for *Agrilus* beetles at international scale

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Jewel beetles (Coleoptera: Buprestidae) of the genus *Agrilus* includes over 3000 species at the global scale. Larvae feed on the living subcortical tissues of trees and shrubs, and feeding activity can kill the host, especially when it has already been weakened by other abiotic or biotic factors. Many *Agrilus* species have a proven invasive potential, being easily transported within nursery plants and wood products. As a consequence, some species have become important invasive pests into new geographic areas, leading to wide environmental, economic and social impacts. Hence, trapping strategies to be used at entry points are of utmost importance to intercept incoming individuals. Reliable trapping protocols exist so far only for the emerald ash borer *Agrilus planipennis*, one of the most destructive species existing nowadays, while only a few and scattered studies targeted other *Agrilus* spp. can be found. The Euphresco project "Developing and assessing surveillance methodologies for *Agrilus* beetles" aimed to assess the effectiveness of the trapping protocols commonly used for *A. planipennis* also for other *Agrilus* spp., testing 1) green sticky prism traps vs. green multifunnel traps, 2) green sticky and multifunnel traps with and without the leaf alcohol 3Z-hexenol, 3) multitraps vs. multitraps on which adult beetles of a target species were attached. These results will contribute to aid in surveillance and monitoring approaches for the early detection of these wood-boring insects.

An unexpected interaction between pollen and nicotine

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Honeybees (*Apis mellifera*) play a vital role in the ecosystems and contribute to the conservation of plant biodiversity and agricultural production. Honeybee survival depends on a convenient supply of essential nutrients, indeed pollen-the only source of proteins for honeybees-is necessary for growth and development. Lack of pollen may therefore be regarded as an important stress for honeybees. However, pollen may contain toxic compounds such as plant's secondary metabolites or residues of pesticides used in agriculture. Nicotine is a alkaloid which can be found in both nectar and pollen of some plants, and it has a chemical affinity with some insecticides. We decided to investigate the interaction between pollen deprivation and nicotine. Furthermore, to study if seasonality can influence this interaction, our investigation was carried out both early in the season when the prevalence of a common honeybees viral pathogen (Deformed wing virus-DWV) is low and later when DWV is widespread. We found that pollen deprivation decreases survival whereas nicotine (50 ppm) appeared to be harmless because of an effective detoxification system which we impaired with piperonyl butoxide. Moreover, DWV causes a reduction in bee survival while, generally, the effect of the other factors remains similar. However, interestingly, the effect of nicotine changes, suggesting that viral infection increases the impact of this stressor; instead, pollen deprivation seems to mitigate the effect of this alkaloid. In conclusion, honeybees seem to have an effective detoxification system which may be sustained by pollen nutrition; however, DWV drastically changes the normal interactions among stress factors.

Fighting the mosquitoes spread: targeting immune genes of *Aedes albopictus* larvae to improve microbial biopesticides performance

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In the Era of globalization and global warming, the spread of insect vectors is increasing enormously in non-endemic regions. Mosquitoes females, through their bites, are one of the most threatening vectors of arboviruses, causing an increasing number of diseases. The highly invasive Asian tiger mosquito, *Aedes albopictus*, has spread worldwide and it has become crucial to control both adults and aquatic larvae. Nowadays, the development of molecular and genetic tools as RNAi and gene-drive set the stage for novel control strategies or the improvement of already used techniques as the sterile insect technique.

We aim to contribute to vector control by developing a novel control strategy against mosquito larvae based on the improvement of microbial entomopathogens effectiveness by immune suppressive RNA interference (RNAi), an approach that resulted effective in lepidopteran crop pests. For this purpose, we have identified and isolated the full-length mRNA of several putative immune genes to be used as target candidates for RNAi in *Ae. albopictus* larvae. Searching in the *Vectorbase* database, we selected putative sequences of *Ae. albopictus* by in *silico* analysis. Then, putative sequences were amplified by PCR, sequenced and confirmed. Real-time qPCR experiments revealed that all selected genes showed increased expression upon larvae exposure to the entomopathogen *Bacillus thuringiensis* var. *israelensis* (*Bti*) suggesting their involvement in immune responses.

