





# European PhD Network **"Insect Science"** IX Annual Meeting

17-19 November 2021

# SCIENTIFIC PROGRAM & BOOK OF ABSTRACTS

CREA Via di Lanciola, 12,

50125 Comune di Impruneta (FI)







### PROGRAMME

#### Wednesday 17 November 2021

- 14:00 Registration
- 15:00 Welcome address

Senior scientist lecture

15:10 Christopher John Topping – Aarhus University ALMaSS use to support a 'Systems Approach' to Environmental Risk Assessment

#### **Oral Presentations**

#### **Chair: Nicola Bodino**

- 15:40 R <u>Tugcan Alinc</u> University of Palermo *Trichoderma harzianum* strain T22 increases egg parasitoids attraction toward tomato plants infested by stink bugs
- 15:55 S <u>Andrea Arpellino</u> University of Turin Investigations on the vine-feeding species complex: insect-microorganism interactions and their implications for vineyard management
- 16:00 <u>R</u> <u>Daniele Bruno</u>– University of Insubria Characterization of the immune response of *Hermetia illucens* larvae to bacteria
- 16:15 Break

#### **Chair: Elia Russo**

- 16:45 R<u>Valentina Candian</u> University of Turin Impact of the diet on the expression of antimicrobial peptide genes in *Tenebrio molitor* (Coleoptera: Tenebrionidae)
- 17:00 R Andree Cappellari University of Padua Functional traits of plants and pollinators explain resource overlap between honeybees and wild pollinators
- 17:15 S Paul Clemencon CNRS / University of Tours The couple Nemobius sylvestris/Pardosa sp.: a model for analysing predator-prey interactions and their neural bases?
- 17:20 Sara D'Arco University of Modena and Reggio Emilia
   Parasitization efficiency in relation to the female age for the pupal parasitoids of *Musca* domestica
- 17:25 S Giovanna De Leva– University of Naples Impact of stress agents on honeybee gut microbiota and immunity







- 17:30 S <u>Gwenaëlle Deconninck</u> CNRS/University of Tours Thermal preference plasticity and fitness of an invasive polyphagous insect in heterogeneous environments
- 17:35 Sara Boschi University of Siena *Popillia japonica* transcriptome analysis
- 17:40 S <u>Rebecca Funari</u>– University of Siena Analyses by conventional microbial methods and metagenomic approach of an enrichment culture obtained from plastic-eating *Alphitobius diaperinus* (Insecta: Coleoptera)
- 17:45 S Marta Ranesi– University of Naples Beauveria bassiana outcompete the growth of a plant pathogen via oxalic acid excretion.
- 17:50 S Onofrio Marco Pistillo– University of Foggia Sex pheromone of the asparagus moth, Parahypopta caestrum: chemical analysis, electrophysiological study and preliminary field tests







#### Thursdays 18 November 2021

#### Senior scientist lecture

09:00 <u>Francesco Nazzi – University of Udine</u> Some little things about research that I've learnt in the past thirty years

#### **Oral Presentations**

#### **Chair: Marco Bonelli**

- 09:30 <u>R Mizuki Uemura</u> University of Padua Division of labour and altruism in tent construction and maintenance in a social caterpillar (*Thaumetopoea pityocampa*)
- 09:45 <u>Martin Dessart</u> CNRS / University of Tours The secret of my success! Visual learning abilities in mosquito larvae (*Aedes aegypti*) are not impaired neither by light deprivation nor by turbidity.

#### 10:00 S<u>Matteo Dho</u>– University of Turin Advancements in symbiotic control of stink bug pests of hazelnut from the local to the global scale

- 10:05 R Elena Eustacchio University of Milan Plant-pollinator interactions: a study along spatial-temporal gradient in different land management types within the Stelvio National Park
- 10:20 R Priscilla Farina University of Pisa Starting with the basics: biology and European distribution of the fig weevil Aclees taiwanensis
- 10:35 S Antonio Franco University of Basilicata Innovative processes for lipid extraction from bioconverter insects, qualitative and quantitative evaluation and industrial applications for the formulation of personal care products
- 10:40 Break
- 11:10 R Elena Gazzea University of Padua Quantifying the effects of insect pollination on crop quality
- 11:25 R Costanza Geppert DAFNAE, University of Padua Herbivory: a hidden player in plant responses to global change
- 11:40 S <u>Anna Guarnieri</u> University of Basilicata The black soldier fly *Hermetia illucens*: an innovative and alternative source of chitin and chitosan







- 11:45 R Mélissa Haouzi CNRS / University of Tours Heterogeneous venom gland composition in females of the invasive Asian hornet Vespa velutina nigrithorax
  12:00 S Giovanni Jesu – University of Naples Distribution of fly species of veterinary importance in Basilicata region (Italy)
  12:05 S Elisa Legoratti – University of Milan Autumn and night: the neglected side of plant-arthropod interactions
  12:10 S Enrico Mirandola – University of Padua Identification of key plant-host volatiles affecting behavior of brown marmorated stink bug Halyomorpha halys (Hemiptera: Pentatomidae) on commercial hazelnuts
- 12:15 R Giovanni Jesu University of Naples Smoke Waters affect behavior in the olive fruit fly *Bactrocera oleae*
- 12:30 R <u>Lucrezia Giovannini -</u> CREA Competition among the egg parasitoids of the invasive stink bug *Halyomorpha halys*
- 12:45 R <u>Mathieu Leclerc</u> CNRS / University of Tours The thermal ecology of a pollinator-thermogenic plant relationship: integrating physics, chemistry and thermal biology in climate change studies
- 13:00 Lunch

Senior scientist lecture

14:30 <u>Samantha Cook – Rothamsted Research</u> Conservation biocontrol in arable ecosystems: can we believe in beneficials?

#### **Oral Presentations**

**Chair: Lucas Sire** 

- 15:00 R <u>Matteo Marchioro</u> University of Padua Evaluation of the dispersal capacity and colonization-risk of the Walnut Twig Beetle, *Pityophthorus juglandis*, in North-east Italy
- 15:15 R<u>Alberto Mele</u> University of Padua Hyperparasitism of *Acroclisoides sinicus* (Huang and Liao) (Hymenoptera: Pteromalidae) on two Biological Control Agents of *Halyomorpha halys*
- 15:30 <u>**R**</u> Davide Nardi University of Padua Landscape predisposing factors to bark beetle attacks after a severe heat wave
- 15:45 <u>R Bianca Orrù</u> University of Turin New insights on symbiotic control of *Halyomorpha halys* and other Pentatomoidea
- 16:00 R <u>Giacomo Ortis</u>– University of Padua Can temperature affect diapause patterns and survival of the outbreak species *Barbitistes vicetinus* (Orthoptera: Tettigoniidae)?







#### 16:15 Break

- 16:45 R<u>Nicola Bodino</u>– CNR-IPSP Matter of time: temporal dynamics of the transmission of *Xylella fastidiosa* by *Philaenus spumarius* to olive plants
- 17:00 <u>R</u> Louis Pailler CNRS/ University of Tours Vibratory behaviours are modulated by social cues in a subterranean termite species
- 17:15 S Carlotta Savio INRAE Impact of probiotic bacteria on *Tenebrio molitor* fitness, gut microbial composition and susceptibility to *Bacillus thuringiensis* serovar *tenebrionis* and *Metarhizium brunneum* infections
- 17:20 R Emiliano Pioltelli– University of Milano-Bicocca Effect of urbanization and environmental stressors on the intraspecific variation of flight functional traits in two bumblebee species
- 17:35 R <u>Nicola Tommasi</u>– University of Milano-Bicocca Investigating The Impact of Anthropogenic Stressors on Pollinator Insects Across Different Continents
- 17:50 S <u>Ilaria Laterza</u>– University of Bari Aldo Moro Halyomorpha halys in newly invaded areas: habitat preference and landscape factors shaping pest occurrence.
- 17:55 S <u>Giulia Magoga</u>– University of Milan A reference database of COI sequences for insect identification through DNA-Metabarcoding







#### Friday 19 November 2021

#### **Senior scientist lecture**

09:00 <u>Davide Rassati– University of Padova</u> Invasions by wood-boring ambrosia beetles: patterns and mechanisms

#### **Oral Presentations**

#### **Chair: Gennaro Volpe**

- 09:30 R Matteo Ripamonti– CNR Scaphoideus titanus feeding behavior and fitness are influenced by different grapevine cultivars
- 09:45 S <u>Elia Russo</u>– University of Naples Host-parasitoid developmental interactions are modulated by a venom component of *Aphidius ervi* (Hymenoptera, Braconidae)
- 09:50 <u>R Giacomo Santoiemma</u> University of Padua Impact of the invasion of *Popillia japonica* on the community of soil entomopathogenic nematodes and native white grubs in Italy
- 10:05 R Lucas Sire CNRS / University of Tours DNA metabarcoding of passive trap collection media for forest insect biomonitoring
- 10:20 <u>R</u> <u>Giorgio Sperandio</u> University of Brescia Modelling the risk of establishment and potential impacts of the fall armyworm (*Spodoptera frugiperda*) in Europe
- 10:35 R Simona Maria Tortorici University of Catania Multilevel analyses on pest primed and not-primed solanaceous plants
- 10:50 Break
- 11:20 S <u>Micaela Triunfo</u> University of Basilicata Extraction and characterization of chitin from different developmental stages of *Hermetia illucens*
- 11:25 S Gennaro Volpe University of Naples RNAi-mediated silencing of an immune gene in *Spodoptera littoralis* (Lepidoptera, Noctuidae) alters its embryonic development
- 11:30 S Ilaria D'Isita– University of Foggia Susceptibility of ancient and modern wheat varieties to the lesser grain borer, *Rhyzopertha* dominica (F.)
- 11:35 R Dona Kireta The University of Adelaide/University of Turin Does revegetation quality matter to Australian native bees?
- 11:50 <u>R</u> Sofía Victoria Prieto UNITo Study of the gut microbiota in *Nezara viridula* and other pentatomids for symbiotic control
- 12:05 Discussion on PhD education and future careers







