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XXVI Italian National Congress of Entomology

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LECTURES AND WEBINARS

PLENARY LECTURE

Novel approaches to Systematic Entomology and state-of-the-art in Italy

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In the last decade insect phylogenetics has made tremendous progresses owing to the combined use of morphological and molecular data, along with new tools for data analysis (Kier et al. 2016, J R Soc Interface, 13: 20160363). Stable phylogenetic reconstructions make classification schemes predictive, besides representing a critical step toward the understanding of evolutionary patterns. The integration of morphological and molecular data plus 'total evidence' approaches to dating phylogenies with fossils have become the most effective means to resolve conflicts in insect systematics. Multi-locus datasets, combining both mitochondrial and nuclear genes - analyzed with evolutionary and coalescence-based models, e.g. species tree approaches - have produced more robust phylogenetic hypotheses. Concurrently, phylogenomics (e.g. anchored hybrid enrichment) and transcriptomics have expanded their realms allowing consolidating the phylogenetic-backbone of insect classification and resolving some main nodes within selected order- and/or family- ranked clades. Besides, in the last 15 years, morphology-based insect systematics has regained momentum, especially thanks to advanced electron microscopy techniques (e.g. LS-FM, FIB-SEM), micro-computed tomography and computer-based 3D reconstruction that greatly accelerated data acquisition of ultrastructural morphological details with an unprecedent high quality. In Italy, systematic entomology has traditionally walked along with faunistics and focused on selected families of several insect orders (Coleoptera, Diptera, Orthoptera, Hymenoptera, Collembola, Neuroptera, Plecoptera, Isoptera and others). Though many studies have relied (and still) on traditional morphology and/or the use of well-established molecular markers, in recent years phylogenetic reconstructions combining different sources of data (i.e. 'integrative taxonomy') have increased. Novel approaches in multi-locus analysis, genomics, ultrastructural morphology and total-evidence dating via fossils have steadily emerged, also in studies with aims going well-beyond taxonomy (e.g. in Medical Entomology). We review some of the most recent and exemplificative innovative works conducted in Italy and remark the need to push future studies toward more updated techniques and analytical approaches.

KEY WORDS: molecular markers, phylogeny and phylogenomics, total evidence, ultrastructural morphology.

PLENARY LECTURE

Towards a landscape-scale management of beneficial insects and pests

Lorenzo Marini

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Insects are integral components of both managed and natural ecosystems. On the one hand, insects are major regulators of ecosystem processes that sustain the delivery of key services such as pollination, pest control and nutrient cycling. On the other hand, insects also produced well-known “disservices” being key crop and forest pests and vectors of plant pathogens. In the last decades, managing insects and landscapes, and their interactions, in ways that ensure the sustainability of ecosystem services and minimize induction of disservices has been the focus of applied entomological research. However, this task is particularly challenging as insects are often mobile organisms that depend on the availability of resources occurring across multiple habitats. Their ability to locate these resources depends on their mobility but also on the landscape composition, i.e. the relative proportion of habitat types, and configuration, i.e. the spatial arrangement of these habitats. To fully understand the processes that govern insect populations and communities across the landscapes, we need to embrace and understand this spatial heterogeneity. Whatever our entomological interests are, landscape processes have a strong influence on the ecology of most insect species that we study. For instance, as pest managers, we need to know how the interplay between crop and semi-natural habitats influences pest population dynamics and their interactions with natural enemies. At the same time, as conservation entomologists, we need to develop strategies to maintain species diversity or associated ecosystem processes across multiple habitats. Here, I will provide an overview of the development of landscape ecology applied to the main entomological fields. Starting from the pioneering work on fragmentation and pests, I will show how the field of landscape ecology has made significant inroads towards understanding beneficial insects’ and pests’ response to land-use. This large body of research has usually investigated the effects of local quality of a focal habitat versus the effect of the surrounding landscape matrix. However, the simple dichotomy of a focal habitat versus the surrounding matrix overlooks the complexity of spatial processes that characterise real-world landscapes, calling for the development of novel analytical approaches. Integrating and analysing species use of multiple habitats may seem a daunting task, especially because the number of species-habitat links scales exponentially with the number of species and habitats considered. Fortunately, tools developed from network theory are now able to analyse and describe complex interactions using unipartite and bipartite networks. Against this background, I will present a novel approach - the species-habitat network- that enables characterization of not only species or habitat-level dynamics, but also the emerging properties of those networks, going beyond the traditional landscape patch-mosaic model. The versatility, visualization power and ease of interpretation of these networks will enable the application of the species-habitat network concept to a wide array of real-world problems concerning species conservation, pest management, ecosystem services, and vector ecology. From a policy perspective, a deeper understanding of how habitat-species networks work can open the door to effective landscape management for both beneficial insects and pests.

KEY WORDS: conservation, ecological interactions, landscape ecology, species-habitat networks.

WEBINAR

Influenza dei cambiamenti climatici sulla vita degli insetti

Luca Mercalli

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Il riscaldamento globale è in atto e condiziona fortemente la biosfera. In base alle scelte di decarbonizzazione adottate dall'umanità, entro la fine di questo secolo gli aumenti termici potranno essere compresi tra circa 2 e oltre 4 °C, con conseguenze inedite.

PLENARY LECTURE

Functional and molecular agroecology: new strategies for insect control

Francesco Pennacchio

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The science of agroecology merges very broad areas of research like, among others, ecology, plant biology, soil science, parasitology, pathology, microbiology to address key-problems facing our society such as food security, food safety and biodiversity loss, as affected by global change. One of the major limits of agroecology is the lack of a consolidated theoretical framework that is required to develop and test generalizable hypotheses, in order to conceive and manage reliable, predictable and safe food production strategies, ecologically and economically sustainable. This problem can be managed only by unravelling the mechanisms underlying the complex network of interactions among organisms inhabiting above- and below-ground agroecosystems, and understanding how these interactions are linked with both crop and off-crop context, at molecular, organism, farm and landscape level. Indeed, a solid background of functional information on agroecosystems is needed to analyze and manipulate the key-elements of complex production systems, through the use of mechanistic models. These tools can allow to study and manage the multifactorial natural processes underpinning agricultural production using a systems biology approach. The current availability of a solid conceptual framework and powerful computing facilities offers unparalleled opportunities to develop new concepts and tools in agriculture. The natural food webs in agroecosystems, even though simplified compared to natural ecosystems, are still incredibly complex. In this context, insects play very important roles, ranging from providers of a wealth of ecosystem services to pests of economic importance. Therefore, insects are key-players in driving the flow of energy generated by primary producers and remarkably contribute to ecosystem stability. The energy made available by plants in natural food chains is allocated through multiple competition processes among organisms at different trophic levels. The myriad of resulting co-evolutionary pathways are driven by a continuous interplay between virulence strategies and immune defence barriers, which are deeply influenced by the microbiota associated with interacting organisms and by the abiotic factors. Understanding the functional bases of these co-evolutionary interactions at metaorganism level, as affected by the environmental global change, is the key for developing sustainable insect control strategies, which have limited or, ideally, no-impact on non-target and/or beneficial organisms. Unveiling the molecular mechanisms which regulate the physiological networks underlying these intricate interactions paves the way towards the development of new bioinspired strategies for pest control and for protection of essential ecosystem service providers.

KEY WORDS: multitrophic interactions, ecosystem services, microbiota, biological control.

PLENARY LECTURE

Reflections on entomological research in Italy from the 18th to the 20th century

Roberto Poggi

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A concise report on the entomological research carried out in Italy in the period between the 18th and the 20th century is presented, referring to the most prominent figures who characterized the development of knowledge on insects in our nation, from the first pioneering authors who adopted the binomial nomenclature of Linnaeus up to the present day, also recalling the most significant bibliographic reference texts. The review closes by highlighting the value of the entomological collections preserved in museums and scientific institutes, which constitute the indispensable basis for anyone who in the future will carry out investigations, from traditional to the most futuristic ones.

KEY WORDS: Entomology, Italy.

PLENARY LECTURE

Insect and entomopathogenic microorganism interactions for pest management)

Luca Ruiu

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Within the complex relationships between the biocenotic components of ecosystems, invertebrates have developed a wide variety of interactions with the microbial communities with which they live and co-evolve. Although in most cases such interactions may appear marginal or of unknown significance, some symbiotic microorganisms have evolved as mutualists of their hosts, while others as pathogens. The sophisticated mechanisms underlying the pathogenetic process, supported by a plethora of molecules of microbial origin, regulate various stages such as invasion of the host, overcoming its barriers and immune system, and infection to death. Knowledge of the biological properties of entomopathogenic microorganisms provide opportunities for their use as biological control agents in agro-ecosystems. After few decades of using synthetic chemicals to protect against agricultural pests and human or animal parasites, this strategy was found to have a significant impact on the environment with deleterious effects on the ecosystems. These include risks for human health through contamination of food and water, and toxic effects on operators and non-target species. For these reasons, the pesticide industrial portfolio is evolving toward the development and authorization of products with progressively lower impact. At the same time, the use of several active substances has been prohibited in different parts of the world and many old generation products have been withdrawn from the market. However, the defence of agri-food crops and against disease-carrying vectors is always necessary, which fosters a continuous search for effective and alternative solutions. Among these, the employment of microbial-derived products in biological control and integrated pest management programmes has proved to be a viable alternative involving living microorganisms and their metabolites. This includes bacteria (eg. *Bacillus thuringiensis* and other *Bacillaceae, Serratia, Pseudomonas, Yersinia, Burkholderia*, *Chromobacterium, Streptomyces*, and *Saccharopolyspora* species), fungi (eg. *Beauveria bassiana, B. brongniartii, Metarhizium anisopliae, Verticillium, Lecanicillium, Hirsutella, Paecilomyces*, and *Isaria* species), baculoviruses (eg. CpGV, LdMNPV), and nematodes (eg. *Steinernema* and *Heterorhabditis* species), acting against pests in different orders. The pesticidal action of these living microbials is related to a strict and co-evolved relationship with their hosts, and often depends on a very specific mode of action. On this base, such microorganisms are generally regarded as safe for the environment and human health. However, this is not the case for any microorganism, given that possible side effects are always to be considered and predicted. It therefore becomes a priority to associate efficient risk assessment systems for newly proposed microbial based products.

KEY WORDS: biocontrol, entomopathogens, bioinsecticides, IPM, agro-ecosystems.

WEBINAR

Forensic entomology: from theory to practice

Stefano Vanin

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Forensic entomology is the application of the knowledge about insects, and more generally about arthropods, in legal matters. The discipline is articulated in three branches: store-products entomology, urban entomology and legal medical/veterinary entomology. The workshop deals with the last one. The attendants will see how the steps from the crime scene to the court of law need some vital prior theoretical components: the knowledge of the species and the ability of a correct identification of them. The workshop is based on four modules. The first one will be a description of the bulk of the discipline and of the investigative questions - known in English as the 6Ws – applied to violent crime and to cases of neglect and abandoned people or animals. It is worth of mentioning that in the last years forensic entomology has been largely applied to veterinary cases and that insects are considered more and more as a strong evidence in cases in which non-self-sufficient people are involved. The second module deals with the description of the developmental stages of the most common species of forensic interest, mainly Diptera and Coleoptera, in Italy. Diagnostic features useful for the identification of larvae and puparia will be presented. The presentation of real forensic Italian cases in which insects were presented in court of law constitutes the topic of the third module. In this module the GIEF (Gruppo Italiano per l'Entomologia Forense) guidelines for insects' collection, preparation and storage will be also described. The fourth module will conclude the workshop introducing the new challenges for Forensic Entomology and the impact that new analytical and biomolecular technologies have on the discipline, enlarging its application field.

KEY WORDS: Post mortem interval, Diptera, violent crime.

SESSION I

Italian apiculture: a journey through the history and honey diversity

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Italy represents probably the country where the bee culture has permeated more than elsewhere in the world the history of people: their traditions, economy, literature and artistic expressions, also thanks to the geographical and cultural diversity that characterize the "Bel Paese", from North to South, until the major islands. The great cultural and environmental diversity has become the basis for a wide variety of melliferous sources, especially with the advent of modern beekeeping. Italy is unique in the world for the extension of honey botanical varieties, their sensory characteristics, properties and composition. Apart from the monumental worldwide historical work of Eva Crane (The world history of beekeeping and honey hunting, 1999), where Italian beekeeping is treated, there is a lack of an extensive discussion/dissertation illustrating and summing up the richness of Italian beekeeping in the different regional cultures. This book represents a first attempt to summarize the broad knowledge of Italian beekeeping and its history, including the diversity of honeys. This is undoubtedly a very ambitious goal, of which this book represents a partial contribution that will hopefully be enriched and updated in the future. However, from this first edition, we can already see the wealth of news and contributions that over time have characterized the evolution of beekeeping in Italy. The features of Italian apiculture are unique and often linked to the many cultural influences that the various regional populations have suffered over the centuries, permeating cultures, behaviours, habits and traditions, which revisited today give us a variegated picture of beekeeping and its relationship with the environment, once again attesting to the richness of Italian culture in all its expressions. The work involved almost all the biggest national experts, not only at academic level, with the coordination of the Italian National Academy of Entomology, and is aimed to enrich the Eva Crane's work and to honour her memory and the memory of the many personalities and researchers that promoted in the past in Italy the modernization of the apiculture and increased the knowledges on honeybees and honey.

KEY WORDS: beekeeping, traditional techniques, Italian regions.

The “Don Giacomo Angeleri” Beekeeping Observatory of the University of Turin

Aulo Manino, Marco Porporato

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The "Don Giacomo Angeleri'' Beekeeping Observatory was set up in 1969, following Maria Grada Angeleri's wish to honour the memory of her brother Don Giacomo, a leading figure in Italian beekeeping in the first half of the 20th century. She gifted properties in Reaglie on the Turin Hill and Pragelato in Val Chisone to the University of Turin to create an institution dedicated to beekeeping research, with a special focus on practical beekeeping. The agreement was signed 12 years after the death of Don Angeleri, thanks to the dedication of Professor Carlo Vidano, who always put his love of research before personal gain. Carlo Vidano, assistant professor of Agricultural Entomology at the University of Turin, after obtaining his free teaching diploma in Agricultural Entomology, taught Bachiculture and Apiculture in the Faculty of Agriculture for nine years, from the 1959/60 academic year to the 1967/68 academic year. At the beginning of the 1968/69 academic year, he obtained the Chair of Bachiculture and Apiculture, which led to the creation of the Institute of Beekeeping at the University of Turin, to which the Observatory was attached. The deed of gift expressly refers to the existence of the chair of beekeeping, with a commitment by the University to provide the Observatory with sufficient means and staff to carry out the planned activities, an obligation which the University has essentially fulfilled to date, even though the means and staff have been formally assigned to the relevant university institutions. Through various organisational changes, the Beekeeping Observatory is currently attached to the Department of Agricultural, Forestry and Food Sciences. In the 1976/77 academic year, Professor Vidano took over the chair of Agricultural Entomology, although he remained in charge of the Observatory until his untimely death; responsibility for the management of the Observatory fell to Professor Franco Marletto, who also became the holder of the apiculture teaching post. Thanks to the existence of the "Don Giacomo Angeleri" Beekeeping Observatory, a research group was formed at the University of Turin, dedicated to the study of apidology, practical beekeeping and crop pollination. Thanks to participation in international, national and local research projects, results have been obtained which have been published in scientific journals and presented at international and national congresses and conferences. A great deal of effort has been put into quality dissemination in favour of beekeepers, also through the publication from 1975 to 1997 of the magazine "L'apicoltore moderno", and participation in numerous public engagement activities aimed at beekeepers, stakeholders and the general public.

KEY WORDS: apidology, education, dissemination, pollination, research.

Entomological research in Piedmont and Aosta Valley

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The north-western corner of Italy is now shared between Piedmont and Aosta Valley regions since the middle of the 20th century. However, the geographical and historical unity of the area comprising most of the Italian side of the Western Alps, the upper Po Valley and the hills descending from the Ligurian Apennines cannot be denied, even though the borders with neighbouring regions are often difficult to define. Similarly, entomological studies did not develop in Piedmont and Aosta Valley separately from what happened in the rest of Italy and neighbouring countries. However, since we have to define the limits of the discussion, we consider those scholars who have carried out part of their entomological research in Piedmont and Aosta Valley and those whose collections are conserved in the area, also mentioning the scientific institutions in which many of these entomologists have worked. Furthermore, in order to ensure sufficient detachment from current events, a time limit is set coinciding with the end of the last millennium, excluding as well all entomologists still living at that date.
The development of entomological studies in Piedmont and Aosta Valley started in the 17th and 18th centuries as a result of the renewed interest in nature, especially on the part of physicians; in the preceding centuries the chronicles reported large infestations of harmful insects from time to time, but without any rational approach. Only in 1811, with the appointment of Franco Andrea Bonelli to the chair of Zoology at the University of Turin, was the separation of zoology from botany and medicine sanctioned. In the following centuries, the Institute and the Museum of Zoology were the setting for the work of distinguished entomologists such as Bonelli himself, Giuseppe Gené, Vittore Ghiliani, Alfredo Borelli, Ermanno Giglio-Tos and Achille Griffini. In the course of the 20th century, scientific interest in the entomological collections waned before their importance as a reference point for environmental studies was understood, and in 1980 the University of Turin entrusted them, perhaps improvidently, to the newly established Regional Museum of Natural Sciences. Alongside the work of academic entomologists, we must not forget the activity of many non-professional entomologists, such as Giuseppe Peiroleri, Eugenio Truqui, Ferdinando di Breme, Luigi Bellardi, Eugenio Sella, Flaminio Baudi di Selve, Mario Sturani, Roberto Pescarolo and many others who contributed to the progress of entomological studies in Piedmont and Aosta Valley. At the same time, many scholars also devoted themselves to applied entomological research, studying insects that are harmful to crops and domestic animals, sericulture and beekeeping; among them we should mention Edoardo Perroncito, Giuseppe Della Beffa, Carlo Vidano and Franco Marletto.

KEY WORDS: applied entomology, beekeeping, insect collections, sericulture.

ORAL PRESENTATION

Origins of urban and commodity entomology in the work of the microscopist Filippo Bonanni (1638-1725)

Rinaldo Nicoli Aldini

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The Roman Jesuit Filippo Bonanni (1638-1725) is an important figure in the European cultural panorama of the seventeenth and eighteenth centuries. Tireless scholar and eclectic writer, over his long life he cultivated both humanistic and scientific interests, ranging from the physical and natural sciences to art, from archeology and numismatics to the history and organization of religious and chivalric orders, also including the study of musical instruments, as can be seen from the titles of his numerous works, almost all written in Latin. As a clerk in Rome at the "Cabinet of Curiosities" known as the Kircherian Museum because enriched by the collections of his famous master, the Jesuit Athanasius Kircher (1601-1680), Bonanni illustrated it in the work 'Musaeum Kircherianum' (Rome 1709) and remained responsible for it until his death. In the field of natural history, Bonanni occupies a non-marginal position as a malacologist ('Ricreatione dell'occhio e della mente nell'osservation delle chiocciole', Rome 1681, re-edited in Latin a few years later) and as a microscopist and insect observer, but he is best known, in the wake of his master, for having been a staunch defender of the theory of spontaneous generation. This has certainly damaged his reputation as a scholar in comparison with more or less contemporary scientists who instead contributed to refuting the 'generatio aequivoca' with accurate experimental research (among the Italians, in this regard, Redi, Malpighi, Marsili, Cestoni and Vallisnieri stand out). The observations by Bonanni of insects and other arthropods, which he carried out above all with the aim of finding arguments in favour of this theory, are found in his work 'Observationes circa viventia, quae in rebus non viventibus reperiuntur. Cum micrographia curiosa sive rerum minutissimarum observationibus, quae ope microscopii recognitae ad vivum exprimuntur' (Rome 1691). These are thus written observations supporting the spontaneous generation, but accompanied by numerous valuable illustrations made with the aid of the microscope that Bonanni, valiant microscopist as well as a skilled draftsman, perfected himself to investigate the microcosm of arthropods beyond the potential of the human eye, apart from the dialectical intent mentioned above. This is indeed the primary objective of the appendix to his work, the 'Micrographia curiosa' which enriches what was presented in the first, greater part of the volume with further observations and tables. A valuable ensemble related above all to urban entomology, given that the author finds in the arthropods of the urban and domestic environment the raw material most at hand as a source of observations. With surprising precision for the time, in the 'Observationes' mosquitoes, sand flies, fleas, lice, booklice, coleopteran larvae, moths, other dipterans and their details, mites, etc. are illustrated. Bonanni is the first descriptor and illustrator ever of certain insects. Noteworthy, among others, is a dermestid larva, probably of the genus *Anthrenus*, with much enlarged details of the typical hastisetae or spear bristles, and some figures of Diptera Brachycera pretarsi. For his activity as a microscopist he can be likened to the Englishman Robert Hooke (1635-1709), almost his peer, who in London (1667) published a well-known 'Micrographia' from which Bonanni seems to have drawn useful insights and with which we can establish appropriate comparisons.

KEY WORDS: history of entomology, Italy, XVII and XVIII centuries, insects of urban and domestic environment, microscopy.

ORAL PRESENTATION

The entomological writings of an eclectic Venetian scholar, Lorenzo Patarol (1674-1727)

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From a wealthy Venetian family, Lorenzo Patarol (1674-1727) stands out for his plurality of interests, ranging from the history and literature of classical antiquity to the natural sciences. His works reflect both humanistic and scientific skills, in accordance with a unifying view of knowledge. His eclecticism was also marked by his habit of collecting coins, minerals, fossils, shells, and plants. The introductory pages of the 'Opera omnia', published posthumously (Venice 1743), which includes many writings previously unpublished, due partly to the premature death of the author, provide us with biographical information about him. His printed works reveal his interests in Latinity especially: Patarol commented on the 'Declamationes' traditionally attributed to Quintilian (1st century AD) alongside his own 'Antilogiae' in which he defends the reasons of opposing sides, and translated celebratory writings in honor of emperors into Italian, illustrating them with series of effigies drawn from ancient coins. He dedicated another of his writings, in Latin, to the numismatics combined with portraits of emperors and sovereigns, illustrated with engravings of coins from the Roman and Byzantine empires, the Middle Ages and the modern age. In addition to these writings, the 'Opera omnia', includes both a poem in Latin on the silkworm, and a series of short heterogeneous works, in the form of 'Epistolae' or, to a lesser extent, of letters in Italian, addressed to contemporary scholars; these include one on the "Cantaride dei Gigli". Patarol's interest in plants should not be overlooked: he personally took care of a botanical garden attached to his palace in Venice, created a large herbarium and left some handwritten botanical works in Latin. On some pages of the herbarium, for decorative purposes he attached butterflies, moths and other dried insects in a manner similar to that used for botanical specimens, that is, slightly flattened and with the wings outstretched. The didactic poem on the silkworm ('Bombycum libri tres') is accompanied by abundant notes, edited by the author, which render it a treatise on sericulture, revealing his undoubted skills; the contribution continues a centuries-old thematic strand that in Italy has counted various other prominent exponents who not uncommonly expressed themselves in poetic form, along the lines of what was in vogue for beekeeping. Furthermore, Patarol dedicates a long letter, addressed to Vallisnieri, to the "Cantaride dei Gigli"; the insect studied is a chrysomelid identifiable as *Lilioceris* sp. (Lily leaf beetle). He describes its morphology, biology and metamorphosis, partly with the aid of a microscope, adding observations on its behavior and the damage it causes, and accompanying this valuable monographic study with excellent engravings relating to both juvenile forms and morphological details of the adult. Vallisnieri enhanced this work by publishing it in his own works, as he would do with correspondence from other scholars. Finally, it is pleasing to recall that in one of his 'Antilogiae' Patarol takes up a court case from the Latin epoch relating to beekeeping: a reason for contention is the fact that in two neighboring country gardens the rich owner of one cultivated flowers, the poor owner of the other raised honeybees that stole from the flowers of the neighbor, who did not appreciate this and demanded the removal of the beehives. When this did not happen, he scattered poison on the flowers, thus killing all the bees. The arguments of the Latin author, and those of Patarol together with his comments, merit careful reading.

KEY WORDS: history of entomology, Italy, 17th and 18th century, silkworm, lily leaf beetle, beekeeping.

POSTER

The entomologist Arnold von Dobeneck (1867-1926) and his wooden washtub

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In the early 19th century there was a heated debate among chemists between supporters of 'humus' or 'mineral' theories. At that time, the scientific community focused on the role in plant nutrition of the chemical elements absorbed through the roots. It was soon discovered that plant growth was determined by the availability of the scarcest resource: the so-called limiting factor. This principle was known as the Law of the Minimum or 'Liebig's Law', from the scientist who popularized it (starting from 1855) even though he was not actually the real discoverer (Carl Sprengel had already put forward the idea in 1828). Later, the law was applied not only to agriculture but also to ecology in terms of the population and community. In 1903, in the Illustrated Agricultural Journal [Illustrierte Landwirtschaftliche Zeitung] in Berlin, the law was represented graphically through the image of a wooden washtub (later became a barrel), in which each stave represented a 'resource' (originally a chemical element). The washtub has a capacity to contain the amount of liquid corresponding to the height of the lowest stave. This graphic representation was taken from an American textbook and had an immediate success. In the original publication the author of the image was anonymous, but Whitson and Walster, the first authors who reproposed it in 1909, attributed the authorship to "Dr. Dobenecks [sic!]", i.e. Arnold von Dobeneck, the editor-in-chief of the journal. However, today it is one of the most famous 'icons' in plant nutrition sciences and ecology: and is known as Liebig's barrel and Liebig's Law. In fact, Arnold von Dobeneck's role was therefore neglected until fairly recently, but above all what has remained almost completely unknown is his activity as an entomologist. Arnold von Dobeneck (May 30, 1867 - September 15, 1926) belonged to a noble and ancient family of landowners who wished him to be educated in agriculture. He first attended the Bavarian Academy of Agriculture in Weihenstephan (Freising) and then the Department of Agriculture of the Munich Polytechnic. He completed his studies with a research doctorate at the University of Rostock, which ended with a thesis on soil physics. From 1891/1892 he spent a few years at the important Agricultural Institute of Jena dealing mainly with entomology as an assistant to the director von Brümmer. In 1895, on the death of his mentor, he returned to Munich where he collaborated with the newly formed Central Station for Plant Protection. In 1899 he moved to Berlin where he took on the role of editor-in-chief of the Illustrated Agricultural Journal. In the decade in which he dealt with entomology, Arnold von Dobeneck mainly produced technical articles on very varied topics, often focused on methods of control (for example against potato wireworms or grain weevils). His work, due to its placement in non-specialized agricultural journals and the practical-applicative content, did not have a wide resonance despite being precise, documented and sufficiently innovative for the time. An exception is a book on caterpillars, set up with criteria that still appear modern today.

KEY WORDS: 19th century, iconography, scientific publishing.

ORAL PRESENTATION

On the Sardinian Musca macedda (killer fly)

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An imaginary animal, the Musca macedda, occurs in the oral traditions of many Sardinian towns. The famous glottologist Gino Bottiglioni (1887 - 1963) collected the more complete set of information about this insect, he provided also the etymological interpretation of its name. The feminine adjective mac(c)edda (also machedda or maghedda) means butcher, but here the meaning is not literal, killer is a more correct reading (killer fly). At least a second variant occurs (for example in Sorgono): manchedda – from mancu, left-handed, sinister – with the meaning of bad, evil, diabolical (diabolic fly). The morphological features of the Musca macedda are: similar to the house fly, very powerful wings, capable of producing a strong buzzing (and, presumably, a sustained flight), and a fatal sting. The testimonies are discordant about the size, following some traditions the size reaches that of a sheep or of an ox head (for example in Nuchis), but these seem recent local variants. Nothing is known about the natural history of the Musca macedda, it was able to kill the people of whole villages – but in one case it has been defeated by a magic prayerful ritual (in Iglesias) – and it was mainly placed in defence of treasures hidden in underground of tombs, castles, and other archaeological buildings. The testimony from Iglesias also cites the maggots, but without giving information about their behaviour. The symptoms of the sting are unknown, but they are always fatal. Usually, to guard the treasures, many flies are closed inside a container (barrel, coffer) exactly identical to that contains the treasure, for example in Alghero, the tradition talks of three big cruses, respectively filled with gold, pearls, flies. Who accidentally releases them opening the wrong container would obviously cause their own death, but also that of the people of their own town and those nearby (“seven towns” in Lotzorai), the claim that the whole world would end (Nuragus) seems a local exaggeration. It is unknown whether the imaginary Musca macedda originates from some real insect pest or it is only one of the many representations of the fly as a satanic manifestation (Baalzebub was the god of flies). Despite the huge amount of materials published by local tradition lovers and available on the web today, almost nothing can be said about this belief, it has been studied too little scientifically. Two are substantially the hypotheses cited by Bottiglioni, one of which is totally baseless but unfortunately still widely accredited because advanced by himself. In fact, he believes that the anopheles, as malaria vector and therefore lethal, contributed to forming and carrying out the beliefs around the Musca macedda. However, he did not ponder that the discovery of the vector role of this mosquitoes has been too recent to influence the legend. Much more credible is the link between Musca macedda and plague, which actually hit Sardinia several times. Actually, it was sufficiently widespread belief that the flies could be the vectors of the plague.

KEY WORDS: etnoentomology, killer fly, traditions, hidden treasures.

POSTER

Paolo de Simone (1859-1906) a forgotten agronomist in Città della Pieve (Umbria), his historic entomological collection

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Paolo de Simone [= De Simone] (1859 - Roma, VI.1906) was the son of Ferdinando De Simone (Intendant / Gardener of the Royal House in Pisa). In 1885 - 1886, he moved near to Città della Pieve (where he held some public offices: Mayor and President of the Agricultural Club since 1894), and here he built an estate, and developed his agronomy studies. However, in 1900 economic problems forced him to move to Rome with his wife Vittoria Guerrieri (1848-1905), an illegitimate daughter of the King of Italy Vittorio Emanuele II of Savoy and Rosa Teresa Vercellana. De Simone, as well as agronomist (degree in Agriculture Sciences, Pisa), and breeder of thoroughbred horses and exotic plants, was also a good collector of local and exotic fauna and flora, whose materials are still preserved in the Civic Museum of Natural History and Territory "Antonio Verri" of Città della Pieve, located in the sixteenth-century Palazzo della Corgna. The materials were donated by De Simone to the “Scuola Tecnica Pareggiata” of Città della Pieve, and after numerous and troubled transfers, only from 1994 did the reconnaissance and cataloging begin. They are exhibited to the public since the opening of the Museum in 2009. Have come to us, a xylotheque, a rich collection of seeds of vegetables and plants, but also taxidermies of birds and mammals (the latter in few specimens compared to the origin), as well as 5 entomological boxes (object of this note) and a display case with silkworm cocoons and other naturalistic finds, which De Simone evidently raised. The insects of Italian origin (without a specific locality, but evidently: Salci, Città della Pieve and Central Italy) and exotic were originally 216, but today are reduced to 185. They are preserved in five entomological boxes with glass lid, and now displayed in a showcase. The entomological collection is basically divided into “Harmful Insects”, “Useful Insects” and “Exotic Insects” and includes various orders, among which: Coleoptera, Hymenoptera, Orthoptera, Dermaptera, Mantodea and in particular the Lepidoptera, which are gathered in two boxes, also indicating the attached plants, denoting and highlighting the nature and agronomic imprint of the De Simone collection. The collection was certainly conceived also for educational purposes. Although limited, it is still well preserved and is one of the few Umbrian historical entomological collections, thus providing us with a small but precious glimpse of the entomological fauna of these areas at the end of the 1800s.

KEY WORDS: De Simone, historical collection, scientific heritage, Insecta, Museum.

POSTER

Insects and archeology. The well of via Satta in Sassari (Sardinia, Italy)

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The well in Sebastiano Satta road, in the historic center of Sassari, is a source of information about the fourteenth-century Sassari. During the excavations, carried out by the Superintendency of Archeology, Fine Arts and Landscape for the provinces of Sassari and Nuoro, faunal and botanical remains, dating back to the second quarter of the fourteenth century were recovered. The lack of oxygen and the presence of water ensured the conservation of otherwise perishable finds, such as wooden and leather objects, seeds and other vegetable elements. Among the organic finds, there are numerous faunal remains of vertebrates and invertebrates, including numerous parts of insects, the presence of which is to be related to the accumulation in short times of animal and plant waste which constituted an excellent nourishment. The lower stratigraphic units are, from the entomological point of view, the most interesting. The ascent of the aquifer allowed the conservation of numerous anatomical parts such as heads, thorax, elytra, abdominal elements and legs of coleoptera, largely carabids and tenebrionids, but without excluding other orders and families. The well environment was also suitable for the reproduction of swarms of dipterans that left a large number of puparia. The extraordinary preservation and quantity of entomological remains from this well make this find unique in Italy and Europe and an exceptional opportunity for the paleo-environmental studies of a medieval city.

KEY WORDS: Archeoentomology, Archeozoology, Middle Ages, Sardinia, Italy.

ORAL PRESENTATION

SESSION II

Natural history and predatory strategies of antlions and allies

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Antlion larvae (Neuroptera: Myrmeleontidae) are among the most iconic of all insect larvae, thanks to their ferocious appearance and their sophisticated hunting technique, i.e. building a funnel-shaped pit trap in the sand to catch other insects. This remarkable behaviour attracted the attention of early naturalists and allowed to these insects to earn their common name and a place in natural history treatises. However, they represent just one of the predatory strategies set up by Myrmeleontiformia, the most species-rich clade within Neuroptera. Traditionally, Myrmeleontiformia comprised five families: Psychopsidae, Nymphidae, Nemopteridae, Myrmeleontidae and Ascalaphidae, though only the latter three groups include more than 100 species and attain a wide distribution. Their larvae are an important source of phylogenetic data, shedding light on the intricate relationships within this clade. However, recent phylogenomic studies challenge the traditional view, revisiting their affinities with related families of Neuroptera and supporting the paraphyly of Myrmeleontidae with respect to Ascalaphidae. Myrmeleontiformia were much more diverse in the Mesozoic, when several extinct groups existed, showing a greater morphological disparity than extant species but at the same time bearing evidence that modern life strategies and behaviours evolved during the Cretaceous. The larvae of these lacewings are refined ambush hunters, characterized by a series of adaptations to a sit-and-wait hunting technique, such as strongly sclerotized head capsule, large and often toothed jaws and specialized body setae and protuberances. The larvae of nymphids and ascalaphids increase the efficiency of their predatory strategy camouflaging with debris, while nemopterids and myrmeleontids are burrowers living in arid, usually sandy habitats. Fossils suggest that Myrmeleontiformia evolved as arboreal predators, a niche still exploited by the members of several families and later independently invaded soil in different lineages. The colonization of soil proved to be the key innovation for Myrmeleontidae, which are the most diverse lacewing family, with over 2000 described species and a key component of the insect communities of deserts and steppes all around the globe.

KEY WORDS: phylogeny, lacewings, larvae, ethology, evolution.

Evolutionary history of Nemognathinae analyzed by a molecular approach

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Blister beetles (Coleoptera: Meloidae), well characterized by both morphological and biological synapomorphies, are divided into three subfamilies: Eleticinae, Nemognathinae, and Meloinae. Nemognathinae is a cosmopolitan subfamily including the only blister beetles distributed in Australia and in some Pacific Islands. Species belonging to this subfamily own a simple sexual behaviour and oviposition can occur either on flowers or around/inside the bee host nest. The first instar larva is highly adapted to phoresy, apart from Stenoderini, which is thought to be the most primitive tribe. Based on morphology, they were divided into four tribes: Stenoderini (1 genus), Palaestrini (1), Horiini (3), and Nemognathini (28). Using two mitochondrial (16S, COI) and three nuclear markers (CAD, 28S, ITS2), this study aims to describe, for the first time with a molecular approach, the evolutionary history of the subfamily and to establish the phylogenetic relationships among tribes and some genera. Analyses were performed on multilocus and single gene datasets with Maximum Likelihood and Bayesian Inference. Resulting trees showed a great distinction between Stenoderini and other tribes, with Stenoderini recognized as the basal tribe of the subfamily followed by Palaestrini, Horiini and Nemognathini. Sitarine beetles, rather than representing a distinct tribe, as considered by some authors, resulted part of the tribe Nemognathini, as previously proposed based on larval morphology. Most of the genera analysed are monophyletic, but *Stenoria*, *Zonitis* and *Nemognatha*; the Nearctic subgenera of these last two taxa resulted distinct genera. The genus *Zoltanzonitis*, recently described and assigned to Nemognathini, represent a very distinct basal lineage that should probably be considered as a new tribe.

KEY WORDS: Phylogeny, systematics, mtDNA, nucDNA, Bayesian inference, Maximum Likelihood.

Applications of Synchrotron Radiation X-ray Phase-Contrast micro tomography in entomological studies

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Micro-computer tomography imaging is a fast low invasive data acquisition technique suitable for three-dimensional visualizations of high contrast images of biological tissues. We proposed an overview of the main applications of the Synchrotron Radiation X-ray Phase-Contrast micro tomography (SR-PhC micro-CT) in the entomological studies. The attention is pointed out on the SR-PhC micro-CT as a method allowing the imaging and exploration of soft anatomical structures in situ, minimizing sample preparation artefacts related to dissections. The results, obtained through studying the general internal morphology of the abdominal organs in a representative coleopteran species, are compared to light and scanning electron microscopy (SEM) analyses. Virtual dissections and 3D rendering provided more accurate information on the real anatomical location, shape and volume of visceral organs than histological and SEM analyses, respectively. The discussion will be focused on the relevance of replacing or integrating the traditional investigation methodologies with SR-PhC micro-CT analyses to study anatomy of organs involved in crucial life history traits such as reproduction, nutrition and excretion in the ecological, taxonomic, phylogenetic and didactic contexts.

KEY WORDS: micro-CT, morphology, histology, Coleoptera.

ORAL PRESENTATION

DNA-Barcoding of insects: available data and efficiency across different orders

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DNA based species identification has become a widely used approach in entomology both for monitoring and conservation purpose. Molecular identification of insects is mainly done through DNA-Barcoding using as marker a segment of the gene Cytochrome c Oxidase subunit I (COI). One of the most used reference databases for species identification is the Barcode Of Life Data system (BOLD), at present consisting of more than 5,800,000 public records of COI sequences of insects. Despite the implementation of quality filters on sequences by BOLD system, some erroneous sequences are present in the database. Moreover, not all the sequences are associated with species level taxonomy. This project aims to i) develop a database of barcode sequences (COI gene) for insects by filtering the sequences of this class present in BOLD through different criteria in order to obtain an accurate reference of insects' barcodes for species level identification; ii) test the efficiency of DNA-Barcoding for species level identification in different insect orders, using the developed database iii) estimate and compare the optimal threshold values for distance-based molecular identification of species among different insects orders. For these purposes, all the barcode sequences of insects present in BOLD database were mined. These sequences were then filtered: i) for length (> 420bp); ii) for complete taxonomy, including species level information; iii) for contaminants; iv) for lack of open reading frame. The mined database after filtering was composed by more than 1,800,000 COI sequences belonging to 27 insect orders. The most represented order resulted Lepidoptera (630,664 barcodes) followed by Diptera (more than 500,000 barcodes), Hymenoptera (~188,000 barcodes) and Coleoptera (152,430 barcodes), while the smallest are Zoraptera, Notoptera and Zygentoma (56 barcodes per order maximum). The filtered dataset was then aligned using the multiple sequence alignment program MAFFT. The huge amount of data for some of the involved orders (e.g., Coleoptera or Lepidoptera) required the implementation of ad hoc software to estimate optimal threshold values and DNA-Barcoding efficiency. Preliminary analyses suggest that optimal threshold values are highly variable across different orders as well as DNA-Barcoding efficiency.

KEY WORDS: DNA-Barcoding, molecular identification efficiency on insects, cytochrome c oxidase subunit.

POSTER

Factors affecting the efficiency of molecular species delimitation in a species-rich insect family

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In the context of global biodiversity loss, molecular species delimitation approaches could be useful for accelerating insects species discovery before it is too late. In this study, the efficiency of various single-marker species delimitation methods is tested on insects data taking into account the effect on the obtained results of some factors intrinsic to the data itself. Specifically, more than 90 empirical data sets, derived from a set of 7,237 COI sequences attributed to 542 leaf beetles species (Coleoptera: Chrysomelidae), were analyzed through six molecular species delimitation methods (fixed and variable nucleotide distance thresholds, ABGD, ASAP, GMYC, mPTP). The factors evaluated for their possible influence on species delimitations results were: (i) species genetic variability (number of haplotypes per species); (ii) species sampling width (geographic distance among conspecific collection localities); (iii) the difficulty related to the morphological identification of the species, and (iv) the taxonomic rank. Distance-based methods (nucleotide distance thresholds, ABGD, ASAP), with on average more than 70% of match with morphological species, showed higher efficiency than methods based on phylogenetic trees (GMYC, mPTP), with only 59% of match. The presence in the dataset of species with high genetic variability was found to have a negative effect on delimitation efficiency, whereas wide geographic sampling for the species to have a positive one. The results of all methods (except for GMYC) were significantly affected by the presence of species difficult to be morphologically identified, decreasing their efficiency. Finally, the only method influenced by the taxonomic rank of the data set was GMYC, showing lower efficiency in data sets at the genus than at higher levels. The biases highlighted in this study could be accounted for when developing input data sets for insects species delimitation analyses to obtain a more reliable representation of their diversity.

KEY WORDS: Molecular species delimitation, cytochrome c oxidase subunit I, Chrysomelidae.

ORAL PRESENTATION

Molecular and morphological analysis on Euro-Mediterranean Myzocallis aphid species

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The Holartic genus *Myzocallis* Passerini (Hemiptera: Aphididae: Calaphidinae) currently includes 43 species and 3 subspecies, ten genera, three of which native to the western Palearctic region. Most of the species inhabit plants belonging to Fagales s.l. such as *Quercus* and *Castanea* (Fagaceae), *Carpinus* and *Corylus* (Betulaceae), as well as the Myricales, with the genus *Myrica* (Myricaceae). Compared to the more numerous and advanced Aphidini, the Calaphidinae are rather primitive aphids, which are, in conformity with the ancient origin of their host plants among the angiosperms. The species of the genus *Myzocallis* are monoecious and mainly holocyclic. To date, twelve species and one subspecies are known in the western Palearctic region, divided into the subgenera *Agrioaphis, Myzocallis* and *Pašekia*. In this study, the molecular and morphological approach was used to define the taxonomic status of the Euro-Mediterranean species, to verify the current subdivision of the genus into subgenera, as well as the interrelationships with their host plants. The research included all the species currently considered as taxonomically valid and concerned 13 taxa (twelve species and one subspecies) recorded for Europe and the Mediterranean basin. The species analysed are *M. (Myzocallis) occidentalis, M. (M.) carpini, M. (M.) boerneri, M. (M.) coryli, M. (M.) glandulosa* and *M. (M.) schreiberi, M. (Agrioaphis) myricae, M. (A.) castanicola* and its subspecies *leclanti*, *M. (Pašekia) komareki, M. (P.) mediterranea* and *M. (P.) cocciferina*, to which was added *M. (Lineomyzocallis) walshii*, a Nearctic species settled in Europe from a few decades. The molecular analyses included other Calaphidinae closely related to *Myzocallis* s.l., namely *Apulicallis trojanae*, *Tuberculatus eggleri, Hoplocallis ruperti, H. picta, Panaphis juglandis* and *Eucallipterus tiliae*. DNA extraction was carried out following a non-destructive protocol and amplification involved a portion of mitochondrial DNA commonly referred as DNA barcode. The sequences (each consisting of about 650 bp) were aligned and the genetic distance was calculated (p-distance) and graphically displayed (NJ) as implemented in MEGA X, while the molecular delimitation of the species was performed using the ABGD (Automatic Barcode Gap Discovery) model. A couple of sequences were retrieved from GeneBank were also included in the analysis. The results allowed to establish (i) the subgeneric subdivision is probably to be reviewed, (ii) *M. boerneri* and *M. schreiberi* should be considered as separate species, (iii) the status of *M. leclanti* should pass from that of subspecies to species, and finally (iv) the taxonomic status of a population found in Puglia, already reported in the literature as conspecific with *M. occidentalis*, should be reviewed.

KEY WORDS: aphids, *Myzocallis*, molecular analysis, taxonomy.

POSTER

Morphological analysis of the male genitalia of chestnut tortrix moths

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Among the main insect pests affecting sweet chestnut (*Castanea sativa* Miller), the tortrix moths *Pammene fasciana* (Linnaeus), *Cydia fagiglandana* (Zeller) and *Cydia splendana* (Hübner), commonly known as early, intermediate and late chestnut tortrix moths are noteworthy. All these moths have fruit-feeding larvae developing in the fruit, causing early fruit drop. They are responsible for great damage in chestnut production in Europe depending on the geographical area and abiotic conditions. In 2018 in some regions of northern Italy (Piedmont, Liguria, Tuscany and Emilia Romagna), chestnut tortrix moths populations were collected by using sexual pheromone sticky traps, within a three-year project funded by Isagro S.p.A. The collected specimens were counted and placed individually for 24 hours in tubes containing xylene in order to remove the glue from the insects' body. The individuals were then transferred to test tubes containing absolute ethanol and subsequently identified by extraction and observation of the male genitalia under a stereomicroscope. The morphological identification of the species occurred by comparing the characters of the genitalia with images already present in the literature. For cryptic species, identification was also supported by molecular analysis. All the genitalia corresponding to different species were prepared on a slide and subsequently photographed. The identification of the adults highlighted that non target species were present in the traps as well. In particular, in the traps for *Pammene fasciana*, several different species were detected: *P. suspectana* (Lienig & Zeller), *Celypha striana* (Denis & Schiffermüller), *Grapholita* (*Aspila*) *funebrana* (Treitschke), and *Oegoconia novimundi* (Busck). In the traps for *Cydia fagiglandana*, various specimens of the congeneric species *C. splendana* (Hübner) and *C. duplicana* (Zetterstedt) were found, besides *O. novimundi*. In the traps for *Cydia splendana*, some individuals of the species *C. fagiglandana* (Zeller), *Idaea rusticata* (Denis & Schiffermüller), and *O. novimundi* were also found. The results obtained highlight how the incidence of non target species can be high and confirm how the morphological observation of genitalia can be a valid tool for discrimination. Photographs of the genitalia were used to set up iconographic tables, so as to facilitate the recognition of those cryptic species avoiding time- and money-consuming techniques as molecular analysis.

KEY WORDS: *Pammene fasciana*, *Cydia* spp., genitalia, taxonomic identification.

New morphological insights on dermestid beetle hastisetae

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Hastisetae are a specific group of setae, characterizing the larvae of Megatominae (Coleoptera: Dermestidae), located on both thoracic and abdominal larval tergites. These setae constitute the primary defense of the larva against invertebrate predators being a biomechanical trap capable to efficiently block and kill predators (Nutting & Spangler, 1969). Thanks to their unique ultrastructure the hastisetae, single or aggregate, are in fact capable to entangle cuticular structures (spines and hairs) and body appendages (antennae, legs and mouthparts) (Mills & Partida, 1976). Although the hastisetae have been tentatively used as diagnostic character to identify the species, very little is actually known about their fine morphology, their diversity and how certain features are associated to ecology, behavior and evolution of the Megatominae. Due to the specific ecological need of Megatominae larvae (relatively dry environment and low in water content feeding substrate) hastisetae are important contaminant of stored products (fabric and food) and work and living environment. In human, the prolonged exposition to hastisetae, has been shown to determine the insurgence of allergic or inflammation symptoms mainly due to the ingestion or inhalation of the setae (Ruzzier et al., 2020). The purpose of this intervention is to preliminarily present to the public a more detailed view of the morphology and ultrastructure of the hastisetae, focusing the attraction on their biomechanical action.

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Ruzzier E, Kadej M, Battisti A. 2020. Occurrence, ecological function and medical importance of dermestid beetlehastisetae. PeerJ 8: e8340

KEY WORDS: allergy, Coleoptera, systematic, zoology, Dermestidae, health, ecology, insects.

ORAL PRESENTATION

Mechanical ecology in plant-herbivore interaction: role of insect claws of different shape in the attachment to hairy plant surfaces

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During the long period of coevolution between insects and plants, these last developed a wide diversity of features not only to attract pollinators but also to defend against herbivores, such as chemical and physical barriers affecting their performance. In this context, the evolution of plant surfaces and insect pads is an interesting example of competition between insect attachment systems and plant anti-attachment surfaces. Among mechanical defenses, plants developed impenetrable barriers, such as bark and waxy cuticles or thorns and spines, to protect them from herbivores. Non glandular trichomes may impale, entangle or impede the locomotion of insects by physical interactions but some insect species developed adaptations to use them to better adhere to the plant surface. In particular, claws are adapted to interlock with rough surfaces, when the distances between adjacent asperities as well as their heights are larger than the claw tip diameter. The aim of the present study is to clarify the role of claws of different shapes (single claw, single claw with basal tooth, double claws with basal tooth) in the locomotion/adhesion to hairy plant surfaces. In particular the attachment ability of three species of Coleoptera, two Coccinellidae, *Harmonia axyridis*, (Pallas) *Chnootriba elaterii* (Rossi) and one Chrysomelidae *Chrysolina herbacea* (Duftschmid) to natural and artificial substrates characterised by hairy and smooth surfaces will be evaluated. Preliminary results reveal a high attachment ability of Coccinellidae claws (single claw with basal tooth, double claws with basal tooth) to natural and artificial hairy surfaces.

KEY WORDS: mechanical ecology, Coleoptera, surfaces adhesion, trichomes.

ORAL PRESENTATION

Morphological investigation of cephalic glands of three Auchenorrhynca vectors of *Xylella fastidiosa*: a possible source of pheromones?

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Xylella fastidiosa is a xylem-limited bacterium, the causal agent of olive quick decline syndrome. The main vector able to acquire and disseminate the bacterium is *Philaenus spumarius* L., an Aphrophoridae present on both cultivated and wild plants. Apart from this species, other xylem feeding insects are considered as potential vectors of *X. fastidiosa*, such as *Neophilaenus campestris* (Fallén) (Hemiptera: Aphrophoridae) and *Cicadella viridis* L. (Hemiptera: Cicadellidae). Here, we described morphology of the cephalic glands in three species by using scanning, transmission and focused-ion beam electron microscope (SEM, TEM and FIB). The glands distribution differs between Aphrophoridae and Cicadellidae. In *P. spumarius* and *N. campestris*, the apical part of the head is devoid of the sensilla and rich with pores of different size. The frontal part of the head is characterized with a characteristic pattern made of areas covered with characteristic grooved bristles and areas devoid of such structures. The lateral parts, positioned below the compound eyes, are mostly covered with bristles and show abundance of large cuticular pores. In *C. viridis*, the area below the compound eye shows numerous elaborate cuticular structures which resemble floral pattern. The ultrastructural investigation conducted by TEM and FIB revealed connections between external pores and their internal structural organization. We discovered and described up to three different types of glands. All of them showed elaborated cuticular ducts of different lengths, and a secretory cell connected to the evacuating duct through an end apparatus. The possible functional implications of these glandular structures, reported for the first time in these insects, is discussed in the context of their biology.

KEY WORDS: Ultrastructure, SEM, TEM, FIB, insect vectors, *Philaenus spumarius*, *Neophilaenus campestris*, *Cicadella viridis*.

POSTER

Preliminary investigation on the morphological differentiation of *Aedes geniculatus* and *Aedes sticticus*

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The genus *Aedes*, widely distributed on our territory, includes a relatively large number of species, some of which are potential vectors of diseases. The adults of some *Aedes* species are not easily distinguishable only through morphology. Moreover,often fragile and subject features are frequently lost especially in specimens collected by aspiration traps such as CDC light trap and BG sentinel®. In this kind of traps, in fact, the mosquitoes can loselegs, bristles or scales essential for identification. An entomological investigation was recently carried out on three very similar species, namely *Aedes (Finlaya) geniculatus* (Olivier), *Aedes (Finlaya) echinus* (Edwards) and *Aedes (Ochlerotatus) sticticus* (Meigen). Among these species *Ae. geniculatus* is mentioned in recent studies as potential vector of viruses such as Chikungunya. Morphological characters of larvae are resistant to deterioration and allow to better determine the species. However as these species occur in woodlands (two of them are dendrolimnic) and the collection of their larvae is very difficult, the identification of adult specimens, often collected by traps, is required. In this preliminary study, with the collaboration of different IIZZSS, a great number of adult specimens, previously identified at the genus level or only as *Ae. geniculatus* has been collected in different locations and regions of central-northern Italy. From the morphological observation it was highlighted that some characters, currently used to distinguish females of these species, often overlap or are not visible due to the deterioration of the sample. For this reason, in order to identify the species with higher reliability, alternative or complementary characters have been selected. To highlight any difference and selecting the better preserved characters, those present on the head, thorax, abdomen and legs were in-depth examined. Furthermore, to strengthen the validity of the selected morphological characters and in order to confirm the morphological identification, a further molecular analysis, using the mitochondrial COI gene as a marker on a sample of mosquitoes (12%), was carried out. Up to now, out of 636 mosquitoes, 248 specimens (39%) have been morphologically analyzed; 66% of them belongs to the species *Ae. sticticus* and the remaining 34% to *Ae. geniculatus*. In light of these preliminary results, it would be useful to deepen this investigation on a larger sample from the whole Italian territory. Furthermore, this data area relevant contribution to the current knowledge on the presence and distribution of these species and to update the identification keys of Italian mosquitoes, currently used. This study was partially funded by the YYY project IRCCS 2017 "Rilevamento precoce di zanzare esotiche e caratterizzazione dei biotipi endemici: citizen science, biologia molecolare ed epidemiologia al servizio di una sorveglianza mirata." Prot. IZS PLV 03/17 RC.

KEY WORDS: Chikungunya, morphology, identification keys.

POSTER

An integrated, morphological, biological and molecular approach reveals four cryptic *Trissolcus* species, egg parasitoids of Pentatomidae. How to identify them?

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A study on Palearctic species of the genus *Trissolcus* (Hymenoptera: Scelionidae) was conducted to support research on the biological control of *Halyomorpha halys* (Stål) (Hemiptera: Pentatomidae). An updated taxonomic review for a cluster of morphologically similar species combined a reassessment of morphological characters and the examination of primary types, with molecular analysis and mating experiments. *Trissolcus semistriatus* (Nees von Esenbeck) sensu Talamas et al. (2017) has resulted including three valid species: *T. belenus* (Walker), *T. colemani* (Crawford) and *T. manteroi* (Kieffer). A morphological analysis of *Trissolcus* specimens emerged from eggs of *Eurygaster maura* (L.) and *Carpocoris* spp. (Hemiptera: Pentatomidae), and the comparison of them with images of species primary types finally reveals new reliable features to distinguish *T. semistriatus* from *T. belenus* [senior synonym of *T. grandis* (Thomson)]: anteroventral area of hind femora (covered or not entirely covered by setae), mesoscutal humeral sulcus (present as a simple furrow or comprised of cells), setation of the first laterotergite (absent or present), medial part of occipital carina (angled or rounded), anteroventral extension of metapleuron (short or extending to base of mesocoxa). Mating tests between *T. semistriatus* and *T. belenus* showed a complete reproductive isolation. Molecular analyses, obtained amplifying the cytochrome oxidase I gene, carried out on the four species plus *Trissolcus rufiventris* (Mayr), reveal genetic distances confirming the validity of all species. Two morphological characters, previously considered as variable within *T. semistriatus*, i.e. the shape of the mesoscutal humeral sulcus and setation on the first laterotergite (Talamas et al., 2017), are revaluated. Additionally, we analysed the primary types of two important, but largely ignored, historical Walker's species: *Trissolcus belenus* and *T. arminon* (Walker), resulting this latter as junior synonym of *T. belenus*. Based on the results, two points were added to the key to species of Palearctic *Trissolcus* (Talamas et al. 2017); all species belonging to *T. basalis* species-group are now identifiable on a morphological basis. Given the difficulties in the identification of the minute characters, the observation must be done exclusively on specimens mounted on card point under a stereoscopic microscope at considerable magnification, shielding the light to avoid reflections. Otherwise, it is easy to fall back into the nebulous specific identification, with a probable only reliable interpretation: *Trissolcus* sp. Identification at a specific level must concern all the specimens emerging from the same egg mass. Also, for some species other than those of this study, identification on a genetic basis is possible, with all the related difficulties.

KEY WORDS: Scelionidae, *Trissolcus belenus*, *Trissolcus colemani*, *Trissolcus manteroi*, *Trissolcus semistriatus.*

POSTER

SESSION III

A salivary chitinase of *Varroa destructor* influences host immunity and mite's survival

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Varroa destructor creates a wound in the host's cuticle through which it feeds on haemolymph and fat body, representing an important stress factor that weakens honeybee colonies and promotes the spreading of diseases. In order to facilitate feeding, this ectoparasitic mite delivers a complex of factors with its salivary secretions. The characterization of these factors is still largely elusive and any progress in this area will offer new insights into the molecular basis of *Varroa*-honeybee interactions, on which to develop new sustainable strategies of mite control. Here, we have used a functional genomics pipeline to identify *V. destructor* salivary proteins putatively involved in the regulation of host physiology, and their expression in salivary glands has been assessed by qPCR and in situ hybridization. This approach allowed the identification of a salivary chitinase, which was subsequently studied from a functional point of view. In vivo studies were based on gene knockdown followed by artificial infestation of honeybee pupae. The effectiveness and the time course of the silencing were assessed by qRT-PCR and associated with the observed level of mortality on experimental Varroa mites. To study the effect of the salivary chitinase of *V. destructor* on honeybee gene expression, we analyzed by RNA seq the transcriptome of worker pupae in response to parasitism by silenced mites. The results obtained indicate an important role of this enzyme in the modulation of host immune response.

KEY WORDS: chitinase, salivary glands, parasitic mite, gene silencing, host- parasite interaction.

ORAL PRESENTATION

Hermetia illucens larvae can exploit substrates with different nutritional quality thanks to the morphological and functional adaptability of the midgut

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The use of insects as a primary agent for organic waste reduction and bioconversion can contribute to the management of organic waste and, at the same time, represent a source of valuable products, such as proteins for feed production, biodiesel, bioplastic and bioactive molecules. In this setting, the larvae of black soldier fly (BSF), *Hermetia illucens* (Diptera: Stratiomyidae), have captured great attention because of their capacity to grow on a variety of waste organic matter and bioconvert this material into valuable products. Even though composition, moisture, and nutrient concentrations of the rearing substrate can significantly affect insect performances and biotransformation processes, the astonishing adaptability of this insect to dietary substrates without any dramatic impact on its development strongly suggests that BSF larvae can finely and profitably regulate nutrient intake and post-ingestion processes to match their nutritional requirement.
Here we address this issue by investigating if and how the midgut of BSF larvae, the organ involved in the digestion and absorption of nutrients, sets in motion post-ingestion responses to compensate variations in nutrient composition of the diet. We chose two rearing substrates: a nutritionally balanced diet for dipteran larvae and a nutritionally poor diet that mimics fruit and vegetable waste composition. Our data show that larval growth performance was affected only moderately by the unbalanced substrate because a diet-dependent adaptation process of the midgut ensured its exploitation; in particular, differences in cell morphology, activity of digestive enzymes, and accumulation of long-term storage molecules were observed. Moreover, transcriptome analysis of midgut samples from larvae reared on the two substrates showed that genes encoding proteins with important roles in midgut function were differentially expressed, confirming the functional adaptation of this organ. Our data demonstrate that the functional plasticity of the midgut has a key role in the ability of BSF larvae to grow and develop on substrates with low nutrient content.

KEY WORDS: *Hermetia illucens*, larval midgut, functional adaptability.

The characterization of cuticular hydrocarbons of *Dasineura oleae* showed differences among sexes and ages

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Insect cuticular hydrocarbons (CHCs) have always been considered essential to withstand desiccation stress and avoid a lethal rate of water loss, since they affect the cuticle permeability depending on their chemical nature. However, in several insect groups, CHCs have also a second functionality concerning the chemical communication as they may serve as sexual pheromone, kairomones, species recognition cues, nestmate recognition, dominance and fertility cues and chemical mimicry. The study of CHCs is an important tool for species characterization, age and sex determination, improving the control strategies of those species that have a high impact on agriculture and are cryptic to identify (e.g. *Ceratitis* species). *Dasineura oleae* (Diptera: Cecidomyiidae) is a leaf gall midge specific of *Olea europaea* and native of the Mediterranean area, generally kept under control by its parasitoid complex. As recorded for several cecidomyiids, outbreaks occur even in *D. oleae*, causing strong foliar infestation and plant stress. In this work, we analyzed the cuticular hydrocarbon profile of virgin D. oleae by gas chromatography coupled with mass spectrometry (GC-MS), characterizing both females and males of different ages (i.e. 0-12 h, 12-24 h, 24-36 h). In D. oleae, females and males can be easily distinguished for the presence of sexual dimorphism (i.e. abdomen color, antennae). A total of 41 compounds were identified, 5 of them specific of the female sex, while other compounds are more representative in one of the two sexes. Moreover, the quantity of fifteen compounds changes in relation to the insect age, and some of these ones (e.g. fatty acids) decreased significantly as age increased, both in females and males. Further studies are planned to investigate also CHCs of mated individuals, and efforts will be made to investigate how the CHCs of D. oleae influence its ecology and sexual behavior.

KEY WORDS: *Dasineura oleae*, cuticular hydrocarbons, GC-MS, olive tree.

POSTER

Olfactory perception of *Hermetia illucens*: a source of inspiration for the development of new biosensors for the monitoring of agri-food products shelf life

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The study of insect physiology at the molecular level and the implementation of innovative technologies, allows the development of applications able to cope with many human issues, through the use of biotechnology. The innovative aspect consists of the ability to exploit natural and physiological phenomena, as a source of inspiration for biotechnological applications, useful for improving the quality of life. An example in this sense is represented by the olfactory perception of the diptera *Hermetia illucens*, a saprophagous insect that has aroused considerable interest in the world for bioconversion of organic wastes. It can be considered a scavenger of organic matter: females oviposit on or near decaying organic matter and, after hatching, larvae feed until the pupal stage. They are able to feed on a wide variety of decomposing organic material, both of animal and vegetable origin. We focused on the Hermetia illucens olfactory perception identifying and functionally characterizing the Odorant Binding Proteins (OBPs) that mediate the chemoreception of the most attractive volatile organic compounds (VOCs) found in decaying organic matter. The use of OBPs specific for VOCs of organic decomposition have been proposed as sensing layers of innovative Quartz Crystal Microbalances (QCMs) sensors, for monitoring food products preservation and shelf-life monitoring, in a new approach to food logistics. The identification of *Hermetia illucens* OBPs (HillOBPs) has been carried out through an "omic" approach, based on RNA-seq, de novo assembly and annotation of larvae, male and female adults transcriptome. We have chosen 15 HillOBPs expressed both in larvae and adults. A bioinformatic approach has solved the HillOBP 3D structures and the molecular docking technique has been used to calculate the interaction energies and the affinities with the chemical structures of 45 identified VOCs, emitted by different decomposing substrates colonized by *H. illucens* larvae. These preliminary in silico analyses have confirmed the ability of HillOBPs to bind VOCs and useful binding sites for the rational design of the QCM-based biosensor have been identified. These data were then subsequently confirmed for 4 HillOBPs with the use of QCMs. The first four HillOBPs cloned, expressed and purified, were immobilized on the golden lamina of QCMs via a self-assembled monolayer and molecular interactions between HillOBPs and VOCs in vapour phase were measured. A QCM device is an extremely sensitive mass balance that measures nanogram to picogram level changes in mass in real time. The heart of technology is a piezoelectric material that oscillates at a defined frequency (in this case 20 MHz). The mass increases due to the protein-ligand complexes leading to a proportional reduction in the resonance frequency of the microbalance described by the well-known Sauerbrey equation. The experiments conducted so far provided useful information about biosensor selectivity, as the ability to discriminate ligands of different nature. Four compounds indicative of lipidic decomposition (Isovaleraldehyde, Isobutyraldehyde, 2-Methylbutyraldehyde, Butyric Acid) have been detected with high affinity in a pool of 45 VOCs tested, known to be spoilage markers. Further tests to verify the QCM-based biosensor stability and its limits of detection are on-going.

ORAL PRESENTATION

Insect antimicrobial defences: attacins from the stick insect *Carausius morosus*

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Insects produce a wide range of proteins and antimicrobial peptides (AMPs) as a first line of defence against pathogens. AMPs are rapidly and transiently synthesised in holometabolous insects by the fat body and secreted into the haemolymph, from where they can easily diffuse toact throughout the whole animal. On the contrary, in heterometabolous insects, AMPs are produced by haemocytes and secreted into the hemolymph upon infection. As for most AMPs, these peptides are produced from a large precursor containing a signal domain which is cleaved in vivo to produce the active protein with antimicrobial activity. Based on their structural features, insect AMPs can be divided into three main groups: 1) cecropins, peptides containing an α-helix structure with no cysteine residues; 2) defensins, containig 6–8 conserved cysteine residues, 3 or 4 disulfide bridges and usually 3 distinct domains; 3) peptides with an over representation of proline/glycine residues. Attacins are glicin-rich peptides that have been discovered in *Hyalophora cecropia*. They are a rather heterogeneous group of proteins usually active mainly against Gram-negative bacteria. The AMP sequence, named carausicin, has been identified recently in transcriptome from the malpighian tubules of the phasmatodean stick insect *Carausius morosus*. Once analysed this sequence, we assigned it to the attacin family. We then synthetized the putative mature attacin peptide to investigate further its structural and biological properties. The peptide was able to cross Gram-negative outer membrane. Moreover, peptide interaction assay with model membranes highlighted a pH dependent behaviour. Preliminary studies did not evidence antimicrobial activity against both Gram-negative and Gram-positive bacteria, and haemolytic activity against rabbit erythrocytes. However, it showed positive effects on viability of mammalian cell in vitro which deserve to be investigated. Also, further studies will be necessary to evaluate the in vivo activity of the carausicin peptide upon insect immune stimulation.

KEY WORDS: antimicrobial peptides, *Carausius morosus*, biological activity, carausicin.