Their functional characterization will be important to validate them as good candidates to develop novel strategies of mosquito larvae control based on the synergy between *Bti* and RNAi.

RNAi-mediated silencing of an immune gene in *Spodoptera littoralis* (Lepidoptera, Noctuidae) alters its embryonic development

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The *102* gene, which encodes a protein essential for the cellular immune response, has been isolated and functionally characterized both in *Heliothis virescens* (Lepidoptera, Noctuidae) and in *Spodoptera littoralis* (*Sl102*). Knockdown of the *Sl102* gene in *S. littoralis* larvae increases the susceptibility to pathogens, providing the basis for the development of new insect control strategies. Here we explore the phenotypic effects of the *Sl102* gene silencing on embryonic development by soaking *S. littoralis* eggs in a dsRNA solution. The experimental results demonstrate that gene expression in embryos treated with *dsSl102* is significantly reduced compared to control embryos treated with *dsGFP*. Furthermore, the gene silencing is associated with a drastic reduction of egg hatching and a very high mortality rate of the few hatched larvae. Structural and ultrastructural analyses showed morphological alterations in treated embryos indicating that the *Sl102* gene, in addition to its immune function, has an important role in the regulation of embryonic development so far unrecognized, which is worth of further research efforts.

SHORT PRESENTATIONS

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ABSTRACTS

Development of innovative methods for the early-detection of the European spruce bark beetle outbreaks

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Increasing frequency and intensity of extreme weather events due to climate change determine great stress to European conifer forests and create favorable conditions for pests' outbreaks. In the Southern Alps, the European spruce bark beetle (*Ips typographus*) shifted from an endemic to an epidemic phase in the last four years, elicited by the dramatic windthrow event occurred at the end of 2018 (Vaia). According to such outbreaks, it is important to understand the spatio-temporal dynamics of the bark beetles' infestations, to better define and manage areas needing pest control. Remote sensing techniques (such as time-series analysis of multispectral imagery acquired by satellites, airplanes, or drones) have a great potential in the detection of bark beetle infestations. However, most of these methods pose several limits. For example, the satellite imagery available on open access platforms can clearly detect the damage only when the tree crowns become red (the so-called red phase), *i.e.*, when trees are dead, and the bark beetles have more likely already left the hosts. This, although useful to quantify damages, does not prove to be an effective tool to detect infestations at their early stages (green phase), which would be needed to promptly map new infestation spots and to take effective actions to contain the outbreak. For this reason, we explored the possibility to develop a method to characterize and detect the symptoms of the infestations at their early stages, employing multispectral images acquired by a drone to identify a spectral signature specific for *Ips typographus* infestations occurrence.

Role of thermal heterogeneity in driving co-existence of competing species

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Intra- and interspecific competition for limited resources exploitation is observed in a wide range of organisms. Costs induced by competition often lead to a decrease in survival and/or reproduction of participants. Temperature, especially in ectotherms, strongly influences individuals' fitness by modifying their metabolic rate, the resource quality and the pay-off of individuals' competitive behavioural strategies. Therefore, in a heterogeneous environment, where microsites of various temperature are present, organisms can compete to access thermal sites close to their thermal optimum. Individuals from a same species or from species with similar thermal performances may exclude each other from microsites presenting thermal optima. In contrast, individuals having wide thermal range or different optima may cohabite within the same heterogeneous habitat. Here, we study two parasitoid species, *Eupelmus vuilleti* and *Dinarmus basalis,* in which females directly compete each other to access their hosts, larvae of the cowpea seed weevil, *Callosobruchus maculatus*. As parasitoid development is often restricted to a narrow range of host species, intraspecific competitive encounters, as well as interspecific ones between species sharing the same hosts, can be frequent. Both *E. vuilleti* and *D. basalis* can be used as biocontrol agents against *C. maculatus*, especially when this pest attacks food stocks in Africa. The study of thermal intra- and interspecific competition between those two species may allow a better understanding of their population dynamics as regard to climate warming. Competitive exclusion or increased costs of competition may reduce their effectiveness as a pest control.