12:50 Meeting closure







# REGULAR PRESENTATIONS ABSTRACTS







# *Trichoderma harzianum* strain T22 increases egg parasitoids attraction toward tomato plants infested by stink bugs

#### T. Alinc

#### University of Palermo - Department of Agricultural, Food and Forest Sciences, Italy

Plants respond to insect herbivory attacks by emitting volatile organic compounds which can be exploited by the natural enemies of attacking herbivores to locate their host (i.e., indirect plant defenses). While a number of studies have explored the interactions among plant, insect and natural enemies in tritrophic systems so far, there has recently been increased emphasis in multitrophic interactions that beneficial soil microbes can also enhance plant indirect defenses through the activation of systemic responses and the attraction of predators and parasitoids. Within this framework, an objective of this study was to investigate whether beneficial soil microbes affect indirect response of plants against stink bugs by recruiting egg parasitoids which are considered as main natural enemies of these herbivores. For this purpose, a multitrophic model system consisting of tomato plant, Solanum lycopersicum cv Dwarf San Marzano, southern green stink bug, Nezara viridula, an egg parasitoid of stink bugs, Trissolcus basalis and plant growth-promoting fungi, Trichoderma harzianum strain T22 was used. The results showed that T. basalis females were significantly attracted to volatiles emitted by tomato plants inoculated with T. harzianum and then infested by N. viridula females compared to those released by infested and uninoculated plants. Furthermore, the inoculation of T. harzianum alone does not modify the response of the egg parasitoid, in fact the inoculated plants did not stimulate a significant response when they were not infested by N. viridula females. The outcomes were discussed in the context of indirect plant defenses against stink bugs by considering the impact of T. harzianum on the attraction of egg parasitoids.







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

#### A. Arpellino

#### University of Turin - Department of Agricultural, Forest and Food Sciences, Italy

Pest management in vineyard requires new eco-friendly control methods. An emerging strategy, namely symbiotic control, takes advantage of insect-microorganism interactions, since manipulating these interactions has an extreme impact on insect life cycle. Grapevine pest insects belong to various orders. In North-West Italy, a major threat is Scaphoideus titanus, the main vector of the phytoplasmal agents of Flavescence dorée (FDp), while an emergent pest is Drosophila suzukii. The aim of this reserch is to provide a contribution to the knowledge regarding the microbial interactions in the pest species S. titanus and D. suzukii, to support the development of sustainable control strategies. Planned activities include in the study of the biological performance of S. titanus in different grapevine cultivars and other Vitis spp., in relation to the acquisition of different resident microbial communities. Moreover, investigations will be conducted on the symbiotic control of FDp, using a symbiont strain in the genus Asaia, which reduces phytoplasma acquisition by vectors. Finally, a survey of olfactive preference toward different microbial agents in D. suzukii will be performed, to develop pre-harvest control protocols. To implement the first objective, longevity will be compared on different S. titanus populations, established on: wild Vitis spp. V. vinifera cultivars and V. labrusca. Furthermore, DNA samples from each population will be destined to throughput sequencing of 16S rRNA gene. To achieve the second goal, plant-mediated acquisition and horizontal transmission of Asaia will be assessed. To complete the third objective, different preference bioassays will be performed using different yeast and bacterial strains.







# Matter of time: temporal dynamics of the transmission of *Xylella fastidiosa* by *Philaenus spumarius* to olive plants

<u>N Bodino<sup>1</sup></u>, V Cavalieri<sup>2</sup>, RPP Almeida<sup>3</sup>, M Saponari<sup>2</sup>, C Dongiovanni<sup>4</sup>, D Bosco<sup>1,5</sup>

<sup>1</sup>CNR - Istituto per la Protezione Sostenibile delle Piante, Torino, Italy; <sup>2</sup>CNR–Istituto per la Protezione Sostenibile delle Piante, Bari, Italy; <sup>3</sup>University of California - Department of Environmental Science, Policy and Management, USA; <sup>4</sup>CRSFA - Centro di Ricerca, Sperimentazione e Formazione in Agricoltura Basile Caramia, Italy; <sup>5</sup>Università di Torino – DISAFA, Italy.

The spittlebug Philaenus spumarius L. is the predominant vector of the exotic bacterium Xylella fastidiosa Wells (Xf) to olive trees in Apulia Region (Italy). The biology of transmission of Xf in the Apulian pathosystem is still poorly known and, consequently, the epidemiology of Xf-associated diseases cannot be properly understood. Here we investigated 1) the variation through time of bacterial multiplication and transmission efficiency by vector, and 2) the spread rate of Xf among olive plants in different seasons and climatic conditions. Xf multiplication in vector and transmission efficiency by *P. spumarius* adults were tested up to 72 days post-acquisition in four separate assays carried out in controlled conditions. Spread rate of Xf infection among olive seedling was tested by microcosm assays to assess the influence of inoculum duration, climatic conditions (semi-field vs controlled), and season. The results show that i) P. spumarius is a competent Xf vector to olive throughout its adult life ii) bacterial load in the vector foregut reaches a plateau after 2-3 weeks from acquisition iii) different climatic conditions and period of the year may result in significant differences in transmission rates and iv) differential survival of vectors — influenced by insect age, season and climatic conditions — may affect the spread of Xf in olive plants. The transmission parameter estimates obtained in this study can be used in modelling the pathogen spread, by explicitly incorporating the effect of insect vectors, with the aim of designing effective control and prevention measures.







### Characterization of the immune response of *Hermetia illucens* larvae to bacteria

<sup>1</sup>D. Bruno, <sup>1</sup>A. Montali, <sup>2</sup>M. Casartelli, <sup>1</sup>G. Tettamanti

<sup>1</sup>University of Insubria - Department of Biotechnology and Life Sciences, Italy; <sup>2</sup>University of Milano - Department of Biosciences, Italy

The larvae of the black soldier fly (BSF), *Hermetia illucens*, can be reared on a wide range of decaying organic substrates rich of potential pathogens which can challenge the health status and growth of the insects. Despite the great interest on the use of these larvae for the bioconversion of organic waste, knowledge about their immune system is still fragmentary.

In the present study, we investigated the main immune mechanisms triggered by the larvae to counteract the infection of *Escherichia coli* and *Micrococcus luteus*, two bacteria which are widely represented in organic wastes processed by BSF. In detail, hemocyte phagocytic activity, and the activity of key components of the humoral response, as phenoloxidase, lysozyme, and antimicrobial peptides, were evaluated over time.

Our data demonstrate different kinetics of the cellular and humoral responses: in particular, phagocytosis was rapidly activated after the immune challenge while the antimicrobial and lysozyme activity intervened later and continued for a longer time. In contrast, the phenoloxidase system was inhibited in infected larvae. Moreover, although a complete elimination of both bacteria from the larval body was obtained within few hours after their administration to the larvae, Grampositive bacteria persisted in the hemolymph longer than Gram-negative.

This study represents the first detailed characterization of the immune response of BSF larvae and this information is a prerequisite to improve the quality of the larvae during mass rearing through modulation of the immune response.







### Impact of the diet on the expression of antimicrobial peptide genes in *Tenebrio molitor* (Coleoptera: Tenebrionidae)

### V. Candian, M. Dho, R. Tedeschi

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

In mass rearing systems it is essential to preserve the insect immune system. The impact of 4 different diets arising from local agro-industrial by-products was assessed in Tenebrio molitor (Coleoptera: Tenebrionidae) larvae. The insects were reared on: i) wheat bran (used as control), ii) wheat bran+brewer's grain, iii) wheat bran+roasted cocoa husks, iv) wheat bran+roasted hazelnut husks. Weight gain and expression level of genes encoding for two antimicrobial peptides (coleoptericin-1 and cecropin-2) were evaluated. Furthermore, the inhibitory activity of the hemolymph extracted from the larvae was tested against *Escherichia coli* DH5a and *Micrococcus* yunnanensis HI55 in diffusion assays in solid medium. The brewer's grain-added diet reduced the larval development time and increased larval weight while the other diets caused a weight decrease. An upregulation of both antimicrobial peptides encoding genes was recorded only in larvae reared on the brewer's grain-added diet. The hemolymph from all the diets reduced considerably the size and the number of *E. coli* DH5 $\alpha$  colonies with a stronger effect with the one arising from the brewer's grain-added diet. No *M. yunnanensis* HI55 colonies developed on the area of the plates with the hemolymph from all diets. According to the obtained results, the brewer's grain-added diet allowed a faster larval development, a higher larval weight and positively affected the immune system opening new perspective in the modulation of insect tolerance towards entomopathogens with the diet.







# Functional traits of plants and pollinators explain resource overlap between honeybees and wild pollinators

<u>A, Cappellari</u><sup>1</sup>, M, Mei<sup>2</sup>, D, Paniccia<sup>3</sup>, P, Cerretti<sup>2</sup>, L, Marini<sup>1</sup>

<sup>1</sup>University of Padua - Department of Agronomy, Food, Natural resources, Animals and Environment, Italy; <sup>2</sup>Sapienza University of Rome - Department of Biology and Biotechnology "Charles Darwin", Italy; <sup>3</sup>Via Colle 13, Frosinone, Italy

Managed and wild pollinators often cohabit in both agricultural and natural ecosystems. The western honeybee, *Apis mellifera* Linnaeus, is the most widespread managed pollinator species. Due to its density and foraging behaviour, it can influence the foraging activity of wild pollinators, but the strength and direction of this effect are often context-dependent.

Here, we observed plant–pollinator interactions in 51 grasslands, and we measured functional traits of both plants and pollinators. Using multi-model inference analyses, we explored the effects of honeybee abundance, temperature, plant functional diversity, and trait similarity between wild pollinators and the honeybee on the resource overlap between wild pollinators and the honeybee. Resource overlap decreased with increasing honeybee abundance only in plant communities with high functional diversity, suggesting a potential diet shift of wild pollinators in areas with a high diversity of flower morphologies. Moreover, resource overlap increased with increasing trait similarity between wild pollinators and the honeybee. In particular, central-place foragers of family Apidae with proboscis length similar to the honeybee (~5 mm) exhibited a higher resource overlap with the latter. Our results underline the importance of promoting functional diversity of plant communities to support wild pollinators in areas with a high density of honeybee hives. Furthermore, greater attention should be paid to areas where pollinators possess functional traits similar to the honeybee, as they are expected to be more prone to potential competition with this species.