POSTER

Energetic budget of the malaria mosquito *Anopheles coluzzii* reared in semi-field environments

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Anopheles coluzzii Coetzee & Wilkerson is a major vector for malaria transmission in Sub-Saharan Africa. Understanding the ecology and physiology of this mosquito in laboratory versus natural conditions is crucial for comprising its vector potential and developing integrated vector management tools. We reared two strains of *A. coluzzii i*n standard small rearing cages and in especially designed large indoor cages to simulate natural environments, and measured nutritional profiles of females and males to assess energetic costs associated with rearing settings. We detected a strong cost of glycogen for adult females reared in semi-field condition, and a similar trend for lipids, but not for males. We speculate that results reflect the insects foraging behavior and reproductive biology, and conclude that rearing mosquitoes in semi-field environments mirror the insect' physiological and behavioral features in natural habitats, thus enabling effective manipulation in controlled environments.

KEY WORDS: malaria mosquito, *Anopheles*, energetic costs, insect reproduction, insect behavior.

POSTER

Mode of action and toxicity of mint, anise, fennel, sage and lavender essential oil nano-emulsions on *Drosophila melanogaster*

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Damages caused by insect pests are among the most challenging threats for food production. Currently they are managed mostly by chemical control, but this approach is not sustainable in the long-term due to concerns regarding the safety of pesticides, their accumulation in the food chain and their impact on non-target organisms. The development of Integrated Pest Management strategies could represent a science-based solution to this challenge. Hence, it is urgent to find environmentally friendly alternatives to integrate them with other approaches, either chemical or biological to reach a successful control of pests. In this scenario, the development of plant derived formulates based on essential oils (EOs), with selective activity against certain pest species, may have a leading role. The aim of this research was to develop stable nano-emulsions based on several EOs and to report their effect on insect physiology investigating their impact on the expression of several genes. *Drosophila melanogaster* (Meigen) (Diptera: Drosophilidae) was chosen as model organism given the vast knowledge of its physiology and because there is full coverage of its genome. Stable nano-emulsions based on mint (*Mentha piperita*), anise (*Pimpinella anisum*), fennel (*Foeniculum vulgare*), sage (*Salvia officinalis*) and lavender (*Lavandula angustifolia*) EOs have been developed to evaluate their insecticidal activity against *D. melanogaster*. The experimental nano-emulsions showed an average negative surface charge of ζ 11.5-23.9 mV and a droplet size within the sub-micrometre range (110.9-141.5 nm). The developed nano-formulations were tested as fumigants and all of them showed acute toxicity against *D. melanogaster* unsexed adults, being anise the most toxic (LD95=0.006 mg/ml of air). To investigate the mode of action of anice EO nano-emulsion on the fly, several genes were selected based on previous knowledge regarding their association with the neurologic function or with detoxification. Thus, the differential expression of Tyramine-beta-hydroxylase, Gamma-aminobutyric acid receptor Gamma-aminobutyric acid transaminase, Acetylcholinesterase, Alcohol dehydrogenase, Arylalkylamine N-acetyltransferase, Glutathione S transferase S1 (GstS1), Microsomal glutathione S transferase (Mgstl), Vacuolar H[+] ATPase 68 kDa subunit 2 and Tubulin (as housekeeping), was assessed via quantitative RT-PCR on *D. melanogaster* treated with anise in comparison with a non-treated control. Our results showed no variation in the expression of genes associated with the neurological function, but did show a significant variation in GstS1 and Mgst1, both involved in detoxification pathways. Further investigations assessing the implication of other key players in the detoxification pathways (like cytochrome P450 monooxygenases) or a full transcriptomic characterization of the response to EOs exposure would increase our understanding of the mode of action of essential oils.

KEY WORDS: botanicals, insect physiology, detoxification.

ORAL PRESENTATION

Antennal transcriptome analysis and identification of candidate olfactory genes in the harlequin ladybird beetle, *Harmonia axyridis*

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In predaceous Coccinellidae, olfaction is extremely important for courtship, mating and for locating suitable habitats for feeding and oviposition. However, despite the huge existing literature, very little is known about chemosensory genes expressed in adults' antennae. The harlequin ladybird, *Harmonia axyridis*, was widely released for classical and augmentative biological control programs against herbivorous pests and is now worldwide distributed. Because of its invasive behaviour and the threat that it can represent for local biodiversity, this ladybird has been adopted as a model species for invasive biological control predators. We assembled the antennal transcriptome of *H. axyridis* male and females using NextSeq 500 Illumina sequencing and Galaxy platform. We obtained 30600 contigs of size higher than 500 bp. The analysis allowed identification of candidate chemosensory genes, including odorant receptors (28 ORs, including the coreceptor, Orco), gustatory receptors and odorant-binding proteins (OBPs). Phylogenetic analyses were conducted including sequences from other beetle species and using maximum likelihood method. The analyses allowed the inclusion of candidate *H. axyridis* chemosensory proteins in subfamilies known for Coleoptera. Differential expression analysis revealed low variability between males and females, possibly reflecting the known absence of sexual dimorphism in the structure of the antennae and in the distribution and abundance of the sensilla. Several transcripts were however differentially expressed, including a male-biased OR and 2 OBPs. Our work brings essential data for understanding the molecular basis of chemosensory perception in Coccinellidae.

KEY WORDS: RNA-seq, chemosensory genes, biological control, invasive species.

POSTER

Evolution of heat shock protein-90 gene structure in nematodes

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Heat shock proteins (HSP) belong to a protein family that are rapidly synthesised in response to a series of environmental stressors. Among all HSPs, HSP90 is the most conserved and abundant in cells and is involved in response to temperature stress. Temperature affects life stage development and behaviour of nematodes, with different species and strains having optimum temperatures for feeding, survival and reproduction. A portion of the hsp-90 gene was amplified and sequenced in free-living, entomopathogenic and plant parasitic nematodes. Sequence analyses of the hsp-90 products showed the existence of two isoforms characterized by presence/absence of introns in nematodes. Furthermore, the exon-intron structure of hsp-90 genes varied among nematodes including the number of introns, intron position and length. In nematodes hsp-90 introns are relatively short compared to vertebrates suggesting that introns are under strong selective pressures. Interestingly, a higher number of introns in hsp-90 genes of parasitic nematodes compared to free-living nematodes is observed. These findings allow us to speculate that introns are gained and loss in the evolution of nematode lineages and may also contribute to adaptation to environmental changes. The duplication of hsp-90 gene in parasitic nematodes may indicate its predisposition to exploit climate changes. Further investigations are ongoing to evaluate their evolutionary implications among different groups of nematodes.

KEY WORDS: exon, intron, sequencing, invertebrates.

POSTER

Preliminary results on the genetic diversity of *Liothrips oleae* in olive growing areas of southern Italy

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In recent years, the use of molecular biology for systematic studies in the *Thysanoptera* order, as well as in the study of particular aspects of the life cycle of pest species, has proved to be auxiliary for the understanding of some biological events occurring within agricultural ecosystems characterizing the crops of the Mediterranean regions. The systematic use of pesticides and current climate changes are the main causes that trigger the alteration of biological balances within complex agroecosystems, including olive groves. The aim of the present study is to investigate the genetic variability within natural populations of *Liothrips oleae* (Costa) living in two of the most intensive southern Italian olive producers (Calabria and Sicily), through the use of molecular methodologies, in order to study the massive infestations that have occurred throughout the Ionian coast over the past three years. The species, known as secondary pest in the olive crop, because its low population densities that have never required specific treatment for their containment. The knowledge of the biological cycle and the genetic identification of the species might provide suitable instruments in order to implement the best sustainable defence strategies from an economic and environmental point of view. In the present work, the results related to the first molecular characterization of L. oleae are reported. Three coding genetic regions already extensively used for the genetic identification of many insect species were employed to detect genetic groups within the olive thrips populations from olive trees. Some differences were particularly obvious in the mitochondrial COI (Cytochrome Oxidase Subunit I) and ITS2 (Internal Transcribed Spacer 2) ribosomal region, while the 28S-D2 ribosomial region was confirmed as the most conserved site in the various populations present in the Calabrian and Sicilian areas. The results obtained question the systematic position of the species that has been poorly investigated to date, as it has always been monophagous on the olive trees and has never caused significant damages to leaves and fruits. Further studies, based on the integration of molecular and systematic data, are needed to provide clearer informations on the species, which is replacing the key-pest of the crop, in some areas of both the regions investigated.

KEY WORDS: *Liothrips oleae*, secondary pest, molecular characterization, genetic variability.

POSTER

The plant-pollinator-nematode relationship: histological observations relating to the *Ficus carica-Blastophaga psenes-Schistonchus caprifici* system

Alessio Vovlas

A.P.S. Polyxena

The present work illustrates the histological observations made on the Hymenoptera Agaonide *Blastophaga psenes* and on the floral tissues of the caprific *Ficus carica* var *sylvestris*. In the context of the species-specific fig-pollinator obligatory mutualistic relationship, it is interesting to underline the important insertion of a third element, the phytoparasitic nematode *Schistonchus caprifici* (Aphelenchoididae), introduced in the syconium, by passive forhesis, using the winged female of the insect pollinator as a carrier. Within the "fig system" framework of this tritrophic mutualistic relationship, flower tissues and individuals of *Blastophaga psenes* were histologically analyzed and microscopically examined. In the present work, the presence of the nematode within the galls of the infructescence and in the abdomen of the females of *Blastophaga psenes* is confirmed for the first time histologically. The vector transfers pollen and nematodes to the ovary, which is subsequently transformed into a gall, ensuring nutrient through cell hyperplasia for the subsequent development of the larva. The wasp lays a single egg per flower, transforming it into a gall and passively dispersing the nematodes. From mature galls the adults of *Blastophaga psenes* emerge, with a ratio of 1: 4 between males and females. Furthermore, evident anatomical floral alterations have been observed in close proximity to single or groups of nematodes. The histological reactions induced by the nematode following trophic activity consist in a unique form in the context of Aphelenchoididae, with ectoparasitism at the epidermal tissue level. These reactions, in which hypertrophic tissues of the first cellular outer layers are formed, are in contrast with the classic manifestations of Aphelenchoididae, which mainly express endoparasitic and necrotic characteristics. Finally, new data relating to phytoparasitism and the consequences related to the presence of the parasite are illustrated and discussed.

KEY WORDS: Agaonidae, Entomogenous Nematode, Fig, Histopathology, Host-parasite relationship.

POSTER

Wolbachia Infection dynamics in the invasive *Rhagoletis cingulata* in Europe

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The Eastern cherry fruit fly, *Rhagoletis cingulata*, is a significant economic pest species in North America and has recently been introduced in Europe. Here, it co-infests cherries with the native European cherry fruit fly, *Rhagoletis cerasi*. This shared habitat has resulted in two independent horizontal transfers of a native *Wolbachia* strain, wCer1, from *R. cerasi* to the invasive *R. cingulata* in Germany and Hungary. Although intraspecific horizontal *Wolbachia* transmissions have been documented in various insects on an evolutionary time scale, direct evidence for the
horizontal transmission of *Wolbachia* on an ecological timescale in nature is rare. Our system provides a unique opportunity to resolve the spatial dynamics of directional horizontal Wolbachia transfers in natural populations. Our system also allows us to investigate the role of *Wolbachia* infection in invasiveness by comparing measured temporal infection dynamics to expected values under classical Wolbachia infection models. Using a combination of short-read (Illumina) and long-read (PacBio and Nanopore) sequencing, we first assembled a dozen *Wolbachia* genomes from individuals of the invasive (wCin2 strains) and the native fly species (wCer1 strains). We compared these strains and found that major structural inversions and duplications have resulted in differences in cytoplasmic incompatibility factor genes between the two strains. These genes are essential as they determine the *Wolbachia* spread dynamics through the invasive *R. cingulata* populations. In addition to these two main *Wolbachia* strains, we also found and characterized three minor strains (wCer2, wCer4, and wCer5) in the native *R. cerasi* species. We hypothesize that these strains might interact and lead to complex dynamics within *R. cerasi* local populations due to multiple combinations of different cytoplasmic incompatibilities factors. After comparing the invasive *R. cingulata* regional German and Hungarian strains to North American strains, we found significantly more SNPs in Germany. These results strongly suggest that the German invasion route is older than the Hungarian route. This work paints a picture of a highly dynamic system where *Wolbachia* plays a significant role in the invasive potential of *R. cingulata* in Europe. It also highlights the role of horizontal Wolbachia transfers between species as a mechanism for ecological novelty.

KEY WORDS: *Wolbachia*, invasive species, horizontal transfer, prophage, *Rhagoletis cerasi*, *Rhagoletis cingulata*.

ORAL PRESENTATION

SESSION IV

Behavioral manipulation of the meadow spittlebug *Philaenus spumarius* by means of vibrational signals

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Pest control strategies involve the manipulation of the sexual behavior of insects, by means of species-specific vibrational stimuli transmitted to plants. It is then possible to control the population of the target pest and to develop either "push-and-pull" or "lure-and-kill" strategies. *Philaenus spumarius* (Hemiptera: Aphrophoridae) is the vector of the causal agent of the Olive Quick Decline Syndrome (OQDS). This insect emits vibrational signals through the host plants to communicate and achieve mating. Three playback trials with mini-shakers were conducted to evaluate if vibrations could affect the behavior of the insect, and therefore their potential application for management practices. In particular, we hypothesized that the transmission of certain playbacks could either lead the insect toward the source of the signal or repel it from the plant (test 1) or disrupt their mating behaviour (tests 2 and 3). In all trials, vibrational signals emitted by the specimens were recorded with a laser vibrometer. In test 1, a *P. spumarius* male or female was tested with two types of male calling signal (MCS), which were transmitted for 15 minutes to a Vicia faba plant. We assessed whether the insects either moved toward the stimulus or left the plant. The MCS were played in two distinct period to evaluate whether the effects of the signal could be different before (June-July) and during (September-October) the *P. spumarius*' mating season, respectively. Test 2 took place between August and September. Pairs consisting of a female and a male were released on a *Helianthus annuus* plant and stimulated for 30 minutes to disrupt the pair formation process. The stimuli consisted in a playback of either two pre-recorded signals or a series of white noises (frequency range 0-2100 Hz). In test 3, in October, pairs were treated for 30 minutes with a continuous white noise to assess whether the signal could disrupt mating. Our results indicate that MCS does trigger neither attraction nor repellent signal was excluded, while the continuous white noise significantly reduced the mating success of *P. spumarius*. Results of the playback trials are presented, as well as a description of the mating behavior of the meadow spittlebug. Insights on the potential future development of vibrational control strategies against this pest are given.

KEY WORDS: vibrations, meadow spittlebug, mating disruption, pest management.

ORAL PRESENTATION

Contrasting effects of exotic plant invasion and managed honeybees on specialization of plant–pollinator networks

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In the last decades, the potential effects of invasive plants on plant–pollinator interactions have been intensively studied, but often in isolation from other biotic and abiotic drivers and without considering the response of the whole pollinator community. *Buddleja davidii* Franch. is a highly invasive perennial shrub native from China that is invading several low elevation areas of the Alps. Its reproduction depends on insects, and it is therefore very attractive to pollinators. The managed honeybee (*Apis mellifera* L.) is considered the most important pollinator in many natural and agricultural ecosystems. However, its potential effects on wild pollinators are still controversial. Here, we explored the potential interaction between the highly invasive plant species *B. davidii* and managed honeybee density on plant–pollinator interactions over the whole elevational range distribution of the exotic plant in the Alps. We selected 9 pairs of sites with/without *B. davidii* over an elevation gradient of 1000 m ca in Veneto and Trentino-Alto Adige, and sampled plant-pollinator networks 5 times between June and September 2018. In each site, we identified all flowering plants and the entire pollinator community (i.e. all the hymenopterans, dipterans and lepidopterans touching the reproductive parts of flowers). We then built the plant–pollinator networks and calculated some of the most commonly used metrics of specialization both at the network- and the species-level. Using linear mixed-effect models, we tested how the invasion of *B. davidii* and the abundance of managed honeybees, in conjunction with the total number of flowering plant species, affected plant–pollinator network topology and species-level specialization of pollinators.
Overall, we recorded 9563 visits between 370 pollinator species and 150 flowering plant species. The honeybee was the most abundant pollinator, accounting for 43% of the whole visits. We found no interactive effect of *B. davidii* and honeybee density on both network- and species-level metrics. However, *B. davidii* presence and honeybee abundance strongly influenced species-level specialization in contrasting ways. Pollinator specialization was lower in sites invaded by the alien plant compared to control sites, while increasing honeybee density generally increased specialization of most pollinator groups. All the observed effects did not change along the elevational gradient. Our results suggest that increasing honeybee density can deconstruct plant–pollinator interactions irrespective of floral resource diversity by forcing species to switch to other plants.

KEY WORDS: *Apis mellifera*, bipartite networks, competition, exotic species, elevation, specialization.

ORAL PRESENTATION

New data about "rubbing behavior" in some species of carabid beetles

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Carabid beetles exhibit many defensive strategies to ward off enemies and among these, gregarious carabids use anti-predator strategies simultaneously against pathogenies and predators. The aggregations of aposematic species with similar color patterns are quite rare in nature but the colonies of *Brachinus* spp. and *Anchomenus dorsalis* are well known. *Brachinus* spp. (bombardier beetles) have aposematic colors and chemical defenses. *A. dorsalis* has a color pattern like *Brachinus* spp. and if "disturbed" it can use undecane as chemical defense. Inside these groups, interspecific interactions between *A. dorsalis* and *Brachinus* spp., known as "rubbing behavior" was described by Zetto Brandmayr et al. (2006). Despite this mechanism, many data must be investigated. The aim of this research is to verify the ability of individuals of *A. dorsalis* to select the species vs which display the rubbing. To investigate the nature of the stimulus that provokes rubbing behavior we selected the morphological characteristics (color and size patterns) and the defensive mechanisms of a total of 22 species of carabid beetles. Rubbing behavior was observed only towards aposematic and protected species while it never occurred towards non aposematic and non-protected species. More work is needed to find an evolutionary significance of this behavior. We believe that this positive interaction should be a vantage for all involved species, but more behavioral and chemical studies will be needed

KEY WORDS: carabidae, behavior, interspecific interactions.

ORAL PRESENTATION

Exploiting chemical ecology to manage hyperparasitoids in biological control

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Insect hyperparasitoids are fourth trophic level organisms that commonly occur in terrestrial food webs, yet they are relatively understudied. There is an increasing awareness that these top-carnivores can disrupt biological pest control by suppressing the populations of their parasitoid hosts leading to pest outbreaks. This is particularly evident in confined environments such as greenhouse crops where augmentative biological control is used. Currently there is no effective eco-friendly strategy that can be used to control hyperparasitoids. Recent advancements in the chemical ecology of hyperparasitoid foraging behavior have opened opportunities to manipulate these top-carnivores in such a way that biological pest control becomes more efficient. In this review, we propose various infochemical-based strategies that can be used to manage hyperparasitoids. In particular, we suggest that a push-pull strategy could be a promising approach to 'push' hyperparasitoids away from the crop and 'pull' them into traps. Infochemicals can also be implemented in biological control: 1) to restrict accessibility of resources (e.g. sugars and banker plants) to primary parasitoid only, or 2) to monitor hyperparasitoid presence in the greenhouse for early detection.

Biodiversity of bees (Hymenoptera, Anthophila) and use of floral resources in sunflower (*Helianthus annuus*) fields in Mediterranean environment

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Blooms of farmed plants can offer important trophic resources for honeybee, wild bees and other pollinifagous and nectarifagous insects when wildflowers presence is limited or during dry seasons. In this research we have studied the community of apiformes bees visiting sunflower blooms in a mediterranean non-intensive agrosystem with the aim to verify the resource (nectar or pollen) used and if these resources are differently exploited by the species. The research has been conducted in 2018 and 2019 in the farm Montepaldi (property of the University of Firenze) on two different cultivars respectively grown following organic and conventional management. Both fields are part of The Montepaldi Long Term Experiment (MoLTE), aimed at studying the long-term effect of organic and conventional management on agroecosystems. The fields are located at a distance of about 400 m, separated by some shrub-tree hedges, and they have the same exposure. Bees were observed by transect method within the sunflower fields and the following data were annotate for each specimen: the floral resource exploited (pollen or nectar), the developmental stage of the visited flowers (which are protandrous) and the size of the selected inflorescence. Some specimens, particularly those which we could not identify while on the field, were collected and others were captured with “pan traps” following the protocol by Westphal et al. (2008). All were identified at the level of species by morphological traits and through DNA barcoding of the COI gene. Data of the sequenced specimens (N=55) are deposited on BOLDSYSTEM platform. We have identified 32 species (included honeybee species) and found that the two cultivars did not differ for the specie-set and for their relative abundance. Regarding the resource used, we observed that most bees forage for nectar on flowers at the male stage for which we found a higher concentration in sugar. Moreover when we categorized bees based on their size and tongue length, we found that small-sized short-tongued bees visited inflorescences of smaller size with respect to those with bigger size and longer tongue. The analysis of the pollen grains within the pollen brushes or in the corbiculae of the most abundant species has shown that while some species (*Apis mellifera*, *Bombus terrestris*, *Halictus scabiosae*), during the same flight, forage only on sunflowers, others *(Lasioglossum malachurum*, *Megachile pilidens*) collect also pollen from wild plants, even if these were very limited during the sunflower bloom. This confirms the importance of wild plants for wild bees, even when trophic resources are abundant thanks to the presence of crop plants.

KEY WORDS: wild bees, agroecology, pollination.

POSTER

A laboratory experience on bee biodiversity to support Biology learning in the Secondary Schools

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In the last decades, the teaching of Science in Italian secondary schools often overlooks the study of organisms and ecosystems, favouring instead that of cell biology and, in the last classes of high schools, of molecular biology.
This trend was further encouraged by the revision of the national guidelines for the various disciplines, following the reorganization of the Secondary Schools curricula in 2010/11; a reorganization that was fully applied starting from 2014/15. Although the National guidelines for Natural Sciences teaching repeatedly stress the importance of the laboratory as a moment of learning, consolidation of knowledges and development of skills, the possibility of carrying out practical experiences of cellular and molecular biology in school laboratories is generally limited, due to the lack of equipment and technical staff that can assist teachers. The National Plan for Scientific Degrees is a program of the Italian Ministry for Education and Research (MIUR) that aims to improve the orientation of high school students towards university studies and to support laboratory activities in collaboration with Universities. In the 2018/19, this program also included Degree courses in Natural and Environmental Sciences for the first time. As part of this national project, the Degree Course in Natural Sciences of the University of Florence designed a laboratory on bees ecology and biodiversity which was attended by 229 students and by 14 Sciences teachers. The experience, lasting a total of 6 hours (for each class), focused on the biodiversity of the Apoidea and alternated very short lectures with practical activities. The ultimate aims were to encourage the integration of knowledges acquired in different school years and to offer a practical experience to follow experimental protocols, which included the use of techniques and equipment generally not available in schools. After a brief introduction to the biology of Apoidea, students were asked to identify, using a simplified key for the bee genera, some specimens and confirm their identification through DNA extraction, amplification of the mitochondrial COX-I (Cytochrome c oxidase I) gene and search of the sequences in the databases. Finally, they calculated a biodiversity index based on experimental data from different environments. Information and materials on this experience are available among the teaching materials listed on the website https://www.pianolaureescientifiche.it/scienze-naturali-e-ambientali/.

KEY WORDS: Biology teaching, educational workshop.

POSTER

Gregarious nidifications of the bee *Megachile parietina* and biodiversity of accompanying entomological fauna

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Megachile parietina (Megachilidae) is a Euro-Asian solitary bee. The species is included in the Red List of Germany, Switzerland and Czech Republic, where its decline is probably due to a reduction in the areas sown for fodder and in particular of *Onobrychis* sp. In Central Italy *M. parietina* forages mostly on *Hedysarum coronarium* and his flight period (less than 2 months) is strictly synchronized with the flowering of this plant. Females build hard and waterproof nests by mixing sand and clay with salivary secretion. Like for most solitary bees, *M. parietina* nests attract several natural arthropod enemies which exploit pollen supplies or feed on the immature brood. Despite being strictly solitary, M. parietina can form large aggregations of nests. A very large nest aggregation of about 650 females was found in the Council of Montespertoli (Tuscany) and it has been observed for three consecutive years (2017-19) with the aim of studying the associated fauna. We have identified 31 insect species associated and verified that about 90% of the cells was parasitized, also by checking nests transferred into the laboratory. These species were classified in 5 categories, according to the biologic interactions with the hosts: I) kleptoparasites, when it lays eggs in nests and its larvae compete with the host brood for the stored resources, II) parasitoids, when it lays in the cells and its brood feeds on the host larvae present in that cell; III) predators, when either at the larval or adult stage it feeds on the host by moving between cells or nests; IV) nest depredator , when it can feed on stored pollen without compromising the offspring development; V) inquiline, when it can use the nests but it doesn't exploit food storages. During the study we found both species already known for being associated with *M. parietina*, such as the cuckoo bees *Stelis nasuta* and *Coelioxys aurolimbata* (Megachilidae), the parasitoid *Anthrax anthrax*, *Spogostylum tripunctatum* (Bombyliidae), *Monodontomerus aeneus* (Torymidae) and *Leucospis gigas* (Leucospidae); and species never reported before for this host, such as *Leucospis dorsigera* (Leucospidae), *Chrysura radians* (Chrysidinae), *Melittobia acasta* (Eulophydae), *Chrysis marginata aliunda*, *Chrysis sexdentata* (Chrysidinae) and *Minettia tabidiventris* a Lauxaniidae species for which we observed behaviours like those reported for other dipterans kleptoparasite of solitary bees. These results show that bee nest aggregations can support a very diverse entomological fauna. This kind of information is important both to clarify aspects of the biology of some species and to predict how the decline of several bees may locally impact on their associated arthropod species.

KEY WORDS: Solitary bess, cuckoo bees, parasites, commensalism.

ORAL PRESENTATION

Vibrational communication of the greenhouse whitefly *Trialeurodes vaporariorum*

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Biotremology is a new discipline that studies the vibrational communication of animals. The applied biotremology investigates the control of insect pests of crops using vibrational signals (VS) to manipulate their behaviour (i.e., attraction, mating disruption, repellence). The greenhouse whitefly (GW), *Trialeurodes vaporariorum* (Westwood) (Homoptera: Aleyrodidae) is considered one of the most harmful insect pests in greenhouses. The GW mating behaviour has been partially investigated and its vibrational communication is mostly unknown. A deeper knowledge of its intraspecific communication is necessary to evaluate the applicability of control methods. In this study, we used the laser doppler vibrometer to record the VS, coupled with video recordings to link the signals production to the insect behaviour. VS were characterized in terms of spectral and temporal parameters and an ethogram was designed. We identified three types of male vibrational emissions ("chirp", "pulses" and a Rivalry Signal), differently arranged according to the stage of the mating process, and one female responding signal here recorded and described for the first time. The GW showed to have a complex mating behaviour, consisting of four different stages: "call", "alternated duet", "courtship" and "overlapped duet". The analysis performed with the Markovian behavioural transition matrix showed that the "overlapped duet" stage seems to cover a crucial role to achieve a successful mating, in that strictly associated to the female acceptance. This study demonstrates that vibrational communication plays an important role in GW mating process, therefore more studies are assessing whether VS playbacks can be exploited to manipulate its behaviour. This would be the first case of pest control with vibrations in greenhouses.

KEY WORDS: biotremology, behaviour, insect, pest, mating.

ORAL PRESENTATION

Interactions between *Nezara viridula* and *Halyomorpha halys* and the parasitoid *Trichopoda pennipes*

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Halyomorpha halys (Stål) is a species native to Asia, now widespread in many countries. Due to its high polyphagy and dispersal capacity, it has become one of the major threats in agroecosystems. The insect spread has also been favored by the absence of specific natural antagonists in the areas of introduction. In some of these areas, a number of indigenous parasitoids have shown to be able to accept *H. halys* as a potential host, although only a few successfully completed their development in the exotic insect. *Trichopoda pennipes* (Fabricius) is a parasitoid originated from the Nearctic region that parasitizes both the adults and nymphs of numerous species of Hemiptera Heteroptera. The females lay macrotype eggs on the host body. This tachinid is a solitary parasitoid, although more eggs are usually laid on one host. It was accidentally introduced in Italy in the late 80s of the last century and it is now naturalized in several Italian areas, where the main host is *Nezara viridula* (L.). Both in North America and Europe, cases of *H. halys* adults with *T. pennipes* eggs on their body have been reported in the literature. The complete development of the parasitoid in this host has, however, never been recorded. Interactions between *T. pennipes* and the two aforementioned Pentatomidae were studied in the laboratory. Exposures of *H. halys* or *N. viridula* adults were performed under no-choice conditions. In addition to evaluating the acceptance and suitability of the two hosts, an anatomical and histological examination was carried out. Although no significant differences were found, the results obtained from the exposures indicated a greater acceptance of *N. viridula* compared to *H. halys* by *T. pennipes*, in terms of both the number of individuals on which oviposition occurred and the mean number of eggs laid on the host bodies. The complete development of the parasitoid, up to the adult stage, was obtained only in *N. viridula*. The anatomical and histological examination showed that the penetration rate of the newly emerged parasitoid larvae was higher in *N. viridula* than in *H. halys.* The penetration, which seemingly occurred due to the enzymes secreted by the larva and to the mechanical activity of the larval hooks, was more difficult in the alien insect, possibly due to a different biochemical composition of the cuticle. When the larvae successfully penetrated the host body, in both species they settled in the indirect muscles of flight. In *H. halys* many *T. pennipes* died as late first or early second instar larvae. In this phase the larvae leave the muscles and become more susceptible to attacks by the host immune system. The average life span of the adults of both species was shorter when they were exposed to the parasitoid compared to the non-exposed individuals, probably due to the stress induced by parasitization.

KEY WORDS: *Trichopoda pennipes, Halyomorpha halys, Nezara viridula*, host acceptance and suitability.

POSTER

A physiologically-based model predicting the potential establishment and impacts of the fall armyworm (*Spodoptera frugiperda*) in Europe

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The fall armyworm (*Spodoptera frugiperda* Smith) is an invasive pest originated from the tropical and subtropical regions of south and north America. The species feeds on more than 300 host plants, including economic valuable crops such as corn, rice and sorghum. It represents one of the major threats for agricultural productivity and food security, especially in countries with poor economies and/or lacking of funds to be allocated to the control of the pest. The high migratory capacity of the species and the risks linked to the introduction of infested products are causing concerns related to the introduction of the pest in Europe. Furthermore, transient populations are able to survive in areas outside their establishment range, causing impacts on the crops. In this work, we present the first physiologically-based model for the assessment of the risks of establishment of *S. frugiperda* in Europe, and for estimating the potential impacts caused by both established and transient populations. The model simulates the stage-specific temperature-dependent physiological responses and accounts for the potential role of density-dependent factors on species mortality and fertility. Parameters related to the density-dependent control factors were estimated through a calibration procedure on 65 locations in central and north America, covering areas where the species is established and areas characterised the seasonal presence of migratory populations. From the calibration procedure we have estimated the uncertainty distributions of model parameters. Combinations of the 10th, 50th, and 90th quantiles of model's parameters were used for generating best, worst and medium scenarios of potential establishment in Europe. The same combination of model's parameters were also used for simulating the impacts of the introduction of migratory population at different times of the favourable seasons (1st of April, 1st of June and 1st of August) in five locations (Agrigento, Terracina and Ghedi in Italy, Ouarville in France and Engelsberg in Germany). The results of the model shows that the coastal areas of southern Europe are at risk of establishment of S. frugiperda. In the worst-case scenario, the species' potential establishment is observed also in limited inland areas of southern Spain and southern Italy. Furthermore, model's outputs showed that the species was able to generate transient populations outside the area of establishment when the introduction occurred at the 1st of June and at the 1st of July, with population abundance highly dependent on local climatic conditions. Only in Agrigento and in Terracina the species was able to generate transient populations also if the introduction occurred at the 1st of April. The model represents a suitable tool for the identification of the areas at major risks of establishment of *S. frugiperda* in Europe. The outputs presented might be used for guiding surveillance and prevention plans towards the reduction of the risks linked to the establishment of the species. The model can be also used at the local level for investigating the potential impacts of populations, thus allowing planning and implementing monitoring and control actions for the local management of the species.

KEY WORDS: *Spodoptera frugiperda*, pest risk assessment, pest management, physiologically-based model.

ORAL PRESENTATION

Ants and plants, a long story of coevolution: case studies with applicative implications in Mediterranean environments

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Ant-plant relationships date back over 100 million years and have led to the development of many adaptations whose knowledge could be extremely important in many fields of basic and applied biology. In this context, most of the information on ant-plant interactions only deals with tropical species. However, it is worth noting that also many plants at temperate latitudes establish relationships with ants, although fewer data on the biology of these interactions are available. Ants may have direct or indirect effects on the plant fitness and this may have important applicative implications in different fields including agronomy. In fact, the tendency of ants to defend their partner plants or the habit of several species acting as generalist predators may give new insights to develop novel systems of biological control. In this context, our research aims to bridge the gap of knowledge regarding the Italian species, on which virtually no detailed study has been conducted so far. Here we report the results of a series of field and laboratory investigations on different model systems mainly focusing on plants of forestry and agronomic interest (e.g. citrus and pear orchards, oaks, wild cherry, fava beans). We showed that ant-plant interactions (including myrmecophilous relationships) are common and may play important roles in the ecological dynamics of agroecosystems. In fact, the impact of ants on the health and fitness of the plant can be significant. The presence of ants limits that of potential predators and / or involves a significant reduction in the damage reported by the plants at various levels (leaves, seeds and fruits). This opens up the possibility to study and discuss new aspects and paradigms of the behavioural biology of animal-plant relationships and at the same time offers the opportunity of using these concepts and new data in an agroecological context.

KEY WORDS: ant-plant biology, mutualism, agroecosystems, applied myrmecology, biological control.

ORAL PRESENTATION

Brassicadiene, a diterpene hydrocarbon of novel identification, is mediating the host plant location process of the herbivorous stink bug *Bagrada hilaris*

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Bagrada hilaris Burmeister, also known as painted bug, is a stink bug native to Asia and Africa and invasive in the United States, Mexico, South America and Italy. The painted bug determines serious damage mainly to various vegetable crops in the genus *Brassica*, with seedlings being particularly susceptible to *B. hilaris* feeding activity. In Italy this pest is confined in the island of Pantelleria where it attacks the cultivation of caper, a local product acknowledged with the protected geographical indication by EU. In this study, the role of volatile organic compounds (VOCs) emitted by seedlings of three *Brassica* species on the host preference of *B. hilaris* was evaluated. Specific objective of this study was to identify the chemicals cues involved in the host plant location processes through laboratory and field studies. Laboratory behavioural bioassays carried out in dual choice arena and olfactometer, indicated that adult painted bugs preferred *B. oleracea* var. *botrytis* and *B. napus* over *B. carinata*. A field traps bioassay, carried out in an infested caper field of Pantelleria, using living *B. oleracea* seedlings as lure, indicated the ability to such seedling to attract a consistent number of individuals while no captures were recorded in the control (empty) traps. Volatiles from *B. oleracea* seedlings were collected and bioassayed with *B. hilaris* adults and late stage nymphs, using electroantennographic and behavioral (olfactometer) techniques. When crude extracts of the VOCs from *B. oleracea* var. *botrytis* seedlings and liquid chromatography fractions thereof were bioassayed, *B. hilaris* adults and nymphs were attracted to the crude extract, and to a non-polar fraction containing hydrocarbons, whereas there were no responses to the more polar fractions. Chemical analysis using GC-MS indicated that the main constituent of the non-polar fraction is a diterpene hydrocarbon of novel identification, with trace amounts of several other diterpene hydrocarbons. The major diterpene occurred in VOCs from both of the preferred host plants *B. oleracea* and *B. napus*, but not in VOCs of *B. carinata*. The structure of this compound was characterized by NMR analysis and named brassicadiene. This compound isolated and bioassayed in olfactometer elicited attraction response versus *B. hilaris* adults, suggesting that it may act as is a key mediator in this insect-plant interaction.

KEY WORDS: painted bug, *Brassica oleracea, Brassica napus, Brassica carinata*, behavioral bioassays, electroantennography.

ORAL PRESENTATION

Response of the jumping spider *Phidippus audax* to the *Halyomorpha halys*'s aggregation pheromone

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The common jumping spider, *Phiddipus audax* (Araneae: Salticidae) is an opportunistic and generalist predator of many invertebrate species in North America. In a previous study on the efficacy of the different lures for the invasive brown marmorated stinkbug, *Halyomorpha halys* (Heteroptera: Pentatomidae), *P. audax* were found living in baited but not in control traps. However, while spiders are present in the habitat all year, they were only found in traps later in the season. The commercial lure consists of two sachets containing different compounds, some produced by North American pentatomids, so it is possible that response is elicited by these cues given that the BMSB is a recently introduced species into North America. The objectives of the current project were: i) to confirm, both in the field and in Y tube laboratory bioassays, that the response of *P. audax* to the pheromone lure differed over the season, and (ii) to determine, in laboratory bioassays, if the response to the lures differed in either spring or fall. Trap lines (each with 4 baited and 4 control traps) were run in three different agroecosystems in southern Ontario, CA from June to November 2019. The traps were checked weekly and their position within the trap line rotated. As seen in the previous study more *P. audax* were captured later in the season. Independent of the pheromone trapping, spiders were captured by hand at different field sites, taken to the laboratory and reared under natural conditions until used in the Y tube bioassay. In both summer and fall. Five different choice assays were conducted: i) air in both arms, ii) air in one arm and complete commercial lure in the other, iii) air in one arm and sachet 1 in the other, iv) air in one arm sachet 2 in the other, and v) Sachet 1 in one arm and sachet 2 in the other. Spiders were fasted for 48 h, placed inside the olfactometer and observed for 10 minutes. There was no evidence of preference during the summer while in the fall spiders showed an increased preference for all components of the pheromone lures. Results indicate that *P. audax* shows a clear seasonal response, only responding to the chemical cues in the fall, suggesting physiological changes in response to environmental conditions such as decreasing daylength and temperature. *Phiddipus audax* overwinters as subadult-adult, and feeds on *H. halys*, so a seasonal response using chemical cues, in particular the aggregating cues, of potential prey would ensure the availability of food sources in both fall and spring.

KEY WORDS: Invasive species, Brown marmorated stink bug, aggregation pheromone, seasonal responses, conservation biocontrol.

POSTER

Using *Sclerodermus brevicornis* to understand sociality in parasitoids

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Sociality is common among the aculeate Hymenoptera, and full range of levels of sociality is exhibited, from solitary to eusocial. Most attention has been paid to eusocial and semisocial species mainly in bees, ants, and paper wasps. There has been far less consideration of sociality in parasitoid species, chiefly because most of them are socially solitary. However, in some taxa of parasitoids, mainly belonging to the aculeate family Bethylidae, a range of degrees of sociality is present. Sub-sociality behaviour is the most primitive level of sociality and is evidenced by species that show single mother maternal care of the brood (from passive egg guarding to an array of complex grooming, feeding, protection, and nesting behaviour). This kind of sociality is typical of species in the genus *Goniozus* Förster in which a single mother paralyses and oviposits eggs in group on a victim and takes care of the offspring from hatching until subsequent emergence. A more complex degree of sociality (quasi-sociality) is represented by a group of females (foundresses) collaborating in the paralysis of a large host that they share as an oviposition site, and they provide mutual care for each other offspring. This is a typical behaviour in the bethylids species of the genus *Sclerodermus* Latreille. Recently, the interest toward these species has increased as they can become active part in the control of xylophagous pest beetles. Adult females are highly active in searching for hosts in the pre-existing galleries bored in the wood, helped by their morphological adaptations such as small size and flattened body. The species *Sclerodermus brevicornis* (Kieffer) has been found in association with the exotic longhorn beetle *Psacothea hilaris hilaris* (Pascoe) in Italy and, as it can be reared in laboratory, it can be used as a model species to study many different characteristics related to quasi-social behaviour. Different behavioural strategies of this species, from host sharing including host paralysis, to egg oviposition, brood guarding until and beyond offspring pupation are described. Also the capability to aggregate and segregate in relation to host availability and dimension has been documented. While these parasitoids exhibit cooperative social behaviour, cooperation is not carried out without reference to the relatedness between social partners. Selected tests are reported, always including a greater degree of complexity and observations on cooperative associative strategies between females, coming from the same brood or different broods, to evidence if females are most willing to take risks attacking hosts when the beneficiaries will include their sisters. Understanding the evolution of quasi-sociality and associated behaviours in *S. brevicornis* will assist in biological control programmes because it allows the design of mass rearing and the optimisation of the mass release technique.

KEY WORDS: quasi-sociality, *Psacothea hilaris hilaris*, xylophagous, biological control, behaviour.

ORAL PRESENTATION

The exploitation of chemical cues left by the invasive stink bug *Halyomorpha halys* affects the reproductive success of the egg parasitoid *Trissolcus basalis*

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Chemical traces left by herbivorous true bugs walking on the leaf surface of field crops represent reliable cues for scelionid egg parasitoids to optimize energy and time by restraining their search to areas where newly laid host eggs are likely to be found. In the field, however, leaf surfaces could be contaminated by a plethora of chemical traces left by several herbivorous insects infesting host plants. As a consequence, scelionid wasps have optimized their ability to exploit these substrate-borne semiochemicals by discriminating between footprints left by associated and non-associated host species. Wasp response towards host footprints is innate, but it can be adjusted according to foraging experience. For example, *Trissolcus* females evolved the ability to distinguish between footprints left by female and male bugs, preferring the former as they are strictly related to the areas where suitable host eggs are present, only in presence of their associated hosts; moreover, parasitoids show a weaker response when they encountered patches without oviposition rewards. Parasitoids can also learn to respond to cues from non-associated hosts such as those related to invasive pests. Recent literature showed that invasive insects could deeply modify semiochemical-mediated tritrophic interactions of local networks. In this study, using a system including the local parasitoid *Trissolcus basalis*, the associated host *Nezara viridula*, and the invasive pest *Halyomorpha halys*, we hypothesized that the adaptive value of response to substrate-borne semiochemicals associated to *H. halys* depends on whether the local parasitoid can recognize the invasive species as a suitable resource for offspring development. Through open arena laboratory bioassays, we show that: 1) in single tests, *T. basalis* innately responded to walking traces left by *H. halys*, and oviposition experience allows the wasp females to learn the hierarchical value of walking traces as they showed a strong preference of chemical cues left by host females over those left by host males; 2) in sequential tests, in which the associated host *N. viridula* co-occurs with *H. halys*, oviposition experience in the low quality invasive host induces in *T. basalis* similar responses to those of the associated host, showing that experience does not lead to the evolutionary trap escape.

KEY WORDS: *Nezara viridula*, oviposition experience, host location, evolutionary trap.

Influence of different feeding sources in *Torymus sinensis* mass rearing

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Torymus sinensis Kamijo (Hymenoptera: Torymidae) is a biocontrol agent released to control outbreaks of the Asian chestnut gall wasp, *Dryocosmus kuriphilus* Yasumatsu (Hymenoptera: Cynipidae). This wasp is mass reared in controlled conditions in order to be released in chestnut orchards and coppices, thus factors such as food quality may deeply influence the effectiveness of parasitoids. To identify different diets that could be used to increase mass rearing, performance, longevity and fecundity of *T. sinensis* were assessed using honey (diet 1), honey plus pollen (diet 2), pollen (diet 3), and water (diet 4), compared to unfed wasps (control) in laboratory experiments. In the present study diet quality greatly influenced survival and reproductive output in *T. sinensis*. Adults using honey plus pollen as food had the longest life span, and significantly increased fecundity compared to pollen alone, water, and unfed wasps. Median lifespan (day at which 50% of the initial number of wasps are still alive) was 31, 31, 2.5, 4, 1.5 days, in the honey, honey plus pollen, pollen, water, control, respectively. The egg production in wasps fed with honey and honey plus pollen observed over lifetime showed that the newly emerged specimens had an average of 2.40 ± 0.21 and 0.14 ± 0.12, respectively at day 0. The number of mature eggs increased rapidly reaching in diet 1 the highest average number, 26.60 ± 2.48, at day 6, and 30.2 ± 1.59 at day 7 in diet 2. No significant difference in the mean number of mature eggs between diets 1 and 2 was observed during all 13 weeks except for week 11.
The cumulative lifetime mean egg load was 1012.67 and 1095.82 for honey and honey plus pollen, respectively, which was significantly higher than 32.40 for pollen, 58.40 for water and 43.57 for control. Based on our studies, feeding the female wasps with honey and pollen increased lifespan by 33-fold, and fecundity by 2.3-fold at day 4 over unfed wasps. This work contributes to a better understanding of the influence of different diets on lifespan and fecundity in the parasitoid wasp *T. sinensis*, to the refinement of mass rearing in controlled conditions, and to the optimization of classical biocontrol programs.

KEY WORDS: Parasitoid feeding, honey-based diet, mass rearing, Asian chestnut gall wasp.

POSTER

Electroantennographic and behavioural responses of *Crioceris duodecimpunctata* to asparagus volatiles

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The spotted asparagus beetle, *Crioceris duodecimpunctata* (L.), (Coleoptera: Chrysomelidae), is one the most damaging pest of asparagus crops in the Palaeartic region. Development of effective monitoring tools and sustainable control means of *C. duodecimpuntata* may greatly benefit from the identification of biologically-active compounds towards adult beetles. In this study, the olfactory responses of *C. duodecimpuntata* males and females to volatile organic compounds (VOCs) of *Asparagus officinalis* L. were evaluated by behavioural and electroantennographic (EAG) tests. Experiments were carried out with adult insects collected in asparagus crops located near Foggia (Apulia, Region) during April - June. Preliminary Y-tube olfactometer bioassays (arm lenght, 26 cm; angle 75°; stem lenght 30 cm; internal diameter, 6 cm) were carried to ascertain the attractiveness of odours emitted by plant cladophylls (Vegalim variety) to adult *C. duodecimpunctata*. In these tests, males presented with cladophylls (treatment) vs. clean-air (control) exhibited a significant preference for the plant material ($\chi2 = 13.32$, GdL = 1, $P < 0.001$) and spent significantly more time in the treatment arm ($t = 0.45$, GdL = 27, $P < 0.001$) than in the control one. On the contrary, host plant odours did not elicit a significant response from females, measured either as first choice ($\chi2 = 0.143$, GdL = 1, $P = 0.705$) or time spent in the treatment arm ($t = 0.95$, GdL = 27, $P = 0.347$). The EAG technique was used to assess the antennal sensitivity of *C. duodecimpunctata* males (n = 6) and females (n = 6) to 31 VOCs (1 mg dose) identified from *A. officinalis* and to calculate the dose-response curves (from 0.0001 to 1 mg doses) to the compounds eliciting the largest EAG amplitudes [heptanal, nonanal, (E)-2-hexenal, (Z)-3-exen-1-ol, 1-octen-3-ol, octanol, 2-heptatone, linalool, terpineol, sulcatone, sulcatolo]. All VOCs tested elicited measurable EAG responses. The most EAG-active compounds were the same in males and females and elicited dose-dependent responses. Male and female EAG responses to compounds tested were not significantly different, except for (Z)-3-esen-1-ol for which the mean EAG response of males (4.22 ± 0.17 mV) was significantly ($t = 2.39$, GdL = 10, $P = 0.038$) higher than that of females (3.53 ± 0.24 mV). The behavioural activity of (Z)-3-esen-1-ol to male and female beetles was investigated in further Y-tube bioassays. Insects were given a choice between an odour (10 µL of a 100 ng/µL, 1 µg/µL, or 5 µg/µL mineral oil solution of (Z)-3-hexen-1-ol) and control (10 µL of mineral oil) stimulus loaded onto filter paper disks (Ø 2 mm) were suspended in the center of the cross section of the each odour chamber. At 1 e 50 µg doses, both sexes did not show any significant preference for either the treatment or the control arm. At the 10 µg dose, (Z)-3-esen-1-ol elicited a significant attraction only in males either as first choice ($\chi2 = 7.53$, GdL = 1, $P < 0.001$) and time spent in the treatment arm of the olfactometer ($t = 3.95$, GdL = 33, $P = 0.001$). In conclusion, the study showed that *C. duodecimpunctata* males and females are able to selectively perceive a wide range of host plant volatiles but the blend of cladophyll VOCs and its main components (Z)-3-hexen-1-ol elicited sexually dimorphic behavioural responses with males being the only attracted. Results provide a basis mainly for developing of semiochemical-based monitoring tools of this pest.

KEY WORDS: spotted asparagus beetle, EAG, Y-tube olfactometer, behaviour, kairomone.

POSTER

Edaphic micro-habitat of arthropods in Oltrepò Pavese vineyards with different inter-row management

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The Oltrepò Pavese is a hilly area particularly suited to viticulture and one of the most important areas for the production of high quality wines in Northern Italy. In the context of the "Oltrepò BioDiverso" project, funded by Cariplo Foundation, we evaluated the effects of tillage in vineyard inter-rows (CV Croatina, Pinot noir, Barbera, Riesling italico, Merlot). Therefore, we used a multidisciplinary approach based on the following characteristics and parameters: a) biodiversity of microarthropod abundance in terms of ecological indices (i.e. Shannon, Simpson, Margalef), QBSar, oribatid mites/other mites, b) communities of soil fungi and bacteria at two different depths (0-30, 30-60 cm), c) soil physical characteristics (texture, porosity), soil chemical parameters (i.e. TOC, TN, S) and soil hydrological indices (SCS, water content, etc.). Three vineyards with minimum inter-row tillage (CT) were compared to 3 vineyards with permanent grass cover (NT). In 2017, 3 soil samples per site were taken in the inter-rows. The microarthropod communities were assessed by their morphological adaptation to the soil (Biological Forms). The total DNA of bacteria and fungi was extracted from the samples. The no tillage inter-row management lead to better surface hydraulics, greater macroporosity and higher organic matter content. In this edaphic context (NT), the biodiversity indices of the microarthropods were higher than in tilled soil (CT), while the values of QBSar were high (> 140) in both cases. The total microarthropod abundances were different between sites ($F_{5.17} = 9.25$ $p < 0.001$) probably due to other environmental and/or management factors such as age of the vineyard or use of herbicides. The oribatid/other mite ratio was higher in NT (O / aA = 0.87), denoting a greater number of ecological niches suitable for this mite group having an important role in the organic matter cycle. A fundamental regulating factor of the microbial community is represented by the water content of the soil: Actinobacteria were particularly abundant in NT; Firmicutes and Bacteroidetes in CT. The fungal community was mainly composed of Ascomycota, Mortierellomycota and Basidiomycota. The soil biological component and its environmental context provided important information for best agronomic practices to preserve soil functionality and biodiversity.

KEY WORDS: soil arthropods, QBSar, mites, fungi, bacteria, inter-row, vineyard.

POSTER

Fitness and feeding behavior of *Scaphoideus titanus* reared on different grapevine cultivars

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Flavescence dorée of grapevine (FD) is a phytoplasma-associated disease present in several European countries and transmitted by the nearctic leafhopper *Scaphoideus titanus* among plants of the genus *Vitis*. The leafhopper spreads FD through primary infections (from gone-wild vines surrounding the vineyard to cultivated grapevines) and secondary infections (from vine to vine within the vineyard). FD transmission experiments to the main Piedmontese grapevine cultivars (cvs) were carried out with the vector *S. titanus*, in order to screen for different levels of cv susceptibility to FD phytoplasmas. We made the hypotheses that tolerance could be due to plant response to phytoplasmas or could be mediated by vector-plant interaction. To test this latter hypothesis, based on the results of the transmission experiments, *S. titanus* fitness and feeding behavior were studied on three cvs with different susceptibility to FD: Barbera (susceptible), Brachetto and Moscato (tolerant). Survival tests revealed a higher mortality rate on Moscato compared to Barbera and Brachetto, while no major differences were highlighted in developmental time measured on the three cvs. Feeding behavior of the vector was investigated with the electropenetrography (EPG) technique. Hemipteran insects insert the stylets inside the leaf to feed in different plant tissues, and this technology allows studying feeding behavior as different electric waveforms and the association with different feeding phases. The EPG experiments on *S. titanus* allowed a precise description of feeding behavior on the above mentioned grapevine cvs, thus shedding light on the mechanisms of susceptibility/tolerance of grapevine to FD eventually mediated by the vector.

KEY WORDS: flavescence dorée, *Scaphoideus titanus*, plant resistance/tolerance/susceptibility, insect fitness, EPG.

POSTER

First evidence of a female-emitted attractant in *Philaenus spumarius*

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The meadow spittlebug, *Philaenus spumarius* L. (Hemiptera: Aphrophoridae), is the main vector of the xylem-limited bacterium *Xylella fastidiosa* subsp. *pauca* strain ST53, known as the causal agent of the Olive Quick Decline Syndrome (OQDS). *Philaenus spumarius* and other Auchenorrhyncha are known to communicate via vibration, however the occurrence of semiochemical communication has been poorly investigated so far. Here, through a multidisciplinary approach involving behavioural assays, headspace volatile collection from insects, chemical analysis and electroantennographic tests, we provide evidence of a female-emitted attractant in *P. spumarius*. In Y-tube olfactometer bioassays, males were significantly attracted to females as well as toward the head-space volatile extracts collected from females. Conversely, females did not respond to male volatiles or their extracts, nor did males and females respond to volatiles from individuals of the same gender. GC-MS analysis of male and female head-space extracts showed qualitative and quantitative differences in their GC profiles. Moreover, female extracts elicited a measurable EAG response in male antennae, suggesting the presence of compounds which are perceived by the male's olfactory system. This is the first record of a female-produced attractant in Auchenorrhyncha, and its possible role as a sexual pheromone is under investigation.

KEY WORDS: Pheromone, Homoptera, *Philaenus spumarius*, *Olea europaea*, insect vector, plant pathogen, *Xylella fastidiosa*, electrophysiology, olfaction.

POSTER

Environmental variables driving habitat suitability for *Popillia japonica* in Lombardy

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The Japanese beetle *Popillia japonica* Newman (Coleoptera: Scarabaeidae) is an invasive species native to Japan and the far east of Russia. The adults of *P. japonica* are polyphagous feeding on over 300 plant species, skeletonizing the leaves and eroding flowers and fruits. The larvae feed on roots, causing yellowing and drying of lawns. Since 1916, the species has been considered established in North America, in the 1970s it was introduced in the Azores and more recently in Northern Italy (2014). From the original area of establishment in few Municipalities in the Northern Ticino valley in 2014, P. japonica is spreading in the Lombardy and the Piedmont regions covering an area that is increasing year by year. The Italian and the regional Phytosanitary Services are implementing the appropriate measures to manage the spread of the species. Understanding the characteristics of the habitat that allows the establishment and the growth of *P. japonica* populations is fundamental for supporting the assessment and the management of the risks linked to this species. In this work, we investigate the habitat suitability for larvae of *P. japonica* populations recently established in western Lombardy (Italy). Data used for the analysis refer to occurrence of larval populations between 2016 and 2019 in 8908 sites sampled in the infested area of the Lombardy Region. The samples sites were characterized on the basis of a set of explanatory variables related to land use, soil geo-pedological characteristics, and meteorology. We performed a stepwise logistic regression analysis for testing the statistical significance of explanatory variables on the occurrence of local larval populations. Then, we implemented a Generalized Additive Model to investigate the possible nonlinear relationships between explanatory and dependent variables. Results show the significant role of soil variables (e.g., soil texture, level of organic substance and soil pH) and weather variables (e.g., cumulated rain, air and soil temperature, soil humidity) on the probability of larval occurrence. The distance from the area of first infestation and number of years passed since the first establishment of the species are also related to the probability of larval occurrence. The proposed modelling framework can be used for evaluating how habitat suitability for *P. japonica* varies across the landscape. The model can be considered as an important tool for generating risk maps helping in prioritize survey and control measures.

KEY WORDS: *Popillia japonica*, habitat suitability model, invasive species, pest management.

ORAL PRESENTATION

Response of microarthropods to plantation of new vineyards in Chiantishire (Tuscany, Italy)

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Conventional agricultural practices for plantation of new vineyards based on deep earthworks in the hilly landscape of the Chianti Classico (Tuscany), may increase soil susceptibility to erosion by modifications in physical, chemical and biological equilibria of soil environment. To evaluate the effect of soil management on its functionality, vine phenology and grape yield and quality in organic farming, the impact was assessed in two different vineyards for six years (2011-2016): a) new vineyard (NV), set in 2010 after soil levelling and deep ploughing b) old vineyard (OV), planted in 2000, characterized both by similar stony-calcareous soil (Cambic Skeletic Calcisol - Loamic, Aric - WRB, 2014), environmental and farming conditions (organic farming, alternate tilled-inter-rows). In this study, soil samples were collected for chemical (TOC, TN, CaCO3, pH, EC), physical (apparent density and aggregates) and biological (microarthropods density) properties. Furthermore, the quality of grapes and vine was characterized by seed maturity, probable alcohol, pH, sugars, etc. The abundance of microarthropods was assessed using a generalized linear model (GLM) to compare time scale (sampling year), age of the vineyard (NV vs OV). The soil biological quality index (QBSar) was also calculated. The results showed differences between vineyards: pre-implantation processes in NV caused negative impact on the abundance of microarthropods and for three years the eu-edaphic forms, such as Diplura, Protura, Symphyla, were absent. Over time, in NV, the structure of the arthropod community became more and more complex and abundant; the stability of soil aggregates increased to get close to that in OV. To confirm this recovery, the QBS indees initially denoted a biological quality halved in NT while, since 2014, these values became similar between vineyards NT and OV (> 150). The year and age of vineyard affected the acarofauna density, the most abundant group (> 50%) of total soil arthropods. Due to low fertility, the young vineyard (NT) showed stunted vegetative growth and no significant production yield until 2015; the yield was 0.4 kg of grapes/vine in NT vs 1.0 kg of grapes in OV in 2016. The chemical properties showed a not clear tendency to recover over time. After 5 years from plantation, the recovery of soil biodiversity was presumably influenced by an increase of aggregate stability (average weight diameter and water stability index). On the whole, the upset of the soil profile led to consequences in terms of loss of nutrients and soil biological activity with changes in chemical and physical features of the root zone.

KEY WORDS: soil biodiversity, resilience, QBSar, above- and belowground interactions.

POSTER

Interactions between the introduced giant resin bee in Europe and the local fauna: insights into nesting competition and biology

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Megachile sculpturalis (Smith) is the first exotic bee species in Europe and its first report in Italy was in Piedmont region in 2009. Its extremely rapid expansion across the continent has generated growing concern about the negative impacts on native bee fauna. To investigate the nature of nesting competition between the exotic and the native wild bees, we set up trap nests for above-ground nesting bees on a semi-urban area of Turin province, Italy. We aimed to 1) identify nesting competition mechanisms at nesting tunnel level, specifically looking for indirect ones and 2) to analyse biological aspects accounting for the exotic bee fitness: sex ratio and presence of natural enemies. We found a significant decrease in native *Osmia cornuta* abundance related to *M. sculpturalis* increase. More troublingly, about half of the tunnels occupied by the exotic bee were already cohabited by the native one. In these cohabitated tunnels, *M. sculpuralis* cells were built in the space between native bee cells and tunnel exits. This implicates that the progeny of the native early-emerging bees is likely to be blocked at the emergence by the subsequently constructed exotic bee brood cells. Here, we described for the first time the emergence block as an indirect and negative mechanism of interaction. On the other hand, the sex ratio of *M. sculpturalis* offspring was strongly biased towards males, indicating a likely temporary genetic poverty of the population in the local invaded area. In addition, we documented for the first time the presence of three local natural enemies (*Cacoxenus indagator*, *Monodontomerus obscurus* and *Ptinus sexpunctatus*) affecting alien bee offspring. Our results provide new insights into the indirect competition between *M. sculpturalis* and native bee species, as well as its performance in the context of new territories.

KEY WORDS: exotic bee, wild bees, *Megachile sculpturalis,* bee invasion, nesting behavior, trap nest, competition, sex ratio, natural enemies.

Impacts of single and repeated heat shocks applied at different developmental stages in an aphid-parasitoid system

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Climate change and associated extreme temperatures will affect many organisms, insects included. How the organisms will respond to this challenge can have great effects in many applied fields, like human and animal health, conservation biology and environmental protection, forestry and agriculture. Extreme thermal events could affect species distributions, life histories, community composition, ecosystem function and interspecific interactions (competition, predation and parasitism). The ability of insects to cope with climate change will also depend on their responses to extreme thermal events, which can have different outcomes if repeated rather than single ones. The timing, intensity and frequency of thermal stress exposure can be very variable in nature, which makes predictive models of insect responses to global change more complex. Exposure to repeated extreme temperature can have important effects on host-parasitoid interactions and, more generally, on the higher trophic levels. In this study we investigate some aspects related to the survival of aphids and parasitoids after single and/or repeated exposure to high temperatures (one mild and two extremes) in the trophic model system *Acyrthosiphon pisum* (the host) - *Aphidius ervi* (the parasitoid). When aphids were exposed to a single severe heat shock, we recorded a higher survival in parasitized aphids compared to the unparasitized ones, and this differential survival was greater according to the greater temporal delay between the parasitization and the heat shock. Survival of unparasitized aphids to a heat shock increased when the aphids were previously exposed to high temperatures. Instead, a previous exposure to high temperatures did not increase the survival of parasitized aphids, and so the overall effect of a previous thermal stress exposure gave similar survival results in the two experimental groups. According to our observations and to previous literature related to other host-parasitoid systems, we think that the parasitization event can induce in our system the synthesis of Heat Shock Proteins by the host, so increasing the survival of the parasitized aphids. Also a previous exposure to a heat stress can induce the synthesis of Heat Shock Proteins to give an increased survival to later heat shocks, a phenomenon known as “hardening”. Even if the previous thermal treatment did not increase the survival of the parasitized aphids (that is, the hardening does not add its effect to the parasitization one), on the long term it increased the parasitoid emergence from the hosts. In conclusion, repeated high thermal shocks induce different changes in both the host and the parasitoid and can affect their population dynamics, possibly affecting in different ways the role of parasitoids in pest control.

KEY WORDS: thermal stress , *Acyrthosiphon pisum*, *Aphidius ervi*, survival.

ORAL PRESENTATION

Possible hierarchy between antennae and forelegs as observed in food handling by *Messor arenarius* ants

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The field observations in this research were done between the years of 1994 and 1997 in different areas in the Coastal Plain of Israel and in the Negev Desert in Israel. In each experiment, a food site was placed, with its closest edge to the nest in a distance of 1m from nest entrance. That food site contained four food points organized in a square with a distance of 10cm between each two neighboring points. In each of these points, a whole-wheat seed and a half of wheat seed cut longitudinally with its cut side facing upward were placed. This food choice was offered to these ants in order to let them handle in each food point two food-particles, which differ from each other. Wheat was offered to these ants since it was observed previously that when *Messor arenarius* ants encounter a wheat seed or a half of wheat seed cut longitudinally they usually collect them. The two seed particles in each food point touched each other or were very close to each other, in order to ensure that while reaching a food point, ants wouldn't ignore any of these food-particles. In experiment-series 1, first collections from food points were used for data analysis, because in further collections there could be a possible effect of pheromone in collection points. In experiment-series 2, data analysis was done on all collection rounds, because after each collection round the sand or the ground from collection points was swept or was changed with sand or with ground from outside the food site, to prevent any possible effect of pheromone. Afterwards, a similar food-particle was placed. It was found, that *M. arenarius* ants touch a food-particle prior to its collection usually first by their antennae, then by their forelegs and afterwards capture it by their mandibles. In experiment-series 1, 26 nests were checked in which 65 first collection rounds were recorded. In experiment-series 2, 10 nests were checked in which 28 collection rounds were recorded. The proportions of touches of ants on both food-particles in first collections from points were significantly higher in touches by their antennae than in touches by their forelegs (0.600 vs. 0.369, P=0.009 in experiment-series 1, and 0.789 vs. 0.368, P=0.010 in experiment-series 2 in one-tailed Fisher's exact tests). The same proportion in experiment-series 2 in further collections was also significantly higher in touches by their antennae than in touches by their forelegs (0.667 vs. 0.111, P=0.025 in one-tailed Fisher's exact test). It was observed, that *M. arenarius* ants handle prior to collection more food-particles by their antennae than by their forelegs. This finding can be characterized as a possible hierarchy between antennae and forelegs in the process of food handling by these ants. It is known that on antennae of ants there are contact chemoreceptors, while forelegs of ants don't contain chemoreceptors but contain mechanoreceptors. It is possible that the touch by the forelegs gives an ant information about the shape of a food-particle in order to collect it afterwards.

KEY WORDS: Antennae, ants, food handling, foraging, forelegs, *Messor.*

POSTER

Possible effect of pheromone as reflected in seed handling by *Messor arenarius* ants

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Ants use pheromone communication for several activities including marking of food sites. In this research, a possible effect of pheromone was observed during food handling by *Messor arenarius* ants. The experiments in this research were carried out between 1994 and 1997 on different areas in The Coastal Plain of Israel and in the Negev Desert in Israel. Near each nest in this research, a food site was located in a distance of 1m from nest entrance. This food site was a square with dimensions of 10x10cm, which contained four food points that were located in its corners. In each of these food points, a whole-wheat seed and a half of wheat seed cut longitudinally were located. These two seed particles touched each other or were very close to each other in order to ensure that while reaching a food point the observed ants wouldn't ignore any of these food particles. A single experiment was done on each nest in which touches of ants by their antennae or by their forelegs were recorded through direct observations. After each collection round the ant that had collected a food item was marked on one of its legs or on some of its legs with correction fluid. Marked ants were not allowed to collect again food items from the food site during the same experiment. After each collection round, a food item similar to the food item that was collected previously, was added to the food point. Experiment series 1 was done without any change in the ground of the food site. However, in experiment series 2, after each collection round the ground of the food point from which collection was observed, was changed with ground from outside that food site. This was done in order to prevent possible effect of pheromone that might have been secreted by ants in the food points from which food items were collected. In experiment series 1, 26 nests were checked in which 65 first collections and 28 further collections were registered. In experiment series 2, 10 nests were checked in which 19 first collections and nine further collections were registered. In experiment series 1, the proportion of touches by ants' antennae on both food items in a same collection point was significantly higher in first collections from food points than in further collections from food points (0.600 vs. 0.369 respectively; Chi-square=3.378; DF=1; P=0.033 in a unilateral test). In experiment series 2, that proportion in first collections didn't differ significantly from the same proportion in further collections (0.789 vs. 0.667 respectively; Chi-square=0.491; DF=1; P-N.S. in a unilateral test). In both experiment-series, no difference was found in the proportion of touches by ants' forelegs on both food items between first collections and further collections from food points. These findings may indicate on a possible effect of pheromone in the collection points on further collections when the ground of these food points wasn't changed.