Characterization of RNASET2 in *Hermetia illucens*

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The larvae of the black soldier fly (BSF), *Hermetia illucens* (Diptera: Stratiomyidae), can be used as a source of protein and lipids for the production of innovative animal feed with high nutritional value. Since they grow on organic waste, decaying substrates can bring the larvae into contact with different potential pathogens that can challenge their health status and growth. These life strategies have likely contributed to shape the evolution of a sophisticated set of antimicrobial molecules in this dipteran.
The aim of this project is to characterize RNASET, a transferase-type ribonuclease with a key role in the immune response of both vertebrates and invertebrates, in *H. illucens* larvae. We investigated the expression of RNASET2 in the fat body and hemocytes - two tissues that are involved in the immune response of insects - in larvae infected with a bacterial mix of *Escherichia coli* and *Micrococcus luteus*. Immunostaining, qRT-PCR, and Western blot analyses of the fat body revealed no variation in RNASET2 expression within 6 hours after the infection, while the expression in hemocytes significantly increased 3 hours after the injection of bacteria in the hemocoel. These preliminary data represent the starting point for an in-depth characterization of this enzyme in BSF and to assess if it plays an antibacterial role as in other animal species.

Symbiotic control of harmful insects as an innovative sustainable method: the case study of *Bactrocera oleae* (Diptera, Tephritidae)

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The olive fruit fly, *Bactrocera oleae* (Diptera, Tephritidae), is one of the main pests of *Oleae europea* that causes significant production and economic losses in the Mediterranean basin. The control of this phytophagous insect has traditionally been conducted using broad-spectrum insecticides (such as dimethoate), which have been revoked by current legislation on sustainable use of pesticides. These restrictions have led the research towards new sustainable strategies to control the olive fruit fly. The symbiotic control, which relies on the elimination of obligate bacterial symbionts of insects, has been proposed as a potential alternative strategy. *B. oleae* is strictly associated with the bacterial symbiont *Candidatus* Erwinia dacicola which is vertically transmitted from mother to offspring. It has important functional roles for *B. oleae* which go from detoxifying toxic compounds related to olives to increasing *fitness*. On this basis, this project is oriented towards disrupting the symbiosis by treating of the eggs surface using antimicrobial products as corroborants and polyphenols since they have been shown to be effective in other studies on the bacterial load of the symbiont. These compounds could open new avenues for sustainable control of *B. oleae*, fostering the development of sustainable strategies.

Plant-mediated effects of the entomopathogenic fungus *Beauveria bassiana* on *Spodoptera littoralis*

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Beneficial microorganisms constitute a significant and promising alternative to agrochemicals thanks to their capacity to promote plant growth and their defence barriers against different biotic stress agents. *Beauveria bassiana* is one of the most important entomopathogenic fungi and can colonize a wide variety of plant species. Experimental evidence has already shown that it can contribute to limit growth and survival of plant pests and pathogens. Here we further contribute to this research topic by studying the effect of tomato plants colonization by *B. bassiana* on the survival, the development, and the immunity of *Spodoptera littoralis* (Lepidoptera, Noctuidae). Unexpectedly, endophytic colonization of plants had no effect survival of larvae feeding on them, which also showed a weight increase. Furthermore, encapsulation and nodulation responses of these larvae were reduced. These larvae proved to be more sensitive to *B. bassiana* or *Bacillus thuringiensis* infection, given their reduced immune competence. These results shed new light on the complex network of plant multitrophic interactions which underlie the evolution of entomopathogenic lifestyle in soil fungi.

Impact of Deformed Wing Virus infection on honey bee gut microbiota

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Honey bee health decline represents a problem of global importance for the remarkable impact of these pollinators on the environment and human economy. The reduced bee survival is the final result of a multifactorial syndrome triggered by several stress factors, that may synergistically interact, inducing a reduction of bee immunocompetence. A common element to all collapsing colonies is the high loads of parasites and associated pathogens, such as Deformed Wing Virus (DVW). DWV is an endemic immunosuppressive virus that generates asymptomatic covert infections, kept in check by the bees' immune system when not exposed to stress agents which weaken antiviral barriers, such as parasitizates, pesticides and poor nutrition. Indeed, nutritional stress may negatively affect the immune system by modulating metabolism or, indirectly, by altering the composition of gut microbiota, which plays a key-role in gut physiology and immunity.
Here we focus on the role that DWV infections can have on the modulation of honey bee microbiota under different nutritional conditions.
The comprehension of the mechanisms underlying these complex immune interactions at metaorganism level will shed light on the key-components of the gut microbiota that are involved in the immune modulation of honeybees and will likely allow to define blends of probiotic microorganisms which may help to rescue the decay of honey bee immune competence.