# The secret of my success! Visual learning abilities in mosquito larvae (Aedes aegypti) are not impaired neither by light deprivation nor by turbidity.

Martin Dessart, Tristan Robineau, Claudio Lazzari, Fernando Guerrieri

Institut de Recherche sur la Biologie de l'Insecte - UMR 7261 – CNRS - Université de Tours - 37200 Tours, France

Living organisms evolve in complex and diverse environments and are therefore submitted to multiple sensory stimulation. In some cases, an individual's behavior, development, and sensory skills can be modified to adapt to the environmental conditions. In this study, we focused on luminosity and turbidity impact on mosquito larvae *Aedes aegypti* sensory and cognitive abilities. We used habituation (a non-associative form of learning) protocol, in which a square cardboard was presented at regular intervals (5 min) under a light spot, projecting a shadow (visual stimulus) on a single larva.

We trained four groups of larvae reared under different luminosity and turbidity conditions: (1) larvae reared in clear water, 12h light/12h dark photoperiod; (2) larvae reared in turbid water, 12h light/12h dark photoperiod; (3) larvae reared in clear water, no light; (4) larvae reared in turbid water, no light. Statistical analyses showed that larvae could learn under each condition, and that rearing conditions did not influence neither the level nor the dynamics of learning.

Physical conditions of the environment during larval development did not modify neither visual abilities nor learning skills in larvae, which would allow individuals to take profit of their visual and cognitive abilities during the imaginal life. Being able to develop under extremely different environmental condition explains some of the worldwide spreading success of *A. aegypti*.







### Plant-pollinator interactions: a study along spatial-temporal gradient in different land management types within the Stelvio National Park

<u>E. Eustacchio</u><sup>1,4</sup>, M. Bonelli<sup>1,4</sup>, E. Legoratti<sup>1</sup>, A. Minici<sup>1</sup>, A. Zanzottera<sup>1</sup>, L. Pedrotti<sup>2</sup>, M. Casartelli<sup>1,3</sup>, M. Gobbi<sup>4</sup>, M. Caccianiga<sup>1,3</sup>

<sup>1</sup>University of Milano – Department of Biosciences, Milano, Italy; <sup>2</sup>Stelvio National Park, Bormio, Italy <sup>3</sup>University of Napoli Federico II - <sup>3</sup>BAT Center-Interuniversity Center for Studies on Bioinspired Agro-Environmental Technology, Napoli, Italy <sup>4</sup>MUSE-Science Museum – Section of Invertebrate Zoology and Hydrobiology, Trento, Italy

Plants and arthropods interact with each other creating complex networks, such as those related to pollination, that are still little known, especially in high-mountain ecosystems. Here, these interactions are expected to be strongly modified by climate change and habitat management. We investigated how the flower-visitor network change along an altitudinal gradient, during the plant growing season and among different land management types (orchards, hay-meadows, pastures, high-altitude grasslands) in order to identify the focal plant and arthropod species responsible of network structure and stability. We selected 14 sampling plots located along the Martello valley (Bolzano, Stelvio National Park, Italian Alps). We focused on flower-visiting arthropods of three main plant groups corresponding to different floral morphology (Asteraceae, Fabaceae, Ranunculaceae/Rosaceae) along altitudinal (from 900 to 2700 m asl) and seasonal (from May to August 2021) range. We used an integrated approach involving manual sampling and simultaneous in-field video observations. The first approach allows the identification of flower visitors at low taxonomic level and the analysis of their pollen load. The second approach allows to describe arthropod behaviour on flowers in order to hypothesize their functional roles. We sampled more than 1900 specimens belonging to 11 orders, of which Diptera, Thysanoptera, Hymenoptera and Coleoptera are the most represented. Flower visitor community change along the altitudinal gradient and during the plant growing season as well as among different land management types. We are confident that this study will be able to provide novel information about plant-arthropod interactions in climate and human driven areas of the Alps.







# Starting with the basics: biology and European distribution of the fig weevil Aclees taiwanensis

<u>P. Farina</u><sup>1</sup>, G. Mazza<sup>2</sup>, C. Benvenuti<sup>2</sup>, I. Cutino<sup>2</sup>, P. Giannotti<sup>1</sup>, B. Conti<sup>1</sup>, S. Bedini<sup>1</sup>, E. Gargani<sup>2</sup>

<sup>1</sup>University of Pisa - Department of Agriculture, Food and Environment; <sup>2</sup>CREA-DC Florence -Research Centre for Plant Protection and Certification

The fig weevil, Aclees taiwanensis Kôno, 1933 (Coleoptera: Curculionidae), is an alien species native to Taiwan, detected for the first time in Europe in 2005 (Tuscany, Italy). It feeds and develops only on plants belonging to the genus Ficus, threatening the Mediterranean production of the common fig Ficus carica. Adults eat buds, leaves, and ripening infructescences, and the xylophagous larvae dig feeding galleries in the wood of trunk and surface roots, slowly bringing the attacked trees to death. Despite its harmfulness, data about the biology, physiology, and distribution of this pest were scarce. Therefore, this work aimed to verify, under laboratory conditions, the duration of all the A. taiwanensis developmental stages (embryonic, larval, and pupal), the oviposition modality and the mean number of eggs laid by each female, the eggs' hatching percentage, and the instars and adults' morphometric measures. Also, we evaluated the susceptibility of three Ficus ornamental species of economic importance (F. benjamina, F. macrocarpa, and F. pandurata). Moving to the field, we registered the presence peaks during 2019 and 2020 using mechanical traps wrapped around the fig plants to catch the adults. In the end, surveying the social networks and naturalistic forums through a citizen science approach, we verified the European distribution of A. taiwanensis that currently includes seven northern and central Italian regions and one French region. The new information provided could help to develop innovative and focused management strategies to avoid the further spread and menace of the fig weevil.







### Quantifying the effects of insect pollination on crop quality

### <u>E Gazzea<sup>1</sup></u>, L Marini<sup>1</sup>

# <sup>1</sup> University of Padova – Department of Agronomy, Food, Natural resources, Animals and Environment, Italy

Insect pollination directly benefits the production of a wide share of crops consumed by humans, significantly contributing to global food security. Pollinator-dependent crops often have high market prices, and their production and marketability have an enormous estimated economic value. Despite the importance of insect pollination in affecting global crop and related markets, the contribution of insect pollination to crop quality and marketability has not yet been quantified. We focus on investigating the effects of the pollination service provided by insects on crop quality by performing a global multi-level meta-analysis. We synthetised 129 articles studying the effects of pollination service, and extracted 970 effects sizes about more than 30 different crops. Preliminary results show that insect pollination contributes to an overall increase of crop quality by 31% (confidence interval: 23%-39%). Results suggest that seed germination and external quality traits, such as crop size, shape and commercial grade significantly improve with pollination. On the other hand, nutritional and biochemical composition do not seem to be affected. Experimental scale at which study is conducted, rather than pollinator species and growing environment, significantly affects the contribution of pollination to crop quality. Understanding the contribution of insect pollination to crop quality highlights opportunities to manage pollinators to ensure crop quality and improve their marketability.







### Herbivory: a hidden player in plant responses to global change

<u>C. Geppert<sup>1</sup></u>, F. Boscutti<sup>2</sup>, L. Marini<sup>1</sup>

<sup>1</sup> DAFNAE - University of Padova, Viale dell'Università 16, 35020 Legnaro, Padova, Italy; <sup>2</sup> Di4A-Department of Agricultural, Food, Environmental and Animal Sciences, University of Udine, 33100, Udine, Italy

Understanding the role of biotic interactions in altering biodiversity responses to global change is a fundamental challenge of ecology and conservation biology. Here, we investigated the effects of arthropod herbivory, climate change, and human disturbance on plant establishment dynamics using two different experiments. First, we performed a semi-field experiment, where we disturbed the soil by tilling it up to 20 cm and we manipulated drought, and nitrogen fertilization. Moreover, we controlled for the effect of arthropod herbivores by installing exclusion cages. We found that herbivores increased the diversity of the plant communities established after disturbing the soil, while they did not affect plant biomass. Second, we addressed similar questions by replicating the experiment under natural conditions in the Eastern Italian Alps along steep elevational gradients. Results from this field experiment showed that natural arthropod herbivory pressure might amplify the negative effects of soil disturbance on resident native plant species and favour exotics. In addition, warm temperatures and disturbance promoted exotic success over natives, suggesting that global change will probably favour the further spread of plant invasions in mountains. Results from both experiments stressed the important role played by arthropod herbivores in plant dynamics and pointed at the urgent need to incorporate herbivory effects when predicting future plant assemblages under global change.







# Competition among the egg parasitoids of the invasive stink bug Halyomorpha halys

#### Lucrezia Giovannini

*Council for Agricultural Research – Research Centre for Plant Protection and Certification, University of Florence – Department of Biology, Florence* 

Halyomorpha halys is an invasive stink bug, native to east Asia, considered a severe agricultural pest of worldwide importance. Chemical control resulted mostly ineffective and the natural enemies, in particular the egg parasitoids, showed interesting perspectives with a high percentage of egg parasitization. For this reason, biological control programs seem to be the most promising method for the long-term management of this invasive bug. In Italy, different species of *H. halys* egg parasitoids are present but the most common ones are the two exotic *Trissolcus japonicus* and *T. mitsukurii*, and the indigenous *Anastatus bifasciatus*. Currently, the two species of *Trissolcus* are considered the main candidates of *H. halys* biocontrol. Recently, an adventive population of an exotic hyperparasitoid parasitoid species, *Acroclisoides sinicus*, was frequently found associated mainly with *H. halys* and its parasitoid guild. The sympatry in Italy of such species makes it important to assess the inter-and intraspecific interactions to understand if and how they can affect the biological control programs. Moreover, the interactions among parasitoids can also negatively affect, the structure and ecology of the communities. With this aim, different experimental designs based on laboratory tests, through multidisciplinary approaches, were performed.