KEY WORDS: Antennae, ants, forelegs, Formicidae, *Messor*, pheromone.

POSTER

Design of an ideal vibrational signal through vibrotaxis experiments to enhance pest control of stinkbugs

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Satisfying the increasing world-population demand for safe and sustainably-produced food is a tough challenge that policy makers, stakeholders and scientist will face in the coming decades. One issue of particular concern is the demand for the reduction of external inputs required for pest control, which currently mainly relies on pesticides, which are threatening the environment and human health. Thus, alternative methods to control insect pests are needed to minimize these risks. Innovative methods aim at developing sustainable pest control methods by means of behavioral manipulation of insects, for instance using vibrational signals. This study aimed to explore the use of substrate-borne vibrations for pest control of the brown marmorated stink bug (BMSB), *Halyomorpha halys* Stål (Heteroptera: Pentatomidae). To this purpose, we first defined the spectral and temporal characteristics that best elicit male responsiveness by means of bioassays conducted with artificially created signals mimicking female vibratory emission. Then, we evaluated male responsiveness to the signals with different features and supposed degree of acceptability by means of a wooden custom-made T stand arena designed to perform single and two choice test experiments. The results from this study showed that males were attracted to female signals along a high range of amplitudes, with higher pulse repetition time and peak of frequency in correspondence to the first harmonic. This allowed us to propose an "optimal" signal to be used to attract males. When exposing males to single and double choice tests in the T arena, males showed a significant orientation towards the "optimal" female signal. Our result brings this signal to elicit searching behavior and attract males of BMSB towards a stimulation point. Furthermore, we confirm the use of vibrational signal as a strong tool for the development of traps for the species and the further management of this pest.

KEY WORDS: biotremology, *Halyomorpha halys*, vibrational communication, playback, brown marmorated stink bug.

ORAL PRESENTATION

SESSION V

First report of *Mamestra brassicae* feeding on the invasive plant *Phytolacca americana* in Campania (Italy)

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Since a few years pokeweed (*Phytolacca americana*) has become invasive at the Astroni Nature Reserve near Naples, Italy. Besides environmental factors which have repeatedly damaged woody plants in this area leaving room to allochthonous weed proliferation, this outburst was considered to derive from the absence of specific herbivorous organisms, which could be related to pokeweed's poisonous properties. Nevertheless, consistent leaf erosions saving the main veins only were observed in the past few years, which resulted to derive from the feeding activity of caterpillars belonging to the Noctuidae. Within this family, the species *Spodoptera eridania* (Stoll) and *S. ornithogalli* (Guenée) have been reported to feed on pokeweeds in the Neartic region; another species, *Peridroma chersotoides* (Butler), is mentioned from Hawaii. Therefore, the unexpected observations at Astroni were at first considered to be related to the possible introduction of one of these exotic species. However, examination of the larvae was more indicative for their belonging to the genus *Mamestra*. In fact, observation of the adults obtained after rearing caterpillars exclusively on pokeweed leaves, coupled with data resulting from cytochrome c oxydase subunit I sequencing unequivocally allowed identification as the cabbage moth, *M. brassicae* (L.). The species develops in two generations per year, one in spring (April-early June), and the other in late summer-autumn (mid August-November), aestivating as chrysalis. To our knowledge, this is the first report of *M. brassicae* on *P. americana*, which seems to confirm the adaptability of Noctuidae to overcome plant resistance conferred by antifeedant metabolites. The occasional finding of another Noctuidae species, *Mniotype solieri* (Boisduval), deserves to be further investigated.

KEY WORDS: Noctuidae, *Mamestra brassicae*, *Phytolacca americana*, plant metabolites, cytochrome c oxydase.

POSTER

Notes on the origin, composition, diffusion and damages of the Auchenorrhyncha of alien origin present in Europe

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In the last 50 years the number of alien species accidentally introduced in Europe belonging to the Auchenorrhyncha (Hemiptera) has considerably increased. Many of them cause substantial economic losses and environmental disturbance. An update on the knowledge of alien Auchenorrhyncha species (Fulgoromorpha and Cicadomorpha) introduced on the European continent is provided. Literature data highlighted the presence of 31 species of the following seven families: Delphacidae (1 sp.), Issidae (1 sp.), Acanaloniidae (1 sp.), Flatidae (1 sp.), Ricaniidae (3 spp.), Membracidae (1 sp.), and Cicadellidae (23 spp.). Furthermore, the presence of the following three species of non-European origin, *Cephalius frontalis* Signoret (Cicadellidae: Dorycephalinae), *Ricania limbata* Lallemand, and *Ricania simulans* (Walker) (Ricaniidae) has previously been erroneously reported, while *Edwardsiana crataegi* (Douglas) (Cicadellidae: Typhlocybinae) has to be considered not an alien species but a native one. In Europe, the countries with the highest number of first records of alien Auchenorrhyncha species are Italy (9 spp.), Portugal (4 spp.), Russia (4 spp.), and France (3 spp.). Based on the native range of the species, the most represented areas are North America (12 spp.) and eastern Asia (14 spp.), followed by tropical Africa (4 spp.), and Oceania (1 sp.). The historical trend shows the following increase: 1800s (1 sp.), 1900-1959 (4 spp.), 1960-1999 (10 spp.), 2000-2018 (16 spp.). Over the past twenty years, the number of taxa has grown by 60% compared to the previous forty years. As regards the host plants, the alien Auchenorrhyncha species are: 12 polyphagous species, 9 monophagous species on a single botanical genus, 5 oligophagous species and 2 species not yet defined. Worrying is the ability of certain species to be vectors of microorganisms such as viruses, rickettsiae, bacteria and phytoplasmas, cause of serious crop diseases, as *Scaphoideus titanus* Ball, known vector of the phytoplasma, Grapevine flavescence dorée (Bacteria: Acholeplasmataceae) in the European vineyards. Although the concrete risk is still to be defined, among the most insidious there are *Jikradia olitoria* (Say), *Orientus ishidae* (Matsumura), *Osbornellus auronitens* (Provancher) and the species of the genus *Hishimonus* (Cicadellidae).

KEY WORDS: Auchenorrhyncha, Fulgoromorpha, Cicadomorpha, alien species, Europe.

Checklist of the Brachycerous Diptera of the Aspromonte National Park (Reggio Calabria)

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In the the research project entitled "First sampling of Brachycerous Diptera in the Aspromonte National Park'' financed by the Aspromonte Park Authority, a sampling of Brachycerous Diptera in the Aspromonte National Park (Reggio Calabria) was carried out over two years. The surveys were carried out in five areas, selected by habitat and altitudinal difference, using three different sampling methods: bait bottle traps, Malaise traps and chromatographic traps. This work made it possible to draw up the first checklist of the species of Diptera Brachycera present in the Park area. Among the species sampled, new elements emerged for the Calabria region and for the entire national territory, as well as elements of medical-veterinary and zoogeographical interest. The most represented families are those of Heleomyzidae, Drosophilidae, Anthomyiidae, Fanniidae, Muscidae, Calliphoridae and Sarcophagidae. From the identification of the collected entomological material, several genera were identified in the family Calliphoridae, of which the most abundant are *Calliphora*, *Lucilia* and *Pollenia*, in the Muscidae the genera *Muscina*, *Thricops*, *Hydrotaea*, *Musca*, *Phaonia* and *Helina*, in the Heleomyzidae the genera *Suillia* and *Tephroclamys* and for the Antomyiidae the genera *Lasiomma*, *Pegomya*, *Anthomyia* and *Hylemya*. For the families Fanniidae and Drosophilidae only one genus was identified, *Fannia* and *Drosophila* respectively. Within the identified genera there are species that play an important role in ecology, fauna, veterinary medicine and forensics. This sampling has made it possible to draw up an initial data set that can be used for surveys aimed at studying the structure of communities of Diptera Brachycerous in other areas of Calabria.

KEY WORDS: Diptera Brachycera, sampling, ecological assessment.

POSTER

The first sampling of Diptera Brachycera in the Aspromonte National Park (Reggio Calabria)

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The Brachycera despite the high number of species, their ecological and medical- and veterinary importance, have been few investigated. Many species play an important role in the recycling of nutrients in terrestrial ecosystems, accelerating the decomposition of organic matter, others are mechanical and biological vectors of myasis, viral and bacterial pathogens and protozoan cysts. In Italy, few data about their abundance, distribution and role as biological indicators are reported. The aim of this work is to investigate on the differences in habitat use by Diptera Brachycera. The study was carried out for two years inside five areas, selected by habitat and altitude characteristics. The sites located between 900 m and 1650 m a.s.l. include forest habitats and wetlands. Sampling was carried out using bait bottle traps, Malaise traps and chromatographic traps. The entomological material were identified at the Laboratory of Applied and forensic entomology of the Department of Biology, Ecology and Earth Sciences (University of Calabria). New data on the diversity, abundance and phenology of the species within the sampled areas were collected. New reports for Calabria and Italy about species of ecological and medical-veterinary importance were provided for the first time. The data reported here suggest differences in phenology and habitat use between the species and lays the basis for more detailed studies on the possible role of some species in ecological assessment and management of protected areas.

KEY WORDS: Diptera, sampling, ecological assessment.

ORAL PRESENTATION

Two ways to be endemics over the Alpine-Appeninic region

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Alps and Apennines are known as an important area of endemism over Europe. Indeed, the high altitude areas currently host many glacial relicts represented by species that were more widely distributed during colder Quaternary periods. In the same geological periods, the southern European peninsula has provided restricted and isolated warm refugia for many species that are now more widely distributed over Europe. There is thus the potential for southern Apennines (mostly the isolated Sicily) and for the areas of Alps to show distinct patterns of endemism. To unravel butterfly endemicity over this region we gathered and analysed a huge dataset consisting of a total of 26,228 COI sequences obtained by specimens of all the butterfly species occurring in the area with comparisons with the entire western Palaearctic. We also collected more than 400,000 occurrence data (0.2 degrees latitude and longitude accuracy) for the Alpine Apenninic region. We constructed phylogenetic trees for the five butterfly families and applied a GMYC procedure to identify genetic entities. By inspecting the distribution of all the sequences belonging to each GMYC species, we identified which entity only occurs on the Alps-Apennines area and marked them as endemics. Based on the concave alpha-hulls defined by the distribution of each endemic entity we attributed the occurrence data to each of them and mapped the fraction of endemic GMYC species occurring over the region. In addition, we calculated for each GMYC species its phylogenetic isolation as the patristic distance with respect to the closest species and a series of functional and ecological traits (size, degree of generalism, distribution area and climatic preferences). We found that the Alps, Sicily and the Southern tip of the Italian Peninsula are the areas showing the highest fraction of endemics. Moreover, GMYC endemics on Alps coincide with ancient taxa, usually representing a single taxonomic entity, with restricted distribution and preference for cold climates. Conversely, the endemics occurring in the South diverged more recently, usually represent genetic lineages of widely distributed species and show ecological traits similar to non-endemic taxa. In conclusion, our results demonstrated for the first time over an entire and large taxonomic group that there are two kinds of endemics in the Mediterranean region, characterized by two contrasting distributions and histories. The first are represented by older taxa, adapted to live in the colder environments occurred in Europe during most of the Pleistocene and now restricted to mountain regions. The second are represented by more recently diverged lineages of species adapted to warmer environments that have diverged at the southern tips of the Mediterranean peninsula and did not moved poleward during the current warm period to cross the Alpine chain. These results have profound implications to explain the historical dynamics of biodiversity in the Mediterranean and to predict the effect or recent climatic changes over the existing butterfly populations.

KEY WORDS: butterflies, endemism, Alps, Apennnines, COI.

ORAL PRESENTATION

Tenuipalpid mites (Trombidiformes: Tetranychoidea) with particular relevance of the Apulian fauna (Southern Italy)

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Mites of the family Tenuipalpidae (Trombidiformes: Tetranychoidea), commonly known as flat mites or false spider mites, are obligate plant feeders. Their importance as agricultural pests has been increasing significantly after the discovery of their association with several relevant plant viruses. Contrary to the trends in systematics and biology of tenuipalpids in the international research community, very little new information on this taxon has been published from Italy during the last two decades. Furthermore, there has been a lack of significant study of this family in Italy and Apulia (southern Italy) except in regard to a few specific crops (e.g. *Olea europaea* L.). This research is aimed at surveying tenuipalpid mites in Apulia on the agricultural, forestry and ornamental trees with the attempt to map their distribution and host association, and provide data for updating the Italian checklist of this taxon. A further aim is to confirm and compare the identity of specimens intercepted from Italy at the airports and ports of the United States of America. The sampling was carried out from March to October 2019 in the Apulian territories (southern Italy), collecting plant materials from various Mediterranean tree species. The mites were slide-mounted in Hoyer's medium and identified using Low Temperature and Conventional Scanning Electron Microscopy (LT-SEM), phase contrast and differential interference contrast (DIC). The data analysis of the new and old samples identifies the presence of four new species belonging to *Cenopalpus* genus (*Cenopalpus apuliae*, *Cenopalpus bauchani*, *Cenopalpus rosaceae*, *Cenopalpus ulmifolius*), new records of the genus *Cenopalpus*, *Brevipalpus* and *Tenuipalpus* in Italy (*Cenopalpus bakeri*, *Cenopalpus lanceolatisetae*, *Cenopalpus longirostris*, *Cenopalpus officinalis*, *Brevipalpus oregonensis*, *Brevipalpus russulus*, *Brevipalpus yothersi*, *Tenuipalpus sarcophilus*). The presence of *Cenopalpus bakeri*, considered a complex species, is relevant. The last Italian check-list (Bernini et al., 1995), of Tenuipalpidae can be updated to 34 species (from the previously 22 listed species), with a largely and spread presence of *Cenopalpus*. It is necessary more investigations about the capacity of the plant pathogen transmission on agricultural and forestry crops, specifically of the genus *Cenopalpus* whom morphological alterations on *Pinus* genus were just reported by Nizi (1963).

KEY WORDS: Flat mites, plant feeders, new species, new reports, viruses, plant protection.

ORAL PRESENTATION

Elusive but still present in italy: two new populations of *Bolbelasmus unicornis* (Schrank) for Northern italy

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Bolbelasmus unicornis (Schrank) is a scarabaeoid beetle belonging to the Geotrupidae family, occurring throughout central and central-eastern Europe. It is an extremely elusive species, which appears to be infrequent and localized all over its range, therefore it has been included in Annexes II and IV of the European Directive 92/43/EEC "Habitat". Up to now few records were known from Italy, mostly from old collections (Piedmont, Lombardy and Friuli Venezia Giulia). Due to the extreme shortage of recent data and the fragmentary state of knowledge of this species in Italy, *B. unicornis* was deemed a marginal presence for the Italian territory and therefore not included in the 3rd National Report of the Habitats Directive (GENOVESI et al. 2014). The discovery of two new populations of B. unicornis in Piedmont and Friuli Venezia Giulia is here reported. The first new population comes from Piedmont, in the surroundings of Lerma (AL), at about 300 m a.s.l. The discovery of a male specimen in 1995 was followed by ten records (4 males and 6 females) in 2014 and 2015, confirming the survival of a population on the site during 20 years. *Bolbelasmus unicornis* individuals were found in the soil during excavation works for the cultivation of a vegetable garden for domestic use or attracted by light (Wood lamp). Those individuals were subsequently released.
The second population, analyzed in more detail, occurs in the province of Pordenone (Friuli Venezia Giulia). Observations were made through the years 2018 and 2019: specimens of *B. unicornis* were found in a meadow located at about 116 m a.s.l. in the extensive steppe-like xeric grasslands of the Magredi del Cellina. A total of 27 specimens of *B. unicornis* were found (and subsequently released), therefore the surveyed population probably represents the largest one in the Italian territory. Soil and vegetation characteristics of the site were analyzed. Three sampling methods (light traps, pitfall traps and active search by torch) were compared in four stations corresponding to different stages of vegetation. The results (GLEREAN & STEFANI 2020) show autoecological aspects that are similar to those known for Central European populations, although with a preference for the *Chamaecytiso hirsuti-Chrysopogonetum grylli* meadows. The data gathered on the habits and ecology of this beetle and its extremely elusive activity suggest that the species is probably less rare and more widespread in Northern Italy than currently known.

KEY WORDS: *Bolbelasmus unicornis*, Geotrupidae, Bolboceratinae, new populations, Piedmont, Friuli Venezia Giulia, Italy, Habitats Directive.

POSTER

Soil macro-invertebrate communities along land use intensity gradients in a mountain region

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Land-use can have profound and diverging effects on the diversity, community composition, and associated ecosystem services of life on and beneath the soil surface of the managed landscape. Ground- and soil-dwelling macro-invertebrates were investigated with both experimental and comparative approaches along land-use change and management intensification gradients in a North-Italian Alpine region. Specifically, we compare low input and traditional management of meadows and orchards with more intense and commonly applied production methods of both land-use types. As expected, the taxonomical richness and diversity were significantly lower under more intensive management (on average 10% less families). The community composition changed, and the direction was governed by the type of management. Furthermore, we tested the effect of specific manure types and quantities in meadows as well as the response of the trophic structure along management intensification gradients. From these results, we conclude that intensification of the current agricultural practices severely alters the soil fauna community and biodiversity. In our view, the extensive management, traditional for mountain areas, favors soil fauna communities that have adapted over a long time and ,thus, can be considered a sustainable reference condition for new production systems that consider the protection of soil diversity in order to conserve its essential ecosystem functions and services.

KEY WORDS: Soil biodiversity, agroecology, macro-invertebrates, land use change, land use intensity.

ORAL PRESENTATION

Note on the presence and distribution of *Parnassius apollo pumilus* Stichel in Aspromonte

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Parnassius apollo (Linnaeus) is a butterfly belonging to the Papilionidae family which has specific ecological characteristics, particularly regarding the trophic needs of the larvae that induce its preference for xerophytic mountain microenvironments, where the caterpillars feed on Crassulaceae belonging to the *Sedum* and *Sempervivum* genera. It is considered an "umbrella species" because its conservation allows the protection of a wide range of species coexisting in the same habitat, which are less known and therefore more difficult to protect. For the same reason, *P. apollo* can be a good bioindicator whose status provides information on the general conditions of the entire ecosystem; in particular, it appears to be a sensitive indicator of environmental quality in the monitoring of xerothermic biotopes at risk of disappearing. It also shows high intra-specific variability due to the distribution of its populations in isolated habitats. It is linked to medium and high-altitude sunny environments with abundance of outcropping rock and occurs in Italy from 400 to 2.500 msl, although it is by far more frequent above 1.000 msl. This species, which requires peculiar climatic conditions (cold winter, sunny summer), as well as open spaces (with a low shrubs cover) and a large surface of meadows, has a habitat characterized by high-altitude sites, where it prefers open areas and flowery valleys, but also rocky slopes. As well as other species within its genus, it is vulnerable due to the isolation of its populations produced by the last ice age, but in the places where it is present it is quite common. The range of the species is gradually decreasing throughout the European territory. In Italy it is quite common in the Alps. On the Apennines its presence is more sporadic, being absent in large sectors, showing, locally, a more or less marked degree of decline. In Calabria it is present in Aspromonte with the subspecies *pumilus*, also reported on the Sila massif, although currently it is quite legitimate to assume that the population present on Monte Botte Donato (CS) has become extinct; in fact, *P. apollo* is a steppe species, so it is possible that the Silane population has shrunk and eventually become extinct due to bioclimatic changes locally tending towards mesophilic regimes or because of other unknown factors. This work is based on the sightings of the butterfly occurring in the studies conducted by the entomologists of the LEEA during their years of research on the Aspromonte arthropods fauna, which allowed the creation of a distribution map based on the real presence of the species and on the identification of potential areas having the characteristics of suitable habitats for hosting it.

KEY WORDS: *Parnassius apollo*, Aspromonte, distribution.

POSTER

Research on Hemiptera Auchenorrhyncha of Aspromonte (southern Italy)

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Hemiptera Auchenorrhyncha are a basic component of the fauna of phytophagous insects in most terrestrial ecosystems and are also characterized by high taxonomic diversity. Despite important recent progress, knowledge about Italian fauna is still partially incomplete, both from a faunistic and taxonomic, as well as ecological point of view. The aim of the research was to investigate the Auchenorrhyncha fauna of the Aspromonte Massif, which is known to be an important biodiversity hotspot in the Apennine biogeographical context. The research, which is still ongoing, began in June 2018 and involved sampling carried out in different types of habitats and in different seasons, taking into account the considerable ecological diversity present in Aspromonte. In particular, forest habitats, mountain and summit grasslands, wetlands, as well as markedly xerophilous and Mediterranean scrub habitats were investigated. The first results confirm that the Auchenorrhyncha fauna of Aspromonte is of considerable interest and needs to be further studied, mainly for the presence of endemic species still unknown and for the faunistic and chorological interest of many findings. Some species new for science have been found, one already published (*Psammotettix aspromontanus* Poggi, Manti et Castiglione) and others whose description is in progress, belonging to the genera *Chlorita*, *Jassargus*, *Aconurella* and *Doratura*, whilst specimens attributable to others probably new taxa are under ongoing studies, belonging to the genera *Florodelphax* and *Arocephalus*. Among the numerous findings of faunistic interest, there is the collection, after forty years from the last published finding, of the endemic subspecies from Calabria *Conomelus lorifer calabricus* Dlabola, however new for Aspromonte. Another example of interesting finding is *Zygina luteipennis* Rey, a species for which there are only old citations from southern France and, generally, from Italy, without further specification; this finding makes possible a taxonomic study of this species, necessary for a long time. These results confirm once again the high interest of the entomological fauna of Aspromonte, characterized by a high rate of biodiversity and endemicity.

KEY WORDS: Hemiptera, Auchenorrhyncha, Aspromonte.

POSTER

Research on staphylinid beetles of the wetlands in the Aspromontano massif

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The Aspromonte massif, due to the peculiar geographical location and to the several geological and climatic features, is widely recognized as a very interessant biodiversity hotspot. In this area, located at the extreme Southern portion of the Apennines, the biodiversity knowledges are increasing continuously. In the district of the Aspromonte National Park many wetland areas with several different features occur, such as small water courses, ponds, swamps, ecc. Among those with highest biodiversity richness, rarest, and often affected by environmental changes, the Sphagnum bogs, the montane water courses, and the riparian lands inhabited by the fern *Woodwardia radicans* must be considered. The primary role played by the wetland areas in the biodiversity conservation is widely recognized even by several reccomendations, laws, and international agreements, all aimed to their protection. They play a primary role in the ecosystems as water regulators of the continental outflow, of the territory hydraulic regime and of the waters quality. This study represents the first contribution to the knowledge of the staphylinids beetles inhabiting the wetlands areas of the Aspromonte National Park. This study has been carried out using different monitoring and sampling techniques, aimed to collect the species living in several microhabitats. The main goal was studying biodiversity (community structure and its relative abundance of species) and to highlight even the characteristic species living in the several wetland areas occurring in the high differentiated environmental district of the Park. Staphylinid beetles, due to their high number of species known for the Italian fauna and to the high capability of inhabiting several macro and microhabitats, from the sea level to the high altitude alpine belts, are very suitable if used as bioindicators, mostly in a very high differentiated environmental district as the Aspromonte massif. The distributional models, the endemic species, and the relevant faunistic and biogeographical data emerging from this study emphasize and confirm the relevance of the montane wetland areas as biodiversity reserves in the Mediterranean basin.

KEY WORDS: Coleoptera, Staphylinidae, Aspromonte, wetlands.

POSTER

Pollinator community composition and network structure in a hydroelectric powerline easement corridor of Manitoba, Canada

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Powerline easement corridors occupy a vast, ever-increasing area with a high ecological potential. If managed accordingly, these lands could provide the full range of pollinator habitat requirements and act as refuges for native insects and wildflowers. In Manitoba (Canada), anthropogenic activity has led to a significant homogenization of the landscape and chronic habitat loss for pollinators and wildflowers. Prairies, meadows, and other pristine early-successional habitats in the province have diminished and become increasingly patchy, threatening the pollination community. Powerline easements may supply many resources for pollinators in these landscapes, however the effects of these corridors on insect communities and the ecosystem services they provide are poorly studied. To understand how powerlines in Manitoba can benefit pollinators, my project involved sampling pollination networks within an easement along a 2.5° latitudinal gradient. My objectives were to determine the biodiversity of insect pollinators within this corridor, and evaluate the effects of vegetation management and other environmental variables at the local and landscape scales on the pollination community. The vegetation management strategies in the corridor included active grazing, mechanical mowing, pasture fallowing, and integrated vegetation management (IVM). Additionally, I planned to identify keystone species in the community by evaluating species' strength (the sum of dependencies; a measure of a species' importance) and their role in the networks. From June to August 2020 I sampled bees, flies, and wasps in 15 sites along the corridor, describing the architecture of the pollination networks by the frequency and patterns of interactions among the insects and plants. Overall, 4959 insects were sampled, representing 259 pollinator species. Both pollinator richness and abundance increased with latitude as anthropogenic disturbance in the corridor and surrounding landscape decreased going northwards. Comparing environmental variables, I found that local vegetation management had the greatest effect on the pollination community. IVM sites harbored greater richness and abundance compared to sites under different management strategies. Furthermore, networks in IVM sites had lower connectance, but were larger and more specialized and modular. Simulating species coextinctions revealed that these networks were also more robust to extinction cascades: an indication of good ecosystem health. Although most keystone species in the networks were bees, several syrphids also had high species strength, with *Toxomerus marginatus* being the strongest pollinator in half of the networks. Most plants in the corridor were native to Manitoba, but three invasive species (*Melilotus albus*, *Cirsium arvense*, and *Sonchus arvensis*) were very strong in most communities, as their super-generalist nature made them central nodes in the networks. Powerline easements in Manitoba support a wide range of native wildflower and pollinator biodiversity, and the networks they form respond positively to targeted vegetation management practices. Management and conservation strategies within these corridors should always consider species' strength and their role in the ecosystem, and act accordingly to promote network robustness and health.

KEY WORDS: pollination networks, powerline easement corridors, vegetation management, landscape homogenization, biodiversity.

ORAL PRESENTATION

Contribution to knowledge of rare saproxylic beetles from Pollino National Park (Italy, Calabria)

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The Pollino National Park is an extensive protected area (about 1926 km2) located in the Southern Apennines along the border between Calabria and Basilicata regions of Italy. In this area there are well-preserved Mediterranean ecosystems including a wide range of woodland types. Between 1000 to 1800 m of altitude beech (*Fagus sylvatica*) is the most widespread vegetation type, with silver fir (*Abies alba*) also present as small clusters. This work represents a contribution to knowledge of rare saproxylic beetles from a representative Mediterranean old-growth forest. The sample area was in the locality "Abetina Spinazzeta" of Terranova di Pollino (southern Italy), an ancient forest stand dominated exclusively by silver fir. The specimens were collected by hand on dead wood and under bark. The most representative families sampled are Carabidae, Curculionidae, Elateridae, Prostomidae, Scarabaeidae, Tenebrionidae. Several of them were listed in the IUCN Red List of Threatened Species: *Omoglymmius germari* G., *Clinidium canaliculatum* C., *Prostomis mandibularis* F., *Cetonia aurata pisana* H., *Pissodes piceae* I., *Ampedus sanguineus* L., *Rhyncolus ater* L., *Stenomax aeneus* S., *Helops caeruleus* L. There are numerous rare and threatened saproxylic species associated with old-growth forests. For most of the beetles there is little information available about their life histories. In particular, we have found three saproxylic beetles that may be considered as important bioindicators: *O. germari* - this species is distributed in Central and Southern Europe and lives in old, fallen trunks of deciduous or conifer trees and is considered a virgin forest relict species. This saproxylic beetle is of great conservation value and was mentioned as coleopteran species useful for identifying forests of international importance. In Italy it has been defined as vulnerable in the red list. *C. canaliculatum* - this species exhibits a restricted distribution, and its geographic range covers exclusively Southern Italy and Greece, from Sicily to Taygetos in Morea. It has been defined as “biodiversity indicator” of forest ecosystems with high conservation value and vulnerable in the Italian red list, but the biology of the beetle remains anyway poorly investigated. *P. mandibularis* – the only representative of the Prostomidae family present in Europe, linked to necromass in deciduous forest systems. Considered almost threatened in the European IUCN red list. Aim of this work is to provide a checklist of the saproxylic beetles living in the Pollino National Park, a guild that comprises several rare and poorly known species bound to typical Mediterranean old-growth forests. On this way we try to construct a data base of indicator species, possibly of value on European scale, to be used for the assessment of the conservation status of natural old and well-preserved broad-leaved woodland communities on the Mediterranean mountains.

KEY WORDS: biodiversity indicator, Mediterranean old-growth forests.

POSTER

Status of the entomological species protected under the Habitat Directive 92/43/EEC in Calabria

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The Habitat Directive 92/43/EEC requires member states to draw up a National Report every six years about the conservation status of the species listed therein and the habitats of community interest present both inside and outside the Natura 2000 Network. (Reporting, art.17 of the HD). Through this report, the European Commission verifies the implementation and adoption of appropriate conservation measures by member states. The parameters that contribute to determining the overall conservation status of the species in a given biogeographical region are range, population, available habitat, and future perspectives. Every six years, each member state also adopts a National Monitoring Plan in order to indicate the monitoring priorities, according to the distribution typology of the species, and in relation to a range of methodological protocols (drawn up in Italy by the ISPRA) and to emergencies deriving from the conservation status. With 40 species of insects of community interest (Odonata 9, Orthoptera 3, Coleoptera 13 + 2 considered extinct, Lepidoptera 23), Italy is one of the European countries with the highest number of species listed in the Habitat Directive. The recently concluded PANLife – Natura 2000 Action Programme took stock of the conservation status of the species and habitats in Calabria, and evidenced that, though the region presents 185 sites (179 SCI/SAC and 6 SPA), there are few data available for most invertebrates. Of the 17 insect species reported for Calabria: 2 are not present in any Natura 2000 Network site (*Proserpinus proserpina, Papilio alexanor*); for some, only very few and ancient data exist (*Parnassius apollo, Eriogaster catax*); while for others, like *Cucujus cinnaberinus*, recent researches enabled to update the knowledge even at the population ecology grade, that is the level of accuracy required by the Directive. Drawing up a National Monitoring Plan requires, for many species, to switch from a distribution framework mainly based on “grey literature” to a more detailed one supported by a georeferenced database with cells of km 1x1 (“EEA reference grid”). Such a database, managed through GIS, should be taken into account for the reconstruction of the “chronogeonemies”, an important step for the identification of the conservation status of the single species, i.e. the contraction or expansion of the distribution range in the national and/or regional territory. Many other parameters help define a monitoring “priority”, such as the “rarity” in all its interpretations, the habitat conservation status, and – where possible – the data about population amount and density. The authors suggest new practical solutions to realize a National Monitoring Plan for the insects of the Habitat Directive that take into account the existing knowledge gap, with examples based on ongoing or concluded researches on species at risk of extinction such as the saproxylic beetles *Rhysodes sulcatus*.

KEY WORDS: Natura 2000 Network, National Monitoring Plan.

ORAL PRESENTATION

First data on dipteran of Sila National Park (Calabria, southern Italy)

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The project is titled “Monitoring Diptera species of medical and veterinary importance in the Sila National Park”. In this research, Diptera species were sampled in six areas selected for ecological and vegetational characteristics, using Malaise traps, bait bottle traps and chromotactic traps. The sampling sites include two pine forests, two beech forests and two wetlands in an altitude range between 1300 and 1850 meters above sea level. The sampled insects were kept in plastic tube containing 70% ethyl alcohol for taxonomic identification at Laboratory of Applied and Forensic entomology, University of Calabria. Flies’ composition and abundance data were analyzed and new data on faunistic and ecological aspects have been raised. Many families were identified (Anthomyiidae, Calliphoridae, Heleomyzidae, Lauxaniidae, Muscidae, Sarcophagidae, Drosophilidae) and species of forensic, medical and veterinary and economic importance are discovered for the first time in Southern Italy. *Anthomyia*, *Calliphora*, *Lucilia*, *Pollenia*, *Suillia*, *Homoneura*, *Helina*, *Phaonia* and *Sarcophaga* were the most abundant genus collected using three different traps from six locations inside the Sila National Park.

KEY WORDS: Diptera, sampling, flies traps, Calabria.

Contribution to faunal knowledge of the Neuroptera of Lomellina (Lombardy: Pavia province)

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Current knowledge of the distribution of Neuropterida (Raphidioptera, Megaloptera, Neuroptera) in the Po plain and in the plain of the neighbouring northern Adriatic area is mainly based on a set of data deriving from non-targeted research and which therefore covers the whole geographical area very irregularly. Surveys with faunal or faunal-ecological aims specifically concerning Neuropterida and limited to more circumscribed sectors of the northern Italy plain have so far been very scarce: an isolated exception considered Neuroptera in the area of the Comacchio valleys. For wider plain areas, covering whole regions, recent summary catalogues of Neuropterida for Veneto, Friuli-Venezia Giulia and Romagna have been made, but these lists also include the related Alpine and Apennine areas, without topographical distinction as regards the findings. This contribution provides a preliminary overview of the results of Neuroptera collecting carried out in the lower Lomellina area (northern Italy: Lombardy: Pavia province) over several decades, starting from the late seventies of the last century. Lomellina, the extreme western edge of the Lombardy plain between the lower Sesia and Ticino rivers and bordered by the Po to the south, corresponds to a large stretch of alluvial plain south of the line of resurgences, to which it owes abundant water. The area is mainly agricultural but also characterized by sand dunes unsuitable for many crops and which partly retain traces of the original forest cover. In the district under consideration, some collecting stations (located in the municipal areas of Pieve Albignola and Scaldasole) have been selected as being representative of environmental characteristics common to the entire Lomellina area. Approximately thirty species of Neuroptera were found there: *Nineta flava* (Scopoli), *Chrysotropia ciliata* (Wesmael), *Chrysopa perla* (Linnaeus), *Ch. walkeri* McLachlan, *Ch. formosa* Brauer, *Ch. abbreviata* Curtis, *Ch. viridana* Schneider, *Ch. pallens* (Rambur), *Pseudomallada inornatus* (Navás), *Pm. cf. picteti* (McLachlan), *Pm. prasinus* (Burmeister), *Pm. clathratus* (Schneider), *Chrysoperla* gruppo *carnea* (Stephens) (Chrysopidae); *Hemerobius humulinus* Linnaeus, *He. stigma* Stephens, *He. contumax* Tjeder, *He. micans* Olivier, *Wesmaelius subnebulosus* (Stephens), *Micromus angulatus* (Stephens), *Mi. variegatus* (Fabricius) (Hemerobiidae); *Sisyra nigra* (Retzius), *Si. terminalis* Curtis (Sisyridae); *Coniopteryx borealis* Tjeder, *Co. esbenpeterseni* Tjeder, *Conwentzia psociformis* (Curtis), *Semidalis aleyrodiformis* (Stephens) (Coniopterygidae); *Myrmeleon formicarius* Linnaeus, *My. inconspicuus* Rambur, *My. bore* (Tjeder), *Euroleon nostras* (Geoffroy in Fourcroy), *Megistopus flavicornis* (Rossi) (Myrmeleontidae). Two other ascertained species, *Psectra diptera* (Burmeister) (Hemerobiidae) and *Macronemurus appendiculatus* (Latreille) (Myrmeleontidae), were instead collected in another territory (municipal area of Mezzana Bigli) of the lower Lomellina. Some species, such as *Pm. inornatus*, *Ps. diptera* and *My. bore*, are undoubtedly of faunal interest, due to sporadic reports of them for Italy. For the latter species, which is very circumscribed in distribution, the sandy bed of the Po in Lomellina (Pavia) and in the Piacenza area (Emilia) seems to correspond to its southern distribution limit in Italy and perhaps also in Europe.

KEY WORDS: Neuroptera, northern Italy, Po plain, lower Lomellina, geographic distribution.

POSTER

The Italian Butterfly Monitoring Scheme: a European monitoring network using a Citizen Science approach

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The European Butterfly Monitoring Scheme (eBMS) is an initiative resulting from a collaboration between Butterfly Conservation Europe and the Centre for Ecology & Hydrology, to promote and develop butterfly monitoring through a representative European network involving citizens as main data collectors. The monitoring scheme aims to: 1) develop an online data entry system to grow the network - allowing new countries to submit data efficiently and in a standard format; 2) produce an effective database of repeated semi-quantitative butterfly counts; 3) provide indicators (e.g. Grassland Butterfly Indicator) and tools to estimate supra-national population trends; 4) establish actions to improve butterfly conservation. Butterflies are one of the most well-known groups of insects, and their charisma attracts thousands of passionate citizens; these aspects are essential for the growth of the project around Europe. The eBMS uses the standardized and shared Pollard Walk method, a transect that ensures the collection of replicable and comparable semi-quantitative estimation of the relative abundances of butterflies populations. Today eBMS involves 22 European Countries and more than 10000 transects are walked every year across Europe for a total of 17600 km walked since 1990. In 2019, the Italian BMS (ITBMS) was born thanks to the contribution of the European Pilot-Project ABLE (Assessing ButterfLies in Europe) and to the ITBMS National Committee that includes the University of Turin, University of Florence, and CREA Research Centre of Forestry and Wood of Calabria. The same research group has already carried out the Barcoding of Italian Butterflies project (www.barcodingitalianbutterflies.eu), for the creation of a national fauna database, and the Italian Butterfly Red List project (Bonelli et al. 2018). The ITBMS National Committee (i) develops an input system for transect data following the European guidelines, (ii) organizes training on butterfly identification, (iii) collects detailed spatial information and habitat categorization for transects, (iv) validates the data collected. Italy hosts 2/3 of European butterfly species and 60% of protected species, and such amazing diversity can create difficulties in butterfly identification. Consequently, our scheme requests a robust validation system and exhaustive training sessions. We organized 8 national workshops to engage new volunteers and many others at a local scale to ensure the spread of the project and to reach natural parks but also small associations, municipal administration, and farms. The Italian BMS counts 83 permanent transects of which 26 are located inside Natura 2000 sites. The community of volunteers grows day by day and more than 79 people joined the national scheme, a strong signal of raising awareness for the health state of butterflies and their protection. The involvement of citizens is providing information about population trends for more than 150 species, half of the whole Italian butterfly biodiversity, undoubtedly an unattainable goal using only the scientists' efforts.

KEY WORDS: butterfly conservation, standardized monitoring, citizen scientists, indicators.

POSTER

Saproxylic beetles of the Sila National Park listed in the EU Habitats Directive: updating distribution

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The Sila National Park extends for almost 74,000 hectares in the southern Apennine (Calabria region, southern Italy), covering most part of the territory of the largest European mountain plateau. It comprises the three main plateaus of Sila Grande, Sila Greca and Sila Piccola, with elevations reaching 1,928 m a.s.l. of Mt. Botte Donato. Despite the millennial exploitation of its wood resources, this territory still maintains well-preserved environments, including several old-growth Calabrian pine forests. We report the results, limited to beetles, of a monitoring program focused on the distribution and conservation status of the species of the Park included in the EU Habitats Directive. In order to assess the most appropriate conservation measures for habitats and threatened species of the protected area, several field surveys have been carried out in 2019. Furthermore, ecological and geonemic data on the target insects have been collected. The occurrence of four protected saproxylic beetles, namely *Cerambyx cerdo*, *Cucujus cinnaberinus*, *Osmoderma italicum* and *Rosalia alpina*, has been confirmed with new data inside the boundary of the protected area. Many other red-listed saproxylic beetles have been detected as well.
A detailed geolocalized database has been implemented, and the Standard Data Forms of five Sites of Community Importance of the Sila National Park have been updated.

KEY WORDS: biodiversity conservation, old-growth forest, Coleoptera, Italy.

Contribution to the knowledge of dragonflies of the Matese area

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Matese massif is one of the most representative mountain ranges of the Southern Apennines (Italy). It is located between two regions (Campania and Molise) and covers an area of approximately 1,440 km². The highest peak, Mount Miletto, has an elevation of 2,050 m. The territory is largely protected by the Matese National Park. The vegetation of the area is similar to that of other mountain ranges of the central and southern Apennines, with different bands that follow the altitude gradient. Although important biogeographical and ecological investigations have been carried out in the Matese in the past for various groups of insects, such as Orthoptera, Coleoptera, and Lepidoptera, knowledge of the entomofauna of the area is still very scarce. In this contribution, the authors present a checklist of the dragonflies for the Matese area carried out between 2010 and 2019. The investigations focused mainly on the western side of the massif, from the mountain area to the Volturno river. The reports mainly come from the various bodies of water that characterize the area under investigation, in particular: Gallo Matese, Matese and Telese lakes, Sava and Volturno rivers Le Mortine natural Oasis. The vegetation is made up of Helophytes with a predominance of Cyperaceae and Poaceae. There are also Hydrophytes and arboreal essences typical of riparian environments. From the research conducted, the species of dragonflies found amount to 37. Of this *Coenagrion mercuriale* (Charpentier) was included in Annex II of the Habitats Directive while *Oxygastra curtisii* (Dale) and *Cordulegaster trinacriae* (Waterston) in Annex II and IV. The aforementioned species are included in the Red List of Italian Dragonflies and classified as Near Threatened (NT). Of particular relevance is the discovery of *Coenagrion pulchellum* (Vander Linden) - NT - and *Sympetrum depressiusculum* (Sélys) Endangered (EN), the latter extremely rare in southern Italy. The remaining 32 species are Least Concern (LC) and therefore included in the Red Book of Italian Dragonflies. Furthermore, the checklist of the Matese Regional Park drawn up in 2004 also indicates *Ischnura pumilio* (Charpentier), and *Coenagrion caerulescens* (Schmidt), whose presence is not currently confirmed by the authors. Odonata are excellent indicators of the quality of the environment, especially water, and play an extremely important role in natural ecosystems. The presence of species included in the Habitats Directive and in the Red List confirms the naturalistic value of the mountain range for the conservation of threatened species. We also believe that the study of the distribution of individual species has an important predictive role and provides detailed data on the quality of the habitats. The results obtained increase the knowledge of the entomofauna of this important natural area. Finally, the research was carried out through the photographic research of dragonflies.

KEY WORDS: odonatofauna, checklist, red list, Habitat Directive, Protected Area.

POSTER

Biodiversity Monitoring of South Tyrol - first results with a focus on insect diversity

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A long-term biodiversity monitoring system was recently set up and implemented in South Tyrol. The primary focus of the monitoring is to survey species groups that respond sensitively to climate and land use change. Among others the focus lies on plants, bats, birds, butterflies, and grasshoppers. Further, special attention is also set on the investigation of stream and soil macro-invertebrates. The monitoring encompasses 320 terrestrial survey sites and 120 aquatic ones, of which a subset of 64 terrestrial and 48 aquatic sites are surveyed each year. The sites were selected using a stratified random sampling approach that takes great care in covering a wide range of natural and managed habitat types. The terrestrial monitoring will complete the first sampling after five years and the limnological after four years. For terrestrial insects, a wide range of sampling methods aim at an optimal collection of different groups in various habitats. For butterflies and grasshoppers, we apply a taxon-specific sampling procedure combining transect walks, area-time counts sweep netting, visual and acoustic control. For the other taxa we use sampling methods that aim at sampling a broad spectrum of invertebrate taxa using pitfall traps for ground-dwelling animals, soil core samples for the soil fauna, beating and sweeping of the vegetation for herbivores and pollinators.
We present the preliminary results of the first two years of the monitoring focusing on insect taxa. Specifically, after two sampling seasons we found 130 species of butterflies (corresponding to 70% of the South Tyrolean butterfly fauna) and 60 species of grasshopper (corresponding to 71% of the local grasshopper fauna). Furthermore, considering the pitfall catches we see a trend that intensively used agricultural sites, like vineyards, harbour a lower number of taxa. In future, more specific analyses with a broader data set will allow us to better identify patterns between different habitats, elevational belts, land-use types, and management intensities. In the long run, we expect to be able to detect changes in the insect fauna and attribute them to either climate change or specific land-use changes.

KEY WORDS: Biodiversity monitoring, South Tyrol, insects.

Reverse pollinator decline through a European monitoring scheme and develop targeted protection policies

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Pollinators, in a few decades jumped from being a zoological group of mere wildlife interest to an object of concern, scientific investigation and regulatory provisions at a global level. The fear for a decline in populations, supported by a now wide scientific production, has made us aware of the centrality of their role as suppliers of an essential ecosystem service such as plant reproduction, and raised the level of attention towards possible catastrophic scenarios regarding the security of food supplies, in addition to the fear of a possible decrease in biodiversity in a general sense. The first step necessary to prepare suitable protection measures is to draw up a list of endangered species. In 2014, the first European Red List of Bees was published on behalf of the European Commission and the coordination of the IUCN. Overall, 9.2% of the species are considered threatened and a further 5.2% are considered Near Threatened. However, the most striking result concerns the lack of knowledge: for 1,048 species (56.7%) of the European Union (27) to assess the risk of extinction, there is insufficient scientific knowledge. In 2018, an Italian Red List of Bees was published, assessing a first group of 151 species through IUCN criteria and categories. The report shows that 34 are affected by different threat levels. For the red lists to be updated, the preparation of annotated national checklists of species is needed, that is, highlighting the taxonomic criticalities and indicating to the scientific community dedicated to taxonomy the taxa to be verified through appropriate investigations. The European pollinator Initiative, launched in June 2018, aims to intervene on the decline of wild pollinators in the EU and includes 10 actions falling into three priority themes. Most of the actions concerning scientific research are included under Priority I: "Improving knowledge of pollinator decline, its causes and consequences". In particular, support is requested for monitoring and assess pollinator populations, and it is recommended to facilitate the exchange of knowledge and access to data. Relevant topics for research concern sampling techniques, sampling design and application of new indicators to monitor on the one hand the decline of pollinator populations, on the other to measure the effectiveness of contributions offered through the Common Agricultural Policy. Another crucial aspect to facilitate knowledge sharing and data access concerns data digitization from field and museum collections. On this, our country is not keeping up with the most advanced European research institutions. A decisive boost must be given through the creation of digital platforms able to transfer collection data from the field to the cloud in real time and the digitization of public collections. In order to prepare suitable protection policies, it is urgent to launch an Italian Pollinator Initiative, which integrates all stakeholders and civil society into a community of purpose.i volti a valutare l'attività in vivo del peptide carausicina in seguito a immunostimolazione degli insetti.

KEY WORDS: pollinators, bees, monitoring, red list, conservation policies, biodiversity, digitization.

ORAL PRESENTATION

Phylogeography of the steppe species *Hycleus polymorphus* reveals multiple interglacial refugia in Mediterranean mountains

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Quaternary climate fluctuations have strongly affected species distribution and their genetic diversity, with best documented patterns in the northern hemisphere. According to their response to climate changes, species have been divided into "temperate", "cold adapted" and "continental" elements. Most phylogeographic studies mainly focused on the first category, and far less is known about the impact of glacial cycles on the genetic diversity of continental species. *Hycleus polymorphus*, from the family of Meloidae, represents a good model to investigate the biogeographic history of a continental element, being distributed from central Asia to Europe, with many relict populations in the mountain ranges of the North Mediterranean Area. Using mitochondrial and nuclear markers (COI, CAD and ITS2), together with species distribution modelling (SDM) under the present and past bioclimatic envelopes, we reconstructed a phylogeographic scenario that could explain the current distribution of H. *polymorphus* and its population structure, in particular in the Mediterranean area. Since the taxonomy of the *H. polymorphus* species group is poorly resolved, we first performed a molecular phylogenetic analysis to define species boundaries within this group. Phylogenetic assessment revealed the inclusion within *H. polymorphus* of specimens ascribed to *H. humerosus* and within *H. atratus* of specimens identified as *H. zebraeus*. ITS2 data analysis showed a strong phylogeographic structure, with four main haplogroups distributed in (i) the Italian Alps, (ii) French Alps and Pyrenees, (iii) South Balkan mountain ranges and North East Pontic mountains, and (iv) North Dinaric Alps. These results, together with SDMs, suggests a scenario of glacial expansion of *H. polymorphus* populations from central Asia to south-western Europe, following the spread of steppe environments. During the last interglacial, suitable climatic conditions for this species in the Mediterranean area were confined to higher altitudes in different mountain ranges that acted as refugia and prompted the allopatric divergence into four main lineages.

KEY WORDS: continental elements, Pleistocene climate oscillations, fragmented distribution, species distribution models, ITS2, CAD, COI.

ORAL PRESENTATION

Contribution to the knowledge of calabrian spider fauna

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Spiders are the most abundant predators found in terrestrial ecosystems. More than 49,000 species are known worldwide and almost all are insect predators. The catalog of Italian spiders reports 435 genera, 52 families and 1688 of these species and subspecies of these 516 are mentioned in Calabria. The knowledge about the spider fauna of the region has seen an increase in recent years, however in a recent analysis of the Calabrian spider fauna show that knowledge is still very incomplete, both for regards the study of the different environments and habitats which characterize them both for the number of reported species. In light of this, data on collected spiders are presented about an annual sampling conducted in two olive orchards in northern Calabria, the first site on the Ionian coast of Cosenza in the area of Mirto-Crosia (CS) (39.616N 16.764E), the second located in the area of Rende (CS) (39.365N 16.230E). In the period between June 2019 and June 2020, sampling was carried out every two weeks using different methods (pitfall traps, frappage and sweep net) to collect different structural levels of the olive orchard ecosystem. This work reported a first summary of the data collected which are very interesting from a taxonomic and faunistic point of view. We collected more than 1,000 specimens belonging to 22 families and to over 100 different species. In the present work we report two new records from the Italian spider fauna: *Cyclosa groppalii* Pesarini (Araneidae) reported in Spain and Greece and *Palliduphantes arenicola* (Denis) (Linyphiidae), reported for France and Switzerland. Nineteen species are reported for the first time in Calabria: *Tegenaria hasperi* (Chyzer) (Agelenidae), *Dysdera kollari* Doblika (Dysderidae), *Gnaphosa alacris* Simon (Gnaphosidae), *Setaphis carmeli* (O. Pickard-Cambridge) (Gnaphosidae), *Marinarozelotes mutabilis* (Simon) (Gnaphosidae), *Zelotes fulvaster* (Simon) (Gnaphosidae) (second report for Italy), *Agyneta fuscipalpa* (C. L. Koch) (Linyphiidae), *Agyneta mesasiatica* Tanasevitch (Linyphiidae) (second report for Italy), *Agyneta mollis* (O. Pickard-Cambridge) (Linyphiidae), *Erigone autumnalis* Emerton (Linyphiidae), *Scutpelecopsis krausi* (Wunderlich) (Linyphiidae) species only recorded from the Venetian lagoon, *Trichoncus aurantiipes* Simon (Linyphiidae) (second report for Italy), *Eidmannella pallida* (Emerton) (Nesticidae) alien species native to North America (second report for Italy), *Lasaeola convexa* (Blackwall) (Theridiidae), *Lasaeola coracina* (C. L. Koch) (Theridiidae), *Bassaniodes bufo* (Dufour) (Thomisidae), *Bassaniodes caperatus* (Simon) (Thomisidae), *Ozyptila simplex* (O. Pickard-Cambridge) (Thomisidae), *Ozyptila trux* (Blackwall) (Thomisidae). *Dysdera monterossoi* Alicata (Dysderidae) endemic species of Calabria reported only for Aspromonte and *Dysdera silana* Alicata (Dysderidae) species known only on the basis of a single specimen from Monte Botte Donato of Sila, *Agyneta inermis* Tanasevitch (Linyphiidae), *Metopobactrus verticalis* (Simon) (Linyphiidae) and *Ozyptila pauxilla* (Simon) (Thomisidae). Some specimens collected belonging to the genera *Gongylidiellum* (Linyphiidae), *Palliduphantes* (Linyphiidae) and *Savignia* (Linyphiidae) are currently under study and probably referable to new species for science.

KEY WORDS: Araneae, spiders, checklist, olive, Calabria.

POSTER

First record of *Baryscapus silvestrii* in Calabria

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The authors report for the first time the presence of the eulophid *Baryscapus silvestrii* (Hymenoptera: Eulophidae) in Calabria (Southern Italy). The species has been found in the area of Mirto Crosia (CS) during a survey on the olive fly *Bactrocera oleae* (Diptera, Tephritidae). *B. silvestrii* parasitizing exclusively puparia and develops as primary endoparasitoid and has an endophagous and gregarious behaviour (Sasso et al. 2020). This species has been found for the first time in Italy by Viggiani et al. (2007) in the Salerno province. From there, the species expanded its distribution range to other regions of Italy. However, its biology remains largely unknown, due to difficult to laboratory rearing (Viggiani et al. 2007). Further sampling will be carried out with the aim of monitoring its spread and providing new knowledge on the biology and ecology of the species.

KEY WORDS: parasitoid, Eulophidae, biological control, *Bactrocera oleae*.

Lepidoptera Tortricidae described by P. Trematerra, 1990-2019

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In the present paper are reported a part of results obtained during faunistic and systematic studies realized from 1990 to 2019 on Lepidoptera Tortricidae. Over the years have been studied the materials deposited in various public or private Italian and foreign collections, as well as specimens collected during scientific expeditions in Italy, in other European countries (Portugal, Spain, France, Slovenia, Croatia, Malta, Greece) and in Africa (Mozambique and Ethiopia). Other tortricids were sent to me by entomologists who partecipated in excursions realized in south-eastern Russia, Kirghizistan, Nepal, the Russian Far-East and Argentina. Overall, we had the opportunity to study very interesting materials increasing our knowledge concerning this group of microlepidoptera, especially with regard to the Italian and European fauna. Even with the collaboration of Italian and foreign colleagues, have been reported for first time the presence of some taxa in various countries. Moreover, there was the possibility of discover and describing 2 new genera and 76 new species. In particular, from European territories have been described 12 new species from Italy (including Sardinia and Sicily), 3 new species were found in Central-Southern Portugal, 1 species from Malta and 3 species from Eastern Greece. From Africa one genus and 4 species have been described from Southern Mozambique and one new genus and 50 new species were described from the highlands of Ethiopia. To these are added the descriptions of one new species from South-Eastern Russia, one species from Kirghizistan, one species from the mountains of Nepal, 4 species from the territories of the Russian Far East, and 2 species from Northern Argentina.

KEY WORDS: Lepidoptera Tortricidae, new genera, new species, Trematerra.

POSTER

Butterflies of "Dune costiere da Torre Canne a Torre S. Leonardo" Regional Natural Park: diversity and distribution along a vegetational gradient

Alessio Vovlas, Daniela Clemente, Enrico Altini

A.P.S. Polyxena

The purpose of this contribution is to expose the results of the studies conducted on the butterfly assemblages in the Regional Natural Park of the "coastal dunes from Torre Canne to Torre S. Leonardo", in the territory of Ostuni (Brindisi, Puglia), to evaluate the ecological value of Mediterranean coastal dunes environment. Five different habitat types were investigated bi-weekly from April to September 2018, in order to assess the abundance, species richness and diversity of butterflies: Backdunes (EC Habitat 2250*), Grey Dunes with garigue vegetation (EC Habitat 6220*), Prairie with Grassy Heat, Woodland in a calcareous gorge and a Reconstructed Shrubland Area in an abandoned quarry. A total of 3886 individuals, belonging to 39 butterfly species and five families, were recorded during the sampling period. *Melanargia arge* and *Zerynthia cassandra* are the most important species occurring in the area. Both are Italian endemic species: *M. arge* is listed in Annexes II and IV of the "Habitat Directive" and probably is associated with *Stipa austroitalica*; *Z. cassandra* which was recently split from *Z. polyxena* (Annex IV). Another important species present is *Thymelicus acteon*, listed as near threatened (NT) in IUCN European Red List of Butterflies. The highest Shannon index value (H=2.50) and the highest number of species (N=28) were found in Reconstructed Shrubland habitat. Woodland was the habitat with the lowest Shannons' index value (H=2.05) and with less biodiversity of butterflies (22 different species, as Grey Dunes). The assemblages were also compared with multivariate method. The data obtained were compared with research carried out in previous years (since 2015) in order to obtain a picture of the changes through time. The results of several diversity indexes suggests that the presence of *Stipa austroitalica* in Grey Dunes plays an important ecological role in dunal succession and can promote the persistence of species of conservation importance. Investigations of this type can be of help to the Park Authority to plan management and monitoring actions in the various environments aimed at knowing, preserving, protecting and enhancing the biodiversity present.

KEY WORDS: Butterfly, *Melanargia arge*, *Zerynthia cassandra*, Coastal Dunes, Biodiversity.

POSTER

SESSION VI

Remote monitoring with e-traps: a technological tool to be improved

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The increase in the number of non-native species invading new territories demands the use of more efficient preventive monitoring methods. Among them, remote monitoring by electronic traps is a useful technological tool to integrate traditional pest monitoring and to facilitate and economize farmers' activities, especially in agricultural and forestry contexts difficult to reach. To reach a precision farming system, remote monitoring would provide timely data on monitored sites, making easier the planning of a possible treatment. The e-traps operating principle consists of the catching mechanism through the use of specific lures and the presence of a digital camera or different types of sensors: the specific substances attract the target pest inside the trap, the opto-acustic sensors detect its presence and the camera takes a picture of it in high definition. The images, regularly captured by the camera, are always available on PC or through apps on smartphones, providing real-time data referring to the target pest population. All the pictures taken by the camera are sent via GPRS or LTE connection to a technological platform where the collected specimens' counts are automatically processed by a recognition software that recorded the catches. Five models of electronic traps made by the brands Pessl Instruments and Trapview were tested and compared. These e-traps were experimentally used during usual monitoring activities of oriental fruit moth *Grapholita molesta* (Busk) (=*Cydia molesta*) (Lepidoptera: Tortricidae), whose damages on peach are recently increasing, and of the oriental fruit fly, *Bactrocera dorsalis* (Hendel) (Diptera: Tephritidae), accidentally found in Campania region in 2018 and 2019. Nine e-traps were placed in peach and fruit orchards in the province of Naples, Salerno, and Caserta; the data remotely registered by e-traps were compared with those collected in traditional traps (McPhail and Delta traps). The specific lures (specific sexual pheromone for G. molesta, torula yeast, and methyl-eugenol for *B. dorsalis*) were regularly replaced and the traditional traps in the same sites were weekly monitored. Monitoring activities show the following information about tested e-traps: The e-traps Trapview have regularly sent images of the catches, confirming to be particularly suitable for *G. molesta* monitoring; The Trapview model used for *B. dorsalis* monitoring did not catch any pest, although it was activated with generic food attractant (torula yeast), which usually allowed the capture of specimens belonging to different species of diptera in traditional traps; The models provided by Pessl Instruments showed several problems in data transmission, hence the system did not provide any photos in real time and it was difficult to assess the efficacy of the trap, making it impossible to carry out remote monitoring. The shortcomings found in this experience suggest that, to date, the monitoring activities cannot yet totally entrust to such electronic devices. However, further improvements in this field are needed to make the e-traps valid technological tools to facilitate farmers' and technicians' monitoring activities.

KEY WORDS: automatic pest detection, *Bactrocera dorsalis*, *Grapholita molesta*, precision agriculture.

POSTER

First report of predator *Raphidia mediterranea* in the vineyard

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The presence of Raphidioptera order in Italian vineyards is usually limited to the Inocelliidae family with the species *Parainocellia bicolor* (Costa) in the Peninsula and *Fibla maclachlani* (Albarda) in the Islands. On the other hand, there are no reports for the second family of the order, the Raphidiidae, although this is represented by a significantly larger numbers of species. Although their ecological characteristics make them mainly considered 'forest' insects, some species of Raphidiidae have been well adapted to agroecosystems. In Central Europe, are mentioned on fruit plants, for example species *Phaeostigma major* (Burmeister), *Subilla confinis* (Stephens), *Xanthostigma xanthostigma* (Schummel), *Venustoraphidia nigricollis* (Albarda), while in North America species of the genus Agulla have been adapted to citrus orchards. In the period 2019-2020, larvae and adults belonging to the Raphidiidae family were collected during the observations on Raphidioptera conducted in two organic vineyards (one for table grape and one for wine production) in Valenzano (BA). The larval population sampled during the winter under the rhytidome consisted mainly of *P. bicolor*, but Raphidiidae larvae were also present with 7% and 4% (in table grape and wine vineyards, respectively). The rearing of the larvae in the laboratory allowed their identification, it was *R. mediterranea* (Aspöck, Aspöck and Rausch), not reported so far on grapevines, but already known in Puglia, Basilicata, Lazio and Tuscany. On 2 yellow sticky traps settled in the center of the table grape vineyard for adult flight monitoring, only 3 adults of *R. mediterranea* were captured, compared to 15 of *P. bicolor*. Adults of *R. mediterranea* have been observed more frequently on the edge rows of the vineyard and in particular near the rows that were close to the hedgerow (*Rubus ulmifolius*, etc.). In preliminary tests, carried out in the laboratory, both adults and larvae of *R. mediterranea* shown excellent predatory capacity on 2nd-3rd age nymphs of *Planococcus ficus* (Signoret). Generally, 90% of the nymphs were predated regardless of the frequency of the prey availability. In both tests, the availability of the prey was offered on a weekly basis as well as every three days. In conclusion, the results demonstrate how *R. mediterranea* can constitute a significant share of the Raphidioptera order in the vineyard agroecosystem, even it is considerably smaller than *P. bicolor*. The presence of adults is probably favored by the proximity of hedges. Dissemination partially supported by: Project PRO.S.IT. (PROduttività e Sostenibilità in vITi-vinicoltura). PSR Basilicata 2014-2020. Misura 16 – Cooperazione. Sottomisura 16.1- Sostegno per la costituzione e la gestione dei Gruppi Operativi del PEI in materia di produttività e sostenibilità dell'agricoltura.

KEY WORDS: corticolous predator, organic vineyard, hedge, *Parainocellia bicolor*, *Planococcus ficus.*

POSTER

Preliminary observations on the larval presence of *Parainocellia bicolor* in the vineyard

Ferdinando Baldacchino[1], Marko Bjeljac[2] ,Ons Najar[2], Flutura Lamaj[2], Roberto A. Pantaleoni[3]

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Parainocellia bicolor (Costa) is a Raphidioptera Inocelliidae with corticolous larvae which actively prey on other arthropods. This species is able to colonize numerous tree crops. In the vineyard, where it has been repeatedly reported, it is presumed to be able to attack, and perhaps to contain, some important phytophages such as *Planococcus* spp., *Targionia vitis* (Signoret), the lepidoptera *Lobesia botrana* (Denis & Schiffermüller) (wintering chrysalis) and *Cryptoblabes gnidiella* (Milliere) (larvae and chrysalis). Nevertheless, the knowledge on the presence of *P. bicolor* in the vineyard is scarce. The short flight period of the adults and rather elusive habits of the larvae make their field study laborious. In Italy, there are previous observations on the presence of adults in Romagna vineyards (occasional observations) and Piedmont vineyards (sampling with yellow traps). During winter 2019-2020, in order to estimate the presence of the larvae of this species, surveys were conducted through peeling the bark. A first survey, on territorial scale, was carried out in commercial vineyards with different pest management systems (16 organic vineyards and 14 integrated vineyards) by peeling the trunk of 12 plants on a central row. A second more detailed study was conducted in the experimental vineyard of CIHEAM Bari, managed in organic and infested for few years by *Planococcus ficus* (Signoret). In this vineyard were examined 28 plants (10% of the plants present) distributed along 2 border and 2 central rows. In addition to the trunk, the peeling was also performed on the branches. The territorial investigation highlighted the presence of larvae in 16 vineyards out of 30 investigated, 7 of them are managed in organic and 9 in integrated management system. In vineyards with larvae presence, the number of larvae/plant was very low and without significant differences between organic (0.18±0.15) and integrated management system (0.18±0.09). Sampling in the experimental organic vineyard infested by *P. ficus* showed a density of 0.86±1.18 larvae/plant, considering only the larvae found on the trunk. The density rises to 2.21±1.89 larvae/plant if the presence of larvae is also considered on branches. The comparison between rows (border and central) did not show significant differences in the distribution of the larvae in the vineyard. However, on the edge row close to the hedge, the larval density was double (3.71±2.87 larvae/plant). In conclusion, the preliminary results suggest that the larval density is positively influenced by the availability of prey (*P. ficus*) rather than by the different management of the vineyard. Further investigations would be needed to investigate the role of hedges in the initial colonization of new vineyards.

KEY WORDS: corticolous predator, larval density, organic vineyard, *Planococcus ficus*, sampling.

POSTER

A survey of potential insect vectors of the plant pathogenic bacterium *Xylella fastidiosa* in the Basilicata Region, Italy

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Xylella fastidiosa subspecies *pauca*, responsible for the 'olive quick decline syndrome', is a xylem-inhabiting bacterium, exclusively transmitted by xylem sap-feeding insects. In Apulia, four xylem feeding species were collected: *Philaenus spumarius* L. (Hemiptera: Aphrophoridae), *Neophilaenus campestris* Fallen (Hemiptera: Aphrophoridae), *Cercopis sanguinolenta* (Scopoli) (Hemiptera: Cercopidae) and *Cicada orni* L. (Hemiptera: Cicadidae). Among them, *P. spumarius* is the most efficient and widespread vector species. The plain along the Ionian coast and the Matera hills, in Basilicata, border on Apulia Region and for this reason they are areas at high risk of introduction of this pathogen. The results of a three years study on potential insect vectors of the bacterium *X. fastidiosa* in the Matera area are presented here. Samplings were performed in eight olive groves, all characterized by the absence of insecticide applications: 3 olive groves located on the hill of Ferrandina , 2 olive groves at the bottom of the hill of Bernalda, 3 olive groves in the plane of Metapontum. Adults of potential vectors have been sampled in the first decade of each month starting from June until October of each year. A sweep net was used to collect insects from 10 randomly selected locations per grove. In each location, samples were collected by 30 sweeps on ground vegetation and 30 sweeps on tree canopies. The captured adults were killed in absolute ethanol and, afterwards, in the laboratory, the abdomen was dissected to check for mature eggs and parasitoid larvae. The insect species, potentially vector of *X. fastidiosa*, captured in the Matera area, were *P. spumarius*, *N. campestris*, *Lepyronia coleoptrata* L. (Hemiptera: Aphrophoridae) and *Cicadella viridis* L. (Hemiptera: Cicadellidae). All three spittlebug species completed only one generation a year. *Cicadella viridis*, in the sampled areas, appears to have three generations a year, with adult emergence on May, July and September. Among the four species, potentially vector of *X. fastidiosa* in the study area, *P. spumarius* was by far the most prevalent in 2016. In 2017 and 2018 the number of individuals caught was modest for all species. *Philaenus spumarius* represented 66% of the individuals overall caught, *C. viridis* 16%, *L. coleoptrata* 12% and N. *campestris* only 6%. The presence of *C. viridis*, with very high catch rates in some places, has been strictly linked to the wetlands inside the olive groves. Parasitization was only ascertained for *N. campestris*. In fact, in 2017 and only in the olive groves in Ferrandina, individuals parasitized by pipunculid Diptera larvae were caught. The parasitization rate was on average 38.3%. During the three years study, 4 potential vector species of *X. fastidiosa* were caught in the olive groves of Basilicata. Two of these species, *L. coleoptrata* and *C. viridis* have not been reported for the olive groves of Apulia, where the pathogen is present.