Beneficial fungi in the genus *Trichoderma* are effective control agents of noctuid moth larvae

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Beneficial fungi belonging to the genus *Trichoderma* have well-known effects on plant biology, such as growth promotion, efficiency of nutrients uptake, seed germination rate, and plant defense against both biotic and abiotic stress agents, including phytophagous insects. Here we obtained colonized tomato plants, by coating the seeds with *Trichoderma afroharzianum* strain T22 spores (T22-plant), and scored the plant-mediated effects of T22 on the noctuid moth *Spodoptera littoralis*. Plant colonization increased mortality and reduced larval weight. Furthermore, larvae fed on T22-plants showed an immunosuppressed phenotype which is more susceptible to sub-letal doses of a biopesticide containing the entomopathogen *Bacillus thuringiensis*. The obtained results showed a clear plant-mediated effect of *T. afroharzianum* on *S. littoralis* larvae which could contribute to the reduction of pesticide use in agriculture by suppressing this pest or by enhancing its susceptibility to biocontrol agents.

Microbiome composition and distribution of major endosymbionts in *Scaphoideus titanus* (Hemiptera: Deltocephalinae)

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The Nearctic leafhopper *Scaphoideus titanus* (Hemiptera: Deltocephalinae) is the main vector of *Candidatus* Phytoplasma vitis, the causative agent of Flavescence dorée in Europe. The disease is causing high damage to grapevine production across the biggest viticultural areas of Europe. Despite quarantine control and obligatory management, outbreaks of Flavescence dorée are still ongoing in winegrowing communities. Meanwhile, *S. titanus* is a highly efficient vector due to its capacity to feed exclusively on *Vitis vinifera*. Also, Insects are associated with different microorganisms which are crucial for host survival and reproduction. Several microorganisms provide parasitoid resistance, provide essential nutrients, and infer the transmission of pathogens. Besides several essential microorganisms have been described in leafhoppers, only a few studies investigated the role of specific microorganisms in host diets and their role in the transmission of phytoplasmas in leafhoppers. Here we present the microbial composition of *S. titanus* across various populations in Northern Italy. Specifically, we investigate potential changes of phytoplasma on its microbiome. This study provides a broader understanding of the interactions between core microbes including the relations between harmful and beneficial bacteria and suggesting suitable targets for biocontrol.

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Plant-insect interactions in mountain areas: insights for an analysis of pollen loads comparing light microscopy and ITS2 metabarcoding

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Plants and arthropods interact with each other creating complex networks such as those related to pollination which play a fundamental role ecosystem functioning. Nowadays, these interactions are still little known, especially in high-mountain ecosystems. For a fine reconstruction of plant-pollinator networks, it is necessary to identify and count pollen grains carried by flower-visiting arthropods. Traditionally, pollen grains are subjected to acetolysis for a morphological identification by light microscope, but a possible alternative is to use molecular analysis, such as metabarcoding, for the simultaneous identification of many pollen taxa within the same sample. Here, we aim to compare light microscope and ITS2 metabarcoding (Illumina MiSeq technique) analysis to evaluate pollen loads of insects collected in mountain agroecosystems. In particular, we analyzed pollen loads carried by bees (Hymenoptera Apoidea Anthophila) sampled along an altitudinal gradient (from 900 to 2700 m asl), and among different agroecosystems (orchards, hay-meadows, pastures, high-altitude grasslands) within the Stelvio National Park (Val Martello, Bolzano - Italy). Even though both strategies are reliable to detect plant species at least to family level, light microscope method is more accurate for a quantitative analysis while metabarcoding is less time consuming and easily identifies plant families present in the sample. These two different techniques can be used to achieve different purposes: light microscope can be used for a fine resolution analysis of pollen load by pollinators taking into account both the qualitative and quantitative aspects, while molecular analysis can be used only for a faster qualitative pollen evaluation.