### Heterogeneous venom gland composition in females of the invasive Asian hornet Vespa velutina nigrithorax

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In social insects, chemical signals are essential for communication systems. These signals are implicated in social organisation and can be carried out by pheromones. These volatile compounds play an essential role in interspecific interactions and thus, regulate colony behaviours. To control the spread of invasive species worldwide, synthetic or natural insect pheromones can be used for pest control. Different management's techniques have included lures, baits, and traps. Due to their invasive success and ecological dominance, some asian hornet species are considered as important economic and ecological pests. Native to Southeast Asia, the yellow-legged hornet Vespa velutina nigrithorax was accidentally introduced in France, around 2004, and spread rapidly throughout Europe. This species is of public concern due to damages caused to insects, specifically to honey bee populations. This study aimed to clarify the composition of the venom gland, the organ that produces the alarm pheromone, among Vespa velutina nigrithorax females: queens, foundresses, gynes, and workers. We performed gas chromatography-mass spectrometry to identify compounds on the alarm pheromone. A total of 10 new compounds were identified among the 26 compounds in the venom gland (chain lengths:  $C_8$  to  $C_{12}$ ). The alarm pheromone composition differed quantitatively between female individuals tested, despite the inbreeding situation of the European populations. These results bring new insights on the pest management strategies against the invasive Asian hornet Vespa velutina nigrithorax, with the possibility to develop highly specific pheromone-based traps.







### Smoke Waters affect behavior in the olive fruit fly *Bactroera oleae*

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The smoke-waters (SWs) are secondary products obtained during the pyrolytic production of biochar. The study aimed to record the effects of the presence of SWs in a closed and open environment towards the behavior of the adults of *Bactrocera oleae*. The two more promising between 12 original SWs previously investigated were deeply evaluated in these assays. The *in vitro* assays were conducted in optimal condition of temperature relative humidity and light using a dynamic airflow glass Y-tube olfactometer. The positive controls were carried out using fresh green olives. SWs have shown attractive, repulsive or indecision effects depending on the starting raw materials they were produced of, and their dilution. The field assays have been carried out for two subsequent years in different pedoclimatic conditions (South and Central Italy), using kaolin clay and isopropyl-myristate as additional control due to their known repulsiveness. The SWs application's efficacy was computed based on the number of catches of flying adults in chemochromotropic traps and on the level of infestation of the drupes at the harvest. Open-air trials, on both fields, shown expected attractive and repulsive effects of the SWs, consistently to those obtained in optimal and controlled environment during the olfactometry tests. Such results encourage further deepening studies to find promising new IPM strategies against *B. oleae*.







### Does revegetation quality matter to Australian native bees?

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Habitat loss is causing declines in native bees and reducing pollination services in natural and agricultural systems. Flower plantings have been found to improve conditions for bees in agricultural settings, largely in Europe and America. However, these approaches may not address wild bees outside of agricultural systems, and cannot easily be applied to places such as Australia with vastly different climates and environments. Perennial revegetation that mimics native vegetation can be used to reverse habitat loss and the associated bee declines, and growing efforts and resources are being funnelled into revegetation projects. However, there remains a lack of understanding around the quality of revegetation outcomes. In consideration of this gap, we undertook floral and bee surveys in revegetated landscapes in South Australia. We found that flower and bee diversity were correlated, and there was higher bee diversity in better quality vegetation types. Further, *Eucalyptus* trees had a disproportionate effect compared with other plants on the abundance and diversity of bees. These results indicate that better quality revegetation can potentially restore native bee diversity, and is a step in the right direction towards reversing habitat loss.







# The thermal ecology of a pollinator-thermogenic plant relationship: integrating physics, chemistry and thermal biology in climate change studies

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The pollinator insects are influenced by climatic conditions, and the temperature if flowers can deviate from air temperature. Little is known on the impact of flower temperature patterns on pollinators. Several plant species display the ability to produce metabolic heat (thermogenesis) that make them even warmer than flowers warmed by the sun. We focused on the interaction between thermogenic flowers (Arum maculatum A.italicum), and their pollinators (flies Psychoda). These flowers rely on a deceptive method to attract pollinators, by emitting odors resembling the odor of their breeding site. They trap pollinators in the floral chamber imposing flies the flower temperature. The aim of this study was to discriminate two sympatric species of Arums using the temperature of flower and odor patterns. We quantified the thermal tolerance of two species of Psychoda sp. by determining their maximum Critical Temperature. We looked the impact of acclimatization at a temperature that measured in the flower and the temperature rise dynamics on the CTmax. We found that two plant species differed in their flower temperature and odor profiles. The two species of *Psychoda* had a relatively low thermal tolerance with little plasticity relative to acclimatization and the rate of temperature rise. Taken together, these results suggest that the flower temperature can be risky for the flies in at least one of the two species of arum. In the future, we will detail the impact of the flower microclimate on the thermal biology of these pollinators. This system may be particularly threatened by climate warming.







### Evaluation of the dispersal capacity and colonization-risk of the Walnut Twig Beetle, *Pityophthorus juglandis*, in North-east Italy

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The Walnut Twig Beetle (WTB), *Pityophthorus juglandis* Blackman, is a small bark beetle native to South-Western USA and Mexico recorded for the first time in Europe in Northeastern Italy in 2013. *P. juglandis* attacks walnut (*Juglans* spp.) and wingnut trees (*Pterocarya* spp.) and is the vector of *Geosmithia morbida* Kolarík et al., a pathogen causing the Thousand Cankers Disease (TCD). WTB and TCD represent a serious threat for walnut orchards in Europe. We used spatio-temporal data of the WTB infestations recorded from an 8-year long (2013-2020) survey conducted in 106 walnut orchards in the Veneto Region, to develop a model in order to analyse: (i) the effective dispersal capacity of the insect, (ii) the factors affecting its dispersal, and (iii) the colonization-risk of healthy walnut orchards. A mean annual dispersal of 9.4 km was observed, with peaks of about 40 km. WTB dispersal is affected by distance of suitable hosts from the nearest infested site, number of walnut orchards in the surroundings (both infested and healthy), the orchard size, and the walnut species in the orchard. Using our model, it was also possible to calculate the colonization-risk of a specific walnut orchard according to its characteristics and we found that the current containment measures, based on a 2 km buffer around infested sites, are insufficient to stop the spread of the pest.







### Hyperparasitism of *Acroclisoides sinicus* (Huang and Liao) (Hymenoptera: Pteromalidae) on two Biological Control Agents of *Halyomorpha halys*

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Halyomorpha halys (Stål) is an invasive Asian pest that causes severe losses on various crops. Trissolcus japonicus (Ashmead) and Trissolcus mitsukurii (Ashmead) are two Asian egg parasitoids already present in Europe that are considered the most effective biological control agents of *H. halys. Acroclisoides sinicus* (Huang and Liao) is a pteromalid parasitoid wasp that frequently emerged from *H. halys* egg masses collected in northern Italy. This species has been hypothesized to be a hyperparasitoid of Trissolcus spp. parasitoids. This study was carried out in laboratory conditions where host preference between T. japonicus and T. mitsukurii A. sinicus was investigated in no-choice and two-choice experiments. Olfactory responses of A. sinicus from volatiles emitted from different potential hosts were also studied. In all trials, A. sinicus showed a clear preference for parasitizing H. halys eggs previously parasitized by T. mitsukurii compared to T. japonicus. In no-choice experiments, the impact of the hyperparasitoid on T. japonicus was low, showing an exploitation rate of 4.0%, while up to a 96.2% exploitation rate was observed on T. mitsukurii. Acroclisoides sinicus was also attracted by volatiles emitted by egg masses parasitized by T. mitsukurii, while no response was observed to egg masses parasitized by *T. japonicus* or not parasitized. Therefore, according to the results obtained, A. sinicus could limit the population development of *T. mitsukurii*, while lesser effects are expected on *T. japonicus*.







### Landscape predisposing factors to bark beetle attacks after a severe heat wave

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European spruce bark beetle (Ips typographus L.) is one of the most important pests of European coniferous forests. In the last decades, climate change has caused an increased outbreak propensity causing severe economic damages to spruce forests. Indeed, warmer temperatures usually lead to a faster insect development, but warmer and drier climate also weakens spruce tree defence. Several time-series analyses have demonstrated that after extreme summer drought bark beetle disturbances increased at the regional scale. However, it is still not clear whether different stands across the same region have the same probability of being attacked depending on local conditions such as topography, microclimate, forest quality, and landscape structure. Using a unique dataset of georeferenced bark beetle infestation data, we tested whether the local conditions of forest stands and landscape structure modify the local occurrence of bark beetle attacks after a severe summer heat wave that hit central European forests in 2017 and 2018. We showed that local bark beetle outbreak occurrence depended on both topography and historical climate of the site. Our findings are consistent with the growth differentiation balance hypothesis predicting that trees growing under chronic drier conditions tend to be more resistant against biotic disturbances. In conclusion, our study elucidates how spatial variation in growing tree conditions can explain local bark beetle attacks after a severe heat wave at large spatial scales. Our results suggest that the most productive stands are likely those more exposed to bark-beetle induced mortality.