KEY WORDS: *Xylella fastidiosa*, *Philaenus spumarius*, *Neophilaenus campestris*, *Lepyronia coleoptrata*, *Cicadella viridis*.

POSTER

Insect seasonal assemblages within olive groves in an ecological management system: structure variation between tree canopy and herbaceous cover

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The insect entomofauna of two olive groves was collected in relation to the plant host phenology evolution, in Beni Mered area (Blida) located in the Central Mitidja (Northern Algeria), during 7 months in 2011-2012. We have evidenced the spatiotemporal occurrence of four functional groups relating to their structure and installation in each studied orchard. The seasonal differences in stability of the phytophagous, predatory, and the flower dwelling communities highlight an early or late installation according to the order of the functional groups arrival in both orchards. In the table olive plot, there is simultaneous presence of the 4 communities highlighting a certain homogeneity, while in the oil olive plot, the trophic assemblages settle one after the other: first pollinators and Diptera with an undefined diet, then the predators and the phytophagous insects. The phytophagous within the table olive grove consist of two heterogeneous communities comprising 14 species where the Mediterranean black scale *Saissetia oleae* dominates. Predators and pollinators form well-balanced and highly diverse homogeneous communities consisting of 19 and 9 taxa, respectively. The phytophagous of the olive oil grove constitute a single community of heterogeneous structure, the functional groups of predators and pollinators have 15 and 10 representatives however. The variations of their abundances seem to be dependent on local environmental variables such as the heterogeneity of vegetation cover and seasonal availability of the herbaceous plants layer. The most representative of the spontaneous flora, namely *Inula viscosa* and *Chrysanthemum coronarium i*n the plot of table olive, and *I. viscosa* , *Chenopodium album*, *Crepis vesicaria*, *Convolvulus althioides* and *Bromus mollis* in the oil olive grove, vary in abundance depending on the season and host more insects than the rest of the adventitious flora. It can be seen that insects are attracted for a given season by characteristic plant species; their average abundance on these plants varies between 40 and 80%. The total seasonal ratio is the most important in autumn followed by the spring ratio, in particular on the plant species of *Oryzopsis milliaceae*, *Galactites tomentosa*, *C. althioides*, *Chrysanthemum segetum* and *C. coronarium* as well as *I. viscosa* in the table olive grove. In the olive oil plot, the highest abundance ratios are inferred to the spring season mainly on *Echium plantagineum*, *C. album* and *I. viscosa*. The presence of phytophagous insects in the olive grove is reduced in the spring-summer season on a single plant *C. althioides* while in the table olive grove the phytophagous insects on the adventitious flora are present on a greater number of plants, *Cychorium vitybus*, *C. coronarium*, *I. viscosa*, *C. althioides* and *Medicago hispida*, especially during the automn period. Predator communities are present with abundant populations during the spring-summer period in the olive oil plot and during the autumn summer period in the table olive plot. We have explained the differences in structure and composition of seasonal assemblages at the spatial scale by the fact that different species contribute more strongly than others to the communities of the canopy and those of the adventitious flora.

KEY WORDS: functional diversity, olive grove, vegetation cover, trophic groups, ecological management, metapopulations.

ORAL PRESENTATION

New insights into the current distribution of *Bemisia tabaci* cryptic species in Italy

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Bemisia tabaci (Hemiptera: Aleyrodidae) is a key pest of horticultural and ornamental crops worldwide, primarily owing to its high efficiency in transmitting a large number of plant viruses. In Italy, this whitefly is responsible for severe economic losses as vector of begomoviruses, such as tomato leaf curl New Delhi virus (ToLCNDV) that has recently endangered cucurbit crops. Within the *B. tabaci* complex, the Mediterranean (MED) and Middle East–Asia Minor 1 (MEAM1) species (formerly referred to as biotype Q and B respectively) are known to be well-established in southern Italy as well as in Sicily and Sardinia, with MED populations that progressively increased in those areas where intensive farming is applied. The occurrence of *B. tabaci* populations reported since 2016 in Latium region (central Italy) associated with ToLCNDV outbreaks, has prompted new surveys on the distribution of the *B. tabaci* species in Italy. Several protected and open-field cultivations of both horticultural and ornamental crops as well as weeds were surveyed between 2017 and 2019 in central and southern Italy and in the main islands Sicily and Sardinia. Whitefly samples were collected at all the inspected sites, and most of them belonged to the MED species. The haplotype MEDQ2 prevailed in the samples collected on cucurbit and solanaceous crops in southern Latium (87.5% of the collected specimens) as well as in the samples collected on horticultural and ornamental crops in Molise (75%) and Tuscany (85%). Single and mixed MEDQ1-Q2 populations were found to be present on crops and weeds surveyed in Marche region as well as in the southern regions Campania and Calabria. In general, in continental Italy most of the specimens collected in open-field cultivations and weeds belonged to MEDQ1 (60%) whereas Q2 was prevailing under greenhouses (87,6%), likely due to its highest tolerance to high temperatures and insecticide treatments. The surveys carried out in Sicily confirmed the high incidence of MEDQ2 in protected conditions (95%). A different scenario was observed in Sardinia, where only MEDQ1 specimens were collected on a wide range of crops and weeds at both coastal and inner horticultural sites. Sardinia is particularly isolated from both geographical and commercial points of view and this has likely prevented the introduction of MEDQ2 into the island so far. This survey provides evidence that *B. tabaci* has become established in central Italy, where *Trialeurodes vaporariorum* was thought to be the prevalent whitefly species up until today. The spread of B. tabaci from southern to central Italy as well as the prevalence of the MED species may have been favored by the prolonged mild winters and warm to hot summers occurring in the Mediterranean basin in the last two decades. This northward spread is a great concern for the epidemiology of *B. tabaci*-transmitted viruses already present in Italy and rises the risk of introduction of other alien viruses by international trades. This work was supported by the project EMERAMB “Emergent viruses and virus vectors in Mediterranean Basin crops” that is funded through theARIMNet2 2015 Call. ARIMNet2 (2014-2017) is an ERA-NET coordinated by INRA (France); it has received funding from the European Union’s Seventh Framework Programme for research, technological development and demonstration under grant agreement no. 618127.

KEY WORDS: whitefly, MEAM1 species, MED species, horticultural crops, ornamental crops.

POSTER

Toxic and repellent effects of hydrosols against hemipteran vectors evaluated in laboratory bioassays

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In recent years, several regulations and safety measures were issued to limit the use of conventional synthetic pesticides and reduce their impact on both human health and environment. The essential oils (EO) extracted from aromatic plants are natural compounds that have been widely investigated for their possible use in integrated pest management programs. The hydrosols are by-products of the EO distillation, still containing a certain proportion of the plant bioactive components that dissolve in water phase during distillation. Fungitoxic and antibacterial activities of hydrosols have been recently assessed whereas little is known about their effects on insects. In the frame of a project testing several natural compounds in cucurbit crop protection, hydrosols are under evaluation for their effects against *Aphis gossypii* (Hemipera: Aphididae), a key vector of cucurbit viruses. Hydrosols extracted from oregano (*Origanum vulgare* L.), thyme (*Thymus vulgaris* L.), rosemary (*Rosmarinus officinalis* L.), apple mint (*Mentha suaveolens* Ehrh.) and fennel (*Foeniculum vulgare* Mill.) were tested in laboratory bioassays, and the effects on insect survival, fecundity and settling behaviour were recorded. The toxicity assays against *A. gossypii* were carried out in Petri dishes where ten apterous adult aphids were transferred onto a cucurbit leaf that was sprayed with the hydrosol or deionized water (control); the mortality rate as well as the parthenogenetic reproduction rate were evaluated. The repellence activity was assessed by recording the number of aphids settled on treated or control cucurbit leaves placed together in a Petri dish. The apple-mint hydrosol showed the most promising results in both toxicity and repellence bioassays. The mortality rate recorded on treated leaves was significantly higher than the control even at 24 hours after the treatment, and about 80% of specimens died after 48 hours. The toxic effect also significantly affected the number of offspring recorded on treated leaves. In the repellence experiment, the number of aphids settled on the treated leaf was significantly lower compared to the control within 24 hours after the treatment. The fennel hydrosol also proved to be repellent against A. gossypii, with an average of less than one individual per treated leaf recorded until 4 hours after the treatment. The fennel hydrosol did not significantly affect the rate of aphid mortality and offspring production. The correlation between the biologic activity of the apple-mint and fennel hydrosols and their chemical composition assessed by GC-MS are under evaluation. Anyway, their detrimental effects on *A. gossypii* encourage large-scale experiments towards a possible greenhouse application of these compounds against hemipteran vectors. Further toxicity and repellence assays on potted cucurbit plants kept under controlled conditions in insect-proof boxes are ongoing against *A. gossypii* and *Bemisia tabaci* (Hemiptera: Aleyrodidae). Particularly, the repellent effect can contribute to delay the feeding activity of the vectors and thus reduce the rates of virus acquisition and transmission. The low costs of production further increase the interest for the potential use of hydrosols in pest management programs. This work was supported by Latium Region, Convenzione N. Registro 21706, CUCURBIOMID project (ID 85-2017-15024) within the Call: “Progetti di Gruppi di Ricerca – Conoscenza e cooperazione per un nuovo modello di sviluppo” (L.R. 13/2008 - art. 4).

KEY WORDS: hydrosols, essential oil distillation, *Aphis gossypii*.

POSTER

Multifaceted sublethal effects of neurotoxic insecticide microdoses on the orientation of the spotted wing Drosophila

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Exposure to sublethal doses of insecticides can result in various physiological and behavioral traits of the exposed organisms. Insects have evolved fine mechanisms for host detection and among these the orientation towards host-borne volatiles are of paramount importance for pest reproduction and thus, in the case of insect pests, for yield losses. The spotted wing drosophila, *Drosophila suzukii* (Matsumura) (Diptera: Drosophilidae), is a devastating invasive pest of soft-skinned fruits. In this context, we first characterized the baseline toxicity of four neurotoxic insecticides through ingestion during larval development and via adult residual exposure. Then, the estimated lethal concentrations 10% (LC10) for the four chemicals were used to carry out a multilevel analysis of their sublethal effects on orientation. Electroantennographic (EAG), RT-qPCR for nine odorant receptor genes and behavioral experiments at the olfactometer were carried out on insects survived to the exposure. The resulting LC10s proved to be extremely lower than insecticide label doses and of their maximum residual limit allowed on fruits at harvest. More interestingly, the expression of the tested genes and the EAG responses were affected at varying levels in exposed insects. Such sublethal effects may have been responsible of the impaired capacity of the exposed flies to orient toward volatile cues at the olfactometer in the behavioral experiments. The results strongly support the need to comprehensively study the potential sublethal effects of agrochemicals, even at very low doses, on key biological traits of insect pests. Moreover, these results stress the need to reconsider the real impact of neurotoxic insecticide residues on the biocenoses present in cropping environments.

KEY WORDS: electroantennography, olfactometer, Real Time PCR, odorant receptors, chemical control.

ORAL PRESENTATION

Potential toxicity of plant essential oils toward the brown marmorated stink Bug

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The arrival of the invasive Brown Marmorated Stink Bug, *Halyomorpha halys* Stål (Hemiptera: Pentatomidae), in Italy prompted the need of developing effective and sustainable control strategies. Among these, biopesticides and in particular plant-derived insecticides represent a promising tool. We carried out laboratory bioassays for assessing the potential toxicity of five nanoformulated essential oils (EOs) (anice, garlic, fennel, lavender and peppermint) applied on *H. halys.* The egg masses were topically treated with six increasing concentrations of each EO. The survival from egg to N2 was used as lethal endpoint for calculating the concentration-mortality curves. The estimated lethal concentrations 10% (LC_{10}) ranged between 5 and 30 g of EO/L with anice EO being the most toxic and lavender the least; while the LC_{50} ranged from 87 to 150 g of EO/L for garlic and peppermint, respectively. Two mixes of the five EOs were then tested and none of them caused significant Egg-N2 mortality compared to the control, very likely because of antagonistic effects among EOs' constituents. Then, LC_{10}s and LC_{50}s of each EO were used to assess the sublethal effects on the individuals survived to the EO exposure. Egg-adult survival, development time and weight of the developed adults were assessed. The egg-adult survival, but not the development time, was 70% lower than the untreated control following egg exposure to the LC_{50}s of all EOs and the LC_{10} of peppermint and garlic. The weight of the developed adults was slightly affected only in the case of females independently of the tested concentrations. Altogether these results stress the fact that the EOs' insecticidal property is not represented by direct acute toxicity, but by sublethal effects very likely mediated by microbial symbionts. Although further EOs' insecticidal effects could be tested in residual exposure and repellency experiments, the obtained results provide clear evidences that EOs could represent a valid control tool when topically applied to the eggs of this pest. Therefore, the inclusion of these EOs into Integrated Pest Management programs for the control of *H. halys* could be taken into account

KEY WORDS: Botanicals, Invasive species, Nanoemulsion, Real Time PCR, Symbiont bacteria.

POSTER

Dispersal of *Philaenus spumarius*, vector of *Xylella fastidiosa*, in olive grove and meadow agroecosystems

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The spread of insect vectors is a key point in understanding the epidemiology of insect-borne plant diseases and in assessing effective vector control strategies. The transmission of the bacterium *Xylella fastidiosa* Wells in Europe is mainly due to spittlebugs (Hemiptera: Aphrophoridae). The meadow spittlebug *Philaenus spumarius* L. plays the major role in the spread of *X. fastidiosa* ssp. *pauca* ST53 on olives in the Apulia region (Italy). Despite its importance, little information is available on the mobility of *P. spumarius*. Dispersal capabilities of the meadow spittlebug were hence investigated by mark-release-recapture experiments. Several trials were carried out from May to October of 2016–2017 in two agroecosystems: olive grove and grass meadow, in the Apulia and the Piedmont regions (Italy), respectively. Adults of *P. spumarius* of both sexes were collected in natural grassland habitats, marked with an aqueous solution of albumin (10%) and then released at a single point in the centre of the experimental area. A total of ≈6500 and ≈3000 spittlebugs were release during Apulia and Piedmont experiments, respectively. Spittlebugs were recaptured through sweep net in fixed sampling points located up to a maximum of 250 m from release point. Recaptures were conducted every two or three days up to 17 days after the release. Marked insects were identified via an indirect ELISA. The dispersal capacity of *P. spumarius* adults was described estimating the probability density function describing the distribution of the end locations of insects relative to the source point (i.e. the dispersal kernel). Diffusion rates in the two agroecosystems were estimated under the hypothesis of a random walk and applying a Gaussian kernel. Spittlebugs were recaptured up to 155 meters and 200 m in Apulia and Piedmont, with 50% of total recaptures within 30 m and 40 m from release point, respectively. Results showed a high variability in the estimated median distance from the release point, ranging from 25 m (Apulia) to 35 m (Piedmont) per day, and from 356 m (Apulia) to 507 m for the estimated median of adult life-long dispersion. Estimated spread parameters of *P. spumarius* are fundamental information to model *X. fastidiosa* spread in Apulia and other European foci. Further researches are needed to investigate on influence of both vector's physiology and environmental factors on movement tendency and directionality of *P. spumarius*.

KEY WORDS: *Xylella fastidiosa*, spittlebugs, insect vectors, mark-release-recapture, dispersal, olive quick decline syndrome.

ORAL PRESENTATION

Philaenus italosignus: new data for Tuscany

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In a study funded by "Fondazione CR di Firenze" to survey potential insect vectors of *Xylella fastidiosa* in Tuscany, as well as their natural enemies, we recorded, for the first time in this Region, the presence of the spittle bug *Philaenus italosignus* Drosopoulos et Ramane. Like the congeneric *P. spumarius* (L.), *P. italosignus* can acquire this bacterium and transmit it to some plant hosts, among which olive. In this study, we gathered new data about this little-known species to learn more about its role as a vector. Samplings took place in 2019 in an olive grove inside the Maremma Regional Park (Province of Grosseto). The lily *Asphodelus ramosus* L., the only known host plant for *P. italosignus*' nymphs, is common in this park. Spittle bugs were collected from weeds and olive trees by using sweeping nets or aspirators. In the laboratory, all specimens were measured and identified. First, adult specimens were analysed morphologically (e.g. male genitalia), then a sub-sample was used for molecular analysis, amplifying a fragment of the cytochrome c oxidase I (COI) gene. A total of 231 adult specimens of *P. italosignus* were collected, 208 of which were sampled on the lily, 17 on olives, and six on other weeds. Adult size resulted larger in *P. italosignus* when compared to *P. spumarius*, which is in agreement with the few information available in the literature. Females' length (7,1-8,1 mm) was practically identical to that reported by the authors of the species, while for males our measurements (6,6-7,7 mm) were slightly above the reported range. Molecular tests confirmed the identification for *P. italosignus*, whose DNA sequences matched the one available in Genbank with an identity of 100%. In our study period *P. italosignus*' occurence on olives (17 specimens) was even higher than *P. spumarius*' (six specimens). Though preliminary and referring to a partial life cycle period for both species, this result highlights the potential significance of *P. italosignus* in environments where the lily is common. Further research is necessary to study more deeply its biology and ethology to better address its real role as vector in typical environments of southern Tuscany.

KEY WORDS: vectors, *Xylella fastidiosa*, olive.

POSTER

Preliminary data on the phenology and the distribution *Philaenus italosignus* in Apulia (southern Italy)

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Spittlebugs received in the past few years major attention by the research community and farmers, as consequence of the introduction and spread in Europe of the quarantine bacterium *Xylella fastidiosa*. Spittlebugs represent in fact the predominant xylem feeders in Europe. Indeed, both *Philaenus spumarius* L. and *Neophilaenus campestris* (Fallén) have been demonstrated to be competent vector and be quite common in olive groves in the Italian region of Apulia where a highly pathogenic bacterial strain is decimating this important crop species. To further investigate the risks posed by other species of spittlebugs, the phenology, host preference and distribution of *Philaenus italosignus* Drosopoulos & Remane (Aphrophoridae) was also investigated through a large scale field survey carried out in the region. *P. italosignus* was first described in Apulia in 2000 and then also reported from Sicily, Lazio and more recently from Tuscany; in the latter case associated with an outbreak of *X. fastidiosa*. Given the continuous expansion of the *Xylella*-epidemic in Apulia towards areas where beside olives different crops are cultivated, and the notion that *P. italosignus* is also a competent vector of *X. fastidiosa*, from 2017 to 2019 surveys were focused to document the presence and occurrence in different areas, to characterize the phenology and juvenile morphometry. Areas with abundant presence of *Asphodelus* spp., on which juveniles develop, were prioritized for surveys. The data collected showed that (i) the density of individuals per m2 can be particularly high, reaching even 400 individuals/m2; (ii) even if *Asphodelus* spp. was abundant in some locations, no specimens of this species could be recorded, as for example in the province of Lecce. Moreover, the results of periodic monitoring carried out in locations at different altitudes have also shown, as expected, that the juveniles appeared about 7 days earlier in the coastal area than those located in the inland of the region. The comparison of the phenology of *P. italosignus* with that of the most common species, *P. spumarius*, showed that juveniles of *P. italosignus* appear 20-30 days earlier than *P. spumarius*, and consequently adults are captured 10-25 days earlier (depending on the vintage). With reference to the adults, unlike the juveniles, they are relatively polyphagous, feeding in particular on cherry trees. One aspect to highlight is that although there are ecological niches with a very high population density of juveniles, no correlation was retrieved between juvenile population density and capture of adults. Generally, a very low number of adults (1 individual/sample unit) if not any could be collected by sweep net. It would therefore be interesting to study the population dynamics of adults, in order to better understand their dispersion or disclose the "refuge" sites. It will also be useful to determine the climatic and/or ecological conditions that delimit the areas of diffusion of this species.

KEY WORDS: *Xylella fastidiosa*, insect vectors, olive quick decline syndrome, *Asphodelus*, spittlebug.

ORAL PRESENTATION

Evaluation of sticky traps for monitoring spittlebugs, the main vector species of *X. fastidiosa* in Europe

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Upon the discovery of several European outbreaks of the quarantine bacterium Xylella fastidiosa, intense research programs and extensive surveys have been settled to tackle the insect vector populations responsible for the spread of the infections in the outbreaks. These investigations have clearly showed that the spittlebugs *Philaenus spumarius* L. and *Neophilaenus campestris* (Fallén) are the most common vector species, being frequently captured in crops and natural environment. Moreover, transmission experiments demonstrated their ability to acquire and transmit the most aggressive bacterial strain so far known in Europe, which is causing the severe epidemics on olives in the Apulia region (southern Italy). Given the detrimental impact of this epidemic spread, monitoring the presence of spittlebugs in different crops in this region, is relevant for the implementation of vector control strategies. To this end, between 2018 and 2019, sticky traps were evaluated as tool for monitoring the insect populations in olive, almond and cherry orchards from late spring to late summer when adults are known to occur. In 2018, different coloured sticky traps (white, red, blue, yellow, yellow with circle-pattern and yellow with line-pattern (Russel IPM, UK) were initially tested to determine the most attractive colour. Overall, the results showed that the yellow sticky traps were the most attractive for both spittlebugs when compared to white, red and blue traps. Thus, a large field validation was then carried out in 2018 and 2019, to further compare the efficiency of these three yellow-coloured traps, to assess the effects of trap height. Traps were inspected and changed every two weeks and concomitantly sweep netting was also carried out for a comparative assessment. The results indicated that an integrated use of both approaches can be very useful for monitoring the presence and the dynamics of the adult populations in crops and natural environment. More specifically, sweep net were more efficient in capturing insects soon after the emergence of the adults, whereas later on in the season the yellow sticky traps yielded higher performance in capturing the adults moving from one vegetation compartment to another (i.e. from the ground vegetation to tree canopies). Yellow sticky traps were more efficient than sweep net in orchards with medium population density, whereas no significant differences were recorded between the two applied methods in orchards with high population density. In situation with low population density, traps were less efficient although still provided evidence on the presence and occurrence of the target species. Overall, the combination of both approaches can effectively support monitoring programs for vectors controls, in particular to support surveys for spittlebugs in the so called "demarcated areas" where control actions are mandatory or highly recommended to control the vector populations.

KEY WORDS: *Philaenus spumarius*, *Neophilaenus campestris*, olive grove, almond grove, cherry grove.

POSTER

Observations on the presence of *Philaenus spumarius* in cork oak forests and olive groves in Sardinia (Italy)

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Xylella fastidiosa is a quarantine pathogen reported for the first time in Europe in 2013 in a delimited area of Apulia (Italy), in which is the major causal agent of the olive quick decline syndrome. The bacterium has been later isolated also in Tuscany, Corsica, southern France, Spain, and Portugal on ornamental plants and Mediterranean maquis species. In view of its wide distribution in the adjacent island Corsica, the Sardinia regional plant protection service, within the European program "Monitoring Pest Survey", has funded a project aimed at studying the distribution of insect vectors in Sardinian environments. The main vector of *X. fastidiosa* in Europe is *Philaenus spumarius* (L.) (Hemiptera: Aphrophoridae), ubiquitous and extremely polyphagous species developing on more than 300 species, even though the pre-imaginal development is completed only in some host species. The population density of *P. spumarius* in a monitoring site can be affected by the abundance and diversity of host species in the surrounding environment. This requires further observations at the landscape level in order to identify reservoir habitats and potential infestation foci in the adjacent areas. Two environments with olive and cork oak, which are susceptible to *X. fastidiosa* subsp. *pauca* and subsp. *multiplex*, respectively, were monitored in Sardinia in 2019. The pest density and its host preference were assessed in olive groves and cork oak forests in different areas of Sardinia by counting the number of foams inside quadrants of 0.25 m2, and collecting adults with a sweep net. Observations carried out in cork oak forests showed the preference of P. *spumarius* nymphs for *Cistus monspeliensis*, *Carlina corymbosa*, *Daucus carota*, *Scandix pecten-veneris*, *Hypochaeris* sp. and *Erigeron canadensis*. Adults were captured mainly on *C. monspeliensis* and *Erica arborea*. Olive groves showed a higher diversity of host species compared to cork oak forests, and the preferred hosts of nymphs were *Pallenis spinosa*, *Dittrichia viscosa*, *Galactites tomentosa*, *Scabiosa atropurpurea, D. carota* and *Foeniculum vulgare*. In olive agroecosystems, *P. spumarius* adults were collected mostly from *Olea europaea*, *Olea oleaster*, *Diplotaxis erucoides* and *D. viscosa*. Real time PCR of 414 meadow spittlebug adults and 418 samples of host plants of *X. fastidiosa* did not highlight the presence of the bacterium in Sardinia. However, the proximity between Sardinia and Corsica and common forest environments require a continuous monitoring of *P. spumarius* in those environments representing potential reservoir habitats of the bacterium.

KEY WORDS: meadow spittlebug, *Xylella fastidiosa*, reservoir habitat.

POSTER

Laboratory and field evaluation of propolis and rock powder in preventing *Bactrocera oleae* infestation

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The olive fruit fly *Bactrocera oleae* (Rossi) (Diptera: Tephritidae) is the key pest of olive trees in several areas of the world. Pest management has been extensively studied for this pest; however, the application of pesticides, both as bait sprays and cover sprays, is still the most common control method despite recognized drawbacks. Therefore, given the needs for sustainable and eco-friendly methods, preventive tools must be considered. Here, we tested the efficacy of two products, rock powder and propolis, in preventing *B. oleae* infestation and compared them with a copper formulate. A field trial was conducted from July till November 2020 in an olive orchard in Spello (Perugia, Italy). Three olive varieties were sprayed with four treatments (rock powder, propolis, the mixture of both, and copper oxychloride) and the control (water). In the laboratory, we conducted no-choice tests to clarify the effects of the products on *B. oleae* female behavior, evaluating a possible deterrence response. All the products, but propolis alone, assured reduction of *B. oleae* attacks in field conditions. Additionally, results show a synergistic effect of propolis and rock powder, and their mixture allowed at least 36% reduction of the attacks on table olives throughout the season. In laboratory conditions, all treatments proved to be deterrent to oviposition.

KEY WORDS: Olive fruit fly, copper, preventive methods, insect behaviour.

Carpophilus: a new or ancient threat to Campanian walnut production?

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Invasive species introductions are increasingly frequent and threaten constantly agriculture activities. In Italy these invasive processes concerned many agroecosystems including walnut trees, which, recently, have been affected by the arrival of at least three invasive species. In 2019, CNR-IPSP staff, involved in the URCoFi project (Regional Unit for Phytosanitary Coordination), found a large number of sap beetles (larvae and adults) on stored walnuts. Following the first finding, in Campania region in 2019 and 2020, a monitoring and sampling campaign in walnut fields and warehouses was carried out. The morpho-molecular identification, performed through systematic keys consultation and Cytochrome c oxidase I gene sequencing, has highlighted the presence of different species belonging to the genus *Carpophilus* (Coleoptera: Nitidulidae). The identification of the most frequently recorded species is still incomplete due to the current uncertain taxonomy of the genus. In fact, the taxonomy of the whole group is in continuous and rapid evolution and several new species were recently described. The species distribution is also an issue: in Argentina on stored walnuts, damages similar to those found in Italy were reported in 2018 and the same attacks were recorded a few years earlier in Australian fields on almond seeds. Argentinian colleagues identified their specimens as *C. dimidiatus* while the Australian ones identified the specimens as belonging to a close but distinct species (*C.* near *dimidiatus*). Studies conducted in Australia with a morpho-molecular approach confirmed that individuals found in both countries are conspecific. Therefore, the detection of the species in Campania highlights how this species is harmful in at least three continents. Specimens found in Campania, indeed are both morphologically and molecularly (at least in part) identical to *C. dimidiatus* found in Argentina and, thus to *C.* near *dimidiatus* from Australia. Some peculiar and common biological characteristics also confirmed that the Campanian specimens belong to the same species. Hence, it is conceivable that this species or population has simultaneously started an invasive process in three different continents. Investigations, still ongoing in Italy, have confirmed its presence and harmfulness in several sites almost all over the Campania region with damage percentages up to 60% of the examined material. Moreover, this sap beetle damages also fruits directly in fields, hence it is not solely associated to stored material. Further studies are in progress to: complete the morpho-molecular characterization to obtain a more precise and stable identification of the species; understand the route of invasion; improve information about its biology, its host range, and find any possible natural enemies. This information could promote the development of effective and eco-sustainable methods to control the pest and its spreading.

KEY WORDS: almond, dried fruit, invasive species, stored products, walnut.

POSTER

Aclees taiwanensis: biology and spread of the invasive species recently introduced in Europe

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Aclees taiwanensis, the fig weevil, is a species native to Taiwan, accidentally introduced in Europe through the bonsai trees' trading. It has been first detected in 2005 in Tuscany, in the nursery district of the Pistoia province. The pest is oligophagous on plants belonging to the genus *Ficus*, so it represents a threat primarily for the common fig *Ficus carica*. Adults feed on leaves and ripening infructescences, making them unsellable. The major damage is though caused by the xylophagous larvae which, digging alimentation galleries inside the trunk and surface roots, compromise the sap flow of the tree. The difficulty in promptly detecting and reaching the preimaginal stages inside the plants is one of the main limits to the control of the fig weevil. The biological and synthetic products tested so far have not been decisive and, despite its harmfulness, there are currently no regulations to limit its spreading. Therefore, specific strategies to detect and control *A. taiwanensis*, developed based on its biology and physiology, are needed. This research aimed to determine, under laboratory and field conditions, the main biological traits of the species. In the laboratory, the duration of the embryonic, larval, and pupal stages was verified. The pre-imaginal instars and adults' morphometric measures were taken, and a reliable characteristic to discern the sexes was identified. We collected data about female fertility and fecundity and on the modalities of oviposition. Moreover, the susceptibility of *Ficus benjamina*, *F. microcarpa*, and *F. pandurata*, three ornamental species of economic value, was evaluated. In the field, through surveys carried out during 2019, the population dynamic was determined. In the end, by a citizen science approach, the current presence of the fig weevil in Southern Europe and its spreading from 2008 to 2020 were verified. Based on our results, the embryonic stage lasts about 10 days, the larval development almost 80, and the pupal about 23. Regarding the adults, females usually have longer rostra and bigger bodies than males, but the sexes can be discerned based on the different shape and position of the last abdominal tergite. Each female lays more than 100 eggs (in the soil or cracks of the bark), of which, in the laboratory, almost 70% hatch. The three ornamental *Ficus* species tested have been defoliated, although in different manners and timing. Among these, *A. taiwanensis* was able to reproduce only on *F. macrocarpa*. In the field, adults have been captured all-year-round with three population peaks. The current distribution of the pest in seven northern and central Italian regions and the Provence-Alpes-Côte d'Azur region in southern France (identified through the citizen science approach) suggests that, without effective control strategies, the further spread of *A. taiwanensis* is predictable. This especially in the Southern Italian regions and the Mediterranean basin where fig cultivation is one of the most ancient and characteristic.

KEY WORDS: *Ficus carica*, alien insect, physiology, population dynamic, citizen science.

ORAL PRESENTATION

Volatile response of olive trees after the attack of the gall midge *Dasineura oleae*

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Dasineura oleae (Angelini) (Diptera: Cecidomyiidae) is a gall midge specific of *Olea europaea* L. in the Mediterranean area, where in the last years several outbreaks occurred. The insect attack induces the formation of galls on leaves during the larval development, thus reducing photosynthetic activity. Despite gallers can usually cope with the plant response by suppressing the plant defence and the release of volatiles, we investigated whether the parenchymal tissue modification and the larval trophic activity may alter the plant emissions. The volatile profiles emitted from infested plants were compared with those of healthy plants under both laboratory and field conditions. Additionally, the volatiles emitted from mechanically damaged plants were considered. A blend of 12 volatiles were distinctive of olive trees infested by *D. oleae*. Of these, β-copaene, β-ocimene, cosmene, unknown 1 and unknown 3 were found to be exclusively emitted in infested plants. The emission of germacrene-D, (E,E)-α-farnesene, and (Z,E)-α-farnesene, α-copaene, DMNT, (E)-β-guaiene and heptadecane significantly increased in infested trees. Linalool, β-copaen-4-α-ol, β-bourbonene, β-cubebene, β-elemene, β-copaene and δ-amorphene were found only in the field trial and showed differences depending on the level of infestation and the plant stage. The green leaves volatiles (GLVs) (Z)-3-Hexenol, (E)-4-oxohen-2-enal, and 2-(2-butoxyethoxy)-ethanol, were exclusively emitted from the leaves after mechanical damage. In the present study we show first evidence of herbivore induced plant volatiles (HIPVs) emitted from infested olive leaves. Future experiments will explore the ecological role of these molecules and their implications in the plant-host-antagonist tritrophic interaction.

KEY WORDS: Herbivore Induced Volatiles, green leaves volatiles, mechanical damage olive tree.

POSTER

Olive fly(*Bactrocera oleae*) olfactory responses towards olive midge (*Dasineura oleae*) induced volatiles in olive leaves

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Dasineura oleae (Diptera: Cecidomyiidae) is a midge responsible for galls formation on olive leaves. To date, it is considered as a secondary pest of olive groves since it has no direct damage on olive production. However, it has recently gained researchers attention due to its severe outbreaks in the Mediterranean Basin. This study aimed at the understanding of *D. oleae* infestation effect on the olfactory behavior of *Bactrocera oleae* (Diptera: Tephritidae), the main key pest of olive groves worldwide. In this context, recordings of olive fly olfactory responses towards infested olive leaves by olive midge were performed and analyzed, to determine the capability of *D. oleae* to aggravate *B. oleae* infestation. Using young males and females (0-5 days old), GC-EAD recordings were carried out on volatiles from field and lab infested plants, as well as healthy leaves, from different olive groves in Italy. The results revealed significant responses to nine volatiles from which three were induced by *D. oleae* infestation. Among these three, only beta-ocimene could be identified. In addition, sulcatone, myrcene and alpha-farnesene were recognized as shared volatiles between the infested and healthy leaves. Furthermore, EAG recordings were conducted to investigate dose-response activity and flies sensitivity to the four identified compounds at four concentrations (10^{-6} to 10^{-3}). In fact, females were highly responding compared to males, presenting a dose-response dependency, with higher responses for the highest concentrations. Moreover, sulcatone elicited the strongest responses among the four volatiles, followed by alpha-farnesene, beta-ocimene and myrcene. This study shows the olfactory sensitivity of *B. oleae* towards three induced volatiles by *D. oleae* which may affect the olive fly infestation potential. Indeed, further behavioral experiments are suggested, both in lab and field conditions, to confirm the synergistic effect that might be induced by the olive midge to increase olive fly infestations in olive groves.

KEY WORDS: olive pests, olfaction, synergistic effect, HIPVs, electroantennography.

POSTER

Biodiversity and restoration of soil ecosytems in organic vineyards

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In Italian vineyards, some areas are characterized by problems in vine health and grape production. Among the causes for this situation there may be included: poor organic matter content, limited contribution of the soil fauna to the ecosystem services (i.e. nutrient cycles), altered pH, water deficiency, soil compaction and/or scarce oxygenation. ReSolVe was a transnational and multidisciplinary 3-years research project (Core-Organic+ program) started on March 2015, aimed at testing the effects of selected agronomic strategies for restoring optimal soil functionality in degraded areas within organic vineyard. Italian experimental plots were situated in two commercial farms in Tuscany: i) Fontodi, Panzano in Chianti (Firenze) and ii) San Disdagio, Roccastrada (Grosseto). In each site, using gamma-ray spectroscopy proximal sensing to define topsoil homogeneity, three plots (250 m2/plot) of degraded areas and three relative non-degraded control areas were selected, The different restoring strategies implemented in each plot were: a) compost produced on farm by manure, pruning residue, grass, b) faba bean, barley green manure, c) sowing, dry mulching with *Trifolium squarrosum*. Each plot has been evaluated regarding soil physics, chemistry and biology with reference to microarthropods. The mesofauna was extracted by Berlese-Tullgren funnels. The abundances of microarthropods were assessed by time scale (year), sites, inter-row. For biodiversity analysis, ecological indices (i.e. Shannon, Simpson, Margalef), Biological Soil Quality (BSQar), oribatid mites/other mites were calculated. On the whole, chemical analysis confirmed the soil stress: lower organic matter content and higher total carbonates were registered in degraded plots in comparison with control plots. After two years, the compost supply was efficient to increase soil nitrogen and organic matter content. Concerning microarthropods, before treatments, abundance and biodiversity of Fontodi was higher than San Disdagio but not significant differences between degraded and non-degraded areas in each vineyard. Overall, the functional recovery strategies determined different effects: dry mulching provided an increase in terms of arthropod abundance in San Disdagio vineyards, while compost in Fontodi's ones. The BSQar values were high (>150) according to data registered in similarly managed vineyards or stable ecosystems. In degraded areas, reduced densities of euedaphic forms were registered, based on the maintenance of natural fertility. It has been confirmed that an integrated approach to physical, chemical, biological and production assessments would be advisable for adoption of sustainable agriculture and preserving soil ecosystem services.

KEY WORDS: good agronomic practices, soil microarthropods, compost, green manure, dry mulching, biodiversity.

POSTER

Effects of *Vitis vinifera* volatile compounds on the sex pheromone attractiveness in *Lobesia botrana*

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In the search for food, mating, and oviposition by phytophagous insects, host plant volatiles play a major role. Laboratory behavioral studies have shown that, at certain doses, some *Vitis vinifera* L. volatile compounds, including hexanal, 1-octen-3-ol, ß-caryophyllene, and farnesene are attractive to mated females and/or increase male attraction to the sex pheromone in *Lobesia botrana* (Den. et Schiff.). To gain information useful for possible practical applications of the above-mentioned compounds, their effects on male attraction to sex pheromone dispensers was evaluated in field experiments. Reservoir polyethylene dispensers (ML30 Braun®, BASF) were loaded with 2 ml of individual test compounds (Sigma-Aldrich) and field applied, singly or in different combinations, in pagoda traps baited with commercial *L. botrana* sex pheromone dispensers (Novapher). Trials were carried out in vineyards located in the Pescara province from July to September 2018 and 2019. Moreover, the release curves of different compounds from dispensers maintained in a wind tunnel in controlled wind speed (10 cm/sec) temperature (29±2°C), relative humidity (60±5%), and photoperiodic (L0:D24) conditions were gravimetrically determined. During 56 days of observation, the release rate (mean±SD) of hexanal, 1-octen-3-ol, ß-caryophyllene, and farnesene was calculated to be respectively 14.14±0.31, 10.24±0.59, 13.70±0.37, 8.34±0.19 µg/min. For both years, mean male catches in traps baited with individual dispensers of hexanal, 1-octen-3-ol, or ß-caryophyllene were significantly ($P<0.05$, Tukey-test) lower than those recorded in control traps baited only with sex pheromone dispensers. Reduction of male attraction was even more evident for traps baited with three dispensers each containing one of these three compounds. In this case, a complete inhibition of male catches was achieved. In field conditions, it was not possible to evaluate the biological activity of farnesene due to its isomerization in the polyethylene reservoir dispensers. The reduction of male attraction to sex pheromone by hexanal, 1-octen-3-ol, ß-caryophyllene appears in contrast with results of previous studies showing an increased male attraction to sex pheromone in the presence of the same compounds. However, the amounts of plant compounds released by dispensers in the present study (µg/min) were about 6-order of magnitude higher than those used in previous ones (pg/min). Therefore, it is conceivable that at least at short distance, such as the active space of a trap, high dosages of the plant volatiles tested are able to effectively interfere with the perception of sex pheromone by male antennal receptors and/or with the integration and coding of intra- and interspecific chemical signals. Otherwise, the high dosage release of these compounds could have resulted in a real repellent effect to males which could have perceived the environment unsuitable for mate seeking.

KEY WORDS: European Grapevine Moth, semiochemicals, field trapping test, VOCs, controlled release.

POSTER

A physiologically-based model predicting phenology and abundance of *Philaenus spumarius*, the main vector of *Xylella fastidiosa* in Europe

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The meadow spittlebug *Philaenus spumarius* (Linnaeus) is a common and widespread xylem-sap feeder in Europe. The species has been recently discovered as the main vector of the gram-negative bacterium *Xylella fastidiosa* (Wells) known as the causal agent of the olive dieback in Apulia (southern Italy). Since then, the vector became one of the major concerns posed to European agriculture. Only the adult stage of *P. spumarius* is able to acquire the bacterium *X. fastidiosa* through feeding on the xylem sap of infected host plants. Once the adult acquired the bacterium, it is persistently infected and can immediately transmit the pathogen to healthy plants, as latency in the vector is not required. Therefore, the capacity to predict the population phenology and abundance of *P. spumarius* is a key element for understanding the epidemiology of *X. fastidiosa* as well as for the development of targeting control interventions for rational management strategies against both the vector and the bacterium. Here, we present a physiologically-based mechanistic model predicting the spatio-temporal variation of phenology and population abundance of *P. spumarius*. The model describes the life-history strategies of the different biological stages by means of stage-specific development, mortality and fertility rate functions. The rate functions describe the physiological responses of the species to local environmental forcing variables (e.g. air temperature) and resources (e.g., host plant availability). The rate function's parameters have been estimated through data obtained from laboratory and field experiments. The model has been calibrated and validated using independent population dynamics datasets from Apulia and Liguria regions (Italy). Model outputs can be used for assessing the risks linked to *P. spumarius* population dynamics and *X. fastidiosa* epidemiology. Model simulations can also support the design and implementation of monitoring and control activities for managing the *X. fastidiosa*-plant-vector pathosystem.

KEY WORDS: meadow spittlebug, vector abundance, *Xylella fastidiosa* epidemiology, vector control.

POSTER

Molecular gut content analysis for host plant identification in the polyphagous xylem feeder *Philaenus spumarius*

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Philaenus spumarius L. (Hemiptera: Aphrophoridae) is the main vector of *Xylella fastidiosa* in Europe, where it is responsible for the spread of the etiological agent of the olive quick decline syndrome, which had a devastating effects on olive production in the Apulia region of southern Italy. Considering the huge number of potential host plants for both the vector and the pathogen, several wild and cultivated plant species may represent an infection reservoir for *X. fastidiosa.* This is of extreme relevance for the epidemic progression of the olive quick decline syndrome, as well as of other diseases possibly related to unintended introduction events of new pathogenic strains in Europe. Therefore, it is very important to increase the current understanding of trophic interactions between *P. spumarius* and its host plants, in order to promptly recognize potential acquisition sources for *X. fastidiosa.* Sources may be asymptomatic wild host plants that may not be intercepted during the common plant protection monitoring procedures. The identification of these essential hubs for the disease spread is extremely relevant for a rational containment of epidemics. A molecular method was set up to identify the host plants of single adult *P. spumarius* specimens by analyzing their gut content. Spittlebug populations of northwestern Italy were considered in this study, as in that area *X. fastidiosa* is not recorded yet, but it is regarded as a primary threat. The designed analysis targets the identification of chloroplast DNA by means of specific nested PCR on total DNA from the insect body, followed by amplicon sequencing. Plant DNA concentration is extremely low in the digestive tract of a xylem feeder, as it is only marginally ingested during the feeding events, through stylet contamination. However, with this method it was possible to identify up to two distinct plant species in the same specimen. The presented protocol represents an innovative tool to support research on the life cycle of xylem feeding insects, which most often display a polyphagous feeding behavior. Additionally, by applying this technique it is possible to assist the monitoring of the infection status of *P. spumarius* field populations in those areas where *X. fastidiosa* has not been introduced yet, both by studying vector ecology in different environments and providing new elements to the epidemiological analysis of pathogen diffusion.

KEY WORDS: meadow spittlebug,vector, polyphagy, chloroplast DNA, *Xylella fastidiosa*, host plant preference.

POSTER

Toxicity, repulsiveness and attractivity of smoke waters on *Bactrocera oleae* (Rossi): lab and field assays

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Smoke waters are secondary products of the biochar production obtained by pyrolysis of biomasses and some of them are already marked as plant growth promoters. In this study, SWs obtained from pyrolysis at 300 °C and 500 °C of alfalfa, paper, *Quercus ilex* L. wood, maize, and olive mill waste were prepared to investigate their toxicity and behavioural activity towards adults of the olive fruit fly (OFF), *Bactrocera oleae* (Rossi). Afterwards, a first mixture was prepared using equal volumes of olive mill waste SWs (OLI) and a second one using equal volumes of the SWs from the remaining substrates (MIX). In laboratory tests, no toxic effects by contact were recorded for both mixtures. On the contrary, ingestion of OLI or MIX caused increasing mortality. Moreover, MIX significantly altered the adult fertility with 64,28% fewer stings, than control. Electroantennographic (EAG) tests demonstrated male and female antennal perception of volatile compounds present in MIX and OLI. Therefore, Y-tube olfactometer bioassays were carried out to assess the behavioral responses of OFF adults to each SWs mixture. In both sexes (30♂ + 30♀), a significant repellent effect of MIX (80% of insects in the control arm) was recorded whereas OLI elicited a significant attraction that was comparable to that of fresh olives (92,5% and 91,8% of insects in the arm with OLI or olives, respectively). Field trials were carried out in 2 different geographical and pedoclimatic condition, in Crotone (Calabria) and Isernia (Molise), in order to confirm the MIX repellency. In both sites, 3 homogenous groups of trees (n=3) have been identified and, for each, 3 tubes containing 10 ml of MIX were uniformly arranged on each plant, about 2 meters above the ground. The control consisted in 3 groups of 3 untreated trees. A yellow sticky trap activated with sex pheromone and ammonium carbonate dispensers was put in the middle of each group of trees. Repellent effectiveness of MIX was evaluated comparing trap catches and the number of stings on fruits of treated and control trees. MIX application led to a decreasing number of total catches by 85,63% and 82,89% compared to control in Crotone and Isernia, respectively. Similarly, the number of stings on fruits did not reach 10% of the number found in control groups. The collected data show a strong effectiveness of SWs to interfere with orientation of OFF adults towards chemio-chromotropic stimuli.

KEY WORDS: Olive Fruit Fly, biochar, bioassays.

POSTER

Use of hydro-alcoholic propolis solution to control some pests citrus

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Propolis is a complex compound derived from buds of some plants and then reworked by bees. Its chemical composition is highly variable depending on the botanic origin, the harvest period, the area of origin and bees secretions. Propolis antibacterial, antiseptic and healing properties are known and exploited by man; this has led to its use in agriculture where it is mainly applied for the inhibitory effects against phytopathogenic fungi and bacteria. However, there are very few studies of its specific use as an insecticide. To this regard, we report data obtained from applications of an hydro-alcoholic propolis solution in semi-field trials, to evaluate the effects on two pests: *Aleurothrixus floccosus* (Maskell) and *Aphis gossypii* (Glover), reared on *Citrus aurantium* (L.) in pot. Propolis extract was obtained after a steeping at 20% w/v in a hydro-alcoholic solution (50% ethyl alcohol), properly diluted for the applications. The selected pests were reared in controlled conditions (22 °C ±0,3 °C, light-dark 12:12) and used to infest plants which were first grouped in clusters homogeneous per number of leaves and vigour and then used in the tests. Preliminary phytotoxicity tests on the plants used were necessary due to the presence of alcohol in the solution. For this pourpose, four weekly applications were disposed using the maximum dose of use without record symptoms of plant suffering at the end of the test. Three different dilutions were used in the trials, corresponding to doses of 1.2, 1.4, 1.6 L/hl. Two semi-field tests were carried out for each pest using 12 plants per trial, divided in 3 replications and an untreated group as control. Checks on the pests' populations were performed at the moment of application and 7 and 14 days later. Due to the non-homogeneity of the starting infestations, the degree of incidence of the infestation was calculated using the Henderson & Tilton formula. The insecticidal efficacy recorded against *A. floccosus* reached average values of 68.8% and 62.2% respectively 7 and 14 days after the treatment regardless of the age of the individuals, without statistical significance among the three dosages. Mean effectiveness against *A. gossypii* were 81% and 67.2% compared to control after 7 e 14 days, reaching a statistically significant peak (87.7%) after 7 days and with the more concentrated dose (1.6 L/hl). The low persistence of the product allowed the treated pests populations (mainly the aphid one) to recover in short time. A possible pest management program could need more applications of propolis extract to control the target pests. Further studies are desiderable to obtain a deeper knowledge about insecticidal activities of propolis hydro-alcoholic solution, crucial to identify bioactive molecules contained in it.

KEY WORDS: *Aleurothrixus floccosus*, *Aphis gossypii*, apiculture.

Susceptibility of stone-fruit rootstocks to *Capnodis tenebrionis* larvae

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Capnodis tenebrionis (L.) is one of the major stone-fruit pests. Larvae develop into roots and adults feed on buds and twigs affecting the productivity of different species of stone-fruit: apricot, peach, plum and cherry. Few tools are available to manage infestations by *C. tenebrionis* adults and larvae. The control of adults is mainly based on chemicals, while control of larvae is problematic due to their localization in the roots. Less susceptible *Prunus* rootstocks offer a promising eco-friendly and complementary tool, and they might be applied as a preventative tool against the larval infestation. The current study aims investigating the susceptibility to *C. tenebrionis* larvae of the rootstocks most commonly used by growers. Two different bioassays were carried out in two years trials. Larval development was studied on semi-artificial substrates containing rootstock bark flour of Adesoto, CAB6P, Colt, Garnem, GF677, MaxMa60, Montclar and Myrabolan 29C. Infestation by neonate larvae was investigated in specific microcosms on twigs of Adesoto, Barrier, CAB6P, Colt, Garnem, GF677, Marianna 26, MaxMa60 and Myrabolan 29C. Larvae developed faster and heavier on Montclar and GF677 treatments from which bigger adults emerged, whereas Adesoto, CAB6P, Colt and MaxMa60 treatments supported a less efficient larval growth resulting in adults with smaller size. Colt was the most infested rootstock in twig infestation assay, while Barrier, MaxMa60 and Marianna 26 were less infested. Combining the results of both assays, GF677 and Myrabolan 29C appear to be more susceptible and Adesoto and MaxMa60 less susceptible to C. tenebrionis larvae, although Barrier and Marianna 26 require further investigations. The experimental model used in this study allows processing a large number of tests on different rootstocks, accumulating a large amount of data on the potential susceptibility of rootstocks. Additionally, rearing larvae on artificial substrate can also lead to assess the influences of additional compounds, for quantity and quality, on the larval growth.

KEY WORDS: peach flat headed root borer, larval rearing, semi-artificial diet, twig infestation assay.

ORAL PRESENTATION

Flight distance of *Popillia japonica*: use of an indirect marking technique

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The Japanese beetle *Popillia japonica* Newman (Coleoptera: Scarabaeidae), an Asian species introduced into the US since 1911, has been detected in European mainland in 2014. At present, it is restricted to an area surrounding the Ticino river, in Lombardy and Piedmont (NW Italy). *P. japonica* is a quarantine pest included in the Priority Pest list of the European Union, according to regulations 2016/2031/EU and 2019/1702/EU. In application to the current rules, the Regional Plant Protection Services (RPPS) of Piedmont and Lombardy have put in place the required actions, including the definition of infested and buffer areas. To support this decision, in 2017-2019 a research to quantify the dispersal of Japanese beetle adults has been conducted in Lombardy, using a protein indirect mark-capture technique. Water solutions of either chicken egg white (marker: albumin) or cow milk (marker: casein) were sprayed in two different areas, on plants infested by adults of *P. japonica* (e.g. hornbeam, dog rose, oak, and so on), for 3 - 5 times per year during June and July. After 24 hours and seven days, potentially marked Japanese beetle adults were captured at 23 points displaced at different distances from the treated areas (up to 12 km far away). A double lure-baited trap (attractive for both males and females) was moved from point to point and let in place for either 10 minutes or until 10 beetles were captured. In the laboratory, markers were identified with two indirect ELISA (anti-albumin, and anti-casein). Flight distance data were analyzed with geostatistical methods (indicator kriging), and female beetles were dissected to relate egg count to flight distance. Overall, more than 1600 adults were captured and analyzed. The marking rate was 7-15% for egg and 2-22% for milk, respectively. The mean flight distance from the treated areas was between 1 km and 7 km, however some specimens were found up to 12 km far away. Spatial correlation occurred up to 2.3 km distance, whereas egg load had no influence on flight dispersal in females. The results obtained, along with the surveys conducted by the RPPS, are useful to define the extension of buffer zones for the regulation of *P. japonica* in Northwestern Italy.

KEY WORDS: alien species, quarantine pest, dispersal, mark-capture, ELISA, indicator kriging.

POSTER

Field observations on damage caused by *Acanthiophilus helianthi* to safflower (*Carthamus tinctorius*) in Sardinia (Italy)

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Safflower (*Carthamus tinctorius* L.) is an annual species belonging to Compositae family that is mainly cultivated in India, Mexico and the USA to product edible and industrial oils. Safflower, which can grow even in dry conditions, can be seriously damaged by several pests and diseases. The main harmful pest to safflower is the tephritid fly *Acanthiophilus helianthi* Rossi, which is a polyphagous species whose larvae feed on ovaries and achenes of many wild plants. As part of a project on the valorization of Mediterranean autochthonous plants using advanced green chemical technology, entitled "COMETA - Colture autoctone mediterranee e loro valorizzazione con tecnologie avanzate di chimica verde" (POR FESR 2014/2020 - ASSE PRIORITARIO I "RICERCA SCIENTIFICA, SVILUPPO TECNOLOGICO E INNOVAZIONE), the harmfulness of *A. helianthi* on safflower growing in Mediterranean conditions was investigated. In particular, larval infestation was estimated on two safflower varieties (Oscar and Novamont) sown both in November (S1) and January (S2). Field observations were carried out at the experimental farm of the Department of Agricultural Sciences of the University of Sassari (Sardinia) from May to August 2019. A randomized complete block design with 4 replications (plot size: 45 m2) was used. Adult fly populations were monitored using 4 yellow chromotropic traps placed in the experimental field. A sample of 8-10 flower heads, randomly selected from different plants (1 head/plant), was collected from each experimental plot weekly. In the laboratory, the flower heads were dissected and the achenes were examined with a stereomicroscope to count the number of achenes infested with larvae or pupae of *A. helianthi* and the number of non-infested achenes. The infestation level was estimated as the percentage of infested heads in relation to the total of heads sampled, whereas the damage was calculated as the percentage of ovaries and/or achenes feed by larvae in relation to the total. During the entire period, no adults of *A. helianthi* were captured by the chromotropic traps, which proved to be an unsuitable method for monitoring adult flights. The flower heads showed a progressive increase of larval infestation in all the experimental plots over time, with a maximum value of approximately 50% and 60% infestation in the plants sown in November and January, respectively. Flower head infestation did not differ significantly between the two cultivars, whereas harvesting damage was the lowest (21%) in Novamont safflower sown in January and the highest in Oscar safflower sown in January (27%). Contrary to what suggested by literature, early sowing did not reduce the incidence of fly infestation and damage seemed to be more affected by the variety than sowing time. Our findings underline the need to conduct studies on suitable methods for monitoring adult *A. helianthi* populations such as the use of attractive substances.

KEY WORDS: safflower, *Acanthiophilus helianthi*, oilseed crops.

Ricania speculum (Walker) in Italy: an update of the distribution, host plants, monitoring and natural enemies

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Ricania speculum (Walker) (Hemiptera, Fulgoromorpha: Ricaniidae) is an alien invasive species reported for the first time in Europe in 2009. This species feeds and lays eggs on a wide range of host plants and its large polyphagy has immediately been observed by several authors. The fast-spreading of *R. speculum* in Italy makes it a possible new threat to native plants and human activities, such as agriculture and plant nurseries with consequent economic damage. The species is usually monitored through classical entomological methods but, in Italy, the reports concerning its distribution have come mainly from visual inspection. To develop effective management strategies to prevent further spread and to control the already established populations, this contribution aims to update: 1) the distribution of *R. speculum* and 2) the host plants in Italy. Moreover, we evaluated 3) the use of the sticky colour traps to capture and 4) the in-depth study of the phenology and activity rhythms of this species. Finally, the effectiveness of the predator *Chrysoperla carnea* complex and four coccinellid adult species was estimated in the laboratory and semi-field conditions. We verified that this species is currently distributed in seven regions and we added 33 new host plants, belonging to 29 families, to the list of plants attacked by this pest. Moreover, we confirmed that this species is univoltine in the newly infested area and is mainly crepuscular, with a high intersexual activity overlap. According to our study, the species showed a flight period during the summer months and the adults were mostly caught by green traps, particularly in September, which is the period of egg-laying inside the leaves. Concerning the biological control, the results of our study showed that the larvae of *C. carnea* complex have the potential to control *R. speculum* under laboratory and semi-field conditions compared to the coccinellid predators. These findings can help to set up a control strategy to manage this pest in the Italian territory.

KEY WORDS: invasive species, biological control, *Chrysoperla carnea* complex, Ricaniidae, sticky colour traps.

ORAL PRESENTATION

Insecticide "target-site" resistance in Italian population of the cotton-melon aphid *Aphis gossypii*

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Aphis gossypii Glover (Homoptera: Aphididae) is a cosmopolitan polyphagous pest causing considerable economic losses due to its feeding damage and plant disease transmission. For several years in Northern Italy *A. gossypii* populations have been monitored mainly in potato crops for the incidence of the main potato virus disease, but in recent years, the attention has shifted on insecticide resistance phenomena. The efficacy of insecticide products can be affected by several insecticide resistance mechanisms but the greatest impact is given by "target-site" resistance. This work was performed on 23 different population of *A. gossypii*, collected especially in Northern Italy, to identify point mutations linked to cause resistance to pyrethroids (kdr and s-kdr), carbamates (MACE) and neonicotinoids (R81T). This research, carried out in the framework of the regional project "PSR 2014-2020 Op. 16.1.01 - GO PEI-Agri - FA 4B, Pr. "5004934 - Resistenze avversità e malerbe - Tecniche diagnostiche, distribuzione territoriale e gestione di resistenze dei principali patogeni, fitofagi e malerbe ai prodotti fitosanitari", adds new data to the presence of the main target-site mutations in Italian population of *A. gossypii*: a high percentage of specimens with carbamates and pyrethroids resistance have been found meanwhile neonicotinoids resistance was not detected.

KEY WORDS: insecticide resistance, pyrethroids, neonicotinoids, Emilia Romagna.

Preliminary data of the synergic effect of kaolin and spintorfly in organic olive groves against the olive fly

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The olive fly, *Bactrocera oleae* (Rossi), the most dangerous and harmful parasite for the olive tree and key phytophagus of the ecosystem, causes qualitative and quantitative deteriorations of an amount proportional to the level of infestation. The recent regulations issued by the EU, with which many active substances, normally used against the main phytophages of the olive tree, have been revoked, have constituted a motivation to implement this type of research, both in support of integrated and organic olive growing. The researchs were carried out in 2018 in an olive grove located in Rende consisting of olive trees of the Carolea cultivar, bred in intensive cultivation with a density of about 400 plants/hectare and in 2019 in an olive grove located on the northern Ionian coast of Calabria (Mirto-Crosia), consisting of young olive trees, in intensive cultivation with a density of about 400 plants /hectare with the presence of the most representative cultivars of Calabria (Carolea, Cassanese). Spinosad, a molecule with an attractive and insecticidal action (obtained from the fermentation process of *Saccharopolyspora spinosa* Mertz & Yao) and kaolin (p. Kaolin) were tested. The kaolin, clay with repellent action, is not included among the antiparasitic substances but admitted as a biostimulant in organic legislation. For the test, the plants of both olive groves, except for the plants constituting the control thesis, were treated in alternate rows with spinosad and kaolin. The plants treated with spinosad were sprayed at a dose of 1 liter of product /ha, diluted in 4 liters of water, every 8-10 days from June to October, while the plants treated with Kaolin were sprayed at a dose of 5 kg / hl with three treatments (end of July, mid August and mid October) defined on the basis of the rains that occurred in the two vintages. The infestation percentages were determined by examining samples of 100 olives taken at random from the plants treated with spinosad and samples of 100 olives taken at random from the plants treated with Kaolin observed under a binocular microscope and distinguished between active infestation (presence of eggs, 1st and 2nd instar larvae) harmful infestation (3rd instar larvae, pupae and abandoned tunnels) and total infestation (all preimaginal stages, alive, dead and parasitized and abandoned tunnels). The effectiveness of the two products and their synergistic action was assessed in terms of active, harmful and total infestation in the treated theses compared to the control theses. The data were subjected to analysis of variance followed by Tukey's post-hoc test. In 2018, the only statistically significant value was the synergistic effect on the total infestation. In 2019 the results, for the overall investigation period, showed a significant reduction in the levels of harmful and total infestation both by examining the synergistic effect and considering the effect of the individual treatments, with the kaolin showing a better performance than the spinosad. Ultimately, the results obtained appear promising for the fight against *B. oleae* in organic and integrated olive growing, especially as regards the potential of the synergic treatment, provided that a constant coverage of the plants is guaranteed. Work supported by the Italian Ministry of agricultural food and forestry policies (Ministerial decree n.–) in the context of the “Piano Olivicolo oleario” (DI.OL. Interministerial Decree n.23774 – 06/09/2017).

KEY WORDS: Kaolin, spinosad, *Bactrocera oleae*, eco-compatible farming, *Olea europea*.

ORAL PRESENTATION

The importance of a network of traps for the preventive monitoring of invasive species: the case of *Bactrocera dorsalis*

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Climate changes and international trade increase are among the main causes of the remarkable gain in the number of invasive insects accidentally introduced and acclimatized in Italy. The EU Commission has listed the priority quarantine pests (Delegated Regulation (EU) 2019/1702) considering the most dangerous among those regulated in the annex II of Implementing Regulation 2019/2072 (EU). The list includes the species *Bactrocera dorsalis* (Hendel) (Diptera: Tephritidae), belonging to the homonymous species-complex, commonly known as "oriental fruit fly" and considered extremely threatful to agriculture due to its great biotic potential and host fruit range. Recently, *B. dorsalis* expanded its distribution area, invading Sub-Saharan Africa and began spreading northward in China, and in western Asia. It has frequently been recorded in the USA where, until now, has been successfully eradicated.
Some adults of *B. dorsalis* were captured in 2018 in Italy during a preventive monitoring with traps in Campanian orchards, as part of the activities of URCoFi project - Regional Unit for Phytosanitary Coordination. These records alarmed the European Union, where *B. dorsalis* had never been collected in traps in field, and other countries, such as the USA, due to the phytosanitary consequences connected with the exportation of fruits or plant materials. Due to the findings, the Italian National Phytosanitary Council has promptly established a work-group that draw up three documents for the 2019 activities: The Surveillance Plan, the Emergence Plan and the Action Plan. All the activities established in the aforementioned plans were carried out in 2019. Two delimited circular areas (about 390 km2) were thus defined around to the two collecting sites of 2018, where about 2100 traps were installed with a density directly proportional to the proximity to the centers. The reinforced monitoring activities included the inspections of host-plant fruits and the use, and weekly check, of McPhail traps baited with methyl-eugenol or torula yeast and Rebell Amarillo sticky traps baited with methyl-eugenol. In 2019, in a single monitoring area, traps caught some males of *B. dorsalis* and some females of *Bactrocera latifrons* (Hendel) (first record in Europe). All collected specimens were identified with a morpho-molecular approach. In 2020, the monitoring continued also using a specific attractant for *B. latifrons*. The total absence of captures and active infestations for a period longer than two insect cycles confirmed that, at present, both *B. dorsalis* and *B. latifrons* are not settled in Italy and have to be considered incursions. The pest interceptions at the Neapolitan airport inspection point, throughout 2019, and the total absence of captures in field after the travel ban in 2020, seem to confirm our hypothesis that the little quantity of fruit imported by travelers represents the main source of exotic species introduction. The achieved data highlight the usefulness of developing a permanent network of traps to early detect allochthonous species and to limit the probability of settlement of populations adaptable to our climate.

KEY WORDS: *Bactrocera latifrons*, emergency action plan, fruit flies, priority pests, quarantine pests.

ORAL PRESENTATION

New insights for the management of chestnut tortrix moths

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In recent years in many Italian chestnut areas the attention is high for some native insect pests characterized by having fruit-feeding larvae, which may represent a limiting factor for chestnut production. In fact, the chestnut tortrix moths [*Pammene fasciana* (L.), *Cydia fagiglandana* (Zeller), *C. splendana* (Hübner)], are pests belonging to the Tortricidae family, whose larvae develop in the fruit and which can lead to huge yield losses depending on the different abiotic factors and in light of current climate change. In 2018 a three-year experimental project funded by Isagro S.p.a. started with the aim of assessing the presence and the abundance of the tortrix species and determining the seasonal flight activity in different chestnut growing areas. Investigations were carried out in 4 regions of Northern Italy (Piedmont, Liguria, Tuscany and Emilia-Romagna). Research has been carried out, in the two-year period 2018-2019, in chestnut orchards located in the municipalities of Villarfocchiardo (TO), Peveragno (CN), Carro (SP), Badia del Borgo (FI), Molazzana (LU) and Montese (MO). At each site, sexual pheromone traps were placed. These traps were placed in a chestnut orchard for each site (3 traps / chestnut groves) and replaced weekly starting from the beginning of June until the end of September. The specimens captured by the traps were counted and identified by extraction of genitalia and molecular analysis. While in 2019 all investigated species were found in the surveyed sites, in 2018 *C. fagiglandana* and *C. splendana* were never found in Peveragno and Molazzana sites. The flight activity of *P. fasciana* adults began in mid-June to end in early September, except in Piedmont where in both Villarfocchiardo and Peveragno sites it was over in late July in both years. The intermediate and late tortrix moths were observed later in the season (early/late August-mid September), with no relevant differences between the sites. In many sites, in the *P. fasciana* traps several non target species, mainly *Oegoconia novimundi* (Haworth) and the plum fruit moth *Grapholita funebrana* (Treitschke), were detected as well. Moreover, in the *C. fagiglandana* traps, several specimens of the congeneric species *C. splendana* were found, both in the same time frame. In the *C. splendana* traps, only in some sites, some individuals of the geometrid Idaea rusticata Denis et Schiffermüller and of the congeneric species *C. fagiglandana* occurred. The results obtained, although still preliminary, show how the flight curves observed do not always overlap with those available in the literature, especially in the case of *P. fasciana*. Furthermore, the finding of high numbers of non target species in the *P. fasciana* traps, as *O. novimundi* and *G. funebrana*, highlights the risk of overestimating the number of individuals captured, also counting non target species.