Reproduction of the black soldier fly, an insect for bioconversion – study of a trade-off between immunity and reproduction

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The black soldier fly (BSF) is a farmed insect, produced on a large scale from co-products and wastes that are very rich in microorganisms. The biology of the BSF is still little known, although the control of reproduction is a key element of the success of the breeding of farmed species. In the meantime, reproduction is a physiologically demanding process and is known to trade-off with immunity. Within the framework of this trade-off, the main goal of my thesis is to understand what parameters define a good BSF male breeder. To do so, I will use an experimental population of BSF to which I will expose different types of stresses of thermal, nutritional and microbial origin. In these various contexts, physiological traits related to reproduction will be measured: quantity of spermatozoa transferred to the female reproductive tract, composition of the seminal fluid (proteins and metabolites) and paternity ratio in a context of sperm competition. At the same time, I will try to understand how an experienced stress can lead to behavioral changes such as a male's ability to mate with females, his number of partners and the duration of copulations. The results of these experiments will give us an insight of the necessary conditions for a good reproduction of BSF, which is of vital importance for us to take full advantage of its bioconversion capabilities.

Evaluation of the efficacy of two different "*Mass trapping*" devices for integrated *Euzophera pinguis* management

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Euzophera pinguis (Haworth,1811) (Lepidoptera: Pyralidae) also called Tabby knot-horn moth, is harmful to many cultivated and spontaneous trees. Females lays eggs on wounds caused by pathogens or mechanical damage caused by climatic events or agronomic practices such as pruning and mechanical harvest. The larvae are xylophagous and feed wood by digging mines.
Since 2005 the pests have been reported as emerging pest on olive tree in the Iberian Peninsula after that, in the Balkans in 2012 and in Lebanon in 2017. The damage, attributable to larval trophic activity, is represented from bulges and subsequent desiccation of branches and large steams. In Italy – *E. pinguis* damages on olive trees were reported for the first time in 2018 in Trentino-Alto Adige, subsequently in Lombardy region in 2019 and in 2020 in Liguria and Veneto.
Considering that the chemical control against larvae is ineffective and the long adult flight period which require many insecticides applications, the aim of this study was to evaluate the efficacy of two "*Mass trapping*" devices in order to develop a sustainable strategy control against Tabby knot-horn moth.
Two different traps, Funnel (Econex) and Pagoda (Isagro), lured with sexual pheromones (Serbios) were investigated in 2022 in different olive growing areas located in Veneto and Lombardy Region.
Preliminary results showed that Pagoda trap was significative more effective than the funnel one.
The integration of "*Mass trapping*" with cultural control technique was discussed for a *E. pinguis* eco-friendly management

Investigation on *Ips typographus* (Coleoptera, Scolytidae) vision and integration of visual cues in trapping systems for plant pests

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Bark beetles play a fundamental role in forest ecosystems. Their attacks on trees promote the opening of forest gaps, degradation of dead plant material, significantly affects nutrient cycling, and soil structure. Their outbreaks can lead to extensive forest mortality, therefore, such aggressive species as the Ips typographus are also defined as “landscape engineers”. Despite their importance from a biodiversity and ecological point of view, they compete with human beings for valued plants and related timber products: for this reason, they are regarded as pests. The trapping methods used for such pest, as many others, are mainly based on olfactory cues through the recognition of chemical compounds, especially aggregation pheromones, while very little is known about their vision, visual perception of the reality and specific targets like host plants. The integration of visual cues in trapping systems of *Ips typographus* is the final goal of this study. In addition, it could open the way to application to different pests affecting both the agricultural and forest sectors, fitting one of the aims of the “Piano Nazionale di Ripresa e Resilienza” (PNRR) in the framework of the “Agritech” program.
The study to characterize *Ips typographus* eyes and vision will include:

- Anatomical and structural research of the eye (retina, interommatidial angle, visual field, number of ommatidia).
- Evaluation of spectral sensitivity, photoreceptors’ dynamic response and adaptation experiments with extracellular recordings.
- Color vision capacity study through Y-maze binary choice experiments.