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

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Obligate bacterial symbioses are common in the order Hemiptera; in Pentatomoidea, bacteria are often related to the genus Pantoea. These symbionts are orally acquired by newborns from maternal secretions smeared on the egg surface, and they are essential for growth, development, and survival of the insect. The manipulation of symbiont transmission, through surface-sterilization of eggs, is regarded as a novel way for the creation of control programs against pentatomid pests. The most studied species is Halyomorpha halys, one of the major damaging agricultural pests, due to its invasive potential and high polyphagy. The application of anti-symbiont products on egg masses resulted in high nymphal mortality in the neonates, and the antibacterial accessory effect displayed by a biocomplex of micronutrient fertilizers (Dentamet SYM®) has been recently included in *H. halys* management measures. Here, the anti-symbiont activity of several active substances was evaluated under laboratory conditions on H. halys egg masses, by assessing mortality rate and fitness of nymphs emerging from treated eggs and untreated control. Molecular diagnosis for the symbiont Pantoea carbekii was done to evaluate if effects are attributable to loss or reduction of symbiont infection. Furthermore, studies were carried out on the interaction between these treatments and the activity of egg parasitoids. Afterwards, field surveys were conducted to assess the effectiveness of symbiotic control in different agroecosystems. Finally, the primary symbiont was identified in another pest in the Pentatomoidea, i.e. Eurygaster maura; a field assessment of the effect of symbiotic control on the populations of this insect was performed in wheat plots.







### Can temperature affect diapause patterns and survival of the outbreak species Barbitistes vicetinus (Orthoptera: Tettigoniidae)?

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Barbitistes vicetinus Galvagni & Fontana was described in 1993 as an endemic bush-cricket of northeast Italy and was considered rare until 2008, when repeated outbreaks occurred in ever-increasing surface areas. This species has become a forest pest, causing severe damages to forests and crops. Despite the occurrence of a multi-year diapause of eggs was observed, there is a lack of knowledge about how temperatures could influence diapause and phenological activity. In this study, we explored the effect of field temperatures on diapause of B. vicetinus, focusing on how summer temperatures can trigger embryos to develop in the same year of oviposition and how winter temperatures affect survival. The effect of temperature on diapause and overwinter survival was also tested in the laboratory, exposing eggs to constant temperatures. B. vicetinus showed a strong thermal sensitivity, being capable to develop in large numbers within a specific thermal range. Two years field observations showed that the proportion of embryos that completed development at the end of summer ranged from zero to nearly 90%, suggesting that more than half embryos of B. vicetinus can develop when mean temperatures are above 20°C. A substantial shift in rates of development from 20 to nearly 80% occurred in a thermal range of about 1°C. However, treatment of eggs at constant temperatures allowed lower number of embryos to develop during summer. Overall, overwinter survival was high (90%) for eggs exposed to natural temperatures but eggs incubated at constant temperatures reported 70% of survival and a prolonged hatching period.







# Vibratory behaviours are modulated by social cues in a subterranean termite species

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Reproductives regulate social interactions among colonies driving the colony's life. These interactions are mainly driven by chemicals, but vibratory communication has also been shown to be efficient, especially for subterranean species. In termites, different vibratory behaviours are involved in the regulation of social interactions. Among them the vibrations produced during bodyshaking events remain poorly explored despite their implication in presence of reproductives. Historically considered as an alarm signal, it seems to be a good candidate to transmit other type of information. We therefore investigated the structure and the occurrence of the substrate borne vibrations produced during body-shaking, in presence of reproductives and/or in presence of a stress event (flashlight), in the subterranean termite Reticulitermes flavipes. Our data show that body-shaking behaviour produces vibrations transmitted through the substrate with different complex frequencies according to the different treatments. We described four types of bodyshaking which were influenced by the presence of the reproductives but not the flashlight. Finally, the number of body-shaking is increased by the presence of the reproductives and only transitory over a short period of time for the flashlight. These results show that vibratory cues are complex in termites which might encode for a plurality of social cues. The potential roles of vibratory signals on social organisation is surprisingly understudied and call for more investigations.







# Effect of urbanization and environmental stressors on the intraspecific variation of flight functional traits in two bumblebee species

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Landscape anthropization represents the principal cause of natural habitats loss and fragmentation with intensive agriculture and urbanization as the major drivers of land-use changes. The impact of these novel anthropogenic habitats on pollinator insects is still poorly understood. Here we investigated how urbanization affects the functional traits of workers in two bumblebee species (Bombus terrestris and B. pascuorum) sampled in 37 sites in the metropolitan area of Milan. The role of multiple environmental conditions and of landscape composition and configuration on morphological traits related to flight performance (i.e., body size, wing shape and size fluctuating asymmetry) was assessed. Our results describes a idiosyncratic response of the two species, with higher temperatures in cities associated with decreasing body size in *B. pascuorum* and increasing fluctuating asymmetry of wing size in B. terrestris. Moreover, flower abundance emerged as a positive predictor of *B. terrestris* workers body size while showing a negative correlation with wing size asymmetry of *B. pascuorum*. These results highlight how environmental variables related to urbanization may impact the functional traits of pollinator insects, with possible consequences on the pollination service they provide. Overall, this study found species-specific variation patterns in syntopic taxa, expanding our understanding about the effects of anthropic disturbance in shaping relevant functional traits of pollinator model species.

Such investigations could inform mitigation policy aimed at ensuring suitable strategies for urban green management and pollinators communities safeguarding.







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

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The family Pentatomidae includes many polyphagous species that are important pests. Pentatomids harbour an obligated symbiont in the gut V4 ventricle which is vertically transmitted via egg surface contamination by the females. Egg surface sterilization disrupts nymphal infection with the symbiont. Symbiotic control programs have been developed against Halyomorpha halys by preventing symbiont acquisition via egg surface sterilization, resulting in high nymphal mortality. Another pentatomid pest is the southern green stink bug Nezara viridula (SGSB). In some cases, eliminating the symbiont shows severe nymphal mortality for SGSB, while in others it causes few fitness defects. This variability may be due to the host/symbiont genetic background. The main aims of this research are to describe the genetic variability of SGSB and symbiont haplotypes in different zoogeographic regions and to assess the host/symbiont interaction. To that end, sampling campaigns were conducted to obtain distinct SGSB populations. Some specimens were dissected, and DNA from the V4 ventricle was amplified by PCR. Amplicons were sequenced and used to build a symbiont phylogenetic tree. Other individuals were reared in climatic chambers, and egg masses were treated with a micronutrient fertilizer, approved for symbiotic control of SGSB, to avoid symbiont acquisition. Symbiont absence was confirmed by RNA extraction and PCR. Preliminary results showed high mortality during first instar with a considerable variability among the treated group. Future activity will include the complete characterization of SGSB's V4 microbiota through Single Molecule Real Time sequencing and the description of the variability in insect haplotypes.







# Scaphoideus titanus feeding behavior and fitness are influenced by different grapevine cultivars

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Scaphoideus titanus (Ball, 1932) is a Nearctic leafhopper, introduced in Europe during the 20<sup>th</sup> century. It is monovoltine and monophagous on Vitis spp., and efficiently transmits the Flavescence dorée phytoplasma (FDp), associated with a severe disease of grapevines (FD), which causes heavy losses to European viticulture. Previous findings suggested a possible role of plant resistance against the vector in grapevine resistance against FD. In order to explore this possibility, S. titanus feeding behavior and fitness were analyzed on three different grapevine cultivars: Barbera (very susceptible to FD), Brachetto, and Moscato (partially resistant to FD). Feeding behavior was studied with the electropenetrography (EPG) technique, while four different fitness parameters were analyzed: nymph developmental time, nymph survival, adult survival, female prolificacy. EPG results showed that S. titanus feeds longer in the phloem of Barbera, with more than 50% of the feeding phases spent in phloem-related waveforms. On the contrary, on Brachetto and Moscato, phloem-related phases were less than 20% and 11% of the total feeding time, respectively, suggesting a possible antixenosis of these two aromatic cultivars. All the different fitness parameters consistently indicated a negative effect of the two FD partially resistant cultivars on S. titanus, in particular Moscato. Nymph developmental time was longer on Moscato, while nymph survival was impaired on both Brachetto and Moscato. Adult survival was negatively affected by Moscato and partially by Brachetto, as well as female prolificacy. These results show a possible mechanism of antibiosis acting against S. titanus. Antixenosis and antibiosis could represent valuable tools in breeding for resistance against S. titanus, thus reducing the spread of FD.







# Impact of the invasion of *Popillia japonica* on the community of soil entomopathogenic nematodes and native white grubs in Italy

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Soil dwelling entomopathogenic nematodes (EPNs) are important natural enemies of insect pests of plant roots. Relationships between nematodes associated with the soil of hay meadows and grubs have been explored in the invasion area of the Japanese beetle *Popillia japonica* Newman (Coleoptera: Scarabaeidae) in northern Italy. Several species of indigenous white grubs are present in the territory and they are assumed to be associated with local EPN strains, mainly belonging to *Heterorhabditis, Steinernema* and *Oscheius* spp. The presence of *P. japonica* could affect the ecological balance between native grub species and the EPNs. To test this hypothesis, the spatial distributions of EPNs, native grub species and *P. japonica* in the laboratory under different stress conditions and all EPN strains showed capacity to exploit *P. japonica* as host. EPNs were more frequently isolated from soil samples collected in the early colonized sites. The native species of grubs did not seem to be dramatically affected by the increase of EPN density associated with the expansion of *P. japonica*.







# DNA metabarcoding of passive trap collection media for forest insect biomonitoring

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Insect decline has been increasingly reported in the past years due to global change. Large-scale biomonitoring has thus become necessary to better understand the dynamics of insect communities and to preserve their essential role in ecosystem functioning. In that sense, coupling high-throughput sequencing and DNA metabarcoding has exponentially increased our potentiality to monitor insect communities over wider geographic regions and time scales. However, biomonitoring of entomofauna using molecular tools often results in destructive DNA extraction through voucher grinding, impeding primordial morphological back-up. Here, we filter unprocessed collection medium to assess insect communities through environmental DNA metabarcoding. We demonstrate that recovered communities are different yet complementary and that insect response to environmental changes remains similar to homogenate bulk metabarcoding. We also show that insect orders—by their contrasting sclerotization ratio—, and collection medium types, are unequal in yielding metabarcoding results. Overall, we believe it as an efficient alternative for biomonitoring insect response to ecological changes while preserving insect vouchers for identification and description, especially in tropical regions were singletons or undescribed species can be very common in trap samples.