KEY WORDS: *Pammene fasciana*, *Cydia fagiglandana*, *Cydia splendana*, flight curves.

POSTER

Seasonal populations abundance and impact of *Halyomorpha halys* in walnut

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The brown marmorated stink bug *Halyomorpha halys* (Stål) causes high losses in agriculture, particularly in fruit crops. Its presence has been frequently reported in walnut (*Juglans regia* L.) orchards for fruit production. The knowledge about the economic impact of this phytophagous on walnut is limited. In this work, the seasonal trend of *H. halys* populations in three walnut orchards was studied with particular reference to the distribution of the pest's population within the orchards and within plants. The surveys were carried out using "low doses" aggregation pheromones traps, placed in different parts of the orchards (edge vs. centre) and the of the canopy (high, medium or low positions). In another experiment, the effect of the duration ad period of infestation was assessed on some production parameters of walnut. During the 2019 growing season, the presence of *H. halys* was detected continuously. An edge effect characterized adults distribution in the orchards, that was stronger at the lower portions of the canopy. A higher number of adults was captured at the top and in the middle parts of the canopy, while the young stages were more abundant in the middle and lower parts. Feeding activity by *H. halys* caused fruit weight reduction, premature fruit dropping, suberification of the husk and anomalies of the kernel. In particular, early infestations were associated with a higher incidence of premature fruit dropping and suberification of the husk, while infestations after the shell formation phase caused more considerable damage to the kernel. The results obtained show that *H. halys* causes severe damage to walnuts and can be considered in designing pest management strategies.

KEY WORDS: Pentatomidae, invasive pest, seasonal abundance, damage potential, integrated pest management.

Preliminary studies on the damages of *Erasmoneura vulnerata*

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Erasmoneura vulnerata (Fitch) is a Nearctic leafhopper rarely harmful to grapevine production in its native area. This specie was first found in Europe in Treviso province in 2004, since then it has spread to all the North-Eastern parts of Italy, Slovenia and more recently Switzerland. Since its appearance in Italy, it has been considered as a secondary pest. From 2016 outbreaks in vineyards has been reported in Veneto region. The leafhopper feeds on the mesophyll of the leaves, causing discolouration which may be followed by early desiccation and leaf drop when infestation levels are higher than 10 leafhoppers per leaf for a few weeks. In this study, we evaluated the level of damage caused by *E. vulnerata* in three vineyards cultivated with Glera, Cabernet Sauvignon and Garganega varieties and located in the provinces of Vicenza (Glera and Cabernet Sauvignon) and Verona (Garganega). In each vineyard, treatments with or without insecticides application were compared. The damage was assessed in terms of leaf chlorophyll content, bunch weight, berries diameter, weight and sugar content. In all the varieties, there were no significant differences between treated and untreated treatments in term of chlorophyll content, bunch weight, berry weight and diameter as well as sugar content. However, on Cabernet Sauvignon, a decrease in sugar content was observed as the density of *E. vulnerata* increased, and on Garganega, a decrease in chlorophyll content was observed as the density of the leafhopper increased. The findings suggest that higher infestations than those reached in these vineyards are necessary to determine the extent of economic damage.

KEY WORDS: leafhoppers, grapevine, integrated pest management, damage potential.

POSTER

Carlina oxide as a new active ingredient for lure and kill against *Bactrocera oleae*? Insecticidal activity and electrophysiological insights

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The olive fruit fly, *Bactrocera oleae* (Rossi) (Diptera: Tephritidae) is a key pest of olive groves worldwide. Recently, a growing research interest about eco-friendly pest management tools has been raised. Developing green pesticides from natural products, including botanicals is a fast-growing research area, contributing to reduce the use of synthetic pesticides, with beneficial effects on human health and the environment. In this scenario, essential oils can represent useful sources of compounds with multiple modes of action, making the development of resistance in targeted pests unlikely. In this scenario, herein we investigated the essential oil (EO) obtained from the roots of *Carlina acaulis* L. (Asteraceae) and the carlina oxide, which represents the major component of the EO as novel potential active ingredients for lure and kill formulations against the olive fruit fly. The EO was obtained by hydrodistillation of the powdered roots by using a Clevenger apparatus for 6 h. The EO chemical composition was achieved by GC-MS and NMR highlighting the presence of the following compounds: carlina oxide (94.6%), benzaldehyde (3.1%), ar-curcumene (0.4%), acetophenone (traces), benzyl methyl ketone (traces), camphor (traces) and carvone (traces). Carlina oxide was purified by silica gel column chromatography and its structure elucidated by 1H- and 13C-NMR spectroscopic analysis. The toxicity of the EO and pure carlina oxide on olive fruit fly adults was tested through ingestion tests carried out incorporating them in protein baits. Eight and nine concentrations, ranging from 156 and 39 to 10,000 ppm, respectively, were used for EO and carlina oxide, and compared with a control containing only the protein substrate. Both *C. acaulis* EO and carlina oxide showed insecticidal activity against the olive fruit fly. A significant dose dependent effect of both EO ($F_{9,49}=65.29$, $p<0.001$) and oxide ($F_{8,44}=47.44$, $p<0.001$) was observed. The LC50 was 706 ppm for *C. acaulis* EO, and 1052 ppm for carlina oxide. Furthermore, to shed light on the potential behavioural effects of the EO and carlina oxide to olive fruit flies, electroantennographic (EAG) tests were carried out to evaluate the capability of the male and female antennae to perceive them. In the dose range of 0.01 to 1000 µg, dose-dependent EAG responses, similar in males and females, were elicited by EO (0.03-5.67 mV) and carlina oxide (0.07-2.11 mV) indicating a strong antennal sensitivity to both stimuli. Behavioral tests investigating the impact of being exposed to EO and carlina oxide subletal doses on *B. oleae* adults are also ongoing. Overall, this contribution sheds light on the possible utilization of carlina oxide for the development of effective eco-friendly lure and kill formulations. However, further studies are needed on the mode of action and field effectiveness, as well as on the side-effects on non-target species.

KEY WORDS: botanical insecticide, EAG, olive fruit fly, protein bait, Tephritidae.

ORAL PRESENTATION

Current distribution in southern Italy and biological data on the priority pest *Aromia bungii*

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The distribution area of the red-necked longhorn beetle (RLB) *Aromia bungii* (Faldermann) (Coleoptera: Cerambycidae) is gradually expanding in the Campania region, the largest outbreak area in Europe. The wood-borer pest is native to East Asia and attacks trunks of healthy plants belonging to the *Prunus* genus many of which are of considerable economic importance, causing gradual decline and tree death. Already present since 2014 in the quarantine EPPO A1 list, it was recently included among the priority pests for the European Union (EU Regulation 1702/2019) for its major threat to agriculture. Starting from 2012 (year of the *A. bungii* presence formalization), in Campanian stone fruit orchards, intense monitoring activities were carried out in order to contain and early detect infestations and, as far as possible, eradicate the invasive species from southern Italy. The containment measures and the infested areas are reported in several Regional Decrees (DDR 134 11/18/2019, the most recent). To date, the phytosanitary inspection activities aimed at searching presences/symptoms ascribable to the pest are carried out by the regional staff, the University of Naples "Federico II" and the CNR-IPSP, cooperating in the URCoFi project (Strengthening of the supervision activities and control of pests). To obtain reliable biological data, laboratory studies were carried out to determine the main biological parameters of this invasive species, essential to predict damage, organising monitoring plans and defining the most adequate control measures to counteract *A. bungii*. Since the first discovery, phytosanitary inspections ascertain that RLB is confined to the territory of the province of Naples, in an infested area of about 250 km2. Only from the monitoring over the last three-years period, 6987 field inspections were carried out throughout the region, followed by 1261 felling of infested plants; the infestations affected 12 municipalities. The study of reproductive parameters showed that *A. bungii* has a high biotic potential. The species is pre-ovigenic and each female is able to lay about 600 eggs on average throughout its life, with a hatching rate of 75% and an egg developmental time of just over a week. The largest number of eggs were laid during the first week after females emerging, while the pre-oviposition and oviposition periods lasted approximately two days and three weeks respectively, with an average of 20 eggs laid per day. The maximum adult longevity was found in fed males at 20°C (little over two months), while starved females survived about two weeks at 25 °C. Obtained results will be useful for a review of the IPM guidelines, in order to contain/eradicate the pest in the outbreak areas.

KEY WORDS: Biotic potential, invasive species, monitoring, red-necked longhorn beetle, reproductive parameters.

POSTER

Invasive quarantine species - The risk of small quantities of plant material transported by passengers

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International trade and passenger air travel play a key role in the accidental spread of invasive alien species (IASs). Their introduction in a new area often involves the alteration of the ecosystem and agroforestry equilibrium with even serious economic consequences. Italy is particularly exposed to this problem due to crucial factors such as geographical location, high number of entry points (ports and airports), recurring migratory flows and favorable climate conditions for the development of several IASs. In terms of phytosanitary protection, the CNR-IPSP technical staff contributes for years to the strengthening of inspections at BIPs (Border Inspection Posts) in the URCoFi project convention, collaborating both with the Phytosanitary Service of the Campania Region and with the Customs Agency of the airport of Naples-Capodichino. Plant material imported into the EU from the third countries is subjected to extensive inspections and laboratory analysis. Checks on imported goods regularly accompanied by plant passport rarely reveal pests. The real threat comes from the passengers' baggage who usually bring with them variable amount of uncertified fruit and vegetables. In the three-years period 2018/2020, at the BIP of the airport of Naples were seized large quantities of plant products from Asian, African and South American countries which, addressed for private consumption or ethnic markets, could have been an alternative route for unintentional introductions. During the checks numerous pests, from different plant species, were intercepted and notified in Europhyt (notification and phytosanitary alert system within the EU). Among these, the following species represent a threat to our agriculture: - *Bactrocera dorsalis* (Hendel) (Diptera: Tephritidae), the oriental fruit fly, in fruits of *Psidium guajava* from Bangladesh, in which live larvae were found and identified by molecular analysis. The pest is one of the most dangerous quarantine organisms, catalogued in the EPPO A1 and the UE priority pest lists, and is characterized by a high polyphagy (over 400 host species, many of which are of agricultural interest); - *Leucinodes africensis* (Lepidoptera: Crambidae), in fruits of *Solanum aethiopicum* from Bangladesh. The species is catalogued in the EPPO A1 list and represents a serious phytosanitary threat to solanaceous crops. The findings of *B. dorsalis* in uncertified material from passengers' baggage could corroborate the hypothesis that specimens found in traps in fields in 2018 have reached Italy through this route. The described interceptions, in association with the awareness of difficulties in carrying out a capillary control on all imported materials, suggest the need to adopt a new regulation on plant material imported in Europe. The introduction of a system of additional financial penalties and the strengthening of the communication campaign to passengers could help in the containment of the IASs spreading in the EU.

KEY WORDS: *Bactrocera dorsalis*, Border Inspection Post, *Leucinodes africensis*, monitoring, plant passport.

POSTER

Bionomics of the grape phylloxera in Tuscany

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For several years in Tuscany, as in other Italian regions, recurrent and heavy attacks of the grape phylloxera, *Daktulosphaira vitifoliae* (Fitch) have been affecting the epigean part of vines in commercial vineyards. The severity of the symptoms due to the leaf gall generations, in particular on the apical vegetation and on the lateral shoots, has induced considerable apprehension in an increasing number of wine growers in some of the most known regional wine-growing areas (Florence, Siena, and Leghorn provinces). As a consequence, several farmers, overestimating the damage of these infestations, have sometimes carried out inappropriate insecticide treatments. The Regional Phytosanitary Service of Tuscany facing this problem has prompted a renewed interest in the scientific world, already stimulated by the recent genetic investigations that allowed to identify and characterize different phylloxera strains in Europe. Therefore, investigations started in 2017 to clarify aspects concerning the distribution, biology, and harmfulness of the species in Tuscany. The following issues were addressed: the possible overwintering forms of the aphid in the vineyard agroecosystem, the dynamics of the population of the epigean generations, the population dynamics of the winged forms and their role in the diffusion of the species as well as the assessment of the damage caused by the presence of leaf galls on the physiology of the plant and on the quality of the final product. In all the vineyards considered, regardless of the area, the grape variety, and the rootstock, it has been found a widespread presence of radicicoles and their typical symptoms. However, surveys carried out on Sangiovese variety showed a considerable variability in the frequency of the knots which apparently does not depend on the rootstock (420 A, Kober 5BB, 1103 Paulsen, 779 Paulsen). Investigations on the population dynamics of the gallecoles were carried out in two Sangiovese vineyards in the wine-growing district of Montalcino. The results showed a remarkable difference between the two vineyards both in terms of average infestation and of the distribution of the affected vines: these parameters, however, remained constant throughout the observation period. Similar differences between the two vineyards were also recorded in the study of the population dynamics of the winged forms, carried out for the first time in Tuscany, through the use of yellow sticky traps. In fact, while in a vineyard the catch peak occurred in mid-September with as many as 70 individuals on average per trap per day, in the other the maximum presence of sexuparae occurred in late August with only 17 individuals on average. As for the possible influence of the density of the leaf galls on the physiological parameters and on the qualitative characteristics of the grapes, it has been shown a significant negative effect of the infestation level on the photosynthesis and on the content of sugar, anthocyanins and polyphenols in grapes. In order to have a deeper knowledge on the functionality of galled leaves, in 2019, investigations on vine leaf morphological characteristics have been started and, more precisely, on the relationship between the gall presence on leaves and stomata distribution.

KEY WORDS: grape phylloxera, population dynamic, leaf infestation, damage, spread.

POSTER

Chemical control of *Popillia japonica* adults on high-value crops and landscape plants of northern Italy

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The Japanese beetle (*Popillia japonica* Newman) is a pest native to Japan, recently recorded in Italy. The insect is highly polyphagous and it is causing considerable damage to herbaceous and perennial crops, nursery and ornamental plants. The appearance of an alien pest requires the integration of agronomic, biological and chemical actions to protect the crops. During the early stages of infestation, the use of readily effective insecticides is essential to sustain the immediate needs of plant protection. Here, we presented the effects of chemical and organic insecticides registered in Europe for adult beetle management on high-value crops (vine, peach and corn) and landscape plants (willow and Virginia creeper), evaluated by field trials carried out in 2019 and 2020. Two effects were considered for each product, i.e., topical and residual. Deltamethrin, lambda-cyhalothrin and the neonicotinoid acetamiprid were effective in killing beetles, with similar topical and residual effectiveness. The natural active ingredients/pathogens were ineffective but pyrethrin formulated with paraffinic mineral oil was effective. The data provide a valid support to update the European guidelines aimed at controlling *P. japonica* for growers, landscape managers and homeowners. The low selectivity of effective insecticides requires the integration into management strategies that envisage their use only whether strictly necessary, and in combination with containment measures based on natural enemies and cultural management practices.

KEY WORDS: agricultural crops, invasive species, ornamental plants, pest management, pesticides, survival analysis.

Potential damage caused by *Halyomorpha halys* on citrus fruits: preliminary laboratory assessment

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The Brown Marmorated Stink Bug (BMSB), *Halyomorpha halys* Stål (Hemiptera: Pentatomidae), is an Asian invasive insect pest recently spread in Northern and Southern America, Europe and Oceania. In Europe, BMSB has been detected in several countries including Italy where it was first recovered in 2007 and currently the pest is widespread in all the territory. In Sicily, the pest was reported since 2017 in various urban and rural locations in Palermo, Catania and Ragusa provinces although no crop damage has been recorded so far. BMSB is a polyphagous pest, able to feed on more than 100 plants and may damage several fruit, vegetable, extensive field crops and ornamentals. Citrus were also reported as susceptible plants for BMSB, but the knowledge on the potential and characterization of damage on citrus cultivated in Italy are poor.
To this end, preliminary *host acceptance* laboratory bioassays were conducted *no choice-test* by exposing Femminello lemon fruit to second instar nymphs and adults and *choice-test* only with adults, exposing them simultaneously to lemons, broad bean seedlings, green bean pods and shelled hazelnuts. On the basis of the data obtained in the preliminary tests - which confirmed that BMSB has the capacity to feed on citrus fruits - laboratory bioassays were conducted to evaluate the susceptibility of immature fruits of orange (cv Tarocco Gallo), mandarin (cv Tardivo di Ciaculli) and clementines (CV Comune) by carrying out *no-choice tests* in experimental arenas exposing individually the fruits to adults of both sexes of BMSB for 48 hours. To evaluate the feeding activity, the number of salivary sheaths visible on the outside of the fruits were observed.
Data showed that BMSB young nymphs and adults have similar capacity to feed on citrus fruits in laboratory conditions. BMSB feeding activity have fostered the fruit ripening process. BMSB adults of both sexes can feed on orange, clementine and mandarin unripe fruits, with a slight preference for orange fruits. Damage was observed both in the inner and outer part of the fruits. Data presented are the first assessment of the potential damage caused by BMSB on citrus varieties cultivated in Italy.

KEY WORDS: *Halyomorpha halys*, citrus, damage, fruit varieties.

Modelling diapause and phenology of the Japanese beetle, *Popillia japonica*

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The Japanese beetle (*Popillia japonica* Newman) is a highly polyphagous invasive species originated from north-eastern Asia. Since its first detection in the US in 1916, the species was able to invade vast areas of North America and some areas in southern Canada. In the 1970s, P. japonica was introduced in the Azores and since 2014, the species has established in mainland Europe (Italy). Since 2017, few occurrences of the species were reported in Switzerland. In Italy *P. japonica* is currently distributed along the Ticino Valley on the border between Lombardy and Piedmont Regions and, since its first detection, the infested area has increased over time. *P. japonica* is considered a quarantine insect pest and thus the Italian National and the two Regional Phytosanitary Services are implementing a set of measures aimed at the management of the species. The effective control of *Popillia japonica* populations can be facilitated by the use of models predicting the time of emergence of susceptible life-stages (i.e. larvae and adults). In this work, we present a temperature-driven, mechanistic model predicting the beginning of adult emergence and the overall phenological curve of adult individuals of *P. japonica*. Time-series adult trap catches data collected by the Phytosanitary Service of the Lombardy Region (Italy) are used for calibrating and validating the model. The model realistically simulates the influence of soil temperature on the developmental patterns and on the life-history strategies of the species. The application of the model at the local level (i.e. point-based simulations) might support decision-makers (farmers, farming cooperatives etc.) in planning and implementing pest monitoring and control actions based on the estimated time of emergence of the species. The model can also be applied at the regional scale (i.e. area-wide simulations) in order to obtain phenological maps, thus allowing to identify the areas where an early emergence of the species is expected. This might support decision-makers acting at the Regional level (e.g. National and Regional Phytosanitary Services) in prioritising the areas of intervention, in the implementation of early-warning systems, and in planning area-wide pest management plans. Funding: The Project GESPO is funded by “Direzione Generale Agricoltura - Regione Lombardia - D.d.s. 28 marzo 2018 - n. 4403D.g.r. n. X/7353 14 novembre 2017”.

KEY WORDS: *Popillia japonica*, phenological model, invasive species, pest management.

ORAL PRESENTATION

New methods for the rational and sustainable management of *Popillia japonica* - The GESPO Project

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The invasive Japanese beetle (*Popillia japonica* Newman) is a highly polyphagous agricultural pest originated from north-eastern Asia. The larval trophic activity may impact the root system of various graminaceous plants while adults may cause damage to leaves, flowers and fruits of cultivated and spontaneous plants. More than 100 economically valuable plant species have been reported as potential hosts including among others, maize, soybean, grapevine, apple, peach and kiwifruit. In 2014 *P. japonica* was detected, for the first time in Continental Europe, in the Ticino Valley on the border between Lombardy and Piedmont regions (Italy). Since then, the National and the two Regional Phytosanitary Services have adopted a set of measures aimed at monitoring and managing *P. japonica*. The GESPO Project, funded by the Lombardy Region, aims at developing rational (cost-efficient) and sustainable (low impacts) solutions for the integrated management of *P. japonica*. In particular we aim at: i) exploring the influence of biotic and abiotic drivers on the species' life-history strategies (development, survival and fertility), ii) investigating the environmental factors influencing the suitability of the habitat for the species, iii) developing rational and cost-efficient protocols for the management of the species and iv) developing models describing the species' phenology, spread and impacts. The main results of the field experiments and the main outputs of the models will be provided to relevant stakeholders through a web-based tool that will support the implementation of knowledge-based management strategies against *P. japonica*. We present the overall framework of the Project and the results achieved. Funding: The Project GESPO is funded by "Direzione Generale Agricoltura - Regione Lombardia - D.d.s. 28 marzo 2018 - n. 4403D.g.r. n. X/7353 14 novembre 2017".

KEY WORDS: *Popillia japonica*, invasive species, integrated pest management.

POSTER

Frankliniella occidentalis on table grapes in Sicily. A really harmful pest?

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Frankliniella occidentalis (Pergande) is considered one of the most damaging pests of table grapes in Sicily. The Sicilian regional disciplinary for the IPM, sets the intervention threshold at 2-3 thrips/bunch during flowering, recommending a pre-flowering chemical spraying and a second one, if necessary, at the end of this period. Sicilian growers carry out at least 2-3 sprayings in the period between the first week of May and the second week of June; for this we decided to verify the real need of chemical control of this pest, considering also some cultivation techniques, such as the closing of bunches into paper bags and the manual bunch thinning during the phenological stage 71 of BBCH-scale, commonly adopted in some Sicilian provinces. The experimental vineyard, cultivar Italia, is conventionally cultivated with pergola training system. Observations were carried out during 2018 and 2019. Four tests were taken into account: a) Ordinary conduction with one chemical spraying at the beginning of flowering (CondO); b) Bagging of the inflorescences at the beginning of flowering (INSAC); c) Releases of *Amblyseius swirskii* Athias-Henriot (BioC); d) Untreated control. The above mentioned intervention threshold was exceeded in the control in both years, with a peak of about 8 thrips/bunch in the phenological stage 65 of the BBCH scale (flowering 80%). On the other hand, in the “BioC” plants, the thrip population remained below the intervention threshold in 2018 (2 thrips/bunch in the phenological stage 65), while in 2019 it was abundantly exceeded with an average of 10.8 thrips/bunch during the fruit set (stage 71). In the "CondO" thesis, after a spraying with spinosad at the beginning of flowering, the population was low (less than 2 thrips/bunch) for the whole period, while in 2019 the intervention threshold was exceeded (5.22 thrips/bunch) after the fruit set. On the other hand, the population of thrip had an identical trend to that of the control on bunches no bagging in the "INSAC" thesis. During the veraison (stage 81), before the manual bunch thinning, the damage caused by *F. occidentalis* in the four thesis was assessed. As expected, in the "INSAC" plot the paper bag avoided the presence of thrips, but rubbing damage due to the paper bag on the outer berries was registred. On the other hand, the damage caused by the thrips in the control, have been similar trend in both years and was scarse: 2 damaged grapes/bunch on an average of 145 berries/bunch. However, during 2018 damage was statistically higher in the control, than in "BioC" and "CondO" plots; in 2019 the damage increased to 2 and 5 berries/bunch, for the two latter plots respectively, due probably to the late increase of thrips population. In 2019, the influence of the manual bunch thinning on the damage caused by the thrips in the control plot was assessed. On average, 12.9% of the berries per bunch have been cut off (19.7 berries/bunch); of these 16.2% were damaged by *F. occidentalis*. The experiments will continue in the current year, but the data up to now obtained, question both the need for chemical sprayings every year for controling *F. occidentalis*, and the economic thresholds adopted by the Regional IPM disciplinary.

KEY WORDS: *Frankliniella occidentalis*, table grapes, Sicily.

POSTER

Susceptibility of five olive cultivars to the attacks of *Bactrocera oleae*

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The susceptibility of five Calabrian olive cultivars was compared (Carolea, Cassanese, Ottobratica, Dolce di Rossano, and Tondina) was compared to the attacks of the diptera *Bactrocera oleae* (Rossi), the main problem in the protection of the olive tree. The test was carried out in an olive grove located on the Ionian coast of Cosenza in the municipality of Mirto-Crosia (Cosenza) characterized by high levels of infestation. For four years (2016-2019) for each cultivar, placed in the same pedo-climatic conditions, and for five repetitions a monitoring was carried out, every 8-10 days, of the phytophagous flights (male and female) with attractive chromotropic traps. There were 25 traps (5 for each cultivar) in the period from June to November. The percentage of active infestation, (presence of eggs, 1st and 2nd instar larvae), harmful infestation (3rd instar larvae, pupae and abandoned tunnels) and total infestation (all preimaginal stages, alive, dead and parasitized and abandoned tunnels) were determined by microscopic analysis of samples consisting of 100 drupes taken, on the cultivars under study at decadic intervals. No type of pesticide treatment against olive fly has been carried out in the study olive grove. The data collected and analyzed indicate that the cv Carolea is in general the most sensitive to infestation by this diptera, followed by the cv Dolce di Rossano, cv Cassanese, cv Tondina and finally cv Ottobratica. Notoriously, the fly shows a preference for larger olives, green rather than dark and spherical rather than elongated. In addition, several studies have shown that the different phenological conditions of the cultivars also show differences in the susceptibility to attack by *B. oleae*. The comparison between varieties with different tolerance levels can shed light on the genetic-molecular mechanisms underlying the process of response of the olive tree to the stress induced by *B. oleae*, providing useful information for genetic improvement for the creation of new tolerant varieties. With phenotyping, a quantitative and qualitative enhancement of plants of agricultural interest is possible, especially for aspects related to the sustainability of production (resistance to pathogens). Being able to identify cultivars less sensitive to phytosanitary problems, such as the olive fly, is both a guarantee for producers interested in the construction of new olive plants and an adequate solution to save the profitability of their companies.

KEY WORDS: *Bactrocera oleae*, susceptibility, Calabria.

POSTER

Liothrips oleae: emerging pest in South Italy

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Olive growing has always been one of the main productive sectors of the Calabrian agricultural sector. The systematic use of pesticides and the current climate changes are the main causes that trigger the alteration of biological balances within complex agroecosystems, including olive groves. Due to its low population density, the olive thrips, known as a secondary pest, never required ad hoc interventions for its containment. Since 2016, however, in Calabria, the olive groves of the Ionian coast were affected by massive infestations of the thrips species, which led to significant production losses due to the damage caused to the vegetative-productive apparatus of the plants. Field monitoring and evaluation of symptoms, in relation to different phytosanitary management systems, can provide useful information relating to the life cycle of the species, as well as the appropriate tools useful for implementing the best economically sustainable defence strategies and environmental. The study was conducted in the 2018-2019 period, through the sampling of fruits and sprouts, in olive-growing plots characterized by different phytosanitary management (Organic and IPM). The evaluation of the infestations was performed through a visual analysis and counts of the damage caused by the species. The estimate of some biometric parameters such as drupe diameters and the number of symptomatic leaves have been related to the damage comparison model adopted. The level of damage counted as the number of bites on fruits was greater in the fields where phytosanitary management is biological. For both years of study, the diameter of drupes is greater in integrated management olive groves. The results also show that the olive thrips is able to damaging leaves and fruits independently, in conditions of high populations density. These preliminary results highlight how a species considered as a secondary pest can become a key-pest for the olive crops in some Mediterranean areas. Further investigations will be necessary in order to understand the eco-biological factors behind this emergence.

KEY WORDS: *Liothrips oleae*, secondary pest, damage, phytosanitary management.

SESSION VII

Le difese antimicrobiche negli insetti: le attacine dell'insetto stecco Carausius morosus (Phasmatodea)

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Plant growth-promoting fungi (PGPFs) belonging to genus *Trichoderma*, are well-known to have beneficial effects on plant growth, yield and plant survival. *Trichoderma* spp. are also widespread biological control agents of plant pathogens with their antagonistic activities. Despite the effect of this genus on direct defenses against pathogenic microbes have long been known, we have limited knowledge regarding their impact on plant responses towards insect herbivory. Here, we aimed to establish whether *Trichoderma harzianum* colonization enhance the direct plant defense responses against above-ground piercing-sucking insects. For this purpose, we used the model system consisting of tomato plant, *Solanum lycopersicum* (cv "Dwarf San Marzano"); PGPF, *T. harzianum* strain T22 and polyphagous pest, *Nezara viridula* (L.) (Hemiptera: Pentatomidae). We observed a reduction in grow rate when *N. viridula* 3rd instar nymphs were feeding on tomato plants colonized by *T. harzianum* compared with control plants. Our finding suggests that soil-born beneficial microbes trigger direct plant defense response to herbivory. Herein, we further plan to investigate the impact of *T. harzianum* in mediating egg parasitoids attraction to herbivore oviposition in order to understand their indirect role in plant defense response system.

KEY WORDS: *Trichoderma harzianum*, *Nezara viridula*, beneficial microbe, tomato, multitrophic interaction.

Role of woods on the association between insects and oenologically relevant yeasts

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Several yeast species are present in many natural substrates (e.g. soil, tree barks, fruit) and hold the potential to influence the process of wine production [1]. In fact, vineyard fungal populations present on grapes drive the spontaneous fermentation of must, hence conferring characteristic traits to the final product [2]. As a consequence, winemakers and researchers are currently considering the microorganisms present in the vineyard as an active and fundamental component of the "terroir". Recently, an important role in defining the composition of vineyard fungal populations has been ascribed to some social insects. Indeed, wasps (*Polistes* spp.) and hornets (*Vespa crabro*) are capable of maintaining oenologically relevant yeasts within their gut, as components of their microbiota and vectoring them on grapes in the vineyard [3]. Preliminary studies showed that the composition of intestinal fungal populations of wasps changes according to the sampling area. Alas, the factors responsible for this variability are still unknown. The presence of woods close to the vineyard could favour not only insect colonization, but also the diversity of yeast populations vectored by insects, and hence the fungal populations capable of contributing to the fermentative process in the winery. To verify this hypothesis, we have caught insects in organic vineyards, close and distant from woods, and assessed, through culturomic and molecular identification approaches, the composition of yeast populations of the insects' gut. Preliminary data, relative to a single vintage, have highlighted significant quantitative and qualitative differences between the fungal population associated with insects caught in vineyards close and distant from woods, hence allowing us to confirm the initial hypothesis. Knowing the natural factors favouring the spread of yeasts in vineyards could allow us to tune the composition of fermentative fungal populations enhancing their contribution to the typicization of the final product.

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POSTER

Hints from the microbiota of Chrysomelidae: does the breadth of the diet spectrum affect the diversity of leaf beetle's microbiota?

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Most of the insects are colonised by a multitude of microorganisms, prevalently living as commensals, but that can also confer either beneficial or detrimental effects to their host. Those effects are of considerable importance in herbivorous insects, where symbiont mediated traits are often crucial in the interaction with the plant. Leaf beetles (Coleoptera: Chrysomelidae), an almost exclusively phytophagous group consisting of species with variable degrees of trophic specialization, represent a particularly interesting model to study the microbiota associated with herbivorous insects, especially regarding the effect of the breadth of the diet spectrum on the composition of the microbiota. We hypothesize that generalist species, feeding on different plants have microbiotas that are more complex than those of specialist species. This could be due to the interaction with more sources of bacterial contamination (different plants and microenvironments) but also to the improved metabolic potential of more complex microbiotas that allow the insect to exploit a higher variety of food sources. Some leaf beetle species (in the subfamilies Donacinae, Cassidinae and Eumolpinae) evolved specialized organs, associated to gut and genitalia, that host vertically transmitted bacterial symbionts mediating fundamental traits for the host (e.g., essential nutrients, digestive enzymes), but not all these symbioses have been well defined. The present study aims to characterize the microbiota associated to a selection of leaf beetle species, representative of the taxonomic diversity and the various degree of trophic specialization. The aims of this study are: i) to determine the main bacterial taxa that characterize the microbiota of the selected leaf beetle species, also detecting the presence of important insect symbionts (e.g., *Wolbachia*) and symbionts typically present in some Chrysomelidae subfamilies; ii) to test the hypothesis that the microbiota of generalist phytophagous species is more complex than the microbiota of more specialist species. Total genomic DNA was isolated from thirty selected species and sequenced with the Ion Torrent platform (Life Technologies) targeting two hypervariable regions of the 16S bacterial rRNA gene (V1-V2 and V4). The most represented bacterial classes were Alphaproteobacteria (~39%), Gammaproteobacteria (~45%) and Bacilli (~14%); while the most abundant and widespread species belong to *Wolbachia*, *Rickettsia* and *Pseudomonas*. We confirmed the presence of vertically transmitted symbionts in Cassidinae and Donacinae species. Interestingly, we also found two symbionts, previously isolated only from specialized gut organs of *Bromius obscurus*, in two other species in the same subfamily. Thus, suggesting the presence of specialized symbioses widespread also in the Eumolpinae subfamily. To provide insights into the role of these symbioses, we planned to sequence the bacterial genomes of candidate primary symbionts. Moreover, our results clearly showed that the microbiota of generalist insect species (feeding on several plant species) has significantly higher diversity when compared to specialists (whose diet spectrum is composed by only few plant species, often in the same genus/family). This result supports the hypothesis that the microbiota of generalist phytophagous species is more complex than the microbiota of more specialist species.

KEY WORDS: Chrysomelidae, phytophagous insects, host plants, microbiota, trophic specialization, symbiosis.

POSTER

Microbial interaction across multiple trophic levels: tomato plants treated with *Trichoderma harzianum* alter the microbiota of *Spodoptera littoralis*

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Organisms living in natural and agricultural ecosystems build complex networks of multitrophic interactions. Mechanistic insights are often lacking, but microorganisms are known to play key roles in mediating this interplay that can affect multiple trophic levels. For example, soil microorganisms exerting beneficial effects on plants can affect also additional trophic levels, such as herbivores feeding on the plant and the associated microbiota. Spodoptera littoralis, a pest of various crop widely adopted as a model organism of phytophagous chewing insects, experience a reduced fitness when fed with tomato plants treated with *Trichoderma harzianum* T22, a fungal strain used as a plant beneficial microorganism. Here this multilevel interaction was investigated leveraging physiological, transcriptional, metagenomic approaches, with a focus on possible alteration of the insect microbiota. Larvae were fed with leaves collected from tomato plants grown from seeds coated with *T. harzianum* T22 or from control plants, IV and VI larval instars were examined. Larvae were dissected to isolate the midgut and separate the gut content within the peritrophic membrane (for enzymatic assays, and microbiota metatranscriptomic and metagenomic) from the midgut tissues (for enzymatic assays and the transcriptomic of the insect host). A partial taxonomic shift in the composition of the microbiota is observed because of the treatment, with a more substantial effect in VI instar larvae. We observed the appearance of taxa almost absent in the control group (e.g., *Flavobacterium*) and also shifts within the most abundant bacteria, known to be important symbionts of *S. littoralis* (i.e., *Enterococcus* spp.). Furthermore, the microbiota of the treated group shows higher values of alpha diversity, providing further support to the evidence that the treatment causes a dysbiosis in the insect gut. When the functional role of the core microbiota is considered, the treated group shows a strong perturbation in transcription activity. Most of the activity reduction is associated to *Enterococcus casseliflavus*, a previously overlooked component of *S. littoralis* core microbiome. Strikingly, in IV instars we observed a consistent reduction of expression in pathways associated to the production of essential amino acids, ribonucleotides and sugar metabolism; these processes represent key contributions of the microbiota to the fitness of the insect host. The observed effect of the treatment to the core microbiota activity and composition is mirrored at the host level: gene differential expression in the gut of the treated group larvae highlights the activation of defense response to bacteria, slower developmental pace and disrupted nutritional uptake for carbohydrates and proteins. Moreover, an alteration of digestive enzyme activities was observed. Our preliminary results support the hypothesis that the microbiota can represent the connection between the T22 treatment and its effect on *S. littoralis* larvae, with a dysbiosis leading at the same time to the proliferation of potential pathogenic strains and an impaired nutritional supply. These data provide an example on how a complex multitrophic interactions can be hijacked for efficient and specific biocontrol strategies.

KEY WORDS: *Trichoderma harzianum*, *Spodoptera littoralis*, pest, gut microbiota, biocontrol, multitrophic interaction.

ORAL PRESENTATION

Evaluating changes in insect-microorganism relationships along an altitudinal gradient

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Alpine pastures are one of the most common landscapes in the Alps even though the climate change and the progressive abandonment of extensive farming are serious threats to this kind of environment. Moreover, pastures harbour a highly specific biodiversity and it is considered that the great amount of biomass is present below-ground instead of above-ground, including livestock and plants. Several aspects of the soil organisms living in Alpine pastures are almost unknown, especially the interactions between microorganism and soil fauna are still undiscovered. Naturally, a great number of Prokaryota and Fungi are predated by soil animals, but they are also symbiotic with them, for example as part of their gut microbiota. Besides, the microbiota associated with soil fauna is paramount for the health of soil fauna itself. Considering that this kind of interactions are almost unexplored, the aim of the study is to evaluate the relationship between microorganism and soil fauna including abiotic factors (temperature, soil moisture, soil organic content, etc..) as well as abiotic factors, in order to elucidate the main drivers of soil biodiversity. The study is carried out along an altitudinal gradient in order to test the effects of climate change on soil fauna and its associated gut microbiota as well as soil and rhizosphere microbiota. Regarding soil fauna, we selected different taxa: 1) Nematodes as component of soil microfauna and as important predators of microorganisms. 2) Collembola as component of mesofauna, including several detritivores species. 3) Earthworms, the soil engineering that affect the soil structure. 4) Beetles belonging to two different families: Ground Beetles (Coleoptera: Carabidae) and Rove Beetles (Coleoptera: Staphylinidae) as important predators of invertebrates. Thanks to the Next Generation Sequencing methods such as metataxonomics, we can identify entire microbial communities. The analyses are performed on sequence 16S rDNA gene (V4-V5 regions) for the Prokaryota and the Internal Transcribed Spacer 1 for Fungi. Since the DNA analyses are still in progress, we are going to present the preliminary results of the study at the Congress.

ORAL PRESENTATION

Population genomics of factors influencing phytoplasma transmission

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Phytoplasmas are obligatory endosymbiotic bacteria that colonize the phloem of many plant species and cause hundreds of plant diseases worldwide. In nature, phytoplasmas are primarily transmitted by hemipteran vectors of the families Cicadellidae, Cixiidae, and Psyllidae. While, in principle, all phloem sucking insects could transmit phytoplasmas, only a limited number of species have been confirmed as vectors. This is because the transmission efficiency of phytoplasmas is a function of the complex tripartite association between the insect vector, the host plant, and the phytoplasma itself. Apple proliferation is a disease caused by the phytoplasma "*Candidatus* Phytoplasma mali", causing proliferation of auxiliary shoots and a decrease in fruit size and quality. '*Ca.* P. mali' is mainly transmitted between apple trees by two psyllids, *Cacopsylla picta* and *Cacopsylla melanoneura*, with regional differences. *C. melanoneura* is considered the main vector of AP phytoplasma in Northwestern Italy, but it is a poor transmitter in Northeastern Italy and other parts of Europe. In contrast, *C. picta* is the primary vector in most European populations with variable transmission rates among populations. Other occurring *Cacopsylla* species do not transmit phytoplasmas. Knowledge about factors influencing the transmission efficiency of "*Ca.* P. mali" is currently scarce.
To compare the acquisition efficiency of different populations of *C. melanoneura*, we collected several individuals in orchards in South Tyrol, Trentino and Aosta Valley. We reared them on plants infected by "*Ca.* P. mali" strains from Aosta Valley and subtypes AT1, AT2 from South Tyrol. After an inoculation period of several weeks, F1 individuals will be tested for the presence of phytoplasma to verify if they were able to acquire it from the infected trees. By sequencing the genome of various vector populations, we aim to determine key factors affecting phytoplasma transmission in different *Cacopsylla* species. Our results will provide novel insights into the complex biology of "*Ca.* P. mali" transmission and will be therefore an important milestone in combatting this disease.

KEY WORDS: apple proliferation, *Candidatus* phytoplasma mali, *Cacopsylla melanoneura*.

ORAL PRESENTATION

Gut microbiome compartmentalization in honey bee foragers

Elena Crotti[1], Matteo Callegari[2], Marco Fusi[2], Ramona Marasco[2], Elena Gonella[3], Ivano De Noni[1], Diego Romano[1], Sara Borin[1], George Tsiamis[4], Ameur Cherif[5], Alberto Alma[3], Daniele Daffonchio[2]

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The gut microbiome of the adult honey bees includes nine dominant bacterial phylotypes for which considerable information on genomic and phenotypic features and evolution has been accumulated. Conversely, limited information is available for the less abundant environmental bacterial phylotypes, as well as for the fungal microbiome. Here, we showed that in the gut of honey bee foragers exists a compartmentalization of the dominant and less abundant bacterial and fungal communities, driven by the local physico-chemical and metabolic conditions. High-throughput amplicon sequencing and quantitative PCR were used to investigate the diversity, abundance and distribution of the three microbial components (i.e. core bacteria, minor environmental bacterial phylotypes and fungal members) along the insect gut (and specifically in the compartments of crop, midgut, ileum and rectum). Significant changes of oxygen concentrations, redox potential and pH, as well as metabolites, were measured along the gut through microsensors and HPLC. All the gut compartments showed a positive redox potential and were anoxic in their centre with a radial oxygen gradient from the epithelia, indicating a role of the microbial distribution in oxygen scavenging. Gut lumen pH significantly decreased from the crop to the rectum according to the acidogenic metabolisms of the inhabiting microorganisms. Our data provide evidence of a physicochemically-driven gut compartmentalization of the three microbial components associated with the honey bee foragers.

KEY WORDS: Microbiota, mycobiota, gut, honeybee.

ORAL PRESENTATION

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

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Beneficial fungi belonging to the genus *Trichoderma* have well-known effects on plant biology, such as promotion of their growth, efficiency of nutrients uptake and use, seed germination rate, and influence plant interactions with the environment, by promoting defense barriers against both biotic and abiotic stress agents. There is a consolidated body of information on the capacity that many *Trichoderma* strains have to activate plant resistance mechanisms targeting a number of phytopatogens, against which they can also directly act as biocontrol agents. Comparatively much less is known on the effects that the plant metabolic changes induced by these beneficial fungi can have on insects. A growing experimental evidence has already shown that they can contribute to limit growth and survival of aphids and can enhance the impact of their natural enemies, even though these results appear to be strain specific.
Here we further contribute to this research topic by studying the effect of *Trichoderma harzianum* strain T22 (T22) on the noctuid moth *Spodoptera littoralis*. Larvae fed with tomato plants obtained by seed coating with spores of T22 showed a marked and significant reduction of their growth, delayed development and a much higher mortality, compared to controls. These effects observed on insects are corroborated by alterations of tomato transcriptomic asset. Among differentially expressed genes, the up-regulation of transcripts associated to different aspects of plant defences may account for the observed negative effects on *S. littoralis* larvae. These latter are currently being investigated at physiological and molecular level, by evaluating the gut enzymatic profile and its transcriptome, and assessing the impact of plant metabolic changes on gut microbiota, which deeply influences gut physiology. Our results corroborate the emerging evidence that beneficial fungi in the genus *Trichoderma* are true plant symbionts, that offer protection against a wealth of biotic stress agents. This intriguing evolutionary hypothesis has important implications from an applied perspective, paving the way towards the definition of broader biocontrol strategies, effective against both pathogens and pest insects.

KEY WORDS: Beneficial microorganism, plant resistance, *Trichoderma harzianum*, *Spodoptera littoralis.*

ORAL PRESENTATION

Variability of *Trichoderma* efficacy against insect pests

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Soil microorganisms living as root symbionts can promote plant growth (PGPR, PGPF) resulting in better yield, so their use in agriculture is steadily increasing. Such application is even more interesting when they are also active in plant protection from pathogens, as many Trichoderma spp. can do. In spite of their diffusion, there is no general consensus yet whether Trichoderma strains effective against plant pathogens are efficient also in plant defence from insect pests. The genetic analysis of plants colonized by this fungus and by feeding insects is a tool that may be of help in clarifying this constraint.
Here we present data about the interactions between tomato plants and Trichoderma strains and the resulting impact on defence from aphids and caterpillars, at controlled environmental conditions. Abiotic factors are essential in determining the outcome of such interactions, since they regulate plant gene expression and consequently the herbivore performance. We observed that the expression of defence-related genes, as well as the insect response, is dependent both on the strain and the temperature. Our conclusion is that, for a successful application of Trichoderma in the control of insect pests, a careful selection of the strain adapted to local/seasonal environmental conditions is mandatory.

KEY WORDS: induced plant defence, root symbionts, beneficial microorganisms.

Long-read genome sequencing of two co-occurring primary endosymbionts in the psyllid *Cacopsylla melanoneura*

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Hemipteran insects are well-known for their ancient and intimate associations with beneficial bacterial endosymbionts. These bacteria are generally nutritional symbionts providing the host with essential nutrients such as amino acids or vitamins lacking from the host's diet. Hence, they enable the exploitation of nutrient-poor food sources such as plant sap or vertebrate blood. In the Auchenorrhyncha (cicadas, planthoppers, spittlebugs), this host-symbiont collaboration has become even more complex and specialized, as at least two different bacterial endosymbionts are needed to jointly produce the complete set of essential amino acids required by the host, resulting in an intricate metabolic interdependence between the three partners. Similar multipartite symbioses may occur in psyllids, since microscopy observations revealed the presence of two types of bacteria in the bacteriomes of various psyllid species. 16S rRNA gene sequencing further showed that all investigated psyllid genera harbour the primary endosymbiont *Carsonella* (which has one of the smallest bacterial genomes sequenced to date), whereas the second endosymbiont appears to be variable depending on the host species. Here, we present the complete genome sequences of both *Carsonella* and the as yet uncharacterized second endosymbiont of *Cacopsylla melanoneura*, an important vector of Apple Proliferation Phytoplasmas. Unlike co-occurring endosymbionts in other psyllid species, the genome of the second endosymbiont is also very small, indicating either extreme genome streamlining or genomic degradation. We will discuss the potential co-evolution of these two symbionts and their metabolic potentials in light of the nutritional requirements of their host.

KEY WORDS: Symbiosis, host-symbiont co-evolution, genomics.

Susceptibility of *Culex pipiens* to entomopathogenic *Brevibacillus* and *Pseudomonas* species

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Mosquitoes are rather common insect pests with a very high reproductive potential and represent major arthropod vectors of human diseases worldwide. Among a wide variety of species, Culex pipiens L. (Diptera: Culicidae) represents a major concern in the Mediterranean area. The limitations of traditional insecticide-based strategies, the related risks for the environment, and the possible development of resistance, support the development of alternative and eco-friendly methods. Among these is the employment of entomopathogenic bacteria, such as Bacillus thuringiensis subsp. israelensis (Bti) that finds successful use against this pest. However, the availability of other microbial products would be very useful due to a limited efficacy of Bti and the possibility of resistance development. In this study, specific isolates of Brevibacillus laterosporus and Pseudomonas protegens were assayed in the laboratory against C. pipiens larvae. As expected, laboratory bioassays showed a higher susceptibility of younger larvae and mortality levels of approximately 100% achieved with 1 billion bacterial spores or cells/ml after 24 h. Estimated median lethal concentrations (LC50s) were in the range of 5-6 and 50-70 million CFU for young larvae exposed to B. laterosporus and P. protegens, respectively. Preliminary bioassays allowed to predict also possible sub-lethal effects of these bacterial species on the development of mosquito larvae.

KEY WORDS: biocontrol, mosquitoes, pest, bacteria, IPM.

Gut microbiota of *Pseudophonus rufipes* showed differences on bacterial communities' composition between organic farming and Integrated Pest Management in agricultural systems.

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Recently, the role of intestinal microorganisms has been demonstrated for an array of functions, including the synthesis of compounds essential for insect life, such as the production of vitamins, micronutrients and sterols as well as food detoxification. Many factors may alter the composition of bacterial communities (e.g. pH, insect stage, diet and environment) with strong repercussions on survival, development and fecundity of insect hosts. However, the majority of studies on this topic show the lack of information on some trophic guilds (e.g. predators) and the overrepresentation of some insect orders (e.g. Rhynchota and Hymenoptera). The main aim of this study was to assess the composition of bacterial communities living in the gut content of carabids. Specimens were collected in farms under different management conditions (ORGANIC vs IPM) by using modified "semi-dry" pitfall traps activated in the afternoon and emptied the morning after. Carabids were individually stored and frozen at -80° until the next molecular assays. In order to assess only the microbiota present in the gut content of carabids, the alimentary canal of insects was isolated from the insects with a microsurgical technique before proceeding to DNA extraction. Finally, quantitative Real-Time PCR (qPCR) and Next Generation Sequencing (NGS) were used for determining the microbiota composition of *Pseudophonus rufipes*, the most abundant species occurred in our samplings. q-PCR didn't show any differences for Eubacteria, while strong differences emerged by NGS output. In particular, the abundance of Bifidobacteriaceae, Xanthomonadaceae and Spiroplasmataceae was higher in *P. rufipes* collected in organic farming than in IPM while, on the contrary, Anaplasmataceae, Enterobacteriaceae and Enterococcaceae were predominant in carabids living in IPM system. In conclusion, to the best of our knowledge, this study was the first of its type on gut microbial communities harbored by carabid beetles under different agricultural management practices. This is the first study recording the presence *Bifidobacterium* spp. in carabids. Further studies are needed for a better understanding of the role of the gut microbiome on predator fitness and the role of agricultural practices on microbiome diversity.

KEY WORDS: *Pseudophonus rufipes*, carabids, gut microbial communities, q-PCR, Next Generation Sequencing, agricultural systems, integrated and organic production.

ORAL PRESENTATION

Does the immune answer to *Bacillus thuringiensis* infection is the same in larvae, females and males of *Rhynchophorus ferrugineus*?

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Bacillus thuringiensis is considered a potential useful entomopathogen of *Rhynchophorus ferrugineus* (RPW), a quarantine pest that attacks the palm trees. We compare the effects of Bt in mature larvae, females and males. The pathogenicity activity of Bt was evaluated estimating: Median Lethal Dose (LD50), Median Lethal Time (LT50), and Total Haemocytes Count (THC) and Differential Haemocytes counts (DHC), and finally the expression of the stress protein Heat Shock Protein 70 in the haemocytes and in the brains. RPW mortality exhibited a positive trend with the dosage and duration of exposure to Bt. The larvae were more susceptible then adults and the LD50 of females was even the double value of the one of larvae. Similarly, the LT50 value was higher for females than males and larvae. Treatment with sub-lethal dose of Bt induces a decrement in THC in larvae, females and males. In treated larvae plasmatocytes decrease while oenocytes and spherulocytes increase. In treated females all type of haemocytes decrease, while in males the number of plasmatocytes decrease and the granulocytes increase. We also registered the stress answer directly on hemocytes, showing that already at 3, hours after eating Bt, it modulates the expression of the stress protein Hsp 70. This effect was also observed in the brain tissue at 6 hours after treatment.
The results confirm that the Bt treatment induce a pathogenic state in RPW larvae and adult of both genders, that answer only after few hours from ingestion, however the effects are different in magnitude and in type of target. The highlights can be useful to understand better the relationship pathogen-host and to implement the integrated control of phytophagous.

KEY WORDS: median lethal dose, median lethal time, stress answer, haemocytes, brain, HSP70.

POSTER

The association between Halyomorpha halys and its primary symbiont "*Candidatus* Pantoea carbekii": a useful tool for studying invasion routes

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Intimate associations between insects and bacteria are widespread in nature. Numerous benefits are provided by symbiotic bacteria to their host and they seem to play an important role in biological invasions of exotic pests. The close association between primary symbionts and their host can be used as a marker to study the phylogeographic structure of the insect. The brown marmorated stink bug, *Halyomorpha halys* (Heteroptera: Pentatomidae), is an exotic species causing economic and ecological damage in invaded areas. Native to Southeast Asia, *H. halys* was accidentally introduced into North America in 1996 and Europe in 2004, causing substantial damage in agriculture and in urban areas. The species is associated with an heritable symbiotic bacteria, '*Candidatus* Pantoea carbekii' which inhabits specialized midgut ceca of the host. In the present work, we investigated the significance of the host-symbiont association and the possibility of using it as a tool to elucidate the routes of invasion of the pest. Special attention was given to the northeast of Italy, where the pest causes heavy damage and high levels of genetic variability of the insect host have been recently reported. The genetic variability of both native and newly introduced populations were studied through the analyses of one bacterial marker (the putative pseudogene ΔybgF) and one mitochondrial gene (cytochrome c oxidase). A total of seven haplotypes of *P. carbekii* were identified, five of which for the first time. Moreover, an association pattern between host-symbiont haplotypes was observed. The results highlight that vertical transmission is the main mechanism for acquisition of this bacterial symbiont, despite its extracellular occurrence. Populations from the northeast of Italy showed the highest values of genetic diversity with both markers, suggesting that this area could be the result of multiple ongoing introductions of the pest from the native regions. Specimens from north-eastern Italy not only shared haplotypes with samples collected in China, as already reported, but also with samples from Japan: so the latter country might likely be a source of further introductions into north-eastern Italy. Overall, our findings highlight that host-symbiont associations could be used as a tool for understanding the invasion patterns of *H. halys*, and they also provide some cues potentially useful for improving pest control programs, especially towards the two Asian countries identified as a possible source of introduction of such harmful insect species.

KEY WORDS: psymbiosis, Brown Marmorated Stinkbug, exotic pest, vertical transmission.

ORAL PRESENTATION

First detection of xylem-sap feeding insects infected with *Xylella fastidiosa* subsp. *multiplex* in the Monte Argentario area

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Xylella fastidiosa Wells et al., is a Gram-negative bacterium which is exclusively transmitted by xylem feeding insect belonging to the Auchenorrhyncha. After its first outbreak in Europe, in 2013 in Apulia (Italy), *X. fastidiosa* has been included in the EPPO A2 list of quarantine pest and it is regulated by specific control measures. In Italy, several surveys have been carried out by the Regional Phytosanitary Services, as emergency action aimed to early detect *X. fastidiosa* in non-infected areas. Since 2014, the Phytosanitary Service of Tuscany has carried out surveys by sampling host-plants and potential insect vectors of the bacterium. Later on, more attention was paid to the true spittlebugs *Philaenus spumarius* L. and *Neophilaenus campestris* (Fallen) since in Apulia they were proved to be vectors of *X. fastidiosa* subsp. *pauca* (ST53) with different role in spreading the olive quick decline syndrome. In 2018 some *Spartium junceum* plants, collected from Monte Argentario area, have been resulted infected with *X. fastidiosa* multiplex, an extremely polyphagous subspecies so far recorded in France, Spain and Portugal. Subsequently, the territory of the Monte Argentario municipality was included in a delimited area, as mandatory by law (EU Commission Implementing Decision 2015/789 of 18 May 2015). Consequently, more intense surveys were planned and conducted in the Monte Argentario area in order to detail the spread of the pathogen. In the same area, at the end of 2018 two specimens of *N. campestris* were tested positive to *X. fastidiosa* subsp. *multiplex* (ST87) (out of a sample of 40 individuals which included two adults of *P. spumarius* and 13 of *N. campestris*). Since there is no specificity in the relationship plant-pathogen-vector, in 2019 faunistic studies were carried out on this biotope to have a deeper knowledge on Auchenorrhyncha and to assess the presence of potential vectors of *X. fastidiosa*. *Philaenus spumarius* and *N. campestris* resulted widely distributed in all the delimitated area, with 258 and 117 specimens respectively collected from May to November out of a total of 662 Auchenorrhyncha caught. The biomolecular analyses allowed to detect *X. fastidiosa multiplex* (ST87) DNA in seven *P. spumarius* pooled samples (33 specimens overall) and in one single and two pooled samples (14 specimens overall) of *N. campestris*. The Mediterranean species P*hilaenus italosignus* Drosopoulos & Remane, also reported as a vector of *X. fastidiosa* subsp. *pauca* (ST53) but with a secondary role, so far has not been collected in the Monte Argentario area, although it was recently recorded in Tuscany in the Grosseto province. The detection of positive specimens of *P. spumarius* and *N. campestris* allow us to hypothesize their involvement too in the transmission of *X. fastidiosa* subsp. *multiplex*.

KEY WORDS: Tuscany, Aphrophoridae, *Philaenus spumarius, Neophilaenus campestris*.

POSTER

Symbiotic control, a sustainable tool against *Halyomorpha halys*

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The interaction between insects and microbial symbionts is a component of insect life cycle deserving a growing attention because of its influence on their metabolism, reproduction, and behavior, which contributes in determining important traits from the human perspective, such as invasive potential of pests – intended as the capability to colonize new environments – or the vector competence for several phytopathogens. The increasing knowledge on symbiotic associations can be applied to insect management in agriculture, considering these relations as an instrument or a target for developing new control approaches, according to the symbiotic control strategy. Symbiotic control can be focused on direct suppression of a pest insect or to block its vector competence for pathogens. It can be operated in different ways, such as genetic transformation of microbial symbionts (paratransgenesis), set up of heterologous associations, or disruption of existing symbioses. Symbiotic control techniques based on disruption of existing symbioses are especially effective for extracellular symbionts that are transmitted from the mother to the progeny by passing through an environmental phase. Gut symbionts of the Heteroptera in the family Pentatomidae are the optimal target for this approach, since they are transmitted by egg smearing of symbiont-containing maternal secretes, which are ingested by newborns after hatching. Surface treatment of egg masses with substance showing antimicrobial activity, such as micronutrient fertilizers, have been proposed for the brown marmorated stink bug *Halyomorpha halys* Stål. Product application results in high nymphal mortality in the first instar, proving a considerable contribution to the reduction of stink bug field populations. Even though this technique is still under field validation in different crops, the exploitation of the accessory effect displayed by biocomplexes of micronutrient fertilizers represents a valid control option. This strategy widely compatible with all the current control measures, including both insecticide sprays and biological control, without interfering with the other agroecosystem components and providing a sustainable alternative to *H. halys* integrated pest management.

KEY WORDS: Halyomorpha halys, Candidatus Pantoea carbekii, vertical transmission, symbiotic control.

POSTER

Survey on *Xylosandrus compactus* associated fungi in Italy

Salvatore Vitale, Laura Luongo, Fabrizio Pennacchio, Mariangela Petrucci, Massimo Galli, Giovanna Tropea Garzia, Antonio Gugliuzzo, Valeria Francardi

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The black twig borer, *Xylosandrus compactus* (Eichhoff), is an invasive ambrosia beetle (Coleoptera: Curculionidae: Scolytinae) globally recorded on more than 220 host plants (EPPO 2020). Several fungi have been described as associates of *X. compactus*, three of them have been reported most consistently: *Ambrosiella xylebori*, *Ambrosiella macrospora*, and *Fusarium solani* (Brader 1964, von Arx and Hennebert 1965, Batra 1967, Bhat and Sreedharan 1988). Since 2011, *X. compactus* has been recorded in Italy on different ornamental plants in Campania, on *Ceratonia siliqua* in Sicily and on *Laurus nobilis* in Tuscany and Liguria. More recently, in 2016, a widespread dieback of branches and twigs of different plant species has been observed in several sites in the province of Latina (Lazio) (Francardi et al.2017). An intensive monitoring has been conducted as part of the Project "Contributions for the control of phytosanitary risk caused by *Xylosandrus compactus*, *Xylella fastidiosa*, *Botrytis cinerea* - CO.XI.BO" (D.M 0037134 of 31.12.2018) funded by the Ministry of Agricultural, Food and Forestry Policies (MiPAAF). In Autumn 2019 and Summer/Autumn 2020, in Lazio, Tuscany and Sicily, samplings were carried out on specific Mediterranean host wood species, namely *Laurus nobilis*, *Ceratonia siliqua*, *Quercus ilex*, *Pistacia lentiscus*, *Arbutus unedo* and *Magnolia grandiflora.* More than 200 sampled branches and twigs with small entry holes (0.8–1 mm in diameter) were observed and associated with external symptoms of wilting. Adult beetles (alive and dead) were found inside infested galleries. Large longitudinal necroses were always associated with the galleriess and extending upward and downward along the branches. Insects and small pieces of necrotic tissues were plated on Potato Dextrose Agar (PDA) and Malt Extract Agar (MEA) media. Over to 15 fungal morphotypes were identified on basis of morphological features and in some cases confirmed by molecular identification of Internal Transcibed Spacer (ITS) rDNA.
Acremonium-like spp., *Geosmithia* spp., *Pestalotiopsis* spp., *Botryosphaeriaceae* spp. and *Ambrosiella xylebori* were more frequently isolated from all monitored sites. *Ambrosiella xylebori* was mainly collected from *Ceratonia siliqua* in Sicily while two species of *Geosmithia* (*G. pallida* and *G. flava*) were mainly collected, in 2020, from *Laurus nobilis* in Latium and in Tuscany respectively. Many other fungal species were isolates in this survey, such as *Phomopsis*, *Alternaria alternata* complex, *Fusarium solani*, *Penicillium*, *Cladosporium*, *Cytospora acacia* and *C. berkeleyi*, *Aureobasidium pullulans*, *Phoma*, *Gliocladium roseum*, *Trichothecium roseum*, *Paraconiothyrium brasiliense* and *Trichoderma*. In addition to the most common symbiotic fungi associated with *X. compactus, Fusarium solani* and *Ambrosiella xylebori* (Ngoanet al. 1976; Bhat and Sreedharan 1988; Bosso et al. 2012; Bateman et al. 2016; Gugliuzzo et al. 2020), other fungi, identified in this study, can be considered as potential causal agents of woody necrosis and cankers. This is the case of *Neofusicoccum parvum* and *N. australe* (*Botryosphaeraceae*) isolated from *Ceratonia siliqua* and *Laurus nobilis*, respectively, that are known aggressive polyphagous pathogens attacking more than 100 hostplants (Aiello et al. 2020; Manca et al. 2020).
Concerning the *Geosmithia* genera, while *G. pallida* has already been reported on *Laurus* (Vannini et al., 2017) little is known about *G. flava.* Associations of *Geosmithia* spp. with various host trees and vectors are relatively well known in Central Europe, more frequently with hardwoods (Jankowiak R and Bilański P. 2018).
In order to better understand the role of *X. compactus* associated with various fungi, as causal agents of primary necrosis on the monitored plant species, pathogenicity tests will be carried out by artificial inoculation.

POSTER

SESSION VIII

Changing in mosquito biodiversity over the years in a wetland reserve in Northeast Italy

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Wetland ecosystems provide ecological niches that shelter a great variety of species, especially those that depends on the water availability, as mosquitoes. Nowadays climate change, pollution, land use change, and environmental management might impact ecosystems and affect biodiversity. The study aims at analysing the mosquito community in a limited area, estimating its composition, and potential changes in time. The study area encompasses a WWF Oasis (Valle Averto, Northeast Italy), a wetland reserve situated in a Site of Community Importance (SCI, EU Habitat Directive 92/43/EEC). We used three methods to collect adults and larvae: (i) CDC-CO2 adult trap, (ii) manual larval sampling in natural and artificial water collection, and (iii) manual adult aspiration. CDC trapping data were available from 2009 to 2020, allowing comparing populations across time. Manual aspiration and larval sampling was performed only in 2019 and 2020. CDC trap was placed with a biweekly timeframe, and both adults and larvae sampling were performed from late spring to autumn. Alpha-diversity indices (species richness, Gini-Simpson's and Pielou's indices; GS and J) were estimated to assess the fluctuations of the mosquito population in 2010-2020, and the potential variations in species dominance. Overall, 18 different species belonging to Culicinae (n=15) and Anophelinae (n=3) were caught during the study period. Although community composition varied, the most represented species across the period were *Culex pipiens*, *Ochlerotatus caspius*, *Aedes vexans* and *Aedes albopictus*. They comprised on average 88.8% (SD ±9.4) of the total mosquitoes collected. Community was strongly dominated by *Cx. pipiens*, except in 2015 when *Oc. caspius* superseded it. Aedes vexans represented between 10.5 and 32.6% of total mosquitoes sampled until 2012 and markedly dropped between 0.41 and 5.95% in 2013-2019, with a slight recovery in 2020. Many other species were observed with very low abundances (0.2-3.6%), although only one was constantly detected (*Coquilletidia richiardii*). Overall, the species richness observed in the area varied over time, showing a peak in 2011 (n=11), and a minimum in 2017-2019 (n=7). The GS and J indices showed similar trends. The higher diversity and equitability levels observed in 2011 (GS=0.58, J=0.45), 2012 (GS=0.58, J=0.45) and 2015 (GS=0.64, J=0.55), were likely associated either with the presence of multiple species with similar abundances (i.e. *Cx. pipiens* and *Ae. vexans*, in 2011-2012) or with a reduction of the *Cx. pipiens* population combined with an raise of other previously unrepresented species (2015). The lower levels recorded in 2013 (GS=0.35, J=0.34) and 2017 (GS=0.34, J=0.33), were potentially related to *Cx. pipiens* population dominating the mosquito community, with a concurrent drop in the abundances of other species. Manual aspiration allowed detecting a species never trapped by CDC, *Cx. hortensis*, while larval sampling confirmed what found by adult sampling. Although to be ascertained, given the differences in mosquito species biology, the fluctuations in population diversity throughout the study period might be influenced by a combination of anthropogenic and natural factors, including environmental/climatic drivers (i.e. precipitations, temperature, water salinity etc.) and management of water levels in the reserve canals and other artificial containers.