The study will be carried out in collaboration with the insect vision unit of the University of Ljubljana Slovenia.

First steps towards an artificial rearing of *Varroa destructor.*

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The ectoparasitic mite *Varroa destructor* is involved in the decline of *Apis mellifera* colonies. A better understanding of *Varroa* biology through the development of an in vitro rearing method would open new possibilities for the control of the parasite. However, a tested protocol for maintaining the mite in the absence of its host is still missing. Moreover, only few and discordant data on the longevity of *Varroa* on its host under lab conditions are available; in other words, even the ultimate target of an artificial rearing method is currently unknown.
Therefore, we set-up an experiment to assess on which pre-imaginal honeybee stage (5th instar larvae vs pupae) *Varroa* would survive longer. Then we used the stage that gave best results to feed three groups of mites of presumably different age and assess their longevity: mites of unknown age from recently sealed honeybees' workers cells, mites of unknown age after a reproductive cycle in artificial cells, newly born mites from artificial rearing cells.
We found that *Varroa* can survive longer on larvae than on pupae (median survival 24 vs 8.5 days, respectively). No differences were found between the longevity of mites according to their presumed age. Overall, *Varroa* can survive up to 6 weeks under lab conditions on honeybee larvae.

Effective and eco-friendly delivery of a *Bacillus thuringiensis*-based insecticide in *Aedes albopictus* larvae

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Mosquito populations are expanding their areal to non-endemic regions due to climate change and trade globalisation, and significant concern is generated by the pathogens that are transmitted by female bites.

To contribute to vector control issue, we have developed "MosChito" raft, a floating bio-based product targeting aquatic larval stages. These innovative floating hydrogel matrixes have been produced using molecules of natural origin (i.e., chitosan and genipin) and their toxic activity was due to the inclusion of a commercial bioinsecticide based on *Bacillus thuringiensis* var. *israelensis*. Moreover, we have tested the inclusion of dead yeast cells (i.e., *Saccharomyces cerevisiae*) in the rafts to evaluate their attractive and fagostimulant potential.

We have measured the susceptibility of larvae of *Aedes albopictus*, the Asian tiger mosquito, to MosChito rafts including different doses of *Bti* and the persistence of toxic activity during time. Larvae were attracted by MosChito rafts, eroded the hydrogel with the mouthparts and ingested released particles. The toxicity was positively correlated with the dose of *Bti*-based bioinsecticide included and their toxic effect was maintained for at least 30 days, a very significant result since *Bti*-based products are easily degraded in a few days after application. MosChito rafts effectiveness was eventually measured in semi-field conditions using different *Ae. albopictus* strains. On the other hand, yeast inclusion had no significant effects on MosChito effectiveness.

Altogether, our results highlight the potential of MosChito rafts as an innovative, biorational and user-friendly solution for the control of mosquito larvae in domestic and peri-domestic breeding sites.

Lethal effects of natural products and dusts on *Trissolcus japonicus*

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The brown marmorated stinkbug *Halyomorpha halys* (Stål) is a worldwide pest, native of Eastern Asia. Its host range includes many cultivated plant species, making this insect responsible for high economic losses on a global scale. Since its first recording in Italy in 2012, *H. halys* has caused severe damage to many crops. To tentatively contain *H. halys* spread, chemical pesticides have been used. More sustainable alternatives have been proposed, such as the use of pesticides of natural origins or dusts as well as the release of biocontrol agents. The exotic egg parasitoid *Trissolcus japonicus* (Ashmead) (Hymenoptera: Scelionidae) is being currently released in Italian regions for classical biocontrol of *H. halys*. The aim of this research is to verify the lethal effects of natural pesticides and dusts on the parasitoid. Using Munger-cell procedure, we evaluated mortality of adult *T. japonicus* during 1-week exposure to commercial products (field doses) containing: zeolite, diatomaceous earth, kaolin, basalt dust, sulphur, potassium salts of fatty acids, azadirachtin, orange oil and calcium polysulphide. Among these products, sulphur, zeolite and calcium polysulphide induced high mortality (80-100%) on *T. japonicus*. Bioassays are in progress aiming at the evaluation of sublethal effects.