# Modelling the risk of establishment and potential impacts of the fall armyworm (Spodoptera frugiperda) in Europe

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The fall armyworm (*Spodoptera frugiperda* Smith) is an invasive pest considered a major threat to agricultural production and food security. The species feeds on more than 350 host plants, including economically valuable crops. The high migratory capacity of the species and the risks linked to the importation of infested products are causing high concerns for the introduction of the species in Europe. Additionally, transient populations might be able to survive and cause impacts outside the area of establishment. In this work, we present a first exploration of the risk associated with established and transient populations of *S. frugiperda* in Europe obtained with a physiologically-based modelling approach. The model allows to realistically represent the effects of environmental drivers, biotic agents, and density-dependent reactions on the life-history strategies of the species. Results show that the potential risk of establishment of *S. frugiperda* is mainly limited to the coastal Mediterranean areas of southern Europe. However, transient populations migrating from suitable areas might represent a risk for vast areas of southern and central Europe. Predictions show that in Europe the species might be able to complete three generations per year in the most suitable areas. The model provides key elements for guiding surveillance and prevention plans towards the reduction of the risks linked to the introduction of the species in Europe.







### Multilevel analyses on pest primed and not-primed solanaceous plants

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Complex plant defense responses are involved in biotic and abiotic stresses, including insect herbivores. In particular, plant volatiles (VOCs) and induced volatiles (HIPVs) can play a key role in indirect plant defense, involving multitrophic levels. VOCs and HIPVs are important to allow plantplant and plant-natural enemy communications. In this context, olfactory bioassays were conducted, VOCs and HIPVs were identified, and gene expression of tomato and aubergine plants primed by Tuta absoluta (Lepidoptera: Gelechiidae) larvae was evaluated. In the tests, seven tomato and one aubergine varieties were used. Adults of T. absoluta and Encarsia formosa (Hymenoptera: Aphelinidae) were used in olfactory bioassays to understand how the key tomato pest and a model parasitoid are attracted to VOCs and HIPVs emitted by primed plants. The evaluation of the expression of three genes were done to highlight defense plant pathways, i.e., jasmonic acid, salicylic acid and B-phellandrene. The results showed that there was a significant increase in the production of primed volatiles. This explains the pest and parasitoid olfactory responses that varied among primed and not-primed tomato and aubergine varieties. The results of the gene expression analyses provided an important evidence of defense priming in the plant. These results are useful for understanding plant defense mechanisms and, above all, can provide volatile compounds involved in the defense of plants that could be used in integrated pest control, for example through attractive dispensers to enhance the biocontrol services of natural enemies, and in multitrophic relationships.







### Division of labour and altruism in tent construction and maintenance in a social caterpillar (*Thaumetopoea pityocampa*)

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Social organisms require organisation to be efficient and functional as a colony. Throughout the whole larval life, Thaumetopoea pityocampa live gregariously in their communal tent with siblings and conspecifics from other colonies. In this study, we explored how T. pityocampa caterpillars determine orientation on the tree and the social behaviour associated with tent construction. The location of the tent built on the host tree and silk application on the tent were both predominant in the south. The caterpillars can determine spatial orientation/direction from early larval instar through a pair of specialised stemmata that detect skylight polarisation patterns, which they used to determine where is south for tent construction and maintenance. When environmental conditions are optimal, the tent is maintained daily at around sunset by a few individuals which are predominantly male. The male caterpillars emerge from the tent first and spin silk on the tent for expansion and strength. During this time, these caterpillars are exposed to parasitoids and consequently, nearly half of parasitised caterpillars from the colony were male caterpillars maintaining the tent. Using non-destructive field collection methods with wildlife cameras and a frass counting apparatus, we were able to determine the foraging behaviour of early instar T. pityocampa colonies. Using wildlife cameras on small-sized insects in the field was effective, and this opens opportunities to other researchers to simultaneously monitor insects over long time periods without disturbance. Exploration into tent construction behaviour of T. pityocampa caterpillars indicated social structure in this species that displayed altruistic behaviours and division of labour to benefit the colony.





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# SHORT PRESENTATIONS ABSTRACTS







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

### A. Arpellino

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Pest management in vineyard requires new eco-friendly control methods. An emerging strategy, namely symbiotic control, takes advantage of insect-microorganism interactions, since manipulating these interactions has an extreme impact on insect life cycle. Grapevine pest insects belong to various orders. In North-West Italy, a major threat is Scaphoideus titanus, the main vector of the phytoplasmal agents of Flavescence dorée (FDp), while an emergent pest is Drosophila suzukii. The aim of this reserch is to provide a contribution to the knowledge regarding the microbial interactions in the pest species S. titanus and D. suzukii, to support the development of sustainable control strategies. Planned activities include in the study of the biological performance of S. titanus in different grapevine cultivars and other Vitis spp., in relation to the acquisition of different resident microbial communities. Moreover, investigations will be conducted on the symbiotic control of FDp, using a symbiont strain in the genus Asaia, which reduces phytoplasma acquisition by vectors. Finally, a survey of olfactive preference toward different microbial agents in D. suzukii will be performed, to develop pre-harvest control protocols. To implement the first objective, longevity will be compared on different S. titanus populations, established on: wild Vitis spp. V. vinifera cultivars and V. labrusca. Furthermore, DNA samples from each population will be destined to throughput sequencing of 16S rRNA gene. To achieve the second goal, plant-mediated acquisition and horizontal transmission of Asaia will be assessed. To complete the third objective, different preference bioassays will be performed using different yeast and bacterial strains.







### Popillia japonica transcriptome analysis

### S Boschi

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Popillia japonica Newman (Coleoptera: Scarabaeidae) is a beetle endemic of Japan unintentionally introduced in North America, The Azores and recently Italy. P. japonica is a serious quarantine pest since it creates enormous damage to agriculture feeding on more than 300 plant species. The study of the transcriptome can help to better understanding how to plan effective strategies to control this pest. Transcriptome is of substantial interest for several downstream applications including the analysis of gene expression during different life stages (larva, pupa and adult) and in treated versus untreated specimens. Total RNA was extracted from two larvae, two pupae and adults male and female of *P. japonica* using a Qiagen RNeasy Mini kit. The RNA extractions were quantified and qualified through Nanodrop, Qubit and Fragment Analyzer and then sequenced by Macrogen Europe using next-generation sequencing technologies. The results of the sequencing are nine libraries that have to be assembled de novo. We performed a preliminary assembly of one of the nine libraries using the TransPi pipeline. There results show a high percentage of complete and single copy genes and a low percentage of missing and fragmented ones. The gene ontology reveals that the majority of the genes that we obtained are typical of Scarabaeiformia and Coleoptera and classifies the resulting genes according to biological processes, cellular component and molecular function.







## The couple *Nemobius sylvestris/Pardosa sp.*: a model for analysing predator-prey interactions and their neural bases?

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For decades, pursuit-and-chase sequences between a predator and its prey have been carefully analysed in order to decipher the preferred strategies of both protagonists, which dictate the outcome of the encounters. One of the most complete approaches to model these interactions comes from game theory and consists in the combination of "pursuit-evasion games" preceded by less-known "hide-and-seek games", describing the searching phase of a predator. Although these theoretical models have given a wide range of testable predictions since the 60s, they suffer from the lack of enough experimental support: most empirical studies record separately the trajectories of both predators and preys and bypass the complex architecture of the environment in which the interaction occurs. We suggest that the couple Nemobius sylvestris/Pardosa sp. may provide an appropriate experimental testbed of the mathematical framework of hide-and-seek games. Moreoever, studying these protagonists may help us decipher the neural circuitry underlying "pursuit-evasion games". When looking at the pursuit-evasion behaviours, one can notice a high rate of successful escapes of crickets. The "dissection" of the cricket escape system has revealed that crickets detect aerodynamic signatures of approaching spiders with filiform hairs located on their cerci; information which is then encoded by a population of giant interneurons, and eventually ascends to higher motor centers triggering an escape response. We are planning to build a new biologically plausible computational model for wind-detection. Such a model may have implications for neuromorphic computing by proposing ways to process information from Microelectromechanical systems (MEMS).





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### Parasitization efficiency in relation to the female age for the pupal parasitoids of Musca domestica

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*Musca domestica* (Diptera, Muscidae) is a major pest for the environments where both humans and farmed animals live. Biological control with the use of pupal parasitoids is the most sustainable strategy to manage this pest in livestock farms. The most commonly used biocontrol agents for this purpose are the pupal parasitoids *Spalangia cameroni* and *Muscidifurax raptor* (Hymenotpera, Pteromalidae). The purpose of this work was to obtain the oviposition peak of the females of each parasitoid species in relation to their age, and to verify the sex ratio of the newly emerged adults. Twenty new fresh pupae of *M. domestica* were supplied daily to each fertilized female for 14 days to both species and the parasitoid emergence was subsequently checked. The results showed that the percentage of parasitization differs between the tested species, being higher for *S. cameroni* (57,71 %) than for *M. raptor* (32,41 %). The parasitization trend of *S. cameroni* was almost constant throughout the period, whereas that of *M. raptor* decreased after the 10th day. The sex ratio of the newly emerged parasitoids was always skewed towards females: 81% for *S. cameroni* and 66% for *M. raptor*. This type of information can be useful for optimizing the mass production and the usage times of the two species of parasitoids, in order to implement the biocontrol of houseflies.