KEY WORDS: biodiversity, mosquitoes, wetlands, relative abundance.

POSTER

Morphological adaptations in hippoboscid flies living on different hosts

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Hippoboscidae family belongs to the Diptera order and includes three subfamilies, namely Ornythomyinae, Hippoboscinae and Lipopteninae, with 213 described species overall. Both sexes are obligate hematophagous ectoparasites that infest various species of birds, domestic mammals and wildlife. Hippoboscids live at the expense of few suitable host species able to provide all the requirements they need and to guarantee their long-term survival. Nevertheless, they can also feed on other species, including humans with a relevant risk to public health, since they may transmit micro-organisms responsible of zoonoses. Some species permanently live on the same animal, remaining into its coat, while others are able to switch frequently subject. The association between victim and fly depends on the type of parasitism and affects the adaptation level evolved to efficiently co-exist with the host. Parasites that are restricted to few species are highly adapted morphologically, but also the environment affects the development of their features. In fact, these diptera evolved several structures of body regions as a response to the extreme ectoparasitic lifestyle. Hippoboscids are poorly investigated, especially for the adaptation process that allowed them to live strictly together with their hosts. To highlight the development of morphological characteristics, light and Scanning Electron Microscope microscopy investigations have been conducted on four species belonging to the three different subfamilies: *Lipoptena cervi* (Linnaeus), *Lipoptena fortisetosa* Maa, *Hippobosca equina* Linnaeus, and *Pseudolynchia canariensis* (Macquart). Besides, in order to explain the role of different environments during their adaptation process, ungulate furs and pigeon feathering have been observed as well. The investigation mainly focused on sensory area located on the external surface of antennae, blood-feeding apparatus, wings, and legs with the adhesion organs. Results highlighted that some characteristics are divergent features among the species, suggesting that the different host range and environment could have affected the morphological development of these parasites. On the contrary, mouthparts are strongly adapted to blood-sucking and represent a convergent feature, commonly present in the Hippoboscoidea superfamily. This comparison proved that the morphology of these parasite has been noticeably influenced by the adaptive selection pressure occurred during the evolutionary process. Studying the relationship between ectoparasites and hosts is crucial also to understand the spread of allochthonous species. To hypothesize the spreading of *L. fortisetosa* from Asia to Italy, another morpho-molecular study was conducted in order to verify if samples collected in Italy are phylogenetically closer to those from the original areas rather than the European ones. Outcomes show that Italian specimens are genetically identical to Koreans and just close to Europeans. These results lead to speculate possible routes travelled by *L. fortisetosa* to reach our country, probably together with its original host.

KEY WORDS: ked flies, louse flies, zoonoses, morphology, SEM, host coat.

ORAL PRESENTATION

Archeo-entomological survey of the tomb of King Ferrante II d'Aragona and other Renaissance nobles

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Funerary archeo-entomology analyses the remains of arthropods found in contexts of archaeological interest, such as tombs, sarcophagi or other burial sites. Although it does not fall within the specific topics of forensic entomology, it shares the collection methods and analysis, particularly those focusing on cadavers. In coanceled environments, such as sarcophagi and tombs, the analysis of arthropod communities can provide important information on ecological, taphonomic and cultural events. In the present study, the analysis of four bodies conserved in the sacristy of the Basilica of San Domenico Maggiore in Naples (14th century) is presented. This church houses 38 sarcophagi containing the remains of ten memebers of the royal family belonging to the Aragonese dynasty, together with other Neapolitan nobles. This study focuses on the bodies of the King of Naples Ferrante II d'Aragona (1469-1496), Francesco Ferdinando d'Avalos, Marquis of Vasto and Pescara (1530-1571), the Queen of Naples Giovanna IV d'Aragona (1479–1518) and Caterina di Moncada, Duchess of Montalto (1611-1659). Several fragments of insects (n=842) belonging to the orders of Diptera, Coleoptera and Lepidoptera were collected. The entomological community was represented by species that colonize the bodies in the last phases of the decomposition processes. The complete absence of calliphorid flies was noted. This supports the hypothesis of a sudden storage of the body in a protected environment and a rather long period of conservation in a dry and well-sheltered place, preventing access to these insects. This contributes to depict a historical scenario where the knowledge about body embalming techniques was advanced, and this sophisticated preparation was applied to people of high social rank. Among the entomological samples, dermestid beetles and tineid moths were found, confirming an access to the bodies occurred at a considerable temporal distance from their burial. The complete absence of tineid adults was interesting, as that of dermestid adults. These data are well associated with the hypothesis of late and prolonged opening of the sarcophagi, which allowed these insects to colonize the bodies, complete their cycle and leave the environment. The presence of *Hydrotaea capensis* (Wiedemann) (Diptera: Muscidae) puparia and tineid moth cocoons showing the typical emergence holes of hymenopteran parasitoids also supported our hypothesis of a prolonged late access to the body by a rich and biodiverse community of insects. The best-preserved body was that of Francesco Ferdinando d'Avalos. The largest number of insect remains were found on it (n=814). This high difference in the number of entomological samples found near the different bodies can be linked to the different fate of the nobles. Indeed, a disastrous fire, which developed in 1506, severely damaged the church, including the mummies of the other three Aragonese nobles.

KEY WORDS: Phoridae; Muscidae; *Hydrotaea capensis*; Fanniidae; Italian Renaissance.

POSTER

Efficacy of different mosquito traps for the collection of potential European malaria vector *Anopheles daciae/messeae*

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Reliable estimates of mosquito populations can be obtained using the most suitable sampling methods, specifically designed for target species. Although abundant literature exists on adult mosquitoes catching procedures, no standardized sampling method for adult European *Anopheles* mosquitoes has yet been considered as a gold standard. This study aims at fill this gap by quantitatively comparing the efficacy of two widely used adult mosquito trapping devices. A field study was designed in 2019, in two sites of northeastern Italy: site 1 (Verona Province), in proximity to livestock premises; site 2 (Rovigo Province) located in a rural area with unreported frequentation of animals. At each study site, two traps differently baited were used, according to a 4×4 Latin square experimental design: (i) BG-Sentinel trap baited with BG-lure, (ii) BG-Sentinel trap baited with BG-lure and CO2, (iii) CDC trap baited with CO2, and (iv) CDC light trap baited with CO2 and BG-lure. Traps functioned for 24 h (total 12 days). Collected *Anopheles maculipennis* complex mosquitoes were identified morphologically and identified by species through PCR of the rRNA internal transcribed spacer 2 and sequencing (without discriminating the putative taxa *Anopheles messeae* and *Anopheles daciae*). Differences in the number of *An. messeae/daciae* captured among sites and trapping methods were tested through a negative binomial generalized linear model, accounting for data over-dispersion, and post-hoc comparisons. Overall, 1,721 *An. maculipennis* complex mosquitoes were collected: 25.4% in site 1 and 74.6% in site 2. In site 1 both *An. messeae/daciae* (n=400) and *An. maculipennis* s.s. (n=37) were captured, while in site 2 only *An. messeae/daciae* was detected. Model coefficient estimates for 'site' and 'trapping device' were statistically significant (P<0.001), which is indicative of differences in trapping performances. The post-hoc pairwise comparisons showed that the number of trapped *An. messeae/daciae* was 3.420 times smaller in site 1 compared to site 2 (P<0.0001), likely due to the presence of farmed animals attracting host-seeking mosquitoes, ultimately determining a lower abundance of trapped individuals. BG-Sentinel trap baited with BG-lure+CO2 caught 4.854 (P<0.0001) and 2.272 (P=0.022) times more *An. messeae/daciae* than BG-Sentinel trap baited with BG-lure only and CDC trap baited with CO2 only, respectively, whereas BG-Sentinel trap baited with BG-lure only had the worst performances (ratioi-ii=0.206, P<0.0001; ratioi-iii= 0.467, P=0.038; ratioi-iv=0.327, P=0.002). No significant differences were observed comparing CDC light trap with BG-lure+CO2 versus BG-Sentinel trap with lure and CO2, and versus CDC trap with CO2. The supplement of both the attractants (BG-lure and CO2) considerably improved the overall capacities of trapping devices, with higher effects reported for BG-Sentinel traps. This data could provide suggestion in trap choice in the framework of malaria vectors entomological surveillance and in the cost-benefit analysis.

KEY WORDS: *Anopheles maculipennis* complex, mosquito trap, monitoring, Malaria.

ORAL PRESENTATION

Insecticidal activity of essential oil-based nano-emulsions for sustainable control of *Musca domestica*

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Musca domestica L. is one of the most common insects, intimately associated with every human establishment regardless of the degree of anthropization. This fly, living and reproducing on decomposing organic matter, waste and food, is considered an important vector of numerous human pathogens such as bacteria, fungi, protozoa and viruses that cause diseases such as cholera, shigellosis and salmonellosis. Its control is mainly based on the use of synthetic insecticides which, beyond the recognized problems related to toxicity toward warm-blooded organisms, and therefore also in humans, induce the onset of resistance phenomena. Therefore, the development of alternative and eco-sustainable methods has become necessary. In this scenario, essential oils seem to be promising candidates for the development of innovative insecticide formulations. This study aimed to develop new essential oils (EO)-based insecticide formulations. Fennel (*Foeniculum vulgare*) and lavender (*Lavandula angustifolia*) EOs were incapsulated in nano-emulsion systems, which can guarantee a lower volatility of the active ingredients, while implementing their effectiveness. Different trials were carried out to evaluate: i) the chemical-physical parameters of the nano-emulsions developed; ii) the acute toxicity of the formulations applied as cold aerosol to adults; iii) the residual toxicity to adults and pupae; iv) the oviposition deterrence. Both the developed nano-emulsions had droplet sizes within the nanometric range (<180 nm) and a low polydispersion index. The surface charge of the droplets presented negative values due to the EO chemical composition. Laboratory trials were set up to evaluate the acute toxicity for the nano-emulsions administered as aerosol to 30 *M. domestica* adult flies inside a confined space. Acute toxicity trials were carried out using EO concentrations between 4.16 and 0.34 µL/L of air. The two developed formulations showed good insecticidal activity with LD50 values of 0.41 µL/L and 2.22 µL/L for the lavender and fennel EO-based nano emulsion, respectively. In residual toxicity trials, the adult housefly mortality showed a dose-dependent trend for both tested formulations. In contrast to acute toxicity caused by aerosol distribution, the lavender EO-based nano-emulsion was less effective (DL50 0.35 µL/cm2) than the fennel one (LD50 0.25 µL/cm2). Furthermore, both EO-based nano-emulsions produced higher mortality of males than females. Despite the promising results against adults, the residual toxicity recorded for the housefly pupae was limited or absent. Concerning ovideterrence, the lavender EO-based formulation showed limited repellence, while the fennel EO-based nano-formulation produced high repellence at higher doses, which decreased significantly, following a reciprocal curve, at lower concentrations.

KEY WORDS: Diptera, housefly, insecticide formulation, repellence.

ORAL PRESENTATION

Entomological surveillance in Abruzzo Region, 2019-2020: mosquitoes and ecoregions

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Climate and environment strongly influence the presence and distribution of the vectors responsible for the spread of animal and human diseases. In 2019 and 2020, an intense and systematic entomological surveillance was carried out in Abruzzo Region, as part of the integrated National Plan for prevention, surveillance and response to West Nile and Usutu viruses, and other research projects. The collection sites were selected so to include a climatic-environmental context with the greatest possible variability. A previous research identified 22 ecoregions across Italy, homogeneous for temperature, rain, vegetation and altitude, each one characterized by peculiar ranges of values of the above mentioned variables. In 2019, 51 collection sites were identified in 16 different ecoregions, taking into account also logistical difficulties and their accessibility; 12 of these sites (in 6 different ecoregions) were sampled also in 2020.
The field activity was carried out from June to November 2019 and from May to November 2020; the insect collections were performed weekly, using light traps baited with CO2. Overall, 1240 collections (958 in 2019 and 282 in 2020) were made and a total of 23,335 mosquitoes were morphologically identified, belonging to the 7 genera present in Italy and to 17 different species. The presence of mosquitoes was detected in all ecoregions, although with a different abundance. Among all, *Culex pipiens* s.l. resulted the most widespread and abundant species with 19,493 specimens collected in all 16 ecoregions, followed by *Aedes vexans* with 911 mosquitoes in 8 ecoregions, *Coquillettidia richiardii* with 612 specimens in 9 ecoregions, *Culex mimeticus* and *Aedes geniculatus/sticticus* with 529 and 484 mosquitoes in 7 and 8 ecoregions respectively, *Aedes albopictus* with 372 specimens in 11 ecoregions, and *Anopheles plumbeus* with 149 mosquitoes in 10 ecoregions. Other species were collected, although in low abundance (< 100 specimens): *Culiseta annulata* (in 6 ecoregions), *Culiseta longiareolata* (in 9 ecoregions), *Anopheles claviger* (in 3 ecoregions), *Anopheles maculipennis* s.l. (in 7 ecoregions), and *Aedes caspius* (in 4 ecoregions). Finally, a small number of specimens (≤10) were collected for the species *Uranotaenia unguiculata* (in 4 ecoregions), *Culex hortensis* (in 4 ecoregions), *Aedes pulcritarsis* (in 2 ecoregions), *Culex territans* (in 1 ecoregion), and *Orthopodomyia pulcripalpis* (in 1 ecoregion).,Although the catch intensity was homogeneous throughout the territory, the abundance of mosquitoes found was different, highlighting sites potentially more favorable to the proliferation of vectors and to the possible circulation of mosquito-borne arboviruses. The ecoregions associated with a greater presence of mosquitoes were those characterized by flat areas and prolonged hot season. Moreover, the different ecoregions showed a variability in terms of seasonal abundance, species composition and their relative abundance. Added value of this field activity was the possibility of finding specimens belonging to relatively uncommon species, such as *Aedes pulcritarsis*, *Culex territans* and *Orthopodomyia pulcripalpis*.

KEY WORDS: Ecoregions, mosquitoes, Abruzzo, vectors.

ORAL PRESENTATION

MosquitoAlert: a Citizen Science project to contribute to research on mosquitoes, vectors of human and zoonotic pathogens in Europe

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Mosquitoes are the animals (indirectly) responsible for the highest numbers of human deaths worldwide, due to their capacity to transmit several human and/or animal pathogens, such as malaria parasites and arboviruses. In Italy, the most widespread autochthonous species, *Culex pipiens*, is responsible for the transmission of West Nile virus to birds and of some human cases each year (606, with 49 deaths in 2018). In addition, invasive *Aedes* species are invading our country and, more in general southern and central Europe. Since 1990, *Aedes albopictus* invaded Italy, rapidly colonized the whole country and became responsible for outbreaks of exotic Dengue and Chikungunya arboviruses. More recently, other *Aedes* invasive species (*Aedes koreicus* and *Aedes japonicus*) reached the country and are spreading in northern regions. Finally, concern is raising about the risk of introduction of the major tropical arbovirus vector, *Aedes aegypti*. The national plan of the Italian Ministry of Health for the prevention, surveillance and control of arboviruses (PNA, 2020-2025) prescribes entomological surveillance at region level to prevent risk of pathogen transmission by autochthonous and invasive mosquito species. However, the high costs of entomological surveillance restrict the implementation of these activities to few areas/regions where the risk is higher or the public health system is more efficient. Under this scenario, Citizen Science offers the opportunity to complement current entomological approaches and to provide data from the whole national territory. In October 2020, the European-funded Aedes Invasive Mosquito COST Action (https://www.aedescost.eu/), coordinated by University of Rome SAPIENZA, made available all over Europe MOSQUITO ALERT application for smartphones, with the objective to involve citizen in mosquito surveillance, while educating them on mosquito biology, public health risks and preventive interventions. The task Force di MOSQUITO ALERT ITALIA, also coordinated by SAPIENZA University, benefits from the contribution of the National Institute of Health, the National Health Authority and Research Organization for Animal Health and Food Safety, the Museum of Science MUSE and the University of Bologna Alma Mater. Citizen can send 3 types of data through MOSQUITO ALERT app: 1) photos of live or dead mosquitoes, which are identified by 3 expert entomologists and can be exploited for updating the distribution of autochthonous and invasive mosquito species and for developing maps of probability of their presence; 2) photos of larval sites; 3) records of bites, which can be exploited to map and model human-mosquito contact and arbovirus transmission risk. On the short term, we expect that the large-scale use of MOSQUITO ALERT by citizen will provide relevant data for research and surveillance of mosquitoes in Italy, while on the medium/large term this tool could be exploited by public administrations to guide and optimise interventions aimed at reducing mosquito nuisance and mosquito-related public health risks.

ORAL PRESENTATION

Professional dermatitis caused by *Cephalonomia gallicola* in a herb trading company

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Cephalonomia gallicola is a Hymenoptera Bethylidae, natural parasite of the immature stages of infesting beetles belonging to the families Anobiidae, Ptinidae and Bruchidae. In Italy, *C. gallicola* is mainly found in homes associated with *Oligomerus ptilinoides* (Anobiidae), the common woodworm of furniture. Its presence generally causes highly itchy strofuloid (erythemato-edemato-vesicular) dermatitis in humans from the beginning of springtime (March-April) to the end of summertime (September-October). Although its presence is more often related to woodworm, *C. gallicola* is also a parasite of the larvae of beetles infesting preserved foodstuffs, such as, for example, *Lasioderma serricorne* and *Stegobium paniceum* (Anobiidae). Some dermatitis, therefore, although more rarely, can be related to the handling of old feed, various types of vegetables and preserved starchy foods. This is the case of the episode reported herein, which can be defined as a "professional dermatitis", arose in the workers of a company that produces raw materials for herbalists. As reported by the company, the herbs and spices that were processed had for years been relapsing infestations with *S. paniceum*, but, until then, no dermatological problems had ever been detected. Suddenly, however, in May 2018, employees began to be stung more and more frequently, until, in June, so numerous and itchy multiple stings forced them to abandon the company, with its consequent closure, waiting to understand the possible cause of their dermatitis. We were therefore asked to carry out an entomological environmental assessment through E.D.P.A.® (Indoor Dust Direct Examination) in order to reveal any presence of pathogenic arthropods inside the company premises and in the raw materials that were handled by the workers. The analysis, carried out in the Urania Research Center in Perugia, showed the presence of a large infestation with *C. gallicola*, both in the raw material processing room and inside the sacks stored in the warehouse. Furthermore, both in the environment and in all the collected herbal samples, there were larvae and adults of *S.paniceum*, a trophic substrate of the micro-hymenoptera. The removal of the infested material, the heat treatment of the herbs and spices and a thorough cleaning of the various rooms, carried out by means of a powerful vacuum cleaner, allowed us to quickly resolve the infestation, obtaining the remission of the itchy symptoms of the staff and the immediate restoration of the working activity.

KEY WORDS: *Cephalonomia gallicola*, dermatitis, herbs and spices.

POSTER

Bioinformatic analysis of antimicrobial peptides identified in the Black Soldier Fly *Hermetia illucens*

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Antineoplastic and antimicrobial resistances represent two of the most urgent therapeutic challenges worldwide underlining the need to develop novel alternative treatments within these fields. Natural products derived from insects have been used in traditional medicine for centuries and still provide a valuable supply of cures in less-developed countries. Numerous insect-derived antimicrobial peptides (AMP) have been reported in the literature. These are promising antimicrobial and anti-cancer peptides potentially lacking toxicity to healthy cells and are unaffected by common mechanisms of resistance. AMPs are small molecules ranging from 10 to 100 amino acid residues playing a key role in the innate immunity, the first line of defence against bacteria, fungi, mycobacteria and viruses. Among insects, the black soldier fly *Hermetia illucens* is probably one of the most promising sources of AMPs, since larvae thrive in hostile environments rich in microbes, including pathogenic bacteria and fungi. To identify AMPs, we generated de novo transcriptome assemblies from larval and adult (both sexes) material. Subsequent analysis and functional annotation identified 82 putative antimicrobial peptides from the combined *H. illucens* transcriptomes. All sequences putatively encoding for AMPs were bioinformatically analyzed in order to predict their antimicrobial and anticancer activity and their physico-chemical properties. AMPs were analysed through different machine learning algorithms: Support Vector Machine, Discriminant Analysis, Artificial Neural Network and Random Forest available on the CAMP database to predict their antimicrobial activity. Moreover, the iACP tool, the AVPpred and the Antifp servers were used to predict anticancer, antiviral and antifungal activities. The related physico-chemical properties were evaluated through the Antimicrobial Peptide Database Calculator and Predictor. These analyses allowed us to identify 57 putative active peptides. These preliminary results will allow the selection of the most promising candidates for heterologous expression or chemical synthesis to obtain peptides for in vitro studies on different bacterial strains and tumor cell lines.

POSTER

Experimental laboratory infection of *Culex pipiens* s.l. with Bluetongue Virus

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Bluetongue (BT) is an infectious disease of domestic and wild ruminants and occurs in severe clinical forms in sheep. The aetiological agent (Bluetongue Virus, BTV) is an RNA virus transmitted by some species of biting midges belonging to the genus *Culicoides*. In recent decades, the virus was also detected in other arthropods such as ticks and mosquitoes of the genera *Aedes*, *Anopheles* and *Toxorhynchites* but their vector competence is still not demonstrated. *Culex pipiens* s.l. is recognized as vector of important arbovirus. In Sardinia (Italy) this species is recorded throughout the year, with peaks from July to October, correlated with the onset of BT outbreaks.
The aim of the work was to test the possible role of *C. pipiens* s.l. mosquitoes in the transmission of BTV and their possible vector competence. In the experimental tests, one week-old mosquitoes of *C. pipiens* s.l., from a long established colony, were used. Sixty females were transferred to a cylindrical container, sealed at both ends with a fine mesh net, and starved for 24 hours before infection. The blood meal was performed using an artificial feeding apparatus (Hemotek). BTV Serotype 1 positive ovine blood, collected from an outbreak in 2013 and stored refrigerated in the laboratory, was used. The mosquitoes were kept in contact for 60 minutes with the plate containing infected blood. At the end of this period, 48 fully engorged females were separated and placed in a cage with a sugar solution. Mosquitoes were analyzed by Real Time PCR to evaluate the presence of BTV after 24 hours (7 females) and after 72 hours (12 females).
After 4 days, a small container with water for laying eggs was made available to the remaining females. The following day, eggs were collected and bred until the adult emergence. One week after the blood meal, the remaining 29 females were analyzed to verify the possible virus replication in the insect's body. All mosquitoes were tested individually. The experiment was carried out in a climatic chamber with a temperature of 25 °C, 60% relative humidity and a light-dark cycle of 14-10 hours. Mature larvae and newly emerged adults were also analyzed to evaluate a possible transovaric virus transmission. BTV positivity was found only in mosquitoes collected after 24 hours, in which ingested blood was clearly visible in the abdomen, and in females taken after 3 days, with eggs in formation and small traces of blood visible in the abdomen. Mosquitoes taken after 7 days and larvae and adults newly emerged were negative. These first results suggest that Culex pipiens s.l. does not appear to have vector competence for BTV transmission.

KEY WORDS: Bluetongue, *Culicoides, Culex pipiens*, vector competence.

ORAL PRESENTATION

Preliminary study on the potential role of *Musca domestica* as a mechanical vector of the African Swine Fever Virus

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African Swine Fever is a highly contagious acute hemorrhagic disease affecting swine and it is caused by a DNA virus (African Swine Fever Virus, ASFV) belonging to the Asfarviridae family, genus Asfivirus. In Sardinia (Italy) the disease has been present since 1978 with serious economic losses for the swine industry. Competent vectors of the disease are ticks belonging to the genus *Ornithodoros*, particularly in Europe *O. erraticus*, which is absent on the Island. Previous studies have highlighted the possibility that some species of flies may act as mechanical vectors of the virus. The aim of the study was to evaluate the possibility that adults and larvae of *Musca domestica*, coming into contact with the virus, could keep it viable and act as mechanical vectors. In a first trial, *M. domestica* eggs, from a long established colony, were placed on a larval pabulum mixed with an ASFV positive pig spleen; in a second test, the larval food was spiked with an ASFV viral isolate (8x106.5 TCID50/ml). Virological analyses were carried out on larvae samples, pupae, larval food and newly emerged adults. Three food samples in the first test and 4 food samples in the second test were found positive by Real Time PCR. To evaluate the potential mechanical role of *M. domestica* adults, 70 flies were kept for 24 hours in a box, closed at the top with a fine mesh net, containing a pig spleen spiked with a ASFV viral isolate (8x106.5 TCID50/ml). At the end of this period, the flies were divided into 7 groups of 10 specimens for each group and the spleen divided into 7 portions. Each group of flies was kept in contact with a portion of the spleen. For a week, a group of flies and a portion of the spleen were daily collected and analyzed. All flies were individually analyzed. Four out of 10 flies taken after 1 day, one out of 10 flies and the piece of spleen taken after 2 days, were positive by Real Time PCR. Positive samples were subjected to viral isolation on swine macrophages cultures and they tested negative. Adults of *M. domestica* resulted positive by Real Time PCR pointing out that flies could be a potential mechanical vector but the negative response to culture isolation suggest that no viral replication had occurred in the insect's body and/or that flies are able to inactivate the virus. However, other experiments should be necessary to verify the presence of infectious virus particles within the first 24 hours.

KEY WORDS: African swine fever, *Musca domestica*, Virological analyses.

POSTER

First data on the biology of *Necrodes littoralis* in Calabria

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Necrodes littoralis (Linnaeus) is a species of forensic interest associated with human corpses and large animal carcasses, in Europe. In Italy, the species was not found on remains and no data about its reproductive behavior and its role in forensic entomology were reported. Our study, carried out in Calabria from November 2017 to June 2018, describes for the first time, the presence of Necrodes littoralis on large pig carcasses. The Silphidae has been observed during the decay stage, feed on maggots of *Calliphora vomitoria* (Linnaeus) and *Hydrotaea dentipes* (Fabricius). After 52 days from the start of the investigation, larvae and adults of *N. littoralis* colonized the remains. In conclusion, we confirm the trophic plasticity of *N. littoralis*; the species is associated with Calliphoridae species and with predatory and necrophagous beetles (*Necrobia* spp., *Dermestes* spp.). This Silphidae should be considered with attention in real cases for the listed reasons: i) it can colonize carcasses quickly and for long time; ii) it can remove the pioneer insects of the entomological succession; iii) the species prefers medium-high temperatures and outdoor environments; iv) it is frequently found on large carcasses and associated with other sarco-saprophagous insects (Cleridae and Dermestidae). In the future, development data and ecological informations for this species would be useful for forensic entomologists in Italy.

KEY WORDS: Forensic entomology, Silphidae, biological cycle.

The insect *Hermetia illucens*: a sustainable source of molecules of high biological and economic value

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To date, the worldwide growing demand for protein sources for farm animals breeding can no longer be satisfied by the intensive fishing for production of high-protein fishmeal, and by the intensive use of agricultural land for protein crops (e.g. soy). Insect proteins can play an important role in the progressive substitution of soy and fishmeal proteins, commonly used to feed animals, and prospective in the progressive integration as novel food for humans. The greatest advantage of insects breeding is the lower environmental impact: less greenhouse gases emissions than any other conventional animal farming, lower water footprint per gram of produced protein and, moreover, some insect species are able to consume different types of organic waste. This is the case of the dipteran *Hermetia illucens*, a scavenger insect able to bioconvert organic waste and agrifood by-products, whose larvae, flours and transformed animal proteins can be used in aquaculture feed (Reg. EU 893/2017). The bioconversion process and the valorization of agrifood by.product, also allow to obtain secondary products of high biological and economic value, including lipids and chitin. The larvae of *H. illucens* have a lipid content about 40% of the larval biomass, but it is variable in relation to the food sub-layer. For their composition (saturated fatty acids: lauric, myristic, palmitic and stearic acid, and unsaturated: hexadecenoic and octadecenoic) the lipids deriving from *H. illucens* are used, as they are, in animal feed and properly functionalized for the production of biodiesel, as vegetable additives of plastics and for the formulation of products suitable for personal care (soaps, detergents, shampoos). Chitin, and its derivatives, due to their properties, like biodegradability, bio-compatibility, non-toxicity, adsorption, find many applications in the industrial, and biomedical field. In addition, due to their attractive biological activities (antifungal, antibacterial, antitumor, immunoadjuvant, antithrombogenic, anticholesteremic agent) and bioadhesivity, they are widely used as absorption promoters and hydrating agents, as well as for film production and wound healing. Chitin may be processed, depending on the intended application, into different conformations such as nanofibrils, powders, films, gels and capsules. Their applications include uses in a variety of areas, such as food industry, wastewater treatment, agriculture, tissue engineering, cosmetics, pharmaceutical and medical applications, paper production, and textiles. The bioconversion process of agrifood by-products mediated by *H. illucens* is a valid means for the valorization of by-products and for the sustainable production of alternative and innovative animal proteins to be used as novel feed and, in perspective, novel food, and for the production of molecules (lipids and chitin) of high economic value, with a view to circular economy.

POSTER

Insects an innovative source of chitosan: possible applications

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Chitin is one of the most abundant natural polymers; it is a polysaccharide with a molecular structure consisting of N-acetylated glucosamine and 2-amino-2-D-glucose linked by β-1,4 glycosidic bonds, Chitin is the major structural component of the arthropods exoskeleton and the cell wall of fungi and yeasts. Chitosan is the N-deacetylated derivative of chitin. Due to their properties, like biodegradability, bio-compatibility, non-toxicity, adsorption and antimicrobial activity, chitin and its main derivative, chitosan, have a great economic value and they are molecules of great interest as new functional biomaterials, finding many applications in the industrial and biomedical field. Chitin and chitosan are widely used in drug delivery systems, for wound dressing, as antibacterial coatings, in tissue engineering and for the production of edible coatings for the shelf life extension of agri-food products. Nowadays, the main commercial source of chitin and chitosan is crustaceans' exoskeleton. The extraction of chitin from insects, a promising alternative source, has received relatively little attention. Among insects, Hermetia illucens is a promising source of chitin. This insect is the subject of increasing research in the scientific world thanks to its ability to convert organic waste into larval biomass rich in proteins and lipids of high biological value. H. illucens is finding application for the production of animal feed, mainly for aquaculture as a more sustainable alternative to fishmeal according to the regulation of the European Commission N. 893/2017.
Currently, the chitin contained in the exoskeleton of last instar larvae, pupae and adult flies constitutes a by-product of the bioconversion process aimed at the production of animal feed. Few papers are present in literature concerning chitin and chitosan extraction from insects. All the available protocols for chitin and chitosan extraction from insectsinvolve the use of chemicals, mainly acids (e.g. HCl, Acetic acid) for minerals removal, bases (e.g. NaOH, KOH) for both proteins hydrolyzation and chitin deacetylation, and various reagents (e.g. potassium permanganate and oxalic acid, hydrogen peroxide, acetone) for depigmentation. The present study focuses on chitin extraction, and its deacetylation into chitosan, from the exoskeleton of H. illucens at different developmental stages, in order to make the most of a resource not yet fully exploited. The obtained product will be investigated for different possible applications.

ORAL PRESENTATION

Finding of *Anopheles labranchiae* in the Caffarella valley in Rome

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Mosquitoes are important vectors in the transmission of parasites and other pathogens worldwide. In metropolitan areas, vegetation tends to be heterogeneously distributed as fragmented "green islands" within the urban landscape. Most of these green areas are urban parks designed to provide the inhabitants a place where to spend leisure time and practice physical activities. Since studies on mosquitoes in a wide metropolitan area such as Rome, are fragmentary and often obsolete, an entomological investigation was undertaken in the Caffarella Valley, within the Appia Antica Natural Park, aimed to contribute to the knowledge of the mosquito composition and distribution in the city, assessing the mosquito fauna in a natural park, with particular attention to the genus *Anopheles*.
Anopheles mosquito collections were mostly performed in the Vaccareccia farm, by using both CDC light traps and BG Sentinel®, setted up outside the sheep shelter and operating for one day, approximately from dusk to dawn, every two weeks. Larval catches were carried out near the farm and in other areas of the park. During the study period from April 2012 to April 2013, 1173 mosquitoes, in Caffarella Valley, belonging to 9 species were collected: *Culex pipiens* Linnaeus, *Anopheles maculipennis sensu lato*, *Anopheles claviger* (Meigen), *Culiseta annulata* (Schrank), *Culiseta longiareolata* (Macquart), *Aedes albopictus* (Skuse), *Culex territans* Walker, *Culex hortensis* Ficalbi and *Anopheles plumbeus* Stephens, whose preliminary results were the subject of a previous publication. Among the specimens collected, *Anopheles maculipennis* complex larvae were mainly found in an animal drinking trough very close to the Vaccareccia farm. In total, during the study period, 207 *Anopheles maculipennis sl* (204 larvae and 3 adults) were collected. Larvae were reared until adulthood and adults were first morphologically identified as *An. maculipennis sl.* Molecular analysis was carried out on a sample of these specimens (10% of the total), using ITS-2 of ribosomal RNA as a marker, which led to the identification of *Anopheles maculipennis sensu stricto* Meigen (35%) and *Anopheles labranchiae* Falleroni (65%). Almost eight years later, in October 2020, in the Vaccareccia farm, a further sampling of *An. maculipennis sl* larvae was carried out, confirming by molecular identification, the presence of *An. labranchiae* in this area. *An. labranchiae*, was the main vector of malaria in the Agro Romano area and its last record in the urban area of Rome dates back to 1938. In the years following the Second World War, both the Agro Romano and the Agro Pontino were the subject of an anti-malarial campaign, with a massive use of DDT, which led to the drastic reduction and perhaps the disappearance of the *Anopheles* fauna.
Our findings represent the first finding of *An. labranchiae* since its apparent disappearance and the confirmation of its discovery does not exclude a stable presence of this species over time, with very rarefied larval foci on the territory. These important results lead us to deepen the knowledge on the biology and distribution of this species, formerly vector of malaria in Italy.

KEY WORDS: Culicidae, mosquitoes, *Anopheles labranchiae*, malaria.

POSTER

Tracking positive selection of an emerging SAP2-mediated insecticide resistance mechanism in Anopheles malaria vectors

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Malaria, an *Anopheles*-transmitted *Plasmodium* disease, still inflicts substantial morbidity and mortality around the globe. In 2019, 409.000 deaths and 229 million new cases occurred (WHO, World Malaria Report, 2020). Malaria incidence and mortality have recently decreased thanks to the implementation of integrated control measures, including Long-Lasting Insecticidal Nets (LLINs), the primary strategy in vector control. LLINs act as a barrier preventing human-vector contact and associates the individual mechanical protective effect of the bednet with the lethal outcome of the impregnated insecticide. Yet, most of sub-Saharan Africa continues to carry a disproportionately high share of the global burden of malaria deaths (WHO, World Malaria Report, 2020). The effectiveness of LLIN interventions is hindered by the spreading of different forms of insecticide resistance developed by malaria vectors against pyrethroid active ingredients. The most common physiological resistance to pyrethroids - defined as `knock down resistance (kdr) - is due to several mutations of the gene encoding the sodium channel that constitutes the target site of the insecticide (Martinez-Torres et al. 1998, Ins Mol Biol, 7: 179-184). However, novel mechanisms are emerging, among which the over-expression of a sensory appendage protein (SAP2) enriched in the legs which confers high pyrethroid resistance to *Anopheles gambiae* (Ingham et al. 2020, Nature, 577: 376-380). A selective sweep near the SAP2 locus in the mosquito populations of three West African countries (Cameroon, Guinea and Burkina Faso) was also observed and a steady increase in haplotype-associated single-nucleotide polymorphisms mirroring the increasing resistance to pyrethroids was reported in *Anopheles coluzzii* in South-West Burkina Faso (Ingham et al. 2020, Nature, 577: 376-380). We aimed to analyze *A. coluzzii* specimens gathered from entomological surveys in two (40 km distant) villages of Central Burkina Faso, Goden and Koubri where the protective performance of LLINs is lower than expected, despite the mass distribution occurred in 2010 (Pombi et al. 2018, Sci Rep, 8: 12806). Mosquitoes from temporal series obtained from these two villages from 1 to 9 years after LLINs implementation were genotyped both for SAP2 resistance-derived SNPs (Ingham et al. 2020, Nature, 577: 376-380) and 1014F kdr mutation, i.e. kdr-west or kdr-w (Martinez-Torres, 1998, Insect Mol Biol, 7: 179-184). We observed an increase of SAP2-related resistance alleles in both village across time which closely mirrors previous results (Ingham et al. 2020, Nature, 577: 376-380). Interestingly, we also observed in Goden a concurrent decrease of the kdr-w allele. Our data are consistent with the hypothesis that a SAP2 resistance mechanism could have rapidly swept across anopheline populations in Burkina Faso and it may gradually replace kdr resistance. In fact, due to SAP2 specific expression in mosquito legs, this chemosensory protein might be able to sequester the insecticide directly, thus preventing the pyrethroid to enter the nervous system where the kdr resistance operates. Further data acquired across space and time in Sub-Saharan Africa would contribute to test this hypothesis, track the spread of the SAP2-based resistance mechanism, to ultimately tackle malaria transmission.

KEY WORDS: malaria, insecticide resistance, *Anopheles*, SAP2, positive selection.

ORAL PRESENTATION

Alphitobius diaperinus: the potential role in the transmission of human pathogenic bacteria

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Historically, *Alphitobius diaperinus* has been a widespread, it is a scavenger, mould feeders and minor pest of a wide range of cereals and cereal products, especially if damp. Hagstrum et al. (2013) reported about 90 different commodities that have been found to be infested by *A. diaperinus*, including vegetative products (cottonseed cake, linseed, soybean and tobacco) and animal (fishmeal, animal bones, casein and leather). It will also feed on dried carcasses of animals (mice, birds etc.). *A. diaperinus* is considered as a major problem worldwide in commercial poultry farms, where it reproduces within the litter floor, feeding on poultry manure, spilled feed and other organic material and dead birds. In high populations, its presence in poultry facilities may lead to considerable economic losses due to the reduced feed consumption and the subsequent poor body-weight gain of poultry that prefers to feed on the beetles than on the feed. Moreover, it causes an extensive damage to insulating materials (polystyrene and polyurethane) of the rearing premises. The insect can host and potentially transmit several microrganisms, including human pathogenic bacteria such as *Escherichia coli*, *Salmonella* spp., *Campylobacter* spp., and *Staphylococcus aureus*. Few studies concerning the role of *A. diaperinus* in the transmission of human pathogens have been carried out. Indeed, this is the first study conducted in Italy to evaluate the carriage of *E. coli*, *Salmonella* spp., *Campylobacter* spp., and *S. aureus* in adult *A. diaperinus* in a poultry farm, by detecting their presence in the external surface of body, in faecal and internal content of the beetles. The detection of these pathogens was also performed for broilers, and administered feed and water. During July and September, three samplings of adult *A. diaperinus* were performed in a poultry farm located in Abruzzo region; each sampling aimed at the collection of 90 beetles, 10 rectal swabs from broilers, one sample each of administered feed and water. Detection of microorganisms was firstly carried out through conventional microbial culture, using pathogens specific and selective growth media. The suspected positive colonies were than tested with biochemical analyses. Results were further confirmed through PCR assays on DNA extracts, using specie-specific genes. Our results allow to support the available evidences on the role of A. diaperinus in the spread within the farm environment of bacteria as *Salmonella* spp. and *E. coli*, which are responsible of human foodborne diseases. Conversely, *S. aureus* was not detected, while other *Staphylococcus* species never reported before in these beetles were found. The study findings do not allow confirming the role of the insect in the spread of *Campylobacter* spp., but additional analyses are needed to better elucidate the potential function of *A. diaperinus* in the transmission of pathogenic bacteria (Tamburro et al., 2019). Recently, *A. diaperinus* attracted a lot attention as it was listed in the EU Regulation 2017/893 among the insect species that are allowed to be used for the production of insect meal as aquafeed ingredient, changing our perspective on this insect from a noxious pest to a potential nutrient source.

KEY WORDS: *Alphitobius diaperinus*, transmission, pathogenic bacteria.

ORAL PRESENTATION

Characterization and identification of puparia from forensic contexts

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The availability of clear and accessible identification keys for all the insect developmental stages is of fundamental importance in all the disciplines that use insects to deduce information and reconstruct past events. Diptera puparia, because of the very resistant physical and chemical composition of their cuticle, represent a large fraction of the entomofauna associated with a cadaver, especially in advance decomposition, skeletonization and in mummified conditions. Puparia identification is still a problematic topic due to the lack of identification keys and in several cases lack in the description of the diagnostic features. Furthermore, DNA extraction is extremely difficult for samples badly preserved. Despite some larval characters are maintained on the puparium others, because the harnesses process, differ in the two stages and others, like the oral sclerites, are not always found among the empty puparia remains. A better description of the puparia and their diagnostic features is the c*onditio sine qua non* for their identification. Puparia of some Calliphoridae, Muscidae, Heleomizidae, Milichidae, Spheroceridae and Piophilidae of forensic interest are here detailed and illustrated. Posterior spiracles, anal plate and intersegmental spines have been considered as good diagnostic characters for the identification of these puparia. This work will provide a general overview of the approach that has to be used from the collection to the microscopy for a correct puparia identification from forensic and archaeological contexts.

KEY WORDS: forensic entomology, puparium, Diptera, identification.

ORAL PRESENTATION

Nosocomial myiases in South Europa: 2018-19 cases

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Myiases are parasitic infestations caused by dipteran larvae that develop on tissues, excreta and secretions of vertebrates, human included. Myiasis classification is based on: 1) relation between host and parasite, 2) happening circumstance and 3) body region parasited. Particular relevant for many reasons, sanitary and legal, are the nosocomial myiasis that occur on hospitalized people. Twelve cases, occurred in the period 2018-2019 in public and private hospitals and care centres in Italy and South France have been analysed. In 11 cases larvae were present on previous skin lesions: 8 cases of necrotic lesions due to alteration of the microcirculation and 3 cases of post-operation or post-traumatic lesions with necrotic tissues. One case was related to an infestation of the ear canal on a neoplastic lesion. *Sarcophaga* s.l., (Diptera: Sarcophagidae) was found in 7 cases, whereas *Lucilia sericata* (Diptera: Calliphoridae) in the other 4 cases. All the cases were reported in the period June-September.
The risk factors associated with nosocomial myiases can be summarised as: 1) season (June-September), 2) presence of necrotic tissues and 3) reduced motility of the patients, in some cases associated with neurological problems. Hospitals and care centres play a key role in the prevention of this kind of parasitosis. It is worth mention, in in fact that all the myiases here reported were associated with issues related with the patient assistance but not with poor hygienic conditions of the hospitals. Patients with risks factors need to be admitted in hospital areas with physical barriers able to prevent the insects' arrival (mosquito net on the windows o air conditioned room, UVA lamps, etc.). Inside the hospital the use of insecticides, especially pyrethroids has to be discouraged because, due to the need of repeated applications, could have negative effects as allergy or respiratory reactions on the patients.

KEY WORDS: Diptera, hospital, Calliphoridae, Sarcophagidae, myiasis.

POSTER

Role and importance of the genus *Hydrotaea* in the colonization of bodies from forensic and archaeological contexts

Stefano Vanin[1], Giovanni Benelli[2], Katiuscia Bisogni[3], Valentina Bugelli[4], Cristina Cattaneo[5], Martina Focardi, Antonio Fornaciari[7], Ezio Fulcheri, Mirella Gherardi, Giorgia Giordani[6], Augusto Loni[7], Jennifer Pradelli[8], Chiara Rossetti, Eugenia Tognotti, Fabiola Tuccia, Elvira Ventura Spagnolo

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The insect colonization of a cadaver depends on several factors that are related to the body and to the environment. These factors affect not only the time of colonization but as well the composition and the structure of the entomofauna colonizing the body. It is worth mentioning that the body accessibility, especially suddenly after the death, plays a fundamental role on the structure of the insect community, being the first colonizers in the family Calliphoridae not able to reach concealed or buried bodies. In contrast, in these conditions, species belonging to the families Phoridae and Muscidae are the dominant taxa able to colonize and degrade a body. Among Muscidae, the species of the genus *Hydrotaea* Robineau-Desvoidy, 1830 can colonize exposed bodies in advanced decomposition but as well bodies concealed in coffins, sarcophagi, crypts and other enclosed environments. In Italy, at present, 23 species of this genus are reported. Five of them [*H. aenescens* (Wiedemann), *H. capensis* (Wiedemann), *H. ignava* (Harris), *H. dentipes* (Fabricius) e *H. armipes* (Fallen)], have been collected from decomposed, skeletonized and as well mummified bodies. The most common species in Italy, but not only, is *H. capensis*. Hovewer, we cannot exclude misidentification that occurred in the past when immature stages have been identified. Indeed, the presence of this species, in skeletonized and mummified bodies is confirmed by the abundant presence of puparia. *Hydrotaea capensis* has been found on bodies preserved in coffins, crypts and putridaria in the whole national continental and insular territory. Its presence, in absence of Calliphoridae, is a clear evidence of a burial performed suddenly after the death or in the winter season, when blowflies are not active. It is also worth of reporting that this species has been collected also from cadavers initially treated with volcanic sands. Very interesting is the record from cadavers of *H. aenescens* that has been introduced to Europe in the '60s. Less common is the presence of *H. ignava*, *H. dentipes* e *H. armipes* that when present in association with *H. capensis* are in small number. In this work, we report the list of Muscidae of the genus *Hydrotaea* from forensic and archaeological contexts from the following Italian regions Veneto, Lombardy, Piedmont, Valle d'Aosta, Tuscany, Emilia-Romagna, Marche, Basilicata, Calabria, Sicily and Sardinia.

KEY WORDS: Muscidae, Hydrotaea capensis, forensic entomology, funerary archaeoentomology.

POSTER

SESSION IX

Laboratory rearing of *Xylosandrus compactus* on *Ambrosiella grosmanniae*

Claudia Benvenuti, Gian Paolo Barzanti, Franca Tarchi, Riccardo Frosinini, Francesco Binazzi, Fabrizio Pennacchio, Valeria Francardi

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Xylosandrus compactus (Curculioinidae: Scolitinae) is an invasive and polyphagous bark beetle, native to East Asia, that damages several plant species, all relevant from an ornamental, scenic and economic standpoint. In Italy, it has been recorded mainly on *Laurus nobilis*, *Magnolia grandiflora* and *Ceratonia siliqua*. Dieback of host plants is caused primarily by the digging activity of adult females and by the rapid development of pathogenic fungi associated with them, which may further affect the already compromised plant health status. In the context of the project "Contributi per il controllo delle emergenze fitosanitarie determinate da *Xylosandrus compactus*, *Xylella fastidiosa*, *Botrytis cinerea* - CO.XI.BO'' funded by MiPAAFT, the survival of artificially reared X. compactus specimens was evaluated in laboratory conditions, in order to maintain populations suitable for future behavioural studies. Though *X. compactus* is an ambrosia beetle feeding mainly on *Ambrosiella xylebori* (Ceratocystidaceae, Microascales), in our trials, the congeneric species Ambrosiella grosmanniae was used as an alternative rearing substrate. This fungus had been originally obtained in June 2019 from galleries of *Xylosandrus germanus* colonizing apple trees. It was then isolated, molecularly identified and maintained in pure cultures at the CREA-DC laboratories (Firenze). Conversely, all *X. compactus* adults used in the trials were directly sampled from infested laurel twigs previously collected in the Villa Salviati Park (Migliarino, Pisa) in the Autumn 2019. In the laboratory, adults were placed into Petri dishes containing an agarized medium (Agar-Water) and short laurel twigs previously sterilized and inoculated with *A. grosmanniae* developed on PDA. Petri dishes were then maintained at room temperature (20-24°C). Adults were active for more than 20 days within the twigs and a 100% survival was recorded at the end of the trials. The study evidenced that, at our rearing conditions, the adults of *X. compactus*, are able to resume the digging activity remaining alive and active for a long period of time. With this method of maintenance we were able to provide homogeneous batches of adults as required by the project to carry out the planned tests, the plate period was a sort of quarantine that allowed the screening of active and healthy individuals. Further studies will need to be conducted to improve the technique described above.

KEY WORDS: bark beetles, *Laurus nobilis*, symbiotic fungi, ambrosia beetles.

ORAL PRESENTATION

Effect of trap color on captures of native and exotic longhorn beetles

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Longhorn beetles (Coleoptera: Cerambycidae) are among the most significant groups of invasive forest insects worldwide. Hidden within live plants and wood-packaging materials, these beetles can escape routine inspections at entry points and border crossings and some become major pests in the invaded environment. Traps baited with blends of pheromones and kairomones and set up in and around entry points are commonly used to improve chances of intercepting exotic species soon after their arrival. Factors able to affect longhorn beetle catches in traps have been extensively studied in the last years. Nonetheless, the effect of trap color is still largely overlooked. Currently, black traps are commonly used in almost all monitoring programs worldwide. In a trapping study carried out in 2019 in 16 forest sites located in northern Italy, we investigated the response of longhorn beetles to eight different colors (black, yellow, green, red, purple, grey, brown and blue). All traps were baited with the same pheromone blends and ethanol; 16 traps per color were tested. In total, 6,001 individuals were trapped, belonging to 56 species, 54 native and two exotic (*Xylotrechus stebbingi* and *Neoclytus acuminatus*). At the family level, longhorn beetle species richness and total abundance were significantly affected by trap color; in particular, yellow traps allowed to catch a significantly greater number of species than black traps. At the subfamily level, a mixed response was found: Lepturinae exhibited a significant preference for yellow, while Laminae and Prioninae for black and brown traps. In addition, no clear pattern was found for Cerambycinae. At the species level, flowers-visiting species exhibited a clear preference for yellow, green and blue whereas black and brown were preferentially chosen by species non-visiting flowers. Color preferences of different longhorn beetle species are likely linked to either their adult food habits or their sexual behavior. In general, these results indicate that trap color can strongly affect longhorn beetle catches and suggest that trap of different colors should be used in surveillance programs carried out at entry points.

KEY WORDS: insect vision, longhorn beetles, trap, colors, survey and detection.

ORAL PRESENTATION

Use of pheromones in the management of pine processionary moths, *Thaumetopoea* spp., in urban parks and woodland recreational areas

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This paper describes recent advances obtained from 2015 to 2018 on the use of pheromones [(Z)-13-esadecen-11-yn-1-yl acetate] (pityolure), in monitoring, mass-trapping, and mating-disruption related to management of *Thaumetopoea pityocampa* (Den. & Schiff.) (in Spain and Italy) and *T. hellenica* Trematerra & Scalercio, in Greece in urban parks and woodland recreational areas (Trematerra e Colacci, 2019). To optimize the monitoring of adults of *Thaumetopoea* spp., 10 different models of funnel traps (some commercial, some experimental), all baited with 1 mg of pityolure (Kenogard, Barcelona, Spain), were compared. Statistically better results were obtained from an experimental prototype called "modified G-trap" (Athanassiou et al., 2017). The mass-trapping test was carried out in Molise (central Italy) in a coastal touristic pinewood forest, mainly composed of *Pinus halepensis* and *P. pinea*, by comparing two 1 ha plots: in the mass-trapping plot, 10 G-trap traps (SEDQ, Barcelona, Spain) were placed, in the control plot there were 2 traps of the same model. All traps were baited with 1 mg of pityolure. The inspection of winter nests and the monitoring of adults verified that, after two years of mass-trapping, the presence of *T. pityocampa* in the mass-trapping plot was significantly reduced in comparison with the control plot (Trematerra et al., 2019a). The mating-disruption test was carried out in Molise and in two sites of Greece (composed of *Pinus brutia* and *P. halepensis*) with the aim of verifying the effectiveness of the synthetic pheromone pityolure. The variation of the adult population and number of nests present in two 1 ha plots (mating-disruption plot and control plot) were compared. The mating-disruption plot was treated with 20 5 g of pityolure distributed in drops (Novagrica, Athens, Greece) on the upper part of the plants, the control plot was not treated. After two years, the presence of adults and winter nests of the moth in the mating-disruption plots was reduced in a statistically different way compared to the control plots (Trematerra et al., 2019b). In the context of IPM, according to the results obtained in our experimentation, mass-trapping and mating-disruption can be adopted in combination with preventive techniques and other control methods. Moreover, the application of pheromones against Thaumetopoea spp. moths could be justifiable in urban and suburban parks, private gardens, and recreational areas in terms of economic viability and ease of application. This is particularly important while considering the risks to public health and domestic animals.

KEY WORDS: *Thaumetopoea*, pheromones, monitoring, mass-trapping, mating-disruption.

Analysis of the pedofauna of the "Bosco di Formigosa", Mantua – Comparison with neighboringareas

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During two-year period 2018-2019, the soil fauna of three neighboring Mantuan sites with similar pedological characteristics, was analysed. Through the application of the QBS-ar index, the edaphic communities of the newly forested area "Bosco di Formigosa" - realized in 2011 - were compared with those of the "Vallazza" Site of Community Importance/Special Protection Area (SIC/SPA), and with those of an agriculture-vocated land in order to assess the biological soil quality and hence the soil microarthropod biodiversity in differentially managed areas. This is possible because the arthropods that live in the soil are considered bioindicators and respond rapidly to anthropogenic disturbance (e.g. soil tillage). The preliminary data were presented in Autumn 2018 at the World Forum on Urban Forests (FAO), held in Mantua from November 28th to December 1st. So far, the data collected show that the QBS-ar value of the "Bosco di Formigosa" site falls in between of the arable land and the "Vallazza" SIC/SPA, as expected.

Guidelines for the management of the pine processionary moth in the central-western Alps: the Interreg MONGEFITOFOR project

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The forests of the Alpine valleys have a strong multifunctional value and, in addition to producing timber, they guarantee a series of positive externalities, including the maintenance of biodiversity, the hydrogeological protection of the slopes and the maintenance of the landscape. In 2019 the project "Linee guida per il monitoraggio e la gestione delle emergenze fitosanitarie nelle foreste delle Alpi centro-occidentali (MONGEFITOFOR)" was funded, within the INTERREG V - A Italy Switzerland 2014-2020 cooperation program to address the main phytosanitary emergencies of the forests of the central-western Alps. The partnership consists of the Aosta Valley (RAVA) as leader partner, the Department of Agricultural, Forest and Food Sciences (DISAFA) of the University of Turin, the Swiss Federal Institute for Forest, Snow and Landscape Research (WSL) of Zurich, the Canton of Grisons (GR) and the Canton Ticino (TI) as partners. The three-year project (2019-2022), taking advantage of cross-border skills, aims to deal with some of the main phytosanitary emergencies occurred in recent years in the forests of the central-western Alps: the pine processionary moth, the ash disease, and some diseases affecting chestnut as the chestnut blight and the ink disease. In addition to defining standards for monitoring, the project includes applied research to set up useful guidelines for the management of these phytosanitary emergencies. In detail, the activities related to the pine processionary moth, *Thaumaetopoea pityocampa* (Denis et Schiffermüller), will be presented. Defoliation by the caterpillars of this moth seriously affects host plants in relation to the severity of the damage, and urticating hairs may threaten people and animals, especially in urban areas and woods. The host plants are potentially all those belonging to the genus *Pinus*, with a marked preference for black and scots pines. In consideration of the seriousness of the impact of this pest, the major aims of the project are I) to map the current distribution area and the potential risk of infestation in the cross-border territory; II) to understand the association between infestations and biotic and abiotic factors, with particular reference to climate; III) to define the most effective control strategies. The activity, coordinated by the Entomology Unit of the DISAFA, with the participation of all partners, involves the extension of the phytosanitary monitoring networks, already implemented by RAVA, to the Swiss territory. RAVA and DISAFA researchers have in fact gained important skills with particular regard to the definition of flight periods and migration areas of the pine processionary moth. These previous experiences will be adopted at WSL, GR and TI with the aim of investigating the intensity of the outbreaks in forestry and in urban areas. The expansion of the distribution and host plant range (*Pinus*, *Cedrus*, *Larix*) will also be assessed, in relation to climatic parameters.

KEY WORDS: Pine processionary moth, distribution, control strategies, climate change.

POSTER

The box tree moth: a new emergency in the rocky habitats of Piedmont

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The box tree moth, *Cydalima perspectalis* (Walker) (Lepidoptera: Crambidae), represents one of the recent examples of exotic insect pest native to Asia accidentally introduced into Europe with the nursery trade. After being reported in Germany in 2007, this species quickly spread to Italy in 2010.
C. perspectalis is a multivoltine species, affecting plants of the genus *Buxus*. In its native range, the species feeds mainly on the Japanese box tree (*Buxus microphylla*), while in Europe it develops mainly on *B. sempervirens*.
The box tree is an ornamental species typically used for hedges, borders, and plant sculptures. It is present in the typical Italian gardens, and it is also found in beech wood, while on alpine rocky slopes it forms the rocky habitat "Habitat 5110 - Stable xerothermophilous formations with *Buxus sempervirens* on rock slopes (*Berberidion* p.p.)" according to EU 92/43 Habitats Directive.
Although in the literature some natural enemies are reported for Europe, parasitism rates are still too low to control this exotic pest, and the use of the bacterium *Bacillus thuringiensis* var. *kurstaki* (*Btk*) or var. *aizawai* often remains the only way to carry out an effective and environmentally sustainable control.
However, the *Btk* treatment is not specific for the control of the box tree moth and, if the product is deposited on the feeding plants, it causes the death of the larval stage of other moths.
In this work, in addition to the study of the biology and ecology of *C. perspectalis*, the potential effect of *Btk* treatments on non-target moth species was also investigated. Surveys were carried out in the Vermenagna valley, in the "Comba di Castelmagno" Site of Community Importance (SCI) (IT1160065) in the Grana valley and near the "Monte Antoroto" SCI (IT1160035) in the Tanaro valley.
Investigations were performed to evaluate: (i) the seasonal flight activity with sex pheromone sticky traps, (ii) the effectiveness of microbiological treatments with *Btk* performed in the presence of *C. perspectalis* newborn larvae, iii) the butterfly community through semi-quantitative transects in the areas subjected to microbiological treatment and in control areas.
The results obtained showed the presence of two generations a year and a good effectiveness of the *Btk* treatment, both in open field and laboratory conditions. On the other hand, no significant short-term effects were detected either on the abundance of individuals or on the species richness of non-target butterfly communities. However, this work highlights the need for a rigorous protocol for the application and surveillance of the effects on non-target organisms to be applied in every natural context where the use of *Btk* may help to control the pest. In particular, it is necessary to verify the presence of protected or endangered species of moths, to know the phenology of their larval development and the location of the feeding plants, in order to evaluate the feasibility of the microbiological treatments. Complex coenoses with an abundance of predators must also be encouraged, i.e. insectivorous birds and bats.

KEY WORDS: *Cydalima perspectalis*, exotic invasive pest, *Buxus* spp., rocky habitat.

ORAL PRESENTATION

Establishment of a *Eucalyptus* sentinel garden in Sardinia (Italy)

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The genus *Eucalyptus* includes a large number of species of great importance in forestry, ornamental and industrial fields, which are cultivated globally on about 20 million hectares. *Eucalyptus* trees were introduced in Italy at the end of the 18th century, in the gardens of the Reggia di Caserta, and have not been threatened by pests for about two centuries. The reason is to be found in the trade of the plants, which took place mainly as seed. The evolution of global trade has led to the introduction initially of xylophagous insects (wood borers), as the first eucalypt pest reported in Italy was in fact *Phoracanta semipunctata* (F.) followed by *Gonipterus scutellatus* Gyllenhal. More recently, foliage feeders, such as gall wasps (*Ophelimus maskelli* (Ashmead) and *Leptocybe invasa* Fisher & La Salle), psyllids (e.g. *Blastopsylla occidentalis* Taylor and *Glycaspis brimblecombei* Moore) finally the bronze bug, *Thaumastocoris peregrinus* Carpintero & Dellapé, have been reported in Europe. The rate of increase of new alien insects on *Eucalyptus* is one of the highest ever. The sentinel garden represents an expanding strategy to early warn or identify plant pests in a given region before they spread. This technique makes it possible to monitor insects or pathogens that may develop on a particular plant species and also pose a risk to other species of significant economic interest. It also makes it possible to highlight new plant-pests associations, provide a more realistic overview of potential risks, improve pest risk analysis procedures by reporting little-known invasive organisms, and help to increase knowledge about the biology of poorly studied phytophagous. During 2019, the seeds from the 50 most common species in the 21 Mediterranean countries were acquired from the Commonwealth Scientific and Industrial Research Organisation - Australian Tree Seed Center. The seeds were sown at the Fo.Re.S.T.A.S. regional agency nursery in Campulongu (39°57'50,5 "N 8°36'12,6 "E) and will be transplanted in an adjacent area of 4 hectares for the construction of the sentinel garden, which adhered to the International Plant Sentinel Network, a network of botanical gardens, arboreta, plant protection organizations and researchers.

KEY WORDS: invasive alien species, monitoring, risk assessment.

POSTER

Potential of biopesticides for controlling the ambrosia beetle *Xylosandrus compactus* and its main symbiont *Ambrosiella xylebori*

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Ambrosia beetles (Coleoptera, Curculionidae, Scolytinae) are among the most representative examples of complex insect-fungus mutualism. They are also known as successful widespread invaders and colonizers causing severe damage to woody plants. Several non-native ambrosia beetle species are establishing and spreading in Europe due to global trade and climate change. Among these fungus farming insects, *Xylosandrus compactus* (Eichhoff), also known as the black twig borer, represents a serious pest for various Mediterranean trees and shrubs. The *X. compactus* cryptic habits and its broad host range negatively affect the efficacy of conventional control tools and, in this context, biopesticides could represent sustainable alternatives. We tested, in laboratory conditions, the efficacy in controlling this pest of seven biofungicides (five *Trichoderma* and two *Bacillus*-based commercial products) and of four bioinsecticides (azadirachtin, pyrethrin, spinosad and a *Beauveria bassiana* (Bals.-Criv.) commercial strain). The mortality and/or the suppression of the beetle symbiont and consequently the brood size (number of offspring per foundress) were evaluated as a proxy of pest control efficacy. Preliminary *in vitro* assays were conducted in order to evaluate the antagonistic activity of the tested *Bacillus* spp. and *Trichoderma* spp. toward the insect symbiont *Ambrosiella xylebori*. Beetle bioassays were performed by exposing carob twigs previously treated with both bioinsecticides and biofungicides. The impact in terms of gallery formation, symbiont growth and brood production by foundresses were thus evaluated. Results demonstrate that the tested *Trichoderma* and *Bacillus*-based products can drastically reduce the beetle progeny by outcompeting the symbiont. Moreover, our results highlight the ability of the entomopathogenic fungus *B. bassiana* to kill the foundresses and reduce the *X. compactus* progeny. Similarly, the azadirachtin, pyrethrin and spinosad strongly affected the beetle survival and/or the offspring number. Overall, the obtained results (i) stress the importance of studying sublethal effects in comprehensive toxicity assessments of biopesticides and (ii) provide new data useful for building sustainable organic or integrated management strategies against this invasive ambrosia beetle.

KEY WORDS: biological control, bioinsecticides, biofungicides, black twig borer, invasive pest, symbiosis.

Current status of the eradication program against *Thaumetopoea pityocampa* in Sardinia (Italy)

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The pine processionary moth, *Thaumetopoea pityocampa* (Denis & Schiffermüller), is widely distributed throughout the Mediterranean Basin. This species was first reported in Sardinia (Italy), in the municipality of Sanluri (Central-Southern Sardinia), in 2006. Thereafter, a second infested area was found in 2013 in the municipality of Arzachena (North-Eastern Sardinia) in 2013. Due to health problems related to *T. pityocampa* larval infestations, the Region of Sardinia funded a program to eradicate this insect from the Island that was coordinated by the Department of Agricultural Sciences of the University of Sassari and carried out in cooperation with different regional authorities (Corpo Forestale e di Vigilanza Ambientale, Agenzia Fo.Re.S.T.A.S., Province administrations). The eradication program started in 2011 and included the following actions: 1) studying insect phenology in the invasion area (adult flight and pupation periods); 2) accurately determining the spread of the pest (arrangement of a network of pheromone traps throughout the Island); 3) mapping isolated pine trees and pine forests within the infested area; 4) detecting infested pine trees and forests by visual inspection; 5) applying mechanical (cutting and burning of nests), biotechnological (mass trapping of male adults), microbiological control (aerial application of *Bacillus thuringiensis kurstaki* on pine forests), and chemical control (spraying of piretroids on isolated pine trees). The eradication program was quite effective even though it was not possible to eliminate all infestation foci, due to lack of application of some intervention actions. In Central-Southern Sardinia, interventions eradicated the pine processionary as no male adults were caught by the pheromone traps from 2017 to 2019. In the North-Eastern Sardinia, the population gradually decreased, with a significant reduction in adult males captured in the traps from 760 in 2013 to 30 in 2019. We believe that the remaining T*. pityocampa* populations were located in unmapped trees. For this reason, a further inspection within the diffusion area is required to map and treat all infested pines. Based on the fact that the diffusion of the pine processionary moth is currently limited to approximately 400 hectares, if the monitoring of adult males in 2020 indicate the persistence of infestation, the application of *B. thuringiensis*-based insecticides on the entire area at risk of spreading will be considered.

KEY WORDS: pine processionary moth, eradication program, alien species.

POSTER

Short- and long-term effectiveness of *Bacillus thuringiensis* applications against *Lymantria dispar* in cork oak forests of Sardinia

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The gypsy moth, *Lymantria dispar*, is one of the most harmful defoliators worldwide and represents a major threat to cork oak forests in the Mediterranean area. Gypsy moth population density fluctuates periodically over the years, and this pest can defoliate over thousand hectares during its periodic outbreaks. Defoliation caused by gypsy moth negatively affects the health status of oaks, thus reducing plant growth and cork production. In Sardinia (Italy), the control of gypsy moth populations by aerial application of *Bacillus thuringiensis* subsp. *kurstaki* (Btk) formulations have been performed in cork oak forests since 2001. Data coming from a 20-year monitoring of egg mass density allowed to better understand the short- and long-term effects of Btk applications on gypsy moth population. The annual larval reduction in Btk-sprayed areas was estimated and the temporal and spatial fluctuation patterns of *L. dispar* egg density in cork oak forests treated with Btk were analyzed. Btk was effective in protecting oak foliage in the year of its application, determining an average annual larval reduction up to approximately 90%. Differences in long-term effectiveness of Btk against L. dispar were found according to the gradation phase in which the bioinsecticide was applied. In fact, *L. dispar* infestations decreased gradually in the years following Btk applications when the product was sprayed in the culmination phase, whereas applications performed in the progradation phase led to an increase in the population density after 2-3 years. These results could be useful for decision-making in control programs against *L. dispar* in cork oak forests and indicate that an accurate choice of timing of Btk application is necessary to minimize application costs over the long-term.

KEY WORDS: *Lymantria dispar*, population dynamics, microbiological application.

ORAL PRESENTATION

Comparative efficacy evaluation of aerially applied *Bacillus thuringiensis* kurstaki formulations against *Lymantria dispar* in cork oak forests

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The efficacy of different *Bacillus thuringiensis* subsp. *kurstaki* (Btk) formulations was evaluated against *Lymantria dispar l*arval populations in 2018 and 2019 in North and Central Sardinian cork oak forests, respectively. The following Btk commercial formulations were compared: (1) Foray® 76B AVIO (strain ABTS-351, Sumitomo Chemical Agro Europe S.A.S., 20 BIU/L); (2) Rapax® AS AIR (strain EG-2348, Biogard, division of CBC Europe, 24 BIU/L). The experimental design involved three replicates for each of the following treatments: (1) untreated control; (2) Foray® 76B at the dose of 2.0 L/ha; (3) Foray® 76B at the dose of 2.5 L/ha; (4) Rapax® AS AIR at the dose of 2.0 L/ha. The application of Btk-based formulations was conducted using helicopters equipped with 4 electronic rotary atomizers adjusted to sprinkle 160 micron-sized drops. The field efficacy of Btk treatments was assessed by counting the number of larvae on 40 shoots, randomly-sampled from 10 tagged trees, before and 7, 14 and 21 days after the application. Additional laboratory assessments were conducted by recording the mortality of 100 II-III instar larvae randomly collected from each treated and control plot and fed with foliage from the same plot. Data were recorded until pupation. Results showed that all Btk-based formulations were effective against gypsy moth larvae, as the insect population density observed in the field was significantly higher in untreated than treated areas. The effectiveness was confirmed by laboratory observations for all three Btk treatments. Our results indicate that all formulations and doses tested are suitable for the implementation in gypsy moth management programs. Furthermore, formulations applied at 2.0-2.5 L/ha represent a convenient alternative to Foray® 48B, the reference product normally used in Sardinia at the dose of 4 L/ha. Its replacement with more active formulations, such as Foray® 76B and Rapax® As, would provide considerable economic and logistical advantages, which would translate into lower costs and faster response time for the protection of cork oak forests.

KEY WORDS: *Bacillus thuringiensis kurstaki,* gypsy moth, cork oak.

ORAL PRESENTATION

Distribution area and biological traits of *Pseudocleruchus triclavatus*, newly recorded in Italy as an egg parasitoid of the forest pest *Barbitistes vicetinus*

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Barbitistes vicetinus Galvagni & Fontana is an endemic forest pest described very recently in 1992 and spread almost exclusively in north-eastern Italy (Euganean and Berici Hills). Since 2008, severe outbreaks occurred in Euganean Hills affecting the whole hillside area, causing defoliations especially to forest species and crops cultivated close to the infested wooded areas, such as vineyards, olive groves and cherry orchards. To date, possible control strategies have yet to be determined. In the current study, data were collected 10 years after the first outbreak had been reported in the Euganean Hills. Through a sampling program aimed at monitoring progress of the outbreaks in 2018, we collected eggs laid by *B. vicetinus* in the soil from 40 forest sites across the Euganean Hills. For each site, eggs were divided in i) hatched eggs, ii) flattened eggs, iii) eggs with small holes on the chorion and iv) fertile eggs. A mymarid parasitoid emerged from a batch of fertile eggs that didn't hatch in the spring. It was identified as *Pseudocleruchus triclavatus* Donev & Huber, previously known only from Bulgaria and Romania. We found that up to 140 parasitoids can hatch from a single egg of *B. vicetinus*. Our reconstruction of the distribution area of *P. triclavatus*, based on eggs found in the ground with small circular emergence holes on the chorion, showed that this species was spread in almost all the sites along all elevation gradients. Furthermore, the highest number of parasitized eggs was found in the sites where oviposition density of the bush-cricket was higher. Further studies are needed to investigate the biology of this parasitoid against the pest *B. vicetinus*, which is the first known host record for *P. triclavatus*.

Management and conservation of the saproxylic beetles fauna in the eastern beech forests of the Matese massif

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The importance of forest stands in the most advanced stages of ecological succession has long been recognized. In addition, the growing awareness of their relevance as carbon stocks and landscape elements, forests represent a key element of conservation strategies. Some authors say that forest biodiversity alone accounts for about 80% of global diversity, and 30% is closely linked to deadwood, reaching peaks of 50% in Coleoptera. The aim of the work is to quantify the wealth and abundance of saproxylic beetle species in beech forests of the southern Apennines, regularly managed, to evaluate the conservation status of these ecosystems using functional and ecological traits of the species (e.g. trophic level) and the IUCN threat level. The study area is located in the eastern side of the Matese massif, in the Roccamandolfi (IS) area within the SCI "La Gallinola - Monte Miletto - Monti del Matese" (Cod. IT 7222287). The study activities were carried out in four distinct beech forest stands based on the altitude (High and Low) and exposure (South and North). These are mainly pure beech woods, regularly managed for production purposes. The forest structure shows some differences, especially in the low-end populations where the signs of the old coppice stand management are noted, in fact, they show a high number of trees, around 1500-2000 per hectare mainly of agamic origin, compared to the high-end populations (both with ~ 700 trees per hectare). Each of the four areas is divided into 15 squares, with a side of 70 m, at the center of which the survey was carried out for the dendroauxometric characterization of the areas, in plots with a radius of 13 m. In the same areas, Coleoptera fauna was sampled positioning a total of 60 windows flight traps and the survey of deadwood was carried out. Insect monitoring activities took place from May to October 2018. Altogether 3415 specimens of beetles of 128 species and 34 families were collected. Among the species collected, 41.4% are saproxylic and included in the Red Data Books. Analyzing the IUCN categories, two species are included in the Vulnerable threat class; 7 are Near Threatened; 43 species are Least Concern and one is Data Deficient. Furthermore, the related trophic category has been studied thoroughly. About half of the sample is represented by the categories of xylophagous and saproxylophagous (56.6%), followed by mycetophagous (20.7%). From the analysis of the species sampled, it is noted that the sites exposed to the South host a greater consistency of species (83 and 46 respectively for High and Low) compared to the areas exposed to the North (44 and 67 between High and Low). The predominance of species that characterizes the South High site can be attributed to a condition of maturity to which the site tends. In conclusion, we can say that forest management practices, in addition to influencing the structure of the forest, play a key role for saproxylic biodiversity. It shows that exposure is also an important factor to consider in the planning phase.

KEY WORDS: Southern Apennines, biodiversity indicators, ecology, threatened species, forest structure.

ORAL PRESENTATION

First reports of *Euwallacea fornicatus* in tropical greenhouses in Europe

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Species of the Euwallacea fornicatus species complex are emerging tree pests. Unlike most other ambrosia beetles, *E. fornicatus* is able to infest healthy plants having a broad host range of more than 400 described plant species in 75 families, including important agricultural crops. Native to South East Asia, this species was introduced into various countries including the USA and Israel, where they cause considerable damage to many tree species. The wide host range makes *E. fornicatus* an important tree pest especially in the invasive ranges. In Europe, *E. fornicatus* is currently considered as a quarantine pest. Recently, a few outbreaks of *E. fornicatus* have been reported in Europe. The first individuals were found in 2017 in a palm house of a botanical garden in Poznan (Poland) on a single sacred fig tree (Ficus religiosa). In 2020, an outbreak of *E. fornicatus* was detected in a tropical greenhouse in Merano (Italy) where 28 trees from 20 different families were attacked. The populations of both localities have been subsequently eradicated. Recently, two additional outbreaks have been described in two greenhouses in Erfurt and Berlin (Germany), where *E. fornicatus* individuals were detected on *Teconta grandis*, *Clusia rosea*, and *Ficus elastica*. These outbreaks are currently under eradications. By performing a comparative genetic analysis of the different populations, we aim to reconstruct the invasion routes of the beetle. We discuss the invasion dynamics of the outbreaks and describe the measures and partial eradication of the different cases. Our study will provide novel insights into the invasion history of this potentially newly emerging invasive pest species in Europe.

KEY WORDS: Ambrosia beetle, invasive species, Tea shot hole borer, *Euwallacea fornicatus*.

Assessment of *Tortrix viridiana* defoliations by analysis of remote sensing data

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The aim of this study is to investigate the suitability of spectral indices derived from satellite data (Sentinel-2) to assess the spatial and temporal consistency of the green oak leaf roller moth (*Tortrix viridana* L.) defoliation on Mediterranean deciduous oak forests. Larval attacks *T. viridana* could completely defoliate the infested oaks in early summer, also in large areas and for several consecutive years when outbreaks occurs. Despite the impact defoliations on the forest management activities there is a lack of information on the spatial and temporal patterns of the leaf roller moth outbreak distribution and severity of defoliations, whereas the availability of accurate monitoring data and maps could support the planning of pest management activities and policies. The data collected by the recent remote sensing platforms are characterized by spatial and temporal resolutions adequate for the monitoring of insect disturbances, and could support the development of pest management models and decision support systems. We collected field data on leaf damage during the period 2016–2019, along a set of 35 field plots of pubescent oaks (*Quercus pubescens*) located in the Goceano and Gennargentu mountains (Central Sardinia) where the degree of defoliation was assessed during June, when oaks shown the peak of leaf consumption by the caterpillars. To evaluate the disturbances caused by defoliations, a set of seasonal (spring and summer) Sentinel-2 remote sensing images were analysed and the following spectral indices were calculated: Normalized Difference Vegetation Index (NDVI), Green Normalized Difference Vegetation Index, and Normalized Difference Red-Edge index (NDRE). For each field plot we derived the time series of the spectral indices and the relationship between the degree of oak defoliation and the amplitude of remote sensing signals were analysed using linear regression equations and graphical methods (seasonal trend). Trends of spectral indices were analysed and summarised for each year, study area, class of defoliation, and forest stand. Supervised and unsupervised algorism were applied in order to classify the forest stand status. NDVI and NDRE exhibited the better relationship with the insect defoliations observed in field plots. The data analysis showed a shift of the summer spectral peak and lower values of spectral indices during the season, related to oak leaf damage by the larval feeding. The method appears to be able to provide useful guidance for the assessment of defoliation caused by spring folivores pests and could be applied at large-scale in order to monitor the progress of outbreaks during the year. In addition, the maps of defoliation derived from remote sensing could provide useful data for development and calibration of modelling applications in forest pest management.

KEY WORDS: Mediterranean forests, insect pest outbreaks, *Quercus pubescens, Tortrix viridana*, Satellite data, Sentinel-2, spectral indices, NDVI, gNDVI, NDRE.

POSTER

Entomopathogenic nematodes and fungi as biological control agents of xylophagous pests in agro-forestry ecosystems

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The effects of entomopathogenic nematodes (Steinernematidae and Heterorhabditidae) and fungi strains (*Beauveria bassiana* and *Metarhizium anisopliae*) were evaluated in laboratory and semifield assays against larvae and adults of five xylophagous pests: the asparagus moth *Parahypopta caestrum* (Lepidoptera: Cossidae), the European goat moth *Cossus cossus* (Lepidoptera: Cossidae), the pine longhorn *Arhopalus syriacus* (Coleoptera: Cerambycidae) the black buprestid *Capnodis tenebrionis* (Coleoptera: Buprestidae) and the red palm weevil *Rhynchophorus ferrugineus* (Coleoptera: Dryophthoridae). Due to their biology and ethology, these insects may be included in the category of pests residing in cryptic habitats. Five examples of pests for which the chemical control is not effective and risky, as it can cause problems to individuals as well as to the environment. For this reason, the potential use of entomopathogenic nematodes (EPNs) and entomopathogenic fungi (EPF) as biological control agents was evaluated in laboratory and semi-field conditions. Native strains of EPNs (*Steinernema feltiae*, *S. carpocapsae* and *Heterorhabditis bacteriophora*) and EPF (*B. bassiana* and *M. anisopliae*) were used to control larval (*C. cossus, P. caestrum, R. ferrugineus, C. tenebrionis* and *A. syriacus*) and adult (*R. ferrugineus, C. tenebrionis* and *A. syriacus*) stages. All the entomopathogens demonstrated the ability to control these pests. *Beauveria bassiana* and *S. feltiae* caused significantly greater mortality rate in the adults and larvae than *M. anisopliae, S. carpocapsae* and *H. bacteriophora*. Nematodes and fungi are able to penetrate the cryptic habitats because they are living organisms and may be horizontally transmitted by infected hosts. The distribution of EPF as preventive control method and the injection of EPNs suspensions to reach and infect the larvae inside the wood galleries can be a combined sustainable control system.

KEY WORDS: microbial control, Steinernematidae, Heterorhabditidae, *Beauveria, Metarhizium*.

ORAL PRESENTATION

SESSION X

Bioactivity of a new insect repellent: laboratory evaluation against *Sitophilus oryzae*

Linda Abenaim, Stefano Bedini, Elio Napolitano, Alessandro Mandoli, Francesca Venturi, Barbara Conti

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Essential oils (EOs) have been often proposed as a valid alternative to synthetic pesticides to control stored grain pests. Generally, EOs contain various percentages of cyclic monoterpenes such as menthol, menthone, carvone, and also phenolic derivatives as eugenol with repellent properties. This study aims to evaluate the repellence of 22 main constituents of various EOs through preliminary tests with the "Area Preference" method against *Sitophilus oryzae* (Coleoptera: Curculionidae), one of the main pests of stored products, that causes important economic losses to cereals and their derivatives. Among the substances tested, eugenol (C10H12O2) showed the greatest repellent activity. However, eugenol is poorly suitable for use in stored grains due to its low persistence and strong and spicy odour. In this research, we modified the eugenol structure to obtain a new compound (compound A, C18H20O4) with a higher molecular weight (less volatile and odorous but more persistent) but with still high bioactivity. The bioactivities of compound A and eugenol were tested in a two-way static olfactometer. Compound A showed a greater repellence and persistence than eugenol against *S. oryzae*. Compound A was subsequently evaluated by sensory analysis (Panel Tests) in comparison with eugenol. The results of the Panel Test indicate that compound A is significantly less odorous than eugenol. We can therefore conclude that: i) the modification of the eugenol structure increases both the effectiveness and the duration of the repellent activity; ii) the reduction of the volatility can significantly reduce the strong smell, increasing the compatibility of the compound with food. Overall, the repellency of compound A, its persistence and lower smell intensity indicate that it could represent a valid alternative to other synthetic repellents in food active packaging for the control of foodstuffs insects.

KEY WORDS: eugenol, foodstuff insects, repellence, olfactometer, panel test.

ORAL PRESENTATION

Evaluation of agro-food by-products for feeding of *Tenebrio molitor* larvae

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Insect protein meal production is generally considered more sustainable than meat production. Theconsume of soil and water is very small, while an improvement is desirable for used raw materials and energy consumption. The use of by-products at "km 0" can increase sustainability of rearing diet for *Tenebrio molitor* (L.). The aim of this work is to evaluate the effect on larval survival and larval growth of rearing substrates formulated with by-products from the agro-food industry. The study considered by-products such as wheat bran, coffee silverskin, old bread and spent grain, available locally at mills, coffee-roasting industry, bakers and craft breweries. The by-products were integrated with 5% zootechnical yeast and tested both individually (test A) and in combination each other (test B). The control is represented by the standard diet based on wheat bran and yeast. The water was supplied once a week through 1.5 g / replica of *Opuntiaficus-indica* (L.)'s cladodes. For each thesis 10 replicas of 20 larvae each, one month old, were used. The tests were carried out in a climate chamber (28 ± 0.1°C, 60 ± 5% RH, L: D = 0: 24h) for the *T. molitor* rearing in ENEA Trisaia Research Centre in Rotondella (MT). After 28 days of growth, larval survival and growth rate were detected. Data were analyzed by Kruskal-Wallis test, followed by Mann-Whitney test. The results of test A showed a significantly lower larval survival than the control only in the coffee silverskindiet (85.5%). The growth rate values of by-products tested are significantly lower than the control (3.79 ± 0.09) in the other thesis:in coffee silverskin thesis, growth rate (0.95 ± 0.28) was less than spent grain (3.00 ± 0.08) and bread (2.95 ± 0.10).Test B results showed no significant differences between theses in larval survival. The growth rates, obtained with the different diets, were not significantly different from the control for the theses wheat bran + bread, wheat bran + spent grain and bread + spent grain, while the mixtures between the different by-products and the coffee silverskin always gave rates of significantly lower growth. In conclusion, the old bread and spent grain by-products represent a valid alternative to wheat bran in the rearing of *T. molitor* larvae, while the coffee silverskin did not give the desired results, at least in the conditions tested. Further studies are underway to deepen the aspects related to the conversion efficiency of the diets used.

KEY WORDS: yellow mealworm, edible insect, circular economy, alternative protein.

Impact of the diet on the gene expression of antimicrobial peptides in *Hermetia illucens* (Diptera: Stratiomyidae)

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The role that insects can play in converting different types of low quality biomass, such as urban and agri-food sector wastes, into high-quality proteins, has led to an increasing spread of insect farming. However, high density, sub-optimal abiotic parameters and high consanguineity occurring in mass rearings make insects more vulnerable to various pathogens causing serious economic losses. In such contests, considering the possible use of insects as food and feed and the growing antibiotic resistance concerns, antibiotics should be avoided. Therefore, the implementation of new strategies to safeguard the healthiness of the rearings, such as the modulation of the insect immune system, are required. Recently it has been highlighted how the environment and the diet influence the production of antimicrobial peptides, key components of the insect innate humoral responses. The gene expressions of some of these peptides, in particular a defensin and a cecropin, was assessed by Real-Time qPCR in *Hermetia illucens* (L.) (Diptera: Stratiomyidae) specimens reared on different diets. In particular, their expression level were evaluated, as fold change ($2^{-\Delta\Delta Ct}$), in larvae and prepupae grown on cereal- and municipal organic waste-based diets. Moreover, this analysis was performed in *H. illucens* prepupae reared on a cereal-based diet supplemented with different vegetal oils (sunflower, corn and soy oil). The municipal organic waste-based diet resulted in a significant overexpression of both genes encoding for defensin (fold change: 9.43) and cecropin (fold change: 17.85) peptides only in *H. illucens* prepupae. This diet, characterized by a higher microbial load, may have strongly stimulated the insect immune system resulting in a greater production of antimicrobial peptides. Although the addition of the vegetal oils did not lead to a synergic and homogeneous increase in the gene expression of the two peptides, it contributed, with the exception of corn oil, to an upregulation of at least one of them. The possibility of modulating the production of antimicrobial peptides through the diet opens up new perspectives in the management of the health aspects of insect mass rearings, not only of *H. illucens*, but also of other species of economic interest.

KEY WORDS: municipal organic solid waste, vegetal oils, cecropin, defensin, circular economy.

POSTER

Experimental research on insect pests which may infest stored foodstuffs recently introduced into the Italian market

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The demand from consumers for foods different from those traditionally available on the market due to various needs, has led companies in this sector to import pseudocereals such as amaranth and quinoa, minor cereals such as teff and millet, and oilseeds such as chia and hemp from traditionally producing countries in recent years, offering a wide range of products derived from them on the market. The presence of these products on the Italian market implies the storage of unpackaged and finished products and involves the food industry at all stages of production up to packaging. In all these stages, foodstuffs are potentially exposed to insect attacks and therefore need careful and effective management throughout the supply chain, from field to consumer. The hygiene and protection of these "new" foods therefore involves both insects from the countries of origin and accidentally imported with the seeds, and insects already present or established in Europe which attack indigenous foodstuffs but which could also develop on these new products. Information on this matter is scarce, particularly in European countries.
Some of the insects - known for their high polyphagy - which are more commonly found in Italy and which infest commodities mainly of cereal origin are *Tenebrio molitor* L. (Coleoptera: Tenebrionidae), *Tribolium confusum* J. du Val (Coleoptera: Tenebrionidae), *Oryzaephilus surinamensis* (L.) (Coleoptera: Silvanidae), *Cryptolestes pusillus* (Schönherr) (Coleoptera: Laemophloeidae) and *Plodia interpunctella* (Hübner) (Lepidoptera: Pyralidae). This research was carried out in order to assess the capacity of these species to develop and reproduce on the above mentioned commodities. Tests involving controlled infestation using insects bred at the Di.Pro.Ve.S., kept in air-conditioned cells at 23±2 °C and 65±5% R.H., were therefore carried out under the same environmental conditions. Larval growth, life cycle duration, the number of individuals which reached the adult stage and the weight of these individuals were evaluated. The seeds used were purchased in supermarkets. The results showed a different susceptibility of the foodstuffs considered to infestation by the species under observation. Millet was the only product that allowed larval development up to the adult stage for each of the species examined, while on the other products some of these pests were not able to develop. In particular, no species were observed to develop on amaranth. The species which reached the adult stage on the greatest number of products was *T. confusum*, while *C. pusillus*, *T. molitor* and *P. interpunctella* completed their life cycle on only two substrates.

KEY WORDS: *Tenebrio molitor*, *Tribolium confusum*, *Oryzaephilus surinamensis*, *Cryptolestes pusillus*, *Plodia interpunctella*, pseudocereals, amaranth, minor cereals, quinoa, teff, millet, oilseeds, chia, hemp seed.

POSTER

Sensory quality for the selection of an essential oil and its use in synergy with diatomaceous earth for the control of foodstuff insects

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Essential oils (EO) have aroused an increasing interest, in the recent years, as an ecological and low-toxic alternative to synthetic repellents and insecticides to be used against harmful insects. However, they currently have little practical application in the protection of stored cereals due to their limited effectiveness over time and their interference with the organoleptic properties of the food they come in contact. Therefore, it is essential that the choice of an EO for the protection of food is not detrimental to the organoleptic quality of the final product. In the research we have evaluated the olfactory profile of the EOs of *Foeniculum vulgare*, *Pistacia lentiscus* and O*cimum basilicum* in addition to their toxicity towards one of the main pests of the stored wheat, *Sitophilus granarius* (Coleoptera: Curculionidae). A panel of the expert evaluators indicated, from the olfactory point of view, the essential oil of *O. basilicum* and *F. vulgare* as the most suitable for the treatment of wheat. In the toxicity tests, the most toxic EO was *P. lentiscus* (LC50 = 36.36 µL kg- 1), however it was also the most unpleasant from the olfactory point of view for the consumer. On the contrary, the least toxic OE was *F. vulgare* (LC50 = 77.59 µL kg- 1). On the basis of these results the OE of *O. basilicum* was selected for the trial in the warehouse. The EO was then tested in combination with diatomaceous earth (DE) and the mixture showed synergistic effects (Co-toxicity coefficient equal to 1.36). Overall, *O. basilicum* was the best OE for the treatment of the wheat, considering its toxicity to insects and the olfactory profile. In warehouse conditions, the wheat co-treated with the OE of *O. basilicum* and DE showed a significantly lower average infestation (1.5 insect kg - 1) compared to the untreated wheat (7.0 insect kg - 1).

KEY WORDS: sensory quality, diatomaceous earth, essential oil, synergy, foodstuff insects.

ORAL PRESENTATION

Growth performance of *Hermetia illucens* larvae on mixtures of stabilized vegetable and dairy by-products

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Nowadays the need to find new protein sources for animal feed is imperative. Based on a circular economy concept, the Flies4Value project (POR-FESR Emilia Romagna) was conceived, focusing on the use of a bio-converter insect, the black soldier fly *Hermetia illucens* (L.) (Diptera: Stratiomyidae). The larvae of this species are reared on by-products with different seasonal availability and will be used for the production of feed for laying hens, thus obtaining eggs with a naturally pigmented yolk, replacing the current use of synthetic pigments. The present work aimed at identifying optimal mixtures of stabilized by-products supplied by agro-food companies in the Emilia-Romagna area, in order to maximize the larval growth performance and the pigment content. Vegetable by-products (tomato, pineapple, apple, orange, borlotti beans, cannellini beans, chickpeas, green beans and wheat bran) were mixed with "scotta", a liquid waste from the dairy supply chain, in an initial ratio of 1:2. Initially, the larval growth was evaluated on individual substrates, tested in two different textures: a) dehydrated and in the size as delivered and b) dehydrated and minced. Subsequently, the larvae were reared on 4 mixtures composed by different percentages of tomato, wheat bran, green beans, borlotti beans and chickpeas, always adding the scotta as a liquid fraction. All the trials were carried out in a climatic chamber (27°C, 70% relative humidity) using glass boxes in which 100 second- or third-stage larvae were placed, together with 300g of feeding substrate. The larval weight was recorded every other day and the trial was concluded at the appearance of the first prepupa. The result of preliminary tests showed that: a) the larvae with the highest growth rate (>0.02 g/day) were those fed with wheat bran, chickpeas in both textures, green beans in both textures and minced tomatoes; b) the best substrate conversion was obtained by feeding the larvae with coarse chickpeas, minced borlotti beans, coarse green beans, minced tomatoes and wheat bran. Moreover, these results together with the larvae nutrient characterization and pigment content, led to the development of mixtures tested in a second series of experiments. Overall, the substrate mixtures resulted in a better growing rate (0.0244-0.0273 g/day) and substrate conversion (10.257-12.720) than the single substrates. The mixture to be used in the scale up production for the project will be defined according to the highest growing rate and pigment content of the larvae. FLIES4VALUE is a project co-funded by POR FESR, Program 2014-2020, Axis 1, Action 12.2 Call for Research Grouping.

KEY WORDS: *Hermetia illucens*, circular economy, sustainability, vegatable by-products, chicken feed, pigments, dairy waste, eggs production, hens.

POSTER

Development of an IPM program in the Vatican Apostolic Archives

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The aim of this work is the development of the management tool of IPM (Integrated Pest Management) in the Vatican Apostolic Archives. In this paper, we describe the implemented strategies used in the Vatican Apostolic Archives for the adoption of an IPM project, based on constant pest monitoring followed by a program evaluation. With the passing of time, the presence of different species of insects has been recorded in all the premises of the Vatican Apostolic Archives, as occasional guests and as active biodeteriogens. IPM in the Vatican Apostolic Archives follows clear principles: 1) comprehensive risk assessment; 2) continuous inspection and monitoring; 3) preventive measures aiming at physically blocking pest development; 4) remedial measures, giving priority to non-toxic methods. As part of a strategy of preventive conservation, IPM is an effective system of reduction of damage and costs and of minimization of corrective interventions. IPM is an integral part of risk management within an organization as defined in UNI ISO 31000:2018. Only when all the risk factors caused by the presence of pests have been identified, analyzed and evaluated, the appropriate plan of action can be decided. The aim is to prevent, monitor, and, if required, to treat the infestation. As of June 2016, within the Vatican Apostolic Archives an infestation of *Reticulitermes lucifugus* was found. Further research permitted to locate in the adjacent Vatican Gardens the origin of the infestation: a *Cedrus* trunk situated circa one hundred meters from the Archive premises; and an underground tunnel that termites used as passage. Afterwards, through the use of luring stations, contact with the colony was established and foraging started with IGR product until complete elimination. Entomological monitoring has been activated with the use of passive glue stations distributed in the rooms nearby the light points, in order to record the insects' activity throughout the various periods of the year. This monitoring activity is combined with detailed visual inspection carried out through the various shelves and documents. All data is collected monthly and processed in order of priority using an internally developed software. As soon as an infestation is detected, an anoxia intervention by the internal staff is readily planned. When the infestation is detected on furniture, being it chairs, cabinets or any other item made of wood, the intervention is performed with products based on permethrin, applied with spray or brush and, when possible, also recurring to impregnation techniques. In conclusion, in the Vatican Apostolic Archives, risk analysis is in various ways connected with structural and managerial problems; the evaluation of pests over time allows the development of an increasingly precise database, capable of processing information in its complexity as a whole; this will lead to a more correct risk analysis and therefore define the necessity for specific actions and the development of control strategies.

KEY WORDS: cultural heritage, biodeteriogens, IPM, woodworm and termites.

ORAL PRESENTATION

Parasitological observations on *Necrobia rufipes*infestations in Umbria

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Necrobia rufipes is an emerging cosmopolitan pest, recently reported in Italy as a parasite of animal feed. *N. rufipes* attacks a large number of preserved substrates, both vegetable (e.g. dried coconut, almonds, cashews) and animals (e.g. meat, dried fish, eggs or milk powder) and is frequent in carcasses in the initial phase of their drying, where it can become predator of larvae and eggs of other insects, especially of Coleoptera Dermestidae. Four years ago we had the opportunity to detect this parasite on kibble for animals, both in industrial production companies and in pet shops, and since every year this insect systematically attacks these foods, we want to report here some of our observations: 1) *N. rufipes* introduces itself into the premises of the factory both actively, attracted by the smell of food, and passively, transported through pallets kept outside, especially if they are close to meat waste. The infestation of the pallets is easily recognizable by the presence of ice-white spots, produced by the insect, inside cracks and crevices in the wood. In the finished product, in general, the infestation comes from inside the packages, but we also observed the characteristic ice-white excretions in the folds and welds of the sacks, a sign of the attempt to penetrate from the outside. 2) *N. rufipes*, widely distributed in Africa, Asia and South America, prefers high temperatures, with an optimum around 33-34°C. Our observations in industrial environments showed a high development and pest capacity, where the storage environment of the animal feed was very hot (33°C) and poorly ventilated, with a relative humidity of about 70%. The infestation was evident, also in this case, due to the presence of whitish material around the openings of the jute sacks, which contained the feed before packaging. 3) The infested material, observed by us, never contained primary pests, but only larvae and adults of *Necrobia* that mated continuously. Sometimes only rare specimens of *Oryzaephilus surinamensis* were present inside. This could indicate that *N. rufipes* carries out an important predatory activity, or that it does not need preys in order to survive. 4) *N. rufipes* was never attracted by the adhesive light traps positioned inside, even during massive infestations. 5) The resolution of the infestation has always been obtained as follows: a) destruction of the infested material; b) environmental disinfestation with residual pyrethroids, cleaning by means of an aspirator and subsequent washing; c) elimination of processing residues and waste both from the storage environment and outside the factory / shop.

KEY WORDS: *Necrobia rufipes*, infestation, feed.

POSTER

Food utilization efficiency of adult Sitophilus granarius: evaluation of nutritional indices on pigmented and yellow wheat genotypes

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The granary weevil, *Sitophilus granarius* (L.) (Coleoptera: Dryophthoridae), is one of the most damaging pests of stored cereals worldwide. Development of resistant wheat varieties is one of the most promising alternatives to insecticides in integrated management of stored grain pests. In this perspective, pigmented wheat genotypes are particularly interesting due to the content of phenolic compounds such as xanthophylls, carotenoids, flavonoids, and anthocyanins which are involved in plant resistance to biotic stress. In the present study, the feeding performance of granary weevil adults on two red durum wheat genotypes of Ethiopian origin (Abyssinicum, T1303) and two durum (Primadur, Ofanto) and one bread (Mec) commercially available wheat varieties with yellow kernels were evaluated. For each genotype, a whole wheat flour was prepared by milling kernel samples (500 g) (Tecator Cyclotec 1093). Each wheat flour (10 g) was uniformly suspended in distilled water (50 mL) by stirring. To obtain flour disks to be used as food source in feeding bioassays, aliquots (200 µL) of a suspension were dropped onto holes (Ø 1 cm, height 3 mm) punched in a plexiglass plate and left overnight at 60±5% r.h. to dry. In a pre-weighed glass vial (Ø 2.5 x 4.0 cm) two flour disks and 5 group-weighed weevil adults (two weeks old) were introduced. Each vial was then re-weighed and maintained in the dark for 6 days. The glass vials with flour disks and live insects were weighed again and the number of dead insects recorded. The following nutritional indices were calculated: relative consumption rate (RCR), relative growth rate (RGR), efficiency conversion of ingested food (ECI), feeding deterrence index (FDI). Mean RCR values of insects feed with disks of red wheat genotypes were lower than those of insects feed on disks of yellow wheat varieties but not statistically different ($F = 1.393$; $df = 4$; $P = 0.272$), indicating small changes in the amounts ingested. However, these slight differences lead to a FDI value for the Abyssinicum genotype which was significantly higher than those of yellow varieties ($P < 0.05$, Tukey test). Mean RGR as well as ECI values were negative for red wheat genotypes but positive for the yellow wheat varieties, indicating respectively a decrease and an increase of the insect body weight during the experiments, as a result of a different food conversion efficiency. Lastly, the mortality percentages of insects feed on red wheat flour disks were significantly higher than those observed for insects feed on yellow wheat varieties. Overall, results suggest the presence in the red wheat genotypes of compounds able to interfere with the food utilization efficiency in the granary weevil.

KEY WORDS: granary weevil, nutritional indices, pigmented wheat genotypes, plant resistance.

POSTER

Assessment of the bioaccumulation and excretion capacity of some pesticides and mycotoxins in *Tenebrio molitor* larvae

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Among the species approved for human consumption *Tenebrio molitor* (Coleoptera: Tenebrionidae) is one of the most promising for industrial food and feed production. *T. molitor* is generally reared on cereals based diets which may content pesticides or mycotoxins derived from field practices and that may be accumulated by feeding causing a health risk to insect consumers. Currently, in European legislation there is no complete guideline to define the safety of insects as food, due to the lack of scientific data and clear parameters. Then the present study is a contribution to fill a gap in this sector. Three pesticides, and three mycotoxins were tested to evaluate the uptake and excretion rates in T. molitor larvae fed on contaminated diet. The pesticides were selected on the base of different lipophilicity characteristics, an insectidide deltamethrin (DELTA), a fungicide tebuconazole (TEB) and a plant growth regulator chlormequat chloride (CCC). The selected mycotoxins are aflatoxin B1 (AFB1), ochratoxin A (OTA) and fumonisin B1 (FB1). In this study the uptake of the above mentioned chemical compounds and mycotoxins and their elimination rate were investigated in *T. molitor* larvae fed on wheat flour based diets contaminated with two concentration levels for each compounds. New analytical methods for extraction and quantification of DELTA, TEB, CCC, AFB1, OTA and FB1 residues in larvae and frass samples were developed and validated. The larvae were analyzed at the end of the rearing periods even after 24 hours of fasting and the analysis showed that DELTA and TEB are retained by larvae even after fasting, depending on their lipophilicity, while for highly hydrophilic molecules (CCC), a fasting period of 24 hours is sufficient to ensure the complete elimination of contaminant residues. We also demonstrated that *T. molitor* larvae do not accumulate the three mycotoxins in detectable or dangerous concentrations at the two concentration levels tested and that a 24-hour fasting period ensured a sufficient rate of elimination of AFB1, OTA and FB1 residues.
The study confirmed that only a complete traceability of diet components and adequate chemical controls can guarantee the safety of insects as "novel foods".

KEY WORDS: Edible insects, pesticides, mycotoxins, *T.molitor*, uptake.

ORAL PRESENTATION

Application of lasting cold storage on *Tenebrio molitor* larvae

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The insects are heterothermic organisms and temperature affects their lifespan. In order to reduce development times, the rearing of *Tenebrio molitor* (L.) is carried out in optimal temperatures close to 27°C. On the other hand, the strong slowdown of the lifespan caused by low temperatures can be exploited to keep the larvae alive for long periods. Studies on the influence of temperatures on the development of *T. molitor* larvae have identified optimal parameters to reduce growth times and extreme limits influencing the survival of the different stages. Observations on the influence of low temperatures are known for a 48 hours period, but no information are available for the long-term effect. This work instead aims to test the long term cold storage on mealworms. Larvae of *T. molitor*, reared in ENEA Trisaia Research Centre, have been divided into two groups: young larvae YG (0.030 g/larva) and almost mature larvae OL (0.063 g/larva). Each group was "refrigerated" on sterile bran at 10±1°C and 63% RH, with four theses coded by the different days of storage (T30, T60, T90 and T120). Each thesis consisted of 10 replicas of 20 larvae everyone. At the end of each conservation period, larval survival, weight gain and the presence of pupae were detected. After the cold storage, preliminary observations were carried out on OL larvae brought in standard rearing conditions (28±1°C and 60±5% RH). On these, the ability to complete the cycle was assessed by detecting the number of pupae hatched, deformed and well-formed adults. The statistical analysis of the data shows that the larvae have passed the "cold storage" period with survival between 98% and 100% without significant differences between the four periods for both the YG and OL groups. The growing trend of weight gain has demonstrated a larval activity even at low temperatures in both YG and OL theses; the growth eased after 3 months, with no significant differences between T90 and T120. The presence of pupae was found only in the OL group even if with very low numbers (4 pupae on 800 larvae).
The OL larvae completed the cycle in standard rearing conditions with 17.13% post-refrigeration cumulative losses. Most of the loss was caused by pupae mortality (10.91%), while the deformed new adults were only 4.07%.
In conclusion, our preliminary results clearly show that it’s possible to cold store the live larvae for long time periods, with minimal losses. If the "preserved fridge" larvae have to provide reproducers, it is anyway to prefer an enhancement of the storing conditions of pupae, in order to reduce losses and to carry out a fertility test of the well-formed adults.

KEY WORDS: yellow mealworm, edible insect rearing, low temperature, cold storage.

POSTER

Lab assay of propolis' extracts on storage mites' control

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Storage mites feed on wide variety of substances and they can be found in many different products such as grain, flour, hay and straw, cold cuts, cheese. These organisms, frequently associated to fungi and bacteria, can infest storage stocks and compromise food products. Several species of storage mites have been shown to cause IgE-mediated sensitization as asthma, rhinitis or conjunctivitis to workers exposed. For environmental prevention in food storage, the conventional control of mites has largely depended on broad-spectrum pesticides but mites have been reported to be fairly tolerant to these products (e.g. pyrethroids). In this context, the development of new mite control strategy is a real challenge whereas, moreover, a sustainable approach based on natural products, such as propolis, can be an alternative to chemicals. Propolis is widely recognized and used as antiseptic, antibacterial, antifungal and antiviral, as well as antiparasite (e.g. against the bee parasite mite *Varroa destructor*). A preliminary test showed high mortality of dust mites treated with hydroalcolhic mother tincture of propolis with prolonged bacteriostatic effect. The experiment aimed to assess the effectiveness of propolis treatment on pest storage mites, *Lepidoglyphus destructor* and *Glycyphagus domesticus*, and on the fungal and bacterial communities associated with these lab-reared mites. In lab, the mites were treated in a mini-chamber (3 mm deep, 3 cm diameter) by <1min immersion with two propolis extracts (Propolis Mother Tincture 80° and Propolis Hydroglyceric extract Fontana srl) and with 80° alcohol, with hydroglycerin and water as controls. For the toxicity assessment of propolis extracts on astigmatid mites, chambers were checked at microscope after 3 (t1) and 72 hours (t2) to assess mortality. During the experiment, mini-chambers were maintained in boxes with 85±5% RH at two different temperatures (6°, 16°C). On the whole, *L. destructor* showed higher susceptibility to tested products than G. *domesticus*. The two formulates of propolis caused important mortality on the adult mite species considered: at t1, they were completely immobile; at t2, some individuals newly moved. The hydroglyceric propolis was a bit more toxic than hydroalcolhic one, especially mortality of *L. destructor* was about 88%. At the lower temperature, *G. domesticus* less suffered adverse impact of treatments ($p<0.000$). The tested mites were also processed to have a broad indication of the effect of the substances on microbial communities associated to the storage mites: propolis treatments sensibly decreased bacterial community (CFU/g) confirming effectiveness as natural antibacterial. The results may represent a starting point in screening natural alternatives to traditional chemicals in environmental prevention and storage food mite pest control.

KEY WORDS: biological control, stored product mites, microbial control, propolis.

ORAL PRESENTATION

Le difese antimicrobiche negli insetti: le attacine dell'insetto stecco Carausius morosus (Phasmatodea)

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The biological activity of different loquat (*Eriobotrya japonica* (Thunb.) Lindl., Maloideae: Rosaceae) seed extracts towards *Sitophilus granarius* (L.) adults, one of the most important pest of stored cereals, was investigated. Peeled fresh seeds of the Algar variety were grounded and extracted for 24 h at room temperature with solvents of decreasing polarity: water, methanol, acetone, hexane/ether 80/20. Each crude extract was centrifuged and the supernatant was dried under vacuum in a rotary evaporator (40±5°C) except for the water extract which was lyophilized. Aliquots (150 mg) of the obtained residues were dissolved in the following solvents (1 mL): water/acetone 1/1 (water and acetone extracts), hexane/ether 80/20 (hexane/ether extract), methanol/acetone 1/1 (methanol extract). For each sample, two-fold serial dilutions were prepared. The contact toxicity of extracts was determined by applying 0.5 µL (75.00 – 4.68 µg) of each solution onto the pronotum of an adult weevil (n = 15) in thanatosis. The fumigant toxicity was assessed by using a fumigation chamber made up of a plastic container (135 mL) in which a perforated septum separated a lower chamber from an upper one. The lower chamber has been assigned to contain increasing doses (5 - 150 mg) of each sample or amygdalin (1-7 mg) plus β-glucosidase (1 EU) whilst the upper chamber contained adult weevils (n = 20-30), intact kernels (n = 20), and a picric acid (0.6%v/v) plunged paper (35x27 mm) to detect putative hydrocyanic acid formation. Toxicity values were determined by counting dead insects after incubation in the dark at 28±1°C for 24 h (both contact and inhalation toxicities) or 48 h (contact toxicity). The repellent activity of extracts to granary weevil adults was evaluated in two-choice pit-fall bioassays using a steel arena (Ø 32 cm × 4 cm height) with two diametrically opposed holes (Ø 3 cm) and two glass flasks (500 mL) positioned under each hole. Ten µL (1.5 mg) of an extract (treatment) and the corresponding solvent (control) were respectively laid on two filter paper discs (Ø 1 cm) which were suspended at the centre of each hole. Twenty insects were released in the centre of the arena and their distribution among treatment and control flasks and the base of the arena, after 3 h in the dark at room temperature, was used to calculate a mean (3 replicates) response index (RI). The hexane/ether extract showed a high contact (LD50 49.83 µg/adult, LD90 82.32 µg/adult at 24 h; LD50 43.58 µg/adult, LD90 77.85 µg/adult at 48 h) and inhalation (LC50 113.51 mg/L; LC90 171.16 mg/L) toxicities; the latter almost comparable to that of amygdalin enzymatic hydrolysis (LC50 17.61 mg/L; LC90 30.34 mg/L). Acetone extract induced only a slight inhalation toxicity (LC50 754.14 mg/L; LC90 1131.01 mg/L) whereas no toxicity was found for both water and methanol extracts. The RIs to the highest dose of different extracts were not significant, indicating neutral effects on insect orientation. In conclusion, among different loquat seed extracts here tested, the hexane/ether extract is the most promising in the searching of bioactive compounds against *S. granarius*.

KEY WORDS: loquat seed, inhalation toxicity, contact toxicity, behavioural response, granary weevil.

POSTER

Olfactory responses of *Sitophilus granarius* adults to wheat kernel volatiles of yellow commercial varieties and pigmented genotypes

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The granary weevil, *Sitophilus granarius* (L.) (Coleoptera: Dryophthoridae), is a primary pest of stored cereals worldwide. The identification of sources and mechanisms of host-plant resistance to specific insect pests is a fundamental step toward the breeding of new varieties. Evaluation of the resistance of wheat varieties to stored-product insect pests has most commonly focused on various physio-chemical factors such as kernel size, hardness, protein and glucid content, whereas the roles of the kernel volatile compounds (VOCs) have received little attention. This was probably due to the low genetic variability among commercial varieties resulting in very similar VOC blends. In this study, the olfactory responses of *S. granarius* adults to kernel VOCs of commercial durum (Ofanto) and bread (Mec) wheat varieties with yellow pericarp were compared with those to kernel VOCs of anthocyanin-pigmented durum (ELS6404-149-2, ELS6404-77-2, Mog, Worldseed-3) and bread (Sebesta Blue-2) wheat genotypes of Ethiopian origin that have never entered commercial production. Kernel head-space VOCs of different varieties and genotypes were collected by solid-phase microextraction and identified by gaschromatography coupled with mass spectrometry (GC-MS). The olfactory responses of granary weevil adults to odors of kernels of the different varieties and genotypes and their hexane extracts were investigated in two-pitfall bioassays. Moreover, the perception of VOCs present in the kernel hexane extracts by the adult granary weevil antennae was confirmed by electroantennography (EAG). GC-MS analysis highlighted 17 and 13 kernel VOCs (aldehydes, alcohols, terpenes, benzene derivatives) from durum and bread wheats, respectively. Quantitative and qualitative differences were seen between the odor profiles of yellow and pigmented wheat kernels. In two-choice behavioral bioassays, granary weevil adults were significantly attracted (Response Indices, positive and significant at P = 0.05, t-test) by the odors from the yellow commercial wheat varieties and their hexane extracts, but not by those of the pigmented wheat genotypes and their hexane extracts. EAG tests confirmed the presence of VOCs in all of the hexane extracts that stimulated the olfactory system of the granary weevil males and females. This indicated that differences among the odor blends were responsible for the different behavioural responses of granary weevils to the yellow and pigmented wheat kernels. In further behavioural bioassays, the majority of compounds responsible for odor profile differences among yellow and pigmented wheats (1-pentanol, 1-octanol, isobornyl acetate, aromadendrene), as determined by Principal Component Analysis (PCA), elicited repellent effects (Response Indices, negative and significant at P = 0.05, t-test) even at low doses. Differences in VOC emissions and olfactory responses induced in granary weevils by the yellow and pigmented wheat kernels can be exploited to characterize resistance mechanisms and to incorporate resistance into improved varieties.

KEY WORDS: granary weevil, pigmented wheats, VOCs, EAG, insect behaviour, plant resistance.

ORAL PRESENTATION

Comparison of diets for the rearing of *Acheta domesticus*, species for human consumption

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The consumption of edible insects for human nutrition is gaining an increase interest in the debate on the environmental challenges related to the production of food, especially protein, and on food safety. While the growing population requires more food production, intensive farming and the exploitation of environmental resources causes more pollution, also influencing climate change. The consumption of insects is now considered an alternative to supplement the major requests of proteins for humans and for animals. *Acheta domesticus* (Orthoptera: Gryllidae), known as the house cricket, is among the species of greatest interest for human consumption, thanks to its high protein content. Mass cricket farms, present in different parts of the world, use chicken feed or cricket formulations containing cereals as rearing diet. In the present project (MAIC "Model for the rearing of edible insects" – funded by Fondazione CARIPLO) we investigate the use of low-cost diets, in terms of environmental and economic impact, consisting of waste ingredients and by-products from the agri-food industry, for the rearing of *A. domesticus*. The growth performance of *A. domesticus* on different substrates were assessed as well as the presence of any risks for human health and the environment. The by-products/wastes taken into consideration were the brewery's spent grain, the trub (brewery waste), the maize distiller, the marc, unsold fruit and vegetables coming from the large-scale retailers, the soybean hulls and the rice hulls. The substrates, alone or mixed with other raw materials commonly used in animal feed, were provided to newly emerged crickets until the adult emergence. During the tests, survival, growth rate, weight achieved by crickets and efficiency of biomass conversion were assessed. Chemical and microbiological analyses were conducted on the growing substrates and the cricket meal to evaluate the possible presence of pathogenic microorganisms and the level of contamination. Moreover, nutritional analyses were carried out to assess the chemical composition of the growing diet and the cricket meal. Experimental tests revealed that all diets allowed *A. domesticus* to reach the adult stage, with significant statistical differences among the diets. However, there was a high mortality that influenced the final data. It is therefore necessary to investigate other by-products/wastes and their possible mixture in order to efficiently meet the nutritional needs of *A. domesticus*. Relating to chemical and microbiological safety, all the analyses have led to results below limits expected for feed used in the feeding of animals intended for the production of food. The results acquired within the project contribute to the poor information available to date on the optimization of diets for the mass rearing of *A. domesticus* which include wastes from the agri-food sector able of reducing production costs and the environmental impact of the farm, allowing a recycling of waste and its valorisation.

KEY WORDS: edible insects, crickets, growth performance, by-products.

ORAL PRESENTATION

Suitability of brewery's waste streams for the rearing of the Black Soldier Fly *Hermetia illucens*

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The increasing population and food demand, has led to an increase in the agro-food activities, causing irreversible exploitation of natural resources. Recently, the mass production of edible insects has received a lot of interest as protein source for livestock feed and as a mean for the management and recycling of by-products coming from the agro-food industry. Indeed, the use of food wastes (e.g. vegetables, maize distiller grains, kitchen waste) as growing substrate, in particular for Diptera, has been assessed by numerous studies and features a valuable and sustainable solution for the insect mass rearing: by products are often available in large quantity at low cost and still have a nutritionally valuable composition. The black soldier fly Hermetia illucens (Diptera: Stratiomyidae) is a species of particular interest as its voracious larvae can be mass reared on a wide range of organic substrates and are rich in proteins and fats. In the present study we investigated the suitability of brewery by-products, in particular spent grain and trub, as feedstock for larvae. Feeding trials were conducted starting from young larvae, providing the brewery wastes separately or mixed, and using a standard diet as control. We evaluated the effects of the rearing substrates on larval growth, larval and pupal weight, preimmaginal survival, and adult traits (in terms of sex-ratio, dimension, and female fertility). Feeding diets significantly impacted different H. illucens biological traits. Larvae reared on the brewery wastes provided together, took less time to reach prepupal stage and showed the highest weight, comparable to the control diet; also the mortality was the lowest on this mixed substrate. Adult morphological characteristics confirmed the best results when the two by-products were mixed, while egg clutch weight and the number of eggs per clutch were less influenced by the diet. Our results are in line with the performance obtained on other growing substrates and highlight the feasibility of using brewery wastes as growing media for H. illucens larvae contributing to the recycle of nutrients and valorising these by-products. More investigations are necessary to assess the larval bioconversion efficiency and the waste reduction of these substrates and the final nutritional composition of the larvae grown on brewery wastes.

KEY WORDS: by-products, larval feeding, biological traits, bioconversion.

POSTER

Biocomposting process of various mixtures of agro-food wastes by the black soldier fly *Hermetia illucens*

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Hermetia illucens L. (Diptera:Stratiomyidae) is a cosmopolitan insect native of the American continent, whose biological cycle is short. This insect is greatly adaptable to different climatic conditions and its larva is highly polyphagous and able to feed on almost all kind of waste and by-products from the agro-food industry. Indeed, *H. illucens* has been used for the composting of organic waste, cattle and chicken manure. Several studies showed the biological potential and versatility of *H. illucens* to degrade different kinds of organic materials, reducing it up to 70% of its initial volume. The Project "Biocompostaggio degli scarti dell'industria agroalimentare mediata da insetti: sviluppo di un processo innovativo di bioconversione e valorizzazione" - POR CALABRIA FESR-FSE 2014-2020 Azione 1.2.2 funded by Regione Calabria, aimed to evaluate biocomposting process of different mixtures of agro-food wastes and by-products, like chicken manure, exhausted olive pomace and leaves, carried out by *H. Illucens* larvae. This study focused on the identification, at optimal climatic conditions, of the best mixing ratio of the employed matrices for the biocomposting process, as well as of the biological responses of this insect at the end of the trials. Six different mixtures, apart from exhausted olive pomace and chicken manure alone, were used as trophic substrates for *H. illucens* development. The efficiency of the biocomposting process was evaluated by measuring, according the methodology described by ANPA (2001), the following physicochemical characteristics: pH, humidity, dry weight, conductivity, C and N content. The evaluation of the insect biological parameters was carried out recording the following morphometric and biological parameters: pupae dimension and weight, average days to complete the biological cycle, number of larvae reaching the pupal stage. The results showed that at controlled climatic conditions (light intensity 8000 Lux, and wavelengths between 450 and 700 nm, temperature 26±1 °C; R.U. 65±5%; photoperiod 12/12) the biocomposting process concludes in 15-20 days, and at the end of the trial the majority of the larvae reach the pre-pupal stage.
Furthermore, considering the legal limitations for the physicochemical parameters for composts, the mixture constituted by 25% of chicken manure and 75% of exhausted olive pomace is the best for the biocomposting process performed by *H. illucens* larvae. Among the tested mixtures, according the insect parameters the best one is the mixture constituted by 50% of chicken manure and 50% of exhausted olive pomace. With this substrate, the pupae measured 1.77 cm in length and weighted 0.20 g in average. Lastly, almost all the mixtures showed an emergence close to 100% and the adult flies emerged generally between day 10-15 from the end of the trials.

KEY WORDS: black soldier fly, bioconversion, olive pomace, manure, compost.

POSTER

susceptibility and attractiveness of ancient and modern durum wheat varieties toward *Rhyzopertha dominica*

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The cultivation of ancient wheat varieties has expanded considerably over the last decade, as well as the request from consumers who consider these grains healthy and more authentic. However, the problematics about the conservation and the protection of this kind of food are the same of those linked to the so-called modern varieties. *Rhyzopertha dominica* F. (Coleoptera: Bostrichidae) is a cosmopolitan primary-pest, able to damage intact kernels. It is a polyphagous species, particularly harmful to cereals and legumes, but also to other foodstuff containing starch. *R. dominica* causes different levels of damage depending on the physicochemical characteristics of the trophic substrate, which may affect the progeny production. It has been acknowledged that *R. dominica* females can produce different offspring even on different varieties of the same cereals. This study aimed to evaluate the damage and the progeny production by this insect in 6 varieties of durum wheat, three modern (Antalis, Iride and Simeto) and three ancient (Timilia, Russello and Perciasacchi) and to determine the attractiveness of olfactory stimuli toward *R. dominica* females. Progeny tests were conducted for 63 days in order to evaluate the total progeny produced. For every variety, 10 replicates were provided and the number of adults, weight loss due to *R. dominica* feeding activity and the percentage of damaged kernels were recorded. The results showed that the ancient and modern grains had similar susceptibility to the pest infestation. The greatest weight loss due to the trophic activity of *R. dominica* was reported for Russello and Perciasacchi varieties, although no significant difference was found for the percentage of damaged kernels in all tested varieties. Moreover, considering the progeny production, the modern wheat Antalis and the ancient Timilia were statistically less susceptible than the other four varieties tested, showing that the progeny generated was related to weight loss. Concerning olfactory preferences, the varieties were compared in choice-test arena. R. dominica females were able to choose using only the olfactory stimuli emitted by the different wheat varieties, although generally females did not show preferences for one of the two proposed stimuli. Just when testing Timilia variety, *R. dominica* females preferred the modern varieties Iride and Simeto over this one. In contrast, when Timilia and Antalis, the two less susceptible varieties to pest damage, were compared, no preference was noticed. These results underlined the ability of *R. dominica* females to assess the quality of food substrates for oviposition through the chemical stimuli emitted by kernels.

KEY WORDS: stored products, cereals, internal feeder, progeny, weight loss.

ORAL PRESENTATION

Monitoring and controlling insects infesting the Herbarium of the Palermo botanical garden

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The herbaria are scientific tools of great importance that preserve extinct, rare, endemic, and common plant species, showing a valuable significance for their historical and aesthetic value as cultural heritage. The herbaria can be subject to attack by different insect species that damage dried plants (exsiccata) with their feeding activity. The control of these infestations is often aggravated by the difficulty of being able to put in implement adequate insecticide treatments. In this context, it is appropriate to use successful monitoring and control tools based on pheromonal or food attractants to be included in the integrated herbarium management. In this study, an evaluation of the main insect pests of the herbarium of the Palermo Botanical Garden was carried out, in order to identify the main entomological problem. In addition to the key pest represented by the anobiid beetle *Lasioderma serricorne* F., several specimens of Psocoptera (*Liposcelis* spp.), Thysanura (*Thermobia domestica* Packard and *Lepisma saccharina* L.) and some Hymenoptera belonging to the Formicidae family were found. Two experimental tests were therefore carried out on *L. serricorne*. In the first test, two pheromone dispensers (polyethylene and patch) were tested both in terms of emission over time and in the effectiveness of the traps. In the second field test, a food attractant consisting of chili powder was added to the pheromone, in order to evaluate its effectiveness as a synergist of the pheromone. The test results showed that the polyethylene dispensers released the pheromone more gradually and resulted in a greater number of catches in the traps. Finally, the use of chili powder in pheromone traps has determined a significant increase of catches compared to the catches observed in the traps loaded with pheromone only.

KEY WORDS: herbarium, *Lasioderma serricorne*, pheromone, Chili.

POSTER

Light-traps in shipping containers: a new tool for the early-detection of stored food insect pests

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Insects represent one of the most successful groups of invasive species, and the number of introductions of new alien species has been steadily increased in Europe in the last decades. In particular, stored food insect pests include the largest number of cosmopolitan species whose introduction and spread to new continents is mainly influenced by the increase in the speed and volume of international trade, which takes place mainly through shipping containers. The aim of this study is to test the effectiveness of a sticky trap, made attractive by a battery light, in catching food pests transported inside containers during shipping. In particular, we tested the attractiveness of the light-trap to different orders of insects, and how the conditions of the container (empty or loaded) can affect the trapping performance. The tests were conducted on three seed and fruit pests: *Cadra cautella* (Walker) (Lepidoptera: Pyralidae), *Drosophila melanogaster* Meigen (Diptera: Drosophilidae) and *Sitophilus zeamais* Motschulsky (Coleoptera: Curculionidae) released individually (50 individuals per test) inside a standard shipping container where the trap (TransTrapTM) was tested under empty and loaded container conditions (simulating the real shipment situation). For each test, an active trap (with light on) and a control trap (with light off) were tested in the same container. For each species and container conditions (loaded and empty) 7 tests were performed. In the empty container activated traps (light on) recaptured significantly more released insects than the control trap (light off) for *C. cautella* (27% vs 4%) and *D. melanogaster* (31% vs 2%), while *S. zeamais* were recaptured only in small numbers and without significant differences between the two trap types (<1%). Even in loaded containers, the average recapture rate of *C. cautella* (20% vs 5%) and *D. melanogaster* (27% vs 4%) was significantly higher in activated traps than in the control ones, with no significant differences between loaded and empty containers. As a result of low catches in empty container, *S. zeamais* was not tested in loaded container. In conclusion, results show that even in loaded container conditions this trap is effective in catching this two species of Lepidoptera and Diptera, whereas it does not seem to work as well for this Coleoptera species. However, it still unclear whether the problem in recapturing beetle is related to a lower flying ability of this insect, or to its lower response to light stimuli, or finally to its possible ability to escape from the sticky trap. The low recapture rates of *S. zeamais* seem to be due to the glue in the trap, which should be too weak for trapping this insect. However, this type of trap appears to be a promising tool for the early interception of small insects during the transport of foodstuffs in containers.

ORAL PRESENTATION

Food waste coming from catering services bioconversion by *Hermetia illucens* larvae

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In 2011, it was estimated that nearly one third of the food intended for the human consumption (1.3 billion tons per year) was lost or wasted through the food supply chain. Food loss and food waste differ between the developing and the developed countries and between rich and poor consumers. According to the latest estimates, in the EU countries 173 kg pro-capite are wasted each year. The household sector is even responsible for 53%, while the catering sector has an impact of 15%. The Black Soldier Fly, *Hermetia illucens* (L.) (Diptera: Stratiomyidae) can reduce and convert a significant amount of organic substrates. Food waste could represent a worthwhile alternative for *H. illucens* feeding and, at the same time, negative environmental and economic impacts due to food waste management could be avoided. It is known that sometimes food wastes are incinerated or taken to the landfill, instead of being converted into compost, moreover, from the larvae chitin, proteins, lipids and other products can be extracted, and used, in many cases, as animal feed. In this study, food wastes from the canteen (both from kitchen preparations and left-overs from trays) of the Università Cattolica del Sacro Cuore, Piacenza campus, have been collected in order to analyse and quantify them. Furthermore, a fraction of the waste has been utilized for diet composition in order to test how different feed can influence growth and protein contents in *H. illucens* larvae. Different substrates have been used, even in different composition, such as coffee grounds, melon peel, bread mixed with vegetables, meat, fish and cheese. Trials including whole canteen waste from the trays at the end of the lunch have been carried out too. Newborn larvae were fed on standard diet for 7-8 days and then transferred on the tested substrate where they fed for 10 days. At the end of this stage they were sifted, weighted and dried aiming at detecting protein concentration. Meaningful results in growth and protein concentration, which look similar to the ones produced with standard diet, were obtained from substrates that have a better balance in terms of energy, proteins, suitable humidity and texture, as the whole canteen food waste. In connection with this, catering waste could represent an interesting solution in the future for the breeding of *H. illucens*.

KEY WORDS: *Hermetia illucens*, food waste, by-products, catering, feeding.

POSTER

Animal welfare in Acheta domesticus farms, preliminary study on density

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For more than ten years, the scientific community has continued to provide evidences regarding the reduced environmental impacts generated by the production of insect-based foods. Compared to traditional zootechnical animals, insects are characterized by an extremely efficient Conversion Factor (feed consumed / live weight produced), therefore they require reduced quantities of feed, water and land, to generate a food consisting of a high quantity and quality of proteins, vitamins and microelements. Insect farming at agro-industrial level is fully integrated in the agricultural policies of the European Union and, as a consequence, in the next few years there is expected to be a sharp increase in insect farms and in the market linked to them. With the development of this new sector, in addition to the relative assessments of actual environmental, economic and social sustainability, various questions arise with respect to animal welfare. The high productive efficiency of insects is mainly due to the high breeding density, which is a sine qua non condition for reducing economic-environmental costs and generating virtuous scale production. Although the farmed species are characterized by a reproductive strategy more R than K and do not show marked territorial behaviors, we wondered what was the correct population density for a farm of *Acheta domesticus*, which guarantees high productivity and at the same time respects the second and fourth freedom on animal welfare (Brambell Report, 1965): "freedom from discomfort by providing an adequate physical environment" and "freedom to express normal behaviour". The study was carried out at the Italian Cricket Farm S.r.l., an agro-industrial farm of *A. domesticus*. Nymphs of the first age were raised in four specific tanks; one with the optimal density found in literature (0.4 individual/cm2) and the others with densities higher than this (0.8 ind./cm2, 1.2 ind./cm2, and 3.2 ind./cm2). Throughout the development process, controlled environmental conditions and a constant supply of food and water were ensured. When the adult stage was reached (65 days old), the number of individuals and the average size of the adults were measured for each tank. The low-density condition has been found to be the one in which there is greater uniformity in size, a larger average size and a lower preimaginal mortality rate. While in the tank with a greater density of individuals there was instead a slower growth, and the adults are of uneven size. In all likelihood, a lower breeding density guarantees a reduction in competitive phenomena linked to accessing the resources and a reduction in fights and cannibalism typical of the species.
Although preliminary, this study opens up the issue of welfare/return in insect farming and sets up a rigorous methodology to assess objective parameters.

KEY WORDS: Animal welfare, insects farming, agri-food sustainability, *Acheta domesticus*, house crickets, entomophagy.

ORAL PRESENTATION

Optimization of *Hermetia illucens* larval production on catering and cafeteria waste

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The Italian cuisine and cafes are internationally recognized. Indeed the restaurants and bars are highly frequented places by both Italians and tourists, and this involves an inevitable generation of large quantities of organic waste. For a full valorization of these substrates, they can be employed in a sustainable way for insect rearing. The black soldier fly (*Hermetia illucens* L.) larvae can convert a large volume of waste into a biomass rich in protein and fat, which can be used for feed and other industrial purposes, while the residual substrate can be employed as soil improver. The present study aims to evaluate whether the addition of coffee grounds might improve the texture of the HoReCa (hotel, restaurant, catering) waste, which usually has a high viscosity, making it more suitable for an industrial process of mechanical separation of the mature larvae from the frass. Moreover, there were evaluated the effects of coffee ground addition on the development and production parameters (development time, larvae survival, substrate reduction, conversion efficiency and larval migration rate). The HoReCa waste substrate (S-HoReCa) used in these experiments was provided by a local canteen and its composition varied according to the daily menu and consumer preferences. Therefore, at laboratory-scale three consecutive trials were carried out, each replicated five times. The ratio between S-HoReCa and the coffee grounds used, were established on the basis of volume, taking 50 ml Vol as the measurement unit. In the first experiment two treatments were tested, both with 9 Vol. of S-HoReCa to which were added: a) 4.5 volumes of coffee grounds and b) 3 volumes of coffee grounds. In the second experiment a ratio of 9 to 1.5 Vol. was tested. Finally, to optimize the production yield, in the third experiment a ratio of 16 V S-HoReCa to 2 V coffee grounds was used, while increasing the larval density and decreasing the amount of substrate administered per larva. The results showed that a higher concentration of coffee grounds in the S-HoReCa used as feeding substrate has a negative influence on the weight of the mature larvae, although it has a positive influence on their survival. Besides, while it facilitates the separation process of the larvae from the frass, it also decreases the percentage of larval migration outside the substrate. As foreseen, also the variability of the nutritional composition of the S-HoReCa influenced the larval development. In conclusion, the addition of small quantities of coffee grounds can be a good compromise to obtain a higher percentage of larval survival, especially when the dry mass content of S-HoReCa waste is lower than 16%, and as well, to optimize the separation process of mature larvae from the residual substrate. This work is part of the project SCALIBUR (Scalable Technologies for Bio-Urban Waste Recovery – H2020-SFS-2018-2020), funded by European Commission.

KEY WORDS: coffee grounds, food waste, Black soldier fly, circular economy, larval development parameters, production parameters.