## Susceptibility of ancient and modern wheat varieties to the lesser grain borer, *Rhyzopertha dominica* (F.)

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#### ABSTRACT

The increasing demand for healthy and organic food is a new chance of survival for ancient cereal varieties in marginal agricultural areas and is providing a way to safeguard these precious genetic resources. Rhyzopertha dominica (F.) (Coleoptera, Bostrichidae) is one of the major primary pests of stored cereals worldwide. To identify less-susceptible or resistant genotypes useful in breeding programs, the susceptibility of 3 ancient genotypes (Saragolla Antica, Dauno III, Senatore Cappelli) originating from Southern Italy, 3 modern durum wheat varieties (Ofanto, Svevo, Faridur), and 1 bread wheat variety (Mec) towards R. dominica was assessed. For each genotype, kernel (60 g) samples (n = 5) were placed in cylindrical containers ( $\emptyset$  9.8 x 10.8 cm), infested with 12 two-weekold unsexed adults and maintained in the dark at  $28 \pm 2^{\circ}$ C and  $60 \pm 5\%$  R.H. After 15 days, insects were removed, sexed and the number of dead specimens recorded. Emergence of F1 progeny was monitored periodically. For all varieties, a low adult mortality was recorded after 15 days. The lowest progeny production was obtained from the Dauno III variety (274.6 ± 39.8) and it was significantly lower than that observed for the Faridur variety (539.6 ± 52.8) (P < 0.05, Tukey test). The average development period (D) of R. dominica in kernels of Senatore Cappelli (52.86 ± 0.17) was significantly higher than in those of the remaining varieties. According to the susceptibility index (S.I. = 100 \* InF1/D), the different wheat varieties were classified as "susceptible" (S.I. from 8 to 10). Similar experiments are in progress to assess the susceptibility of the same wheat varieties to granary weevils.







# Thermal preference plasticity and fitness of an invasive polyphagous insect in heterogeneous environments

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Climate change has a strong impact on insects, including changes in phenology, distribution area or population extinction. The capacity for behavioral thermoregulation then becomes a crucial trait when the environment is sufficiently heterogeneous to allow individuals to find areas that are always favorable to sub-lethal. Thermal preference (also called selected temperature) is a central trait in behavioral thermoregulation, but to date very few studies have analyzed in depth the plasticity of this trait and the mechanisms (adaptive or not) underlying this plasticity. The study proposed here consists of analyzing this trait in an invasive insect whose invasion success has been dazzling: Drosophila suzukii. This fly has spread widely in Europe, despite having rather low thermal resistance traits compared to other species. Our hypothesis is that the plasticity of the thermal preference allows this fly to constantly make the "right choices" by selecting favorable microclimates during all seasons. This polyphagous fly causes significant economic damage, and its ability to lay and develop in a wide range of fruits (cultivated and wild) must play a role in this success. The thesis consists in determining the interaction between temperature and the resource (identity of the fruit) in the success of Drosophila suzukii. Are there tradeoffs between host (resource) selection and behavioral thermoregulation? The experiments aim to quantify the plasticity of thermal preference as a function of environmental conditions (thermal and trophic). The thesis will identify the adaptive value (fitness) of the thermal preference plasticity in a context of climate change.







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

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Halyomorpha halys is a phytophagous insect native to Eastern Asia and present as an invasive species in North America and Europe. Due to its exceptionally broad host range, it causes severe economic damages to many cultivated plants, including hazelnuts. Several control strategies have been studied and compared so far, among which symbiotic control resulted to be a promising option. This strategy is based on preventing the vertical transmission of insect's primary endosymbiont Pantoea carbekii. Recently, a micronutrient fertilizer (Dentamet SYM®) showed to effectively eliminate P. carbekii from the egg surface, and it was included in H. halys management measures. This research aims to empower stink bug symbiotic control in the hazelnut grove, dealing with several objectives. The genomes of *H. halys* populations from hazelnut groves, and their *P.* carbekii strains, will be characterized to (i) describe the relation between the genetic structure and possible phenotypic variations, also studying their effect on the efficacy of symbiotic control, (ii) identify genetic markers for fast recognition of different insect functional haplotypes. The evaluation of the real fate of symbiotic control treatments will be performed by using sentinel H. halys egg masses placed in hazelnut groves prior to treatments and then moved to the laboratory for mortality assessment and determination of symbiont elimination. The proportion between resident *H. halys* population in hazelnut groves and fertile females coming from the surrounding environment will be estimated in orchards with different landscape composition, by molecular characterization of the gut content.







# Innovative processes for lipid extraction from bioconverter insects, qualitative and quantitative evaluation and industrial applications for the formulation of personal care products

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The use of insects as a new source of lipids and proteins is a topic of great interest from both environmental and economic point of view. The bioconversion process mediated by the saprophagous insect Hermetia illucens (Diptera: Stratiomyidae) is very advantageous because from substrates of low economic and biological value, for example agri-food by-products or zootechnical and catering waste, it is possible to obtain products of high economical, biological and commercial value. My PhD project proposes the extraction of lipids from *H. illucens* larvae and the use of fatty acids, principally lauric acid, for the formulation of new products for personal care (shampoo, detergent and soap). After the construction of a biofabric (bio-farm) of H. illucens, three types of feeding substrates were evaluated; oranges, tangerines and strawberries (by-products from a factory in Basilicata region) were bioconverted by larvae and a lipids extraction was performed by Soxhlet method. Lipid yield was similar for each substrate (29-30%). The H. illucens ability to bioconvert organic substrates into valuable products represents a valid solution to the request for the implementation of sustainable processes in a circular economy perspective. A massive production of larvae reared on the "best diet/s", in terms of both lipid and lauric acid content of larval biomass, will be carried out. As final experiments, extracted fats will be used for experimental trials on formulation of products for personal hygiene.







# Analyses by conventional microbial methods and metagenomic approach of an enrichment culture obtained from plastic-eating *Alphitobius diaperinus* (Insecta: Coleoptera)

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The unceasing use of plastics for multiple daily-life uses is causing alarming debris accumulation and micro-plastic pollution. Among these synthetic materials, polystyrene (PS) is one of the most employed. For several years, many authors demonstrated that some tenebrionid insects can apparently degrade it. Recently, it was proven that the lesser mealworm Alphitobius diaperinus is capable of feeding on and degrading PS. A previous analysis carried on the gut microbiota of PS-fed larvae, by using metagenomic Next Generation Sequencing (mNGS) method, revealed several microbe taxa as differentially abundant between treatment and control groups, with some of them being previously found associated with plastic compounds and/or proposed as plastic-eaters. This observation offers challenging options for technology innovation in plastic biodegradation. In this perspective, a culture was started from PS-fed larvae of A. diaperinus in liquid carbon-free basal medium with PS film as sole carbon source in order to isolate culturable bacteria which become prevalent under this selective enriched conditions. After two-months the liquid enrichment was analysed by conventional microbial methods and a metagenomic approach. Bacteria isolated were characterized by means of phenotypic and phylogenetic analysis, and they resulted ascribable to three taxonomic groups: Klebsiella, Pseudomonas and Stenothrophomonas. The predominance of these groups in PS-fed larvae was also confirmed by the metagenomic approach, and it resulted consistent with the previously published findings. The ability of all isolates to attach to PS surfaces and produce biofilm-like structures was then evaluated by enzymatic assay and observation with Scanning Electron Microscopy (SEM).







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

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The 102 gene has been functionally characterized in *Heliothis virescens* (Lepidoptera, Noctuidae) as well as in *Spodoptera littoralis* (*Sl102*) and in both species it encodes a protein essential for the cellular immune response (encapsulation and nodulation of non-self organisms). The silencing of this gene by RNA interference (RNAi) increases the noctuid moth larvae susceptibility to *Bacillus thuringiensis* by enhancing the induced septicaemia, and provides the basis for the development of new insect pest control strategies. Here we further explore the phenotypic effects of *Sl102* silencing in embryos of *S. littoralis*. The experimental results demonstrate that gene expression in embryos is significantly reduced when eggs are treated with dsRNA targeting *Sl102* gene, compared to control eggs treated with dsRNA targeting *GFP* gene. There was a drastic reduction in egg hatching, due to the arrest of embryonic development, and the few hatched larvae showed a very high mortality rate. The observed lethal phenotype indicates 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. Our data pave the way towards new insect pest control applications and offer the opportunity to unravel new molecular details of insect development.







### Impact of stress agents on honeybee gut microbiota and immunity

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Honeybees are the cornerstone of the terrestrial ecosystem's biodiversity as providers of the pollination service both for crops and wild plants. The honeybee health decline and associated large-scale colony losses are a big problem of global importance for their remarkable impact on the environment and human economy. The reduced honeybees survival is the final result of a multifactorial syndrome trigged by several stress factors, that may synergistically interact with each other, inducing a reduction of honeybee immunocompetence, not yet fully characterized at functional level. Several monitoring programs indicated that high loads of parasites and pathogens largely contribute to this problem. Among them, the mite Varroa destructor and the Deformed Wing Virus (DVW) stand out by their frequency and abundance. DWV is an endemic pathogen 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 Varroa, pesticides and poor nutrition. Here we focus on the role that the gut microbiota can have on the modulation of honeybee immunity and if and how the stress factors mentioned above can trigger gut dysbiosis which may partly account for their negative effects on immune barriers. 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 stress-induced decays of immune competence.







# The black soldier fly *Hermetia illucens*: an innovative and alternative source of chitin and chitosan

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Chitin is the structural component of the exoskeleton of arthropods and the cell wall of fungi and yeasts and it is the second most abundant natural biopolymer on Earth, after cellulose. Chitin can be deacetylated in a more soluble biopolymer, chitosan. Several properties, like biodegradability, bio-compatibility, non-toxicity, antioxidant and antimicrobial activity, allow chitin and chitosan to be used in various fields. The main source of commercial chitin is represented by waste from the fishing industry, chiefly crustaceans' shells. Insects are a valid alternative to crustaceans as a source of chitin and represent a solution to the current growing demand for the set up of sustainable process. We focused our attention on the insect *Hermetia illucens*, (Diptera: Stratiomyidae), since it is perfectly framed within a system of circular economy, as it feeds on decaying material and organic waste from the agri-food chain. Mainly pupal exuviae and dead adults of *H. illucens*, are a chitin-rich substrate that can be used for chitin extraction and chitosan production.

The aim of this research project is to produce different types of chitosan-based nanoparticles (NPs), usable in pharmaceutical field for the drug delivery and, in this perspective, in biomedical applications, like wound healing. Together with chitosan NPs, chitin nanofibrils have also gained attention as carriers that can be combined with electronegative compounds, carrying various bioactive substances. Therefore, starting from chitin it is possible to obtain chitosan, through a deacetylation process, and to develop NPs.







### Distribution of fly species of veterinary importance in Basilicata region (Italy)

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Livestock farms are prominent realities in Basilicata region (Italy), for which parasitological maps were already completed, in collaboration with the Associazione Regionale Allevatori Basilicata (ARAB), allowing the identification of the most relevant epidemiologic and etiologic aspects affecting production. Here we further expand this effort, focusing on the detection and identification of Diptera of veterinary importance. During 2020, a preliminary mass trapping program was carried out in 217 farms, using specific baits for sponging flies. The identification of captured individuals, carried out on morphological basis, allowed the identification of 7 major species. Among these, Musca domestica was the most widely distributed, while the smaller house fly (Fannia canicularis) was identified in samples from only two farms. Most of the identified flies belong to Calliphoridae family (green- and blue-bottle flies, blowflies). The species found can affect the animal welfare to a various extent, from simply annoying the animals to causing important myiases or other disease, like the Sheep strike. These problems have been largely prevented so far by good practices promoted by ARAB staff, which allowed to keep these insects under control. However, it is essential to implement a continuous monitoring to update population maps of these insects, as a basis for developing targeted and effective IPM (Integrated Pest Management) strategies.







### Autumn and night: the neglected side of plant-arthropod interactions

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Plant-arthropod interactions are an essential component of ecosystems: arthropods have a key role in plant reproduction and dispersal, both as pollen and seed vectors, and plants can provide trophic resources for arthropods as well as shelters, mating and oviposition sites. However, the knowledge about these interactions is still limited, especially in mountain environments. Moreover, most of the literature focuses on diurnal interactions occurring in flowering, while interactions occurring at other times (e.g., seed dispersal, site of oviposition, foraging or sheltering) have been neglected, especially during night. The aim of this work is to investigate the relationship between autumnal visitors and Androsace brevis, a vulnerable endemic Alpine plant, and to identify arthropod taxa potentially involved in seed dispersal. We used an integrated approach: diurnal and nocturnal video observations, and diurnal manual sampling of plant visitors. The two sampling methods highlighted the presence of a diverse diurnal community (video observation: Hymenoptera, Coleoptera, Collembola, Diptera, Lepidoptera, Araneae, Hemiptera; manual sampling: Acari, Hymenoptera, Araneae, Coleoptera, Diptera). Nocturnal video observations revealed the presence of visitors, too (Opiliones, Collembola, Araneae, Hymenoptera, Chilopoda). Seed dispersers were not detected, but the high level of observed biodiversity indicates that A. brevis might represent an important resource for arthropods, in particular for trophic activity and sheltering.







### Identification of key plant-host volatiles affecting behavior of brown marmorated stink bug - *Halyomorpha halys* (Hemiptera: Pentatomidae) on commercial hazelnuts

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Brown Marmorated Stink Bug (BMSB), *Halyomorpha halys* Stål (Hemiptera: Pentatomidae), was accidentally introduced to the United States in the mid of 90's, and later in Europe becoming a major past in hazelnut orchards. In recent years research interest has been focused on the identification and synthesis of organic compounds (aggregation pheromones and attractants), to keep *H. halys* populations under control. These methods have been proven to be extremely useful for monitoring and trapping BMSB individuals. Plants, under certain environmental and stressful conditions express specific VOCs (volatile organic compounds) that can potentially affect the presence of phytophagous pests. These volatiles consist of a complex mixture of chemical compounds, which in combinations have distinctive characteristics. Here we investigated on the BMSB antennal physiological and behavioral responses to VOCs emitted by hazelnut using EAG-GC and Y-tube olfactometer. *Halyomorpha halys* exhibited antennal physiological responses to all tested compounds with different degrees of sensitivity on GC-EAD. It was possible to identify and test with EAG six key plant-volatiles. Behavioral studies showed *H. halys* attraction to two compounds and repellency for a specific one.







# Sex pheromone of the asparagus moth, *Parahypopta caestrum*: chemical analysis, electrophysiological study and preliminary field tests

### Onofrio Marco Pistillo, Giacinto Salvatore Germinara

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Parahypopta caestrum (Hübner) (Lepidoptera, Cossidae) is the most serious pest of Asparagus spp. in the Mediterranean basin. To contribute to the knowledge on the sex pheromone of this pest further chemical, electrophysiological and field tests were carried out during the second year of the Ph.D. In the previous year, gas chromatography coupled with mass spectrometry and electroantennography (GC-MS-EAD) analyses have revealed the presence of four EAG-active peaks in the gland extracts of calling virgin females whose mass spectra matched those of three monounsatured C<sub>14</sub> aliphatic acetates and one monounsatured C<sub>14</sub> aliphatic alcohol, respectively. In the second year, isomerism and double bond position in the structure of these EAG-active compounds were fully elucidated by comparing their GC retention times and MS fragmentation patterns with those of authentic standards. Double-bond position and configuration of sex pheromone candidates were also confirmed by comparative EAG experiments with series of monounsaturated C<sub>14</sub> aliphatic acetates and alcohols. To define the relative ratio of different sex pheromone components in female glands, quantitative analyses of different gland extracts were performed. The sensitivity of male antennae to each component was characterized by calculating the corresponding EAG dose-response curves. In preliminary field trapping trials, individual compounds were not attractive. Some binary and even more ternary mixtures of acetates were attractive to P. caestrum males. In the next year, activities will focus on the optimization of pheromone lure and traps for monitoring and mass trapping applications and on the identification of asparagus volatile compounds possibly involved in insect host-plant interactions.







# Host-parasitoid developmental interactions are modulated by a venom component of *Aphidius ervi* (Hymenoptera, Braconidae)

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The parasitoid Aphidius ervi regulates physiology and reproduction of the host Acyrthosiphon pisum (Homoptera, Aphididae), in order to enhance its nutritional suitability for the developing offspring. Understanding the molecular mechanisms underlying the host-parasitoid interaction may allow the development of novel "bio-inspired" pest control strategies. Using the RNA interference (RNAi) technique, we investigated the functional role in vivo of the main component of A. ervi venom, Aey-glutamyl transpeptidase (Ae-y-GT1), known to cause host castration. Microinjections of doublestranded RNA (dsRNA) into female pupae showed a significant Ae-y-GT1 knockdown in newly emerged females. Parasitism by these wasps allowed to score the phenotypic changes of the host and of its progeny as affected by a venom blend lacking Ae-y-GT1. The effect of gene silencing significantly induced an increase in size of parasitized aphids and of their load of the primary bacterial symbiont Buchnera. Similarly, both A. ervi larvae and teratocytes (cells derived from the embryonic membrane) showed a significant increase in size. The silencing of Ae-y-GT1 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 at the emergence, 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 regulation of the intricate network of interactions among the parasitoid, the host aphid and its symbiont, which finely orchestrate the development of parasitoid's offspring.







### Impact of probiotic bacteria on *Tenebrio molitor* fitness, gut microbial composition and susceptibility to *Bacillus thuringiensis serovar tenebrionis* and *Metarhizium brunneum* infections

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Tenebrio molitor (Coleoptera L.) the yellow mealworm is an insect model for infection and immunity studies and is used in mass-production of insects as feed and food. The industrial rearing of T. molitor on agricultural by-products may expose them to biocontrol residues, like environmental resistant Bacillus spores and fungal conidia, which could impact the fitness of T. molitor. Therefore, my PhD project deals with experiments analyzing different outcomes of single and co-infections of B. thuringiensis, and Metarhizium brunneum on the larval stages of T. molitor. Furthermore, as for other animals, the possible benefits of addition of probiotic bacteria to the feed will be analyzed. The pathogenicity of B. thuringiensis serovar tenebrionis (Btt) and Metahrizium brunneum KVL 12-30 are first tested by single infection on T. molitor to define LD25 and LD50. Then targeted coinfections will allow to determine additive, synergistic or antagonistic interactions between these pathogens. Alongside infections, feed uptake, growth rate etc. are recorded and gut microbiota composition is analyzed by 16s rRNA Mi-sequencing to measure how probiotic and pathogens modify the OTUs' composition. The hypotheses are: 1) M. brunneum and Btt have different mechanisms of infection, therefore dose and timing of pathogen exposure should influence the outcome 2) the presence of probiotics may help the insect to cope with the infection by improved immunity, by presenting a shorter period for pathogen clearance, by expressing better fitness performances. The presentation will include the experimental setup and the preliminary results.







# Extraction and characterization of chitin from different developmental stages of Hermetia illucens

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Chitin is the most widespread biopolymer on Earth, after cellulose. It is the major structural component of the arthropods' exoskeleton and the cell wall of fungi and yeasts. Due to their properties, like biodegradability, biocompatibility, non-toxicity, antioxidant, chitin and its main derivative, chitosan, find many applications in the industrial and biomedical fields, such as drug delivery, cosmetic, wound healing, food preservation, tissue engineering and wastewater treatment.

Nowadays, the main commercial source of these biopolymers is represented by fishing industry waste, mainly crustacean exoskeletons, but this source is no longer sustainable. Among the potential alternative sources of chitin, insects are receiving great attention, particularly bioconverter species, part of a circular economy system.

The present project focuses on the extraction of chitin from the insect *Hermetia illucens* for the production of chitin nanofibrils (CNs), cosmeceutical active compounds, that have the ability to promote skin health. CNs are used both as active ingredients and as carriers capable of accelerating the penetration of emulsions and the skin repair. Chitin was extracted with chemical method from larvae, pupal exuviae and adults of *H illucens*, and then was characterized. The values of chitin content varied depending on the developmental stage of the insect, but were comparable to the commercial one (5- 30%). The highest chitin yield (21-24%) was obtained from pupal exuviae, the insect sample with the highest chitin content. The FTIR and XRD spectra showed similar peaks between the chitin obtained from *H. illucens* and the commercial one confirming the insects as alternative source to crustaceans.