POSTER

Unwelcome guests at farms breeding the black soldier fly, *Hermetia illucens* (L.) (Diptera Stratiomyidae)

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Livestock farms represent a source of attraction for other species, which find food resources both directly on the animals themselves and indirectly in the food supplied to them, in their manure, in other organic substances present, etc. Insect farms are no exception; indeed, the substrates used for feeding juvenile stages and/or adults turn out to be palatable to other organisms.
The breeding of the black soldier fly, *Hermetia illucens* (Diptera Stratiomyidae), whenever it is done in small or large structures, requires the use and presence of organic materials which are attractive for other species of arthropods (commensals, mycetophages, scavengers etc.); furthermore any antagonists of *H. illucens* itself cannot be neglected. During recent years, the breeding system adopted at the Di.Pro.Ve.S. of the Università Cattolica del Sacro Cuore, Piacenza, has shown the presence of the following insect and mite species: *Megaselia scalaris* (Loew) (Diptera Phoridae), *Muscina stabulans* (Fallén) (Diptera Muscidae), *Monomorium pharaonis* (L.) (Hymenoptera Formicidae) and *Caloglyphus berlesei* (Michael) (Astigmata Acaridae).
The use of fermented fruit and vegetables, but also of an artificial diet, consisting of rehydrated chicken feed, in order to attract females and induce them to lay eggs, also entails the attraction of small flies such as *M. scalaris*. Infestations by *M. scalaris* have been rather worrying, since the larvae of this species have shown to be very competitive with respect to those of the black soldier fly in substrate colonization. Moreover, this species takes advantage of the sugar water available to *H. illucens* for adult feeding. The substrate, on which the larvae of this stratiomyid develop, also attracts flies such as *M. stabulans*, although this species has not shown the same high level of competitiveness as the two previously mentioned species. *M. pharaonis* lives in small colonies, growing up in small openings and crevices and also the small bars used for egg collection can be colonised, where it preys on the eggs and newborn larvae of *H. illucens*. Lastly, infestations - in some cases very significant - by the mite *C. berlesei* were detected when conditions for the larval development of the black soldier fly are not optimal; caution should be exercised because this mite could be harmful for the workers involved in the breeding.
The possible presence of insect and mite pests inside the *H. illucens* rearing facilities suggests that a careful analysis has to be made according to the location of the breeding facility and, of course, a series of measures has to be adopted when this kind of structure and activities are designed and realized.

KEY WORDS: *Megaselia scalaris, Muscina stabulans*, *Monomorium pharaonis, Caloglyphus berlesei, Hermetia illucens,* breeding, pest.

POSTER

Could Hermetia illucens' larvae be used as processing tool?

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Insect farming is experiencing great development all around the world, drawing the attention of several operators in the food and feed sectors, but also in completely different industrial sectors. This activity fits perfectly in the context of circular economy.
Hermetia illucens (L.) (Diptera: Stratiomyidae) is the most considered insect species, especially for its high potential in different fields.
SCARABEO project (PSR 2014-2020 Op. 16.1.01 - GO PEI-Agri - FA 5C, Pr. "5015701 - Scarti di CAnapa - Riutilizzi Alimentari e Biovalorizzazione Energetica degli Oli'' coordinated by Azienda Agraria Sperimentale Stuard S.C.R.L. – Parma) is about the use of hemp for textile purpose where *H. illucens* larvae have been used to remove sugars (mainly hemicellulose and pectin) coated on the fibre, as an efficient alternative to traditional technique, which involves a significant amount of water at high pressure, thus ensuring an important reduction of energy used.
In the perspective of circular economy, trials in the SCOOTER project (PSR 2014-2020 Op. 16.1.01 - GO PEI-Agri - FA 5C, Pr. "5015700 Scarti Colture Orticole: Opportunità nella Trasformazione Energetica e nel loro Riutilizzo'' coordinated by Università Cattolica del Sacro Cuore) have been carried out in order to evaluate the waste enhancement in fruit and vegetable companies, through the conversion undertaken by *H. illucens* larvae. They have been able to reduce on average 82% of the waste initial volume, furthermore they have limited the bad smell that wastes could produce if allowed to rot without control or composted.
Another interesting aspect is entomoremediation: Ewuim (2013) describes the use of mass insects farmed and released in the soil as a solution for its decontamination. A hypothetic scenario including *H. llucens* would be different, at least for the temperate climate countries, it would be necessary to breed them in facilities with T and RH controlled on substrates to be decontaminated.
However, it should be noted that, according to the Regulation (EC) No. 1069/2009, insects are considered as "farmed animals" and, therefore, they must only be fed with products permitted in the production of feed.
Within the legislative framework governing insect farming in Europe, if the use of *H. illucens* as processing tool makes its way through, for instance to clean the hemp fibres or to convert substrates contaminated by mycotoxins or heavy metals, it would be necessary to change the rules. For example to include the possibility to feed them with substrates which are outside the feed regulation, acknowledging them as process aid and, of course, maintaining rules that can guarantee their welfare along all phases.
If this happened also in Europe, it would open more possibilities, which could widely contribute to the circular economy and could make the insect farming sector more economically attractive.

KEY WORDS: *Hermetia illucens*, larvae, process aid, hemp fibre, vegetable wastes.

ORAL PRESENTATION

Characterization of fatty acid profiles of *Tenebrio molitor* larvae reared on diets enriched with several edible oils

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Several species of insects have been indicated as suitable and alternative sources of nutrients for humans and animals. However, their nutritional composition and development time can be influenced by the substrate used for rearing. The main objective of this study is to evaluate the fatty acid composition of *Tenebrio molitor* (Coleoptera: Tenebrionidae) larvae as a function of different rearing substrates. For this purpose a standard diet (51.8% wheat bran + 40.7% soybean meal) was alternatively supplemented with addition of 7.5% of sunflower, linseed or fish oils. Percentages of total lipids, protein and fiber, as well as development time of larvae were determined, highlighting no differences between insects raised on different diets. Gas-chromatography was used to evaluate the fatty acids profile of larvae and substrates. A total of 15 fatty acids were identified and quantified. Fatty acids composition was significantly influenced by the rearing substrate, although correlation with diets was observed only for 4 fatty acids. Specifically, the level of eicosapentaenoic acid and alpha-linolenic acid in larvae were strictly dependent by their concentration in the rearing medium. Instead, the palmitic acid, oleic acid and linoleic acid presented a concentration that was not significantly affected by the substrate. Additionally, these were the most abundant fatty acids in mealworm larvae. The n6/n3 ratio was also influenced by the substrate, showing a similar trend between insects and diets but with more equilibrate values in larvae. Principal component analysis on the fatty acid profiles of larvae evidenced several clusters based on different growing media. In conclusion, this study confirms that the fatty acids profile of mealworms can be easily manipulated by the diet, suggesting the possibility of obtaining *T. molitor* larvae with relevant levels of healthy fatty acids, thus being useful to promote human and animal health.

KEY WORDS: Tenebrio molitor, fatty acids, linoleic acid, alpha-linolenic acid.

ORAL PRESENTATION

Damages of *Necrobia rufipes* in pet food packaging and set up of a monitoring trap

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Necrobia rufipes (De Geer), a coleopteran species belonging to Cleridae family, is a new emerging pest of pet food stores. This beetle has a cosmopolitan distribution and attacks generally protein-rich animal products (dried fish, cured ham, bacon, cheese) and, as it can be found on carrions, is also considered in forensic entomology to evaluate the post mortem interval. In Mediterranean countries the presence of this pest has been reported associated with pet food chain starting from 2014, while in Italy it was found in 2016. Pet food products are rich in animal protein content and are particularly susceptible to *N. rufipes* infestation that is able to penetrate the common packaging used. Moreover, in consideration of the aggressiveness of this species, the development of appropriated monitoring instruments is highly needful to reduce chemical treatments for its control. Objectives of this study were to limit *N. rufipes* damages through the packaging improvement and also to set up an appropriate trapping tool for monitoring this beetle. In particular laboratory tests were carried out on woven packaging with *N. rufipes* larvae and adults to check for any insect access point. To develop a monitoring device two types of commercial glue (hot-melt adhesive and polybutene adhesive) were evaluated trough laboratory observations for their ability to retain the caught insect into the trap avoiding escapes and finally behavioral bioassays were carried out in dual choice arena to test several food attractants candidates.
The study evidenced that *N. rufipes* enters in packaging through the air vent valves on its bottom, suggesting that a way to improve the packaging to prevent insect infestation would be to modify these points of weakness. Laboratory tests showed that the different glues bioassayed have strong differences in the ability to retain the caught insects, with the polybutene adhesive more effective than hot melt one. Finally, the results of duals choice arena bioassays showed that among the candidate attractant tested, a mixture of pet food and methylcyclopentenolone elicited the strongest attraction in *N. rufipes* adults. These results are an encouraging first step in the perspective of further experiments to carry out in pet food storages to test the efficacy of this tool for trapping *N. rufipes* adults.

KEY WORDS: red-legged ham beetle, packaging, adhesive traps, food attractants.

ORAL PRESENTATION

Hermetia illucens and microalgae for a quality feed production in a circular economy model

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The SMART-Feed (Sustainable Model for Agroenergy and Feed production by Urban Waste Recycling and Treatment, funded by the CARIPLO Foundation) project proposes a Circular Economy model for the production of a high value-added feed consisting of *Hermetia illucens* larvae (Diptera: Stratiomyidae) reared on the organic fraction of municipal solid waste, and microalgae produced by recovering the nutrients contained in the digestates, following anaerobic digestion of the residual organic fraction. The latter is also valorised for biogas, electricity and heat production used for the energy requested by the production process itself. Finally, the digestate will be used as fertilizer by replacing the use of chemicals through precision farming. The larvae of *H. illucens* are able to grow on different types of wastes, contributing both to their disposal and to their conversion into larval biomass with a high nutritional value, being rich in fats and proteins. *H. illucens* can be used for different purposes, for example as feed, biofuel and biofertilizers. Microalgae, on the other hand, have been identified as a valuable feed supplement for livestock, thanks to their high protein content, the value of amino acids, vitamins, antioxidants and other substances useful for animal health. The SMART-Feed project therefore aims the feasibility of producing a sustainable feed, which includes larvae and microalgae, intended for consumption by broilers. A holistic and multidisciplinary approach will allow to deepen the numerous aspects of the whole process. The objectives of the project include: 1) characterization of the organic fraction of urban waste with particular reference to the biological value of the organic components; 2) production of insects and microalgae on the organic fraction of the municipal solid waste and their characterization; 3) production of the SMART-Feed, evaluation of its quality and safety, in vitro and in vivo digestibility for broilers; 4) production and evaluation of biogas and fertilizers deriving from the organic fraction of the residual municipal solid waste; 5) estimation of the environmental impact of the entire circular production system; 6) economic evaluation of the impact of the SMART-Feed obtained on the regional economy. Particular attention will be paid to the quality of the feed and the hygienic-sanitary characteristics of the final product.

KEY WORDS: bioconversion, black soldier fly, microalgae, organic fraction of municipal solid waste, circular economy.

POSTER

Assessment of quantitative and qualitative response of yellow mealworm larvae (*Tenebrio molitor*) reared on wheat bran or brewer's spent grain

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Mass insect rearing is becoming increasingly pursued by food and feed industry due to its high sustainability and low environmental impact. Yellow mealworm larvae (*Tenebrio molitor*; TM) are conventionally reared on wheat bran (WB), but alternative substrates, such as brewer's spent grains (BSG), a by-product of the brewer industry, have shown good perspectives for insect rearing. The objective of this study was to investigate on quantitative, nutritional and metabolic response of yellow mealworm larvae to dried BSG and WB used as rearing substrates. TM larvae were fed either WB (WB-L) or BSG (BSG-L) in trials reproducing different scale rearing condition. Feed efficiency parameters were determined for WB-L and BSG-L. Proximate composition (water content, protein, fat, chitin and ash) of WB-L and BSG-L were compared. BSG-L group exhibited better feed conversion ratio and efficiency in conversion of ingested food. BSG-L showed almost half fat content than WB-L while chitin was higher than WB-L. No differences were observed in protein contents. These results increasing knowledge about the growth and body composition yellow mealworm larvae reared on different substrates and pave the way for the use of alternative by-products from the agri-food industry e as insect feed.

KEY WORDS: mass rearing insects, food conversion, proximate composition, agri-food by-products.

ORAL PRESENTATION

Screening of some agroindustry by-products as feed substrates for *Tenebrio molitor* larvae

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Yellow mealworm (*Tenebrio molitor* L.) represents an alternative highly sustainable source of proteins for food and feed. Industrial production must count on optimized processing methods also based on a deep knowledge of nutritional and quality aspects of rearing substrates. In this research the effects of different rearing substrates on the development of larvae were compared. These were selected among the main byproducts of the agro-industry. Wheat middlings, wheat bran, rice bran, hemp cake and thistle cake (the latter coming from the green chemistry chain) brewer's spent grains, tomato pomace, grape marcs, and rusk crump remains were compared. Brewer's spent grains, grape marcs and tomatoes pomace were dehydrated and ground to make them suitable for the consumption of mealworms. The test results evidenced three groups of substrates in relation to the growth performance of the yellow mealworm: the most performing group, consisting of wheat bran, wheat middlings and hemp cake showed the best performance in terms of fewer days to reach pupation (about 14 weeks), higher final larval weight (about 0.13 g) and lower mortality (from 83% to 91%); the second, consisting of brewer's spent grains and tomato pomace showed intermediate performances (about 16 weeks to reach pupation, 0.12 g of final larval weigh 78 and 80% of mortality); the third and worst group, consisting of rice bran, thistle cake, grape marcs and rusk crump remains, evidenced a slower larval development (from 19 to 26 weeks to reach pupation), lower final larval weigh (0.11 g) and in some cases high mortality (45-75%) compared to the most performing substrates. Our findings evidenced that the nutritional composition of the substrates (protein, carbohydrate, fat, and fiber content) influences larval mortality, duration of larval development, and average larval weight. Overall, the results obtained encourage further efforts in the valorization perspective of agro-industrial by-products, singly or in mixture, as rearing substrates for the yellow mealworm in order to obtain alternative protein and lipid matrices, to be used for the production of food and feed, safeguarding biodiversity and making more sustainable terrestrial and aquatic livestock productions.

KEY WORDS: yellow mealworm, mass rearing, agri-food byproducts, larval growing, larval mortality.

POSTER

Nanostructured alumina against pulse seed insect pests

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Insect pests constitute a major threat not only to stored grains and legumes for food but also to the seeds for sowing. Seeds, usually stored in relatively small quantities and in separate packages, are particularly susceptible to insect attack because of their long period of storage. Moreover, due to the global market, seeds insect infestation promotes the spread of invasive stored food pests. Numerous studies showed the effectiveness of nanoparticles against various arthropod species of economic importance. Due to their small size (1-100 nm), nanostructured materials, have been showed to have properties that are not shared by non-nanoscale particles with the same chemical composition. In this study, we evaluated the effectiveness of nanostructured alumina particles (NSA) against the main seed-infesting insect pests: *Oryzaephilus surinamensis* (Silvanidae), *Stegobium paniceum* (Anobiidae), and *Tribolium confusum* (Tenebrionidae). After sixteen days, the percentage of insect mortality at the highest NSA concentration tested (400 mg kg-1) was 100% for *S. paniceum* followed by *O. surinamensis* (80.64%) and *T. confusum* (79.41%). The median Lethal Concentration (LC50) values calculated by Probit analysis were 14.87, 61.53, and 127.17 mg Kg-1 for *S. paniceum*, *O. surinamensis*, and *T. confusum*, respectively. Besides, in vitro-tests indicated that nanoparticles have not affected seeds germination, sprout growth and root lengthening, and, by comparing the plants treated with no-treated plants, no differences were recorded in the leaf surface, stoma density and roots length. On the contrary, a positive effect of the NSA was observed on the shoot growth with the treated plants that were about 66% higher than the non-treated plants. Finally, using the EDX system coupled with the Scanning Electron Microscope (SEM), no contamination by alumina nanoparticles was found on the surface of the *P. vulgaris* leaves of plants originated from the treated seed. The efficacy of NSA against the insect pests, and the absence of negative effects on seed germination and plant growth, indicate that NSA could represent a viable alternative to synthetic insecticides, for the seeds coating.

KEY WORDS: Nanostructured alumina, Insect pests, seeds, Scanning Electron Microscope (SEM).

ORAL PRESENTATION

SESSION XI

Induced immunosuppression as a strategy to enhance insect biocontrol

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The reduction of chemical pesticide use highly contributes to food safety and environmental protection. This challenging goal has promoted intense research efforts towards the identification of new bio-inspired tools and strategies of pest suppression, using also novel biotechnologies. In this context, we have recently demonstrated that RNAi-mediated silencing of an immune gene generates an immunosuppressed phenotype and makes insect pests more susceptible to their natural antagonists. In particular, we have recently shown that the RNAi mediated silencing of a gene (102 Sl) controlling the cellular immune response in *Spodoptera littoralis* (Lepidoptera: Noctuidae) enhances the killing activity of the entomopathogen *Bacillus thuringiensis*. Here we explore a new delivery strategy of dsRNAs targeting 102Sl gene by feeding S. littoralis larvae on transgenic tobacco plants, expressing 102 Sl dsRNA. The experimental larvae showed marked immunosuppression associated with a significant transcriptional down-regulation of the target immune gene. Moreover, the resulting immunosuppressed phenotype showed a very high mortality when exposed to sub-letal doses of Bt (Xentari). Therefore, the ingestion of dsRNA molecules, delivered under realistic field conditions, has the potential to enhance the Bt insecticide activity on *S. littoralis* larvae. From a theoretical point of view, the induction of a reduced immune competence in the target pest appears to be ecologically more sustainable as it can enhance the ecological services provided by natural antagonists. Indeed, such an approach will promote the establishment and proliferation of biological control agents, rather than favoring their dispersal as a consequence of a treatment directly killing the target pest and reducing its density.

KEY WORDS: immunity, entomopathogens, *Bacillus thuringiensis*, bioinsecticides.

ORAL PRESENTATION

Carnauba wax as virulence enhancer of entomopathogenic fungi against the blowfly *Lucilia sericata*

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The blowfly, *Lucilia sericata* (Diptera: Calliphoridae), is a problematic synanthropic insect pest cause of secondary myasis, a severe medical and veterinary parasitosis, and vector of microbial pathogens. Fungal biopesticides are considered valid tools, alternative to synthetic ones. However, to date, little is known about their bioactivity against blowflies. In this work we assessed the insecticidal activity of the three entomopathogenic fungi *Beauveria bassiana*, *Akanthomyces muscarius*, and *Beauveria pseudobassiana* against *L. sericata.* In addition, we tested powdered carnauba wax (*Copernicia prunifera*) as an electrically charged carrier to enhance the virulence of the fungal spores. Pathogenicity tests on adult flies by adult immersion in conidial suspension (108 conidia mL-1) indicated *B. bassiana* as the most virulent followed by *A. muscarius*, and *B. pseudobassiana*. In contact test with dry conidia the percentage of mortality estimated by ANCOVA model (controlled factors, time and concentration: time = 7.23 days, concentration = 3.7 X 108 conidia/g) varied from 56.4 to 13.6% for *B. bassiana*, and *A. muscarius*, respectively. Overall, the mean mortality without carnauba wax was 31.9%, while with the carnauba as a dielectric carrier wax 40.3%. Overall, our results indicated *B. bassiana* as the most virulent among the tested fungi and that the use of carnauba wax as carrier is able to significantly enhance the bioactivity of the entomopathogenic fungi.

KEY WORDS: electrostatic powder, *Beauveria bassiana, Akanthomyces muscarius, Beauveria pseudobassiana,* biological control, dry formulation.

ORAL PRESENTATION

Relationships between the olive fly *Bactrocera oleae* and its endosymbiont *Candidatus* Erwinia dacicola: from basic knowledge to practical applications

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Since old times, *Bactrocera oleae* (Rossi) is considered a key pest of the olive crops in the Mediterranean basin. Its relationship with bacteria has been thoroughly studied, especially its endosymbiosis with *Candidatus* Erwinia dacicola. This bacterium is considered essential for the olive fly. It is vertically transmitted through generations and it benefits both larvae and adults in field; whereas, it had been rarely found in lab colonies, probably due to the preservatives added to artificial diets. The aim of this research was to evaluate the possibility of controlling the olive fly through the management of this peculiar symbiosis, with its interruption in the field by the use of several natural substances and favouring its maintenance in lab rearing for sterile insect technique (SIT) purposes. Regarding the first goal, several substances were evaluated in lab trials, such as copper at 5%, copper at 20% and propolis. Regarding the second objective, a horizontal transfer was set out among wild and lab adults, exposing lab flies to different sources contaminated by wild flies with *Ca*. E. dacicola. For the same goal, the effects of lab procedures on the maintenance of the endosymbiosis were evaluated, treating eggs laid by wild females with two different disinfectants: a propionic acid solution, and a mixture of sodium hypochlorite plus Triton X. Also irradiation of wild males at two different doses was evaluated. Concerning the first goal, our findings proved that all the lab-tested substances in this research reduced the symbiosis rate, even if to a different extent. So that, the same substances should be further tested in the field. For the symbiosis maintenance, we can affirm that preservatives in lab rearing significantly decreased the symbiont load on eggs, interrupting the vertical transfer whereas it was demonstrated that a horizontal transfer could occur if a wild population cohabits with flies of a lab strain. Moreover, this research contributed to set up a reliable molecular procedure for *Ca*. E. dacicola screening. Thus, these original results open further outlooks for the control of the olive fly and new opportunities to establish a permanent symbiotic colony, the starting condition for future SIT applications, improving perspectives for a sustainable agriculture and low environmental impact control strategies.

KEY WORDS: *Bactrocera oleae*, endosymbiont, SIT, copper, horizontal transfer.

POSTER

Study of chemoreception in *Capnodis tenebrionis* as a innovative tool in biological control

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Insects use chemoreception to interact with the environment through the perception of volatile molecules that is essential for their survival. Chemoreception is mediated by molecules belonging to heterogeneous families such as olfactory receptors, ionotropic receptors, chemosensory proteins and odorant binding proteins. *Capnodis tenebrionis* (Coleoptera: Buprestidae) is a pest of plant species belonging to the Rosaceae family (apricot, cherry, peach, plum). Larvae are rhizophagous and excavate subcortical galleries in the root, while adults are defoliators, feeding the foliar petioles, the bark of the branches and the base of the buds, compromising plant physiology. *Capnodis tenebrionis*, as other insects, is attracted by chemical signals emitted by plants. The control of this pest lacks of monitoring strategies such as information on insect chemical ecology. The composition of perceived volatile organic compounds (VOCs) play an important role in determining the signal specificity for *C. tenebrionis* repulsion or attraction. Since adults use VOCs emitted from leaves or shoots, volatiles emission profiles of apricot trees have been characterized and several compounds have been identified. To improve the understanding of how VOCs are perceived, putative soluble olfactory proteins involved in chemoreception (odorant-binding proteins and chemosensory proteins) have been identified from de novo transcriptomes derived from antennae and whole bodies of *C. tenebrionis* adult males and females. Transcriptome analysis led to the identification, among the others, of a putative Odorant Binding Protein named CtenOBP7, similar in terms of aminoacid sequence to *Agrillus mali* (Coleoptera: Buprestidae) OBP3, already functionally characterized in another study. CtenOBP7 was cloned, expressed and purified from *Escherichia coli* extracts, using Ni-NTA resin and used for in vitro experiments. Interaction of recombinant CtenOBP7 with different plant VOCs was measured by fluorescence spectroscopy. The adoption of a similar approach could offer guidelines to understand the mechanisms involved in the interaction between selected plants and *C. tenebrionis* adults, to evaluate the potential role of plant compounds as chemical cues that orient and attract the insect. The main objective is to define new strategies to control the proliferation of this pest, starting from the study of insect olfactory perception at the molecular level.

POSTER

Silencing of ATP synthase beta in phytoplasma vectors: effects on pathogen multiplication and egg development

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Phytoplasmas are wall-less bacteria associated with severe diseases in hundreds of plant species, producing in many cases major yield loss in several crops. Phytoplasmas inhabit the phloem elements of infected plants and are transmitted from plant to plant by hemipteran insect species, belonging to the suborders Auchenorrhyncha (Cicadellidae and Cixiidae are the families with the largest number of vector species) and Sternorrhynca (Psyllidae family). Control of phytoplasma diseases is so far mainly based on insecticide treatments against vector populations. Taking into account the impacts of insecticides on agro-economy, public health, and environment, the development of innovative and more sustainable management strategies is becoming urgent. The leafhoppers *Scaphoideus titanus* and *Euscelidius variegatus* are known phytoplasma vectors. The former species is the natural vector of the phytoplasma associated with the grapevine flavescence dorée (FD), 16SrV-C and -D; the latter is the natural vector of the '*Candidatus* Phytoplasma asteris' Chrysanthemum yellows strain, 16SrI-B, and a laboratory vector of FD phytoplasma. Previous studies indicated a crucial role of the insect ATP synthase during phytoplasma infection process. In the present work, the silencing of ATP synthase β was obtained by injection of specific dsRNAs (dsATP) in both *S. titanus* and *E. variegatus*. After the injection of dsRNAs in *E. variegatus* and successive phytoplasma acquisition on infected plants, silenced *E. variegatus* showed significant reduction of the target protein and lower phytoplasma loads compared to the not-silenced controls. An in vivo role for insect ATP synthase β could be hypothesized during phytoplasma infection process. Complete lack of progeny was also observed in several repeated experiments for *E. variegatus* insects following silencing of ATP synthase β. Consistently, altered morphology of ovaries as well as deregulation of genes involved in egg development were observed in ATP synthase β silenced *E. variegatus* adult females in comparison with specimen injected with dsGFP, used as negative controls. Our results are in line with the known role of ATP synthase in promoting germ cell differentiation, as already described in the fruit fly Drosophila melanogaster. The strong sterilizing effect obtained by silencing ATP synthase β could pave the way towards the development of innovative control approaches to limit phytoplasma vector population, although many issues still need to be addressed, such as specificity, to avoid undesired off-target effects, and efficient strategies to deliver dsRNAs to phloem-sucking insects.

KEY WORDS: flavescence dorée, *Scaphoideus titanus, Euscelidius variegatus*, RNAi.

ORAL PRESENTATION

Effects of (Z)-3-hexen-1-ol and sex pheromone combination on adult *Megaplatypus mutatus* trap catches in poplar plantations of Campania - Italy

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Megaplatypus mutatus (Chapuis) is a wood borer native to tropical rain forests of South America. The species is now established in different areas of Central and Southern Italy (Campania, Latium and Molise), starting from the first record in the Caserta province on 1999. *M. mutatus* belongs to the family Curculionidae, subfamily Platypodinae, which groups about 1500 species mainly of tropical and subtropical origin. This polyphagous pest causes considerable damage in poplar plantations and, to a lesser extent, to fruit, ornamental, and forest species, boring reproductive galleries in tree trunks and main branches. The low reproductive success, despite a mean progeny per female of about 100 offspring, is consequence of a rapid dehydration process of adults outside the galleries and a complicated courtship and mating behaviour. In Campania, reproduction starts in May-June with a peak during summer and a decrease in October-November, in relation to the host attacked. As part of phytosanitary monitoring and research activity aiming at the development of sustainable control methods against this xylomycetophagous beetle, laboratory and field trials have been conducted in Campania. To select potential bioactive compounds towards *M. mutatus* adults, electroantennographic (EAG) tests evaluating the responsiveness of male and female antennae to different (n = 26) volatile organic compounds, including the male sex pheromone components (sulcatol, sulcatone, pentan-3-ol), were performed. In addition to the sex pheromone components, strong EAG responses were also recorded on stimulation with (Z)-3-hexen-1-ol in both sexes. The possible effects of (Z)-3-hexen-1-ol on the attractiveness of the male sex-pheromone blend towards females were assessed in field trapping trials carried out in poplar plantations located in the Avellino and Benevento provinces during 2018-2019. Tests were designed to compare adult catches in funnel traps (Mastrap L®, Isagro, Italy) loaded with sex pheromone dispensers alone or in combination with polyethylene (thickness, 100 µm) bags (6 x 4 cm) containing (Z)-3-hexen-1-ol (2 mL). Using the experimental mixture, the percentage of adult catches per trap was significantly lower than that (P = 0.01; test t) obtained with the pheromone blend alone. These results have been found in both experimental areas and both years of application (respectively 30.9% vs. 69.1% at Dugenta and 37.5% vs. 62.5% at Cervinara), with minimal differences between the trapping levels. The disruption of the attractiveness of *M. mutatus* pheromone blend in the presence of (Z)-3-hexen-1-ol, probably due to the interference with antennal perception of pheromone components, has great ecological and practical interests. In fact, it may lead to the inhibition or a delay of the female mate-seeking behavior causing increased female exposure to dry conditions, fitness reduction and a decline of the reproductive success.

KEY WORDS: invasive species, monitoring, wood borer, electroantennography.

POSTER

Using stage-structured matrix models to determine the impact of an elicitor inducing plant resistance on *Myzus persicae*

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Induction of resistance in plants represents a promising approach for the defense against pathogens and pests of cultivated plants. This strategy consists in stimulating the activation of defense mechanisms by means of synthetic or natural molecules that simulate the attack of a pathogen. While a number of studies addressing effects on pathogens have been conducted, very few researches concerned effects on pests. With this study we aim to evaluate the possibility of using elicitor-induced plant resistance as a tactic for protecting plants against arthropod pests. Laminarin, an elicitor of induced responses in plants, was evaluated for the control of the green peach aphid, *Myzus persicae* (Sulzer) (Homoptera: Aphididae) on peach. The bioassay was carried out in controlled environment. Small peach plants received a mixed inoculum (nymphs of different ages and adults of *M. persicae*) and after 7 days, time required to ensure aphid settlement, the peach plants were treated with two different concentration of laminarin. After the spraying the aphid population was quantified every other day on 6 subsequent dates by counting 1) juvenile stages, 2) adults, and 3) winged adults on all the leaves of each peach plant. In this way, time series (average number of juvenile and adult stages present in subsequent sampling intervals) were obtained.
We used a demographic approach that is particularly suitable to study this kind of compounds which are mainly characterized by causing sub-lethal effects rather than acute mortality. A stage-classified population model (Lefkovitch Matrix) was used. Matrix parameters were estimated using two inverse estimation methods that use the time series obtained during the bioassay. Starting from this model, various demographic parameters were then estimated in each treatment and compared. Since all demographic analyses rely on how much the estimated parameters of the matrix are able to represent population dynamics, both a quadratic program method and a genetic algorithm for inverse matrix parameter estimation were used in order to find the best model fit for the observed stage class distributions. These two methods were compared to determining the set of parameters that minimizes the residual between the collected data and the model output.
Populations of *M. persicae* grew significantly more slowly on plants treated with laminarin than on control plants. Although laminarin does not present an acute toxicity towards *M. persicae,* the reduction of the aphid population seems to be mainly linked to an effect towards the younger aphids nymphs whose chances of survival and growth appeared reduced, probably due to the presence of the defence compounds produced by the plant following the stimulation of the laminarin.

KEY WORDS: elicitors, induced defenses, laminarin, matrix population model, genetic algorithm, quadratic programming.

ORAL PRESENTATION

Leafhoppers and Iflaviruses: new insights in virus-host interactions and their potential application

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Insects viruses as biocontrol tool in crop protection have received much attention in recent years due to their narrowed target and their environmentally-safe properties, which make them harmless both to human and non-target organisms. Indeed, baculoviruses have already been used for decades as biocontrol strategies against Lepidoptera. Although many formulates have been successfully developed against Lepidoptera, little attention has been payed to the selection of new suitable viruses able to control insects belonging to other orders (i.e. Hemiptera). In this perspective, the H2020-funded VIROPLANT project aims at identifying potential biocontrol agents from the virosphere of phytoplasma vectors. Phytoplasmas are wall-less bacteria which multiplies in the plant phloem. In particular, Flavescence dorée phytoplasma of grapevine was classified as quarantine pest by EFSA, since its fast spread causes severe damages to the vineyards. Although *Scaphoideus titanus* Ball (Hemiptera: Cicadellidae) is the principal vector of flavescence dorée (FD) of grapevine, *Euscelidius variegatus* Kirschbaum (Hemiptera: Cicadellidae) is used as an efficient vector of FD phytoplasma in laboratory condition, because, contrary to *S. titanus*, it is a polivoltine and polyphagous leafhopper. In this work we present for the first time four novel +ssRNA viruses putatively belonging to the family Iflaviridae found to be naturally present in both the above mentioned leafhoppers populations. Two iflaviruses, named *Euscelidius variegatus* virus 1 (EVV-1) and *Euscelidus variegatus* virus 2 (EVV-2), were serendipitously found in our in-house rearing of *E. variegatus* during an RNA seq analysis. Finally, other two iflaviruses were identified during an NGS analysis of *S. titanus* transcriptome. The latters, named *Scaphoideus titanus* iflavirus 1 (STiV-1) and *Scaphoideus titanus* iflavirus 2 (STiV-2), were discovered in a wild population sampled in Europe and in another sampled in the US, respectively. In particular, EVV-1 was found to be present in our lab rearing with a 100% prevalence. The discovery of a virus-free lab population coming from France allowed us to characterize the transmission routes of this iflavirus. The ability to transmit FD phytoplasma was also tested for both EVV-1 infected and non-infected individuals. Preliminary results showed that the virus was constantly detected both in phytoplasma-exposed and non-exposed vectors, but, interestingly, the virus load was significantly lower in FD-exposed insects, suggesting a potential cross-talk among insects, endogenous bacteria and viruses. Further experiments aiming at setting up an efficient inoculative strategy of EVV-1 in *E. variegatus* virus-free individuals are ongoing. An infectious clone derived from EVV1 was able to infect and replicate in virus-free insects. Even though the infection rates still need to be improved, the EVV-1 infectious clone might provide the unprecedented opportunity to manipulate the expression of endogenous insect genes by promoting virus-induced gene silencing by the iflavirus and interfere with insect ability to transmit phytoplasmas in both leafhopper target.

KEY WORDS: leafhopper, Insect virus, flavescence dorée, Iflavirus, *Euscelidius variegatus*, *Scaphoideus titanus*, infectious clone.

ORAL PRESENTATION

Odorant receptor expression-related modulation in behavior during larval development in African cotton leaf worm, *Spodoptera littoralis*

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Insect larvae rely on sensory input, mainly olfaction, for locating food sources. In several lepidopteran species, foraging behaviour and food preferences change across larval instars. We hypothesize changes in the olfactory system, specifically instar-specific expression of odorant receptors (ORs), as a possible mechanism influencing odor detection and behaviour during larval development. We therefore investigated the expression patterns of ORs in larvae of the cotton leafworm *Spodoptera littoralis* between the first and fourth instar. We provide the first empirical evidence of changes in the expression of a subset of ORs during larval development. Among few ORs differentially expressed, one receptor expression was significantly different in real time quantitative PCR analysis. Focusing on this receptor, deorphanization using heterologous expression coupled with electrophysiological recordings revealed that it responded to β-caryophyllene and its structural isomer α-humulene. Correspondingly, we showed that first instar larvae were attracted towards β-caryophyllene, while fourth instar larvae were not. Furthermore, we successfully disrupted the function of this receptor using CRISPR-cas9 system that induced a targeted heritable mutagenesis. In the behavioral experiments, the mutant S. littoralis larvae were not anymore attracted to β-caryophyllene and α-humulene compared to wild type larvae. Electroantennography recordings on mutant and wild type male and female moths corroborates with larval results, where mutant adults responded significantly lower to β-caryophyllene and α-humulene than wild type adults. We demonstrate that the receptor we identified in *S. littoralis* is essential for host plant location in larvae and adults. These findings improve our understanding of the role of odorant receptors for behavioural responses and plasticity in insects.

ORAL PRESENTATION

Sterile Insect Technique to control pentatomid pest species: irradiation screening on *Halyomorpha halys* and *Bagrada hilaris*

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The Brown marmorated stink bug (BMSB), *Halyomorpha halys* (Stål) and the bagrada bug (BB), *Bagrada hilaris* (Burmeister) (Hemiptera: Pentatomidae) are two invasive alien phytophagous species. BMSB is extremely polyphagous, and for this reason it is considered among the most harmful agricultural pests in Europe, United States and Chile. The small *B. hilaris* has a narrow host range (it is reported as a pest in the Brassicaceae family) and it's a very important pest in the Southern part of the US, Chile, Central Asia, Africa and two European islands (Pantelleria and Malta). In the province of Trento, at the Edmund Mach Foundation, various tools are being tested for the monitoring and control of these two species, to be included in the integrated control strategies in development. In particular, the possibility of using the sterile insect technique (SIT) in combination with classical biological control has being evaluated. The main objective is to use the SIT in eradication programs of BMSB and BB in newly introduced areas and/or to use it as one of the tools for the reduction of their populations in areas where the invasion is already occurring, exploiting the biological characteristics of the species during the period of winter reproductive diapause. Therefore, during autumn 2019 and 2020 for BMSB, and late winter and early spring 2021 for BB, large numbers of adults of the two pest species were captured and then kept in semi-natural conditions for a minimum period, required by the characteristics of each species, to complete the reproductive diapause. After the interruption of the latter, males only were irradiated with γ rays at different doses. For the irradiation we used different approaches: for BMSB the irradiation took place at a health facility (Santa Chiara Hospital, Trento) using an innovative technique that involves the use of high energy photons (6 MeV), while for BB the bugs have been irradiated at the ENEA facilities using the conventional method based on Co-60. Following irradiation, mating and reproductive bioassays were carried out; various physiological parameters have been evaluated such as longevity, sterility and number of eggs produced by each female. Our preliminary results are very encouraging for the feasibility of the sterile insect technique to control these two pentatomid pest species; further studies are in progress to evaluate the competitiveness of the irradiated males. The work has been done with the financial support of IAEA Research Contract No. 23536/R0 and the CRDF Global Grant N. RD-AA3-20-67096-2.

KEY WORDS: BMSB, bagrada bug, SIT, IPM.

ORAL PRESENTATION

In vivo functional analysis of an *Aphidius ervi* venom protein

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To complete their development, many hymenopteran parasitoids finely modulate the physiology of their hosts thanks to regulation factors of maternal (venom and symbiotic polydnavirus) and embryonic (teratocytes) origins released during oviposition. The study of the molecular mechanisms of these virulence factors has allowed to understand the strategies used by parasitoids to exploit their victims, paving the way towards to the development of new bio-inspired strategies for pest control. The venom of the endophagous parasitoid *Aphidius ervi* (Hymenoptera: Braconidae) regulates the physiology and reproduction of the host *Acyrthosiphon pisum* (Homoptera: Aphididae) in order to enhance its suitability for the developing progeny. The major bioactive venom component in *A. ervi* venom is the protein γ-glutamyl transpeptidase (Ae-γ-GT1), an enzyme known to cause host castration. Here, we investigated some functional aspects of Ae-γ-GT1 by observing the phenotypic alterations in the host and in the parasitoid's progeny at different time intervals, through the post-transcriptional silencing of the gene Ae-γ-GT1, by means of RNA interference (RNAi). The suppression of the target gene was obtained through microinjection of dsRNA (double-stranded RNA) in the hemocoel of *A. ervi* female pupae and the relative expression of Ae-γ-GT1 was checked by qRT-PCR. The silenced emerging females have successfully parasitized *A. pisum* third instar nymphs. Parasitized aphids by dsGFP injected females and other non-parasitized ones were used as control groups.
The effect of Ae-γ-GT1 gene silencing induced an increase in the size of the aphids both 48 hours and 5 days after parasitization. The metabolic and physiological alterations in the pea aphid, due to a lacking of Ae-γ-GT1 in the venom blend, have also led to an increase of size of larvae, teratocytes and adult parasitoids at emergence. These latter showed reduced survival rates and no significant difference was found in their developmental times from oviposition to adult stage. The obtained results indicate that Ae-γ-GT1 gene silencing does not interfere with the development of parasitoid's progeny but seems to exert a positive action. Further studies will be needed to investigate the metabolic and biochemical changes in the host associated with the manipulation of gene expression in the venom gland and of its impact on the aphid's reproductive tissues and primary bacterial symbionts (*Buchnera aphidicola*). The methodology developed is per se an interesting novel tool that allows to perform a functional analysis *A. ervi* venom component under very realistic physiological conditions, very useful to explore the molecular mechanisms involved in host modulation and to identify molecules with potential bioinsecticidal activity.

KEY WORDS: Biological control, host-parasitoid interaction, RNAi.

ORAL PRESENTATION

New insights into the main components of the venom of *Torymus sinensis* by a transcriptomic and proteomic approach

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In recent years, venomics, or the study of the entire pool of peptides and proteins of one specific venom, has acquired increasing importance as a source of new bioactive molecules which can find many applications in several fields. For example, in medicine, each year, several molecules obtained from animal venoms are tested for multiple purposes such as the search for new immunotherapy drugs for hypersensitive patients, for new inhibitors platelet aggregation. Another field of application of these interesting molecules is agriculture, since they could constitute a winning weapon in the biological fight against parasitic crop species. In nature these harmful insects are kept under control by parasitoid species. Example of these is *Torymus sinensis*, the parasitoid of the chestnut gallogenic cinipid *Dryocosmus kuriphilus*. To date, *T. sinesis* is considered the only effective means of biological control of the chestnut gallogenic cinipid therefore the knowledge of the protein composition of the venom is extremely useful in order to identify new molecules that can be used as bioinsecticides in biological and integrated pest control. In this work we have identified the main components of the *T. sinesis* venom through a transcriptomic and proteomic approach. We used high-throughput nucleic acid sequencing methods to describe the transcriptome of *T. sinesis* venom gland. From *de novo* assembly of all cDNA 22874 contigs were obtained, all sequences were analysed using Blast2GO software. The transcriptomic information provided an overall picture of the putative proteins present in the venom gland and information on molecular functions, biological processes and putative cellular compartments. The proteomic analysis was carried out on the components of the venom, fractionated by SDS-PAGE electrophoresis. Protein bands were excised from the gel and, after tryptic digestion, were identified by mass spectrometry (MALDI TOF/TOF and LC-MS/MS). The comparison between the protein sequences identified and the transcriptomic data using the Andromeda software, allowed us to identify 206 putative proteins of *T. sinensis* venom. Molecular identification and subsequent characterization of these molecules will be essential to understand the role played by venom in the induction and regulation of the pathological syndrome observed in parasitized hosts.

POSTER

Plant resistance induced by the South American tomato pinworm in wild and commercial *Solanum* species

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Plant-insect interactions have been studied for a long time in both general and applied science, e.g., for optimized integrated pest management. The South American tomato pinworm, *Tuta absoluta* (Meyrick) (Lepidoptera: Gelechiidae), is a key pest of tomato that can although develop on several solanaceous species. This insect has been traditionally subjected to chemical control and, owing to pesticide side effects (e.g. insecticide resistance), alternative control strategies, such as resistant host plants, must be prioritized. Within this context, we assessed the biochemical consequences of *T. absoluta* infestation and the subsequent plant suitability for the pest development in several *Solanum* species and tomato varieties. The accumulation of secondary metabolites, the main enzymatic activities and the emission of organic volatile compounds were identified for both control and *T. absoluta*-induced plants. The host species showing promising results in preliminary experiments and in a literature search, together with wild and further commercial species (e.g., eggplant *Solanum melongena*) grown under protected conditions in the south Mediterranean area were used in biological and biochemical experiments. The results suggest that some *Solanum* species and tomato varieties have potential for induced plant resistance via direct antibiosis and antifeedant activities. These findings represent the first bases for selecting resistant/tolerant tomato varieties under the Mediterranean protected tomato growing conditions.

KEY WORDS: Secondary metabolites, IPM, plant defense mechanism, pest resistance, *Tuta absoluta.*

ORAL PRESENTATION

RNAi silencing of an immune gene disrupts the embryonic development of *Spodoptera littoralis*

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The study of the immunosuppression syndrome in Heliothis virescens (Lepidoptera: Noctuidae) larvae parasitized by *Toxoneuron nigriceps* (Hymenoptera: Braconidae) allowed the isolation and characterization of a host gene that is negatively modulated soon after oviposition. This gene, denoted as 102, has been functionally characterized in *H. virescens* as well as in *Spodoptera littoralis* (Lepidoptera: Noctuidae) (102 Sl); 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) enhances the septicaemia induced by *Bacillus thuringiensis* and the resulting killing activity on noctuid moth larvae, and provides new opportunities for pest control. To further explore the potential of this pest control strategy, here we assess the effect of RNAi mediated silencing of this immune gene in S. littoralis eggs. The experimental results demonstrate that the expression of 102 Sl gene is significantly down regulated in eggs soaked in a solution of dsRNA targeting this gene, compared to control eggs soaked in a solution of dsRNA targeting the Green Fluorescence Protein. The observed gene silencing was associated with a drastic reduction of egg hatching and larval survival. This result appears very promising for the development of new control strategies of pre-imaginal stages of noctuid moths. The observed lethal phenotype indicates that 102 Sl gene, besides its immune function, has an important role in the regulation of embryonic development which has been totally ignored so far. Therefore, our data pave the way towards novel pest control application and offer the opportunity to unravel new molecular details of insect development.

KEY WORDS: bioinsecticides, noctuid moths, plant protection, insect immunity.

ORAL PRESENTATION

SESSION XII

First molecular detection of *Apis mellifera* filamentous virus in *Apis mellifera* in southern Italy

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The global and continuous increasing decline of honey bees over the last decade has driven several research programs to tackle biotic and abiotic causes threatening this important insect species. Among the biotic factors, several positive-sense RNA viruses have been recently associated with colony decline, and a widespread occurrence of the DNA virus "*Apis mellifera* filamentous virus (AmFV)" has also been reported. However, the pathogenicity of this virus on the infected colonies is still questioned, being rarely associated to acute infections or colony losses. In this work we investigated the occurrence of AmFV in the apiaries in the Apulia region (southern Italy) using conventional PCR and high-throughput sequencing (HTS). More than 35 apiaries were visited between 2017 and 2018, and about 380 honey bee specimens consisting of adults, pupae, larvae of the worker bees and in some cases queen pupae were collected and tested by PCR as groups using two diagnostic primers that amplify two partial region of the Thymidylate synthase gene and the BRO-N gene, respectively. From two apiaries where trembling and weak bees were found, specimens were collected and extracted total RNA subjected to HTS. PCR results showed the occurrence of AmFV in all sampled colonies with only two negative samples: (i) one sample consisting of 20 adult bees collected in Copertino (LE) and (ii) one sample consisting of two died pupae collected from queen cell in an apiary located in Lecce. The overall AmFV-infection rate in the Apulian colonies was approximately 91%. The nucleotide sequence determined for a selected representative AmFV positive sample, showed that based on the phylogenetic analysis of the thymidylate synthase gene and Bro gene sequences, the Apulian isolate shared more than 98% of nucleotide identity with isolates previously characterized in Belgium, Switzerland and China. The presence of this virus in the diseased-colonies was also confirmed by HTS: two large contigs (3604bp and 2112bp) were assembled from the sequence dataset and shared up to 99% similarity- at nucleotide level- with sequences of AmFV previously reported from Switzerland. This is the first detection of AmFV infections on *A. mellifera* populations in Italy by HTS and PCR. HTS technology proved to be a rapid tool for identifying viruses in honeybees. Analyses are still ongoing on the recovered sequence dataset to identify other putative viral agents. Although limited information is available on the potential impact of AmFV, the wide distribution of this virus in the Apulian colonies should be of concern for the healthy status of honey bee in the region.

KEY WORDS: Puglia, AmFV, HTS, PCR, *Apis mellifera* filamentous virus.

POSTER

Unexpected interactions between insecticides and parasites can contribute to the decline of honey bee colonies

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The neonicotinoid clothianidin has a negative impact on NF-kB signaling and on immune responses controlled by this transcription factor, which can boost the proliferation of honey bee parasites and pathogens. This has been well documented for the insecticide-induced replication of deformed wing virus in asymptomatically infected honey bees. Here we show that the immune-suppression exerted by clothianidin has a direct effect on the parasitic mite *Varroa destructor*, promoting its fertility as a likely consequence of a higher feeding efficiency.

KEY WORDS: neonicotinoids, clothianidin, *Varroa destructor*, *Apis mellifera*, immunity.

ORAL PRESENTATION

Freeze-drying or microwaves? New techniques for the conservation of honeybee-collected pollen

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Honeybee-collected pollen is gaining food industry attention as a key functional food, showing a high content of essential amino acids, antioxidants, vitamins and lipids (e.g. omega-6 and omega-3 polyunsaturated fatty acids). However, fresh honeybee collected pollen has a high water content (15%–30% wt %), thus representing an optimal substrate for microbial pathogens. Classic conservation methods mainly rely to drying in a hot air chamber or freezing. Both approaches negatively affect the pollen organoleptic properties, as well as its content of nutraceutical compounds. Recently, two novel techniques have been proposed for the conservation of honeybee collected pollen, i.e. freeze-drying and microwave-assisted drying. Their impact on phenols, flavonoids (with special reference to rutin), amino acids and lipids of pollen collected by bees on different botanical species was assessed, and a careful optimization of the technical parameters of both methods was conducted. Results showed that microwave-assisted drying offers key advantages for the conservation of bee pollen. Irrespective of microwave power and treatment time, phenol and flavonoid content did not vary over untreated fresh pollen. Rutin content was not affected by microwave drying. Furthermore, both conservation approaches did not substantially damage pollen lipids; only the microwave treatment showed a damaging action on antioxidant compounds (i.e. reduction of tocopherols). Freeze-drying influenced rutin level in chestnut pollen, depending on the freeze-drying duration. However, the free proline to free amino acid ratio was always <80%, and the free amino acid to total amino acid ratio remained unaltered, allowing us to claim that freeze-drying did not affect the overall nutritional value of this honeybee-collected pollen.

KEY WORDS: *Apis mellifera, Castanea sativa,* freeze-drying, polyunsaturated fatty acids, proline, rutin.

ORAL PRESENTATION

Immune characterization of *Vespa velutina* and *Vespa crabro*: a comparison across caste and sex

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Social insects are known for the noticeable ecosystem services they provide, but, at the same time, some species can represent a threat for ecosystems and human activities (i.e invasive species). A striking example is the recent invasion of the Asian hornet, *Vespa velutina nigrithorax*, in Europe, which represents a considerable threat for apiculture and native biodiversity because of its predatory habits towards honeybees and other pollinators and for the competition with the native hornet, *Vespa crabro*, which shares a similar ecological niche. Deepening the knowledge on the biology of this species is therefore fundamental to stop its spread and protect beekeeping and local biodiversity. In particular, understanding the major mechanisms that allow an alien species to become invasive is crucial for limiting the impact of invasive alien species. An increased resistance to pathogens in the invasive species has often been evoked among the factors which could favour the establishment and proliferation of an alien species in a newly colonized region. Thus, in the present study, we compare the immune competence of different castes and sex of the invasive Asian hornet, *Vespa velutina*, and the native European hornet, *Vespa crabro*, by means of a bacterial challenge to assess if an increased immunocompetence might play a role in the ecological success of *V. velutina* in its invasive range. Our results demonstrate that immune response differ between the two species with an opposite pattern with respect to caste: in *V. crabro*, workers were more immunocompetent than in *V. velutina*; conversely, sexuals of both sexes were more resistant to bacterial challenge in the invasive species compared to the native one. The higher pathogen resistance in reproductives of the Asian hornet, especially in spring queens, might therefore represent a key factor contributing to the ecological success of this invader.

KEY WORDS: Immunity, invasive species, Asian hornet, European hornet.

Vespa velutina impact on native Vespinae in Liguria Region

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An increasing number of invasive alien species (IAS) are spreading worldwide, because of international trade. IAS damage native species trough parasitism, predation, competition, or other mechanisms, often in synergy with other drivers of global change (e.g. climate). Invertebrates are the most successful IAS because of their biological traits and reproductive capabilities. Among invertebrates, the Asian yellow-legged hornet *Vespa velutina*, established in 2004 in France, is a generalist predator of other insects. Its predation is mainly towards honey bees, thus causing production losses to the beekeepers, but the species may feed on other insects. Moreover, its prey spectrum overlaps with the one of other Vespinae species present in Europe. Thus, Vespinae species might also be affected by competition for food resources. In Italy, *V. velutina* is spreading in Liguria region, and populations near the French border are nowadays established. The aim of this study, within the framework of the LIFE STOPVESPA project, is to assess the impacts of *V. velutina* on three native wasp species, in an area where *V. velutina* was well established since at least four years. Furthermore, we compared the abundances of native Vespinae (*Vespa crabro*, *Vespula vulgaris* and *Vespula germanica*), between the invaded area and an adjacent uninvaded valley with comparable environmental features. Through a Bayesian Generalized Linear Models, we found that the abundance of the native *V. crabro* was positively correlated to *V. velutina* abundance, when the latter species was scarce. The positive correlation between the two species disappeared at trapping sites where *V. velutina* was extremely abundant. We did not find considerable differences of the abundance of *V. crabro*, *V. vulgaris* and *V. germanica* between the invaded and the uninvaded area. Overall, our findings indicate that native Vespinae probably avoided or minimised the competition with V. velutina, at least in its invaded range in Italy. The presence of *V. velutina* did not lead to an evident replacement of *V. crabro* and *Vespula* species. In addition, these results can serve as a baseline to assess biodiversity loss due *V. velutina* in a long-term invasion.

KEY WORDS: Asian yellow-legged hornet, European hornet, impacts, invasive species, inter-specific competition, niche overlap.

POSTER

Preliminary results on genetic and faunistic investigations of the genus *Eucera* in Sicily

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The bee species of *Eucera* Scopoli (Hymenoptera: Apidae), are pollinators of numerous wild and cultivated plants, and are richly represented in the Palearctic region and, in particular, in the southern regions of the Mediterranean Basin. The ecological importance of these species has been recognized by several specialists, that however, have not yet managed to resolve the critical taxonomic issues of the genus *Eucera*, defined as a 'problematic taxon'. Recent genetic studies have analysed the phylogenetic relationships between the species of the genus *Eucera*, proposing three new possible classification quite different from each other. Currently, studies concerning the species of *Eucera* present in Sicily are sporadic and a review of the general character of the taxon is lacking. This region is rich in biodiversity, made more complex because influenced by the presence of taxa originating in the African continent. In the years 2018-19, a study was conducted aimed at investigating the faunistic biodiversity of *Eucera* in Sicily, through the analysis of the main morphological characters and the mitochondrial gene cytochrome c-oxidase sub units I, used for discrimination and species recognition (DNA barcode). The specimens studied came from the Di3A collection, University of Catania, and from the samples taken in the Nature Reserve 'Oasi del Simeto' (province of Catania). In the Reserve, five different sites have been identified, characterized by dune and retro-dune environments and by the presence of particular botanical species. During the two years of sampling, 64 specimens of Eucera were recorded, belonging to 6 species: *E. nigrescens* Pérez, *E. proxima* Morawitz, *E. numida* Lepeletier, *E. vulpes* Brullé, *E. eucnemidea* Dours, and *E. oraniensis* Lepeletier. The collection of Di3A includes these species, and moreover *E. aterrima* Friese, *E. caspica* Morawitz, *E. notata* Lepeletier and *E. seminuda* Brullé. In this first phase of research, eighteen sequences were obtained. The five sequences of *E. oraniensis* and the two sequences of *Habropoda tarsata* (used as outgroup) are the first ones studied and deposited in GenBank for these species. Two phylogenetic trees were built using the neighbour-joining algorithm, implemented in MEGA X 10.1 software. The results obtained represent a preliminary step for the phylogenetic study of the Eucerini in the Mediterranean Basin; the investigations continue with an extension of the investigated areas, in the southern part of Sicily and in other sites in Italy.

KEY WORDS: bees, Apidae, sistematics, DNA barcode, *Eucera.*

ORAL PRESENTATION

POSHBEE: a multi – actor approach for the assessment and mitigation of pollinator stress factors

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The Horizon project POSHBEE (Pan-european assessment, monitoring, and mitigation Of Stressors on the Health of BEEs) aims to investigate the effects of crop protection products on bee health. Other Apoidea, essential for pollination of cultivated and wild flora, are considered at the monitoring and experimental level. The consortium, lead by Mark Brown from Royal Holloway University (UK), includes 14 countries and 42 organizations (research institutions, associations, companies) and aims to obtain, within the 5 year duration of the project, a more in-depth and organic knowledge of the effects on bees of exposure to chemical molecules, using new experimental protocols and modelling approaches. CREA has been involved in the implementation of field monitoring for Italy, according to a common protocol applied by the 8 countries in which this first part of the project was carried out (in addition to Italy, Estonia, Germany, Ireland, the United Kingdom, Spain, Sweden and Switzerland were involved in WP1). This Work Package included the deployment in 16 sites, 8 apple orchards and 8 rapeseed fields, of 3 *Apis mellifera* hives of, 3 *Bombus terrestris* nests of and 3 *Osmia bicornis* aggregations. Samples collected from these nests and the measurement of fitness parameters will provide indications on the presence of pesticides, on the incidence of diseases and on the quality of nutrition. An in-depth analysis of the landscape surrounding the sites was also carried out both in terms of land cover (with the use of digital / satellite technologies) and of presence of vegetation and pollinating insects by means of field surveys. The monitoring sites were chosen on the basis of different levels of pesticide use, using the presence of crops around the site as a proxy. The apple orchards sites were located in Trentino, while the rapeseed fields were in Piedmont. COLDIRETTI and UNAAPI were actively involved in the monitoring WP, collaborating in the choice of sites, providing the hives and ensuring their management, as well as providing support in data collection (on bees and on the agro-ecosystem). They were also present during discussion and planning stages, bringing the point of view of farmers and beekeepers. The bee samples of the three species, samples of wax and royal jelly from the monitoring sites of the 8 countries will be analyzed by the accredited laboratory of CREA-AA for the presence of residues of agrochemicals or their metabolites, as well as samples from the semi-field tests and toxicity tests. Experiments are also being carried out at CREA-AA for development of dose-response curves for molecules representative of the 3 classes: an insecticide / acaricide (sulfoxaflor), a fungicide (azoxystrobin) and a herbicide (glyphosate), for 4 European subspecies of *Apis mellifera* (protocols are developed by CREA-AA and experiments carried out for the subspecies *A. m. ligustica*) and for the determination of the toxicokinetics of the selected molecules at the individual bee level. The results obtained so far show divergences compared to official European data, suggesting that the toxicity of the tested molecules is higher than what has been reported so far.

KEY WORDS: *Apis mellifera*, bees, monitoring, agrochemicals, apple orchards, rapeseed.

ORAL PRESENTATION

Parasitism of a *Vespa crabro* honeycomb by *Hermetia illucens*

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Hermetia illucens is a Stratiomyidae dipteran, also called the black soldier fly. It is native of the American continent, and today is a cosmopolitan insect. It is known for its use among edible insects that can be easily bred in large quantities. This fly is also known in forensic entomology, as it is strongly attracted to putrescent organic material. It is frequently found in composting and waste disposal plants, where it plays a useful role in the degradation of organic material. In this context, an episode of parasitism by *H. illucens* is framed, which resulted in the death of larvae and nymphs inside a honeycomb of *Vespa crabro* (Hymenoptera: Vespidae), just collected in Umbria in the summer by a pest control officer and delivered to the laboratory of our Urania Research Center in Perugia. During the phase of extraction of the hymenoptera larvae, we noticed that the full cells of the lower part of the honeycomb were colonized by larvae of *H. illucens* at various stages of development. Most of the larvae and nymphs of *V. crabro* were still perfectly alive, while some had already died long before and in a putrefactive state. The larval or nymphal cell of those already dead generally contained a mature larva of *H. illucens* (or no larva of this dipteran), while every cell containing larvae or nymphs of *V. crabro* still alive had inside 3-4 larvae of *H. illucens* alive, at various stages of development. This would suggest an egg-laying of the fly on the whole honeycomb and, therefore, of a real phenomenon of parasitism. The presence, however, of mature larvae of *H. illucens* inside the cells in which the larva or nymph of *V. crabro* had already died leads us to think that the origin of the infestation may be consequent to the damage of the honeycomb or to the death and putrefaction of some *V. crabro* larva, which then attracted and induced *H. illucens* to deposit also in the intact cells, presumably with subsequent depositions, since many of the larvae were still immature. In conclusion, we believe that *H. illucens* initially needs an attractive stimulus, represented by the smell of rotting organic material and, only subsequently, can actually become a parasite. This possibility allows us to hypothesize the usefulness and use of this insect in the biological fight against *V. crabro* and other Vespidae.

KEY WORDS: *Hermetia illucens*, *Vespa crabro*, honeycomb.

POSTER

Honeybees as environmental samplers: analyses of bee-collected pollen loads to detect pesticide residues and to study the pesticide drift in an alpine valley (Val di Sole, Trentino)

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Val di Sole (Trentino) is an alpine valley which altitude ranges from 500 m in the valley floor up to over 1700 m. The valley floor is about 50 km long including also two lateral deadened valleys of Val di Pejo and Val di Rabbi of the Stelvio National Park. Val di Sole is characterized by apple orchards in the lower part and meadows and pastures in the upper part. In order to monitor the presence and the dispersal of pesticides and heavy metals along the valley, we selected 22 bee apiaries located along the valley floor and we gathered the bee-collected pollen at the hive entrance. The average distance between apiaries was 2.5 km. The pollen loads were collected after apple blossom in May and in July 2019. Each of the 42 pollen samples was made pooling together the pollen collected by two hives of the same apiary, considering two subsequent weekends of pollen collections, in order to mediate bee preference within the foraging area. Pollen samples were then analysed with accredited methods for multi-residuals to determine the level and frequency of about 560 among pesticides (including also dithiocarbamates and glyphosate), their metabolites and three heavy metals: cadmium, lead and copper. In parallel, we also performed the palynological analyses on all the samples in order to understand the relevance of every botanical species on sample composition. In total, we detected of 63 pesticides, among which 15 insecticides, 43 fungicides, 3 herbicides and 2 plant growth regulators. The most frequent pesticides were phosmet, dithiocarbamates, fluazinam and captan. Several pesticide residues have been reported in five of the 22 monitored locations in both the two periods, whereas in 17 locations at least one residue has been found for each period. Five locations of the most upper valley were pesticide free. The most critical situations for the number of molecules has been observed in two stations located in the apple-growing area where about 30 substances per period were detected. Phosmet, dithiocarbamates, fluazinam, captan, and Folpet showed also an extended drift along the valley floor and within Val di Rabbi. Indeed, residues of these pesticides have been found ca. 10 kilometres away from the apple orchards. The toxicity to honeybees has been calculated by using the Pollen Hazard Quotient. Heavy metals are also present in all the monitored areas. Copper settled on 10 ppm, while lead has a very high peak (1680 ppb) in Pellizzano compared to less than 100 ppb in the other areas. The palynological analyses detected the specific botanical composition typical for each period and location. Up to our knowledge, this is the first study that considered such a detailed coverage of the territory for the investigation of pesticide residues in pollen loads. The agricultural use and the topography of the valley allowed to explain the long distance of the pesticide drift along the valley floors, showing that in some cases it can be substantial.

KEY WORDS: *Apis mellifera*, apple, palynology, agricultural landscape, multi-residue analysis, pollen hazard quotient.

ORAL PRESENTATION

Pyrrolizidine alkaloids and honey at risk. Are the honey bees also at risk?

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The hepatotoxicity of pyrrolizidine alkaloids (PAs), often accused for their carcinogenic and genotoxic effects also, has for a long time been verified. These compounds are present in a high number of plants (about 6,000, the 3% of flowering plants) overall in the Boraginaceae, Asteraceae, Orchidaceae families included. The function of PAs is to protect plants from attacks by herbivorous animals and pests. European Food Safety Authority (EFSA) widely studied the PAs (about 600 different structures are known) and the foods containing them, and revealed an alarm on herbal teas and honeys. The allowed daily intake limits of PAs indicated by the German Federal Institute for Risk Assessment are equal to 0.007 µg/kg bw (body weight); however in honeys produced in Veneto (NE Italy) the highest value was 17.6 µg/kg of honey, corresponding to a daily quantity of 0.006 µg/kg bw. Much higher are the PA values in unifloral honeys such as Echium vulgare, up to 2,850 µg/kg. Regarding pollen, it contains higher PA levels than honey. In Italy, the plants causing the greatest risk due to the presence of PA, as they are widespread and assiduously visited by honey bees both for nectar and pollen, belong to the genus *Echium*, essentially *E. vulgare* (viper's bugloss), *E. plantagineum* (salvation Jane), and *E. italicum*, to *Borago officinalis* (borage), and *Anchusa* among Boraginaceae, and to *Senecio* among Asteraceae, that include also *Eupatorium cannabinum* (hemp agrimony). About *Senecio inaequidens* (narrow leaves ragworth), native to South Africa but in the last time largely diffused in Italy in ruderal areas and characterized by a long flowering period, the PA content represents a hazard also for livestock. However, in Italy the production of unifloral honeys of these species occurs rarely; it may concern *Echium*, spring-summer flowering species, and *Borago*, typically early flowering, but hardly detectable in the honeys also by means of melissopalynological analyses due to its pollen strongly under-represented in relation to the floral morphology. Although *Apis mellifera* usually avoids toxic plants, it frequently visits most of the PA-containing species. Little research has been done about the effects of these compounds on the honey bee. Laboratory tests showed mortality effects on the bees as a result from a diet containing a 2% 1,2 unsaturated PA-mix extracted from Senecio vernalis, and deterrence of 2% PA-Nox-mix, as well as the toxicity of 2% single PA (monocrotaline), which caused mortality but not deterrence. The trophallaxis after feeding with high PA content transfers lower percentages of these alkaloids to honey bees involved in this process compared to feeding with lower PA content. The direct action of these alkaloids on the larvae is damaging, but the nursing secretion supplied by the nurse bees appears to reduce the PA content present in the bee bread used for its production. Concerning adult honey bees, they showed a relative tolerance that seems to limit the toxic action of PAs, but further investigations are needed in terms of PA effects on *A. mellifera,* unfortunately already exposed to a large number of threats.

KEY WORDS: pyrrolizidine alkaloids, toxicity, honey bee, accountable flora, honeys.

POSTER

The interaction between a parasite and sub-optimal temperatures contributes to honey bee decline

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Global insect decline and, in particular, honey bee colony losses are related to interacting stress factors, including landscape deterioration, pollution, climate change, parasites and pathogens. However, little is known about the implications of the interactions between different factors and, in particular, between biotic and abiotic factors. Here we exploited the honey bee as a model system to approach this problem and carried out extensive lab and field work aiming at assessing how suboptimal temperatures and *Varroa destructor* infections can alter the homeostatic balance of individual bees and the whole colony, leading to individual death and colony collapse.
We found that mite infestation further than increasing the mortality of bees, induces an anorexia that reduces the capacity of bees to thermoregulate, thus exposing them to the detrimental effect of lower temperatures. In turn, low temperature enhances the negative effect of the increasing mite infestation further reducing the survival of bees; this has got dramatic implications for the colony as a whole. The results highlight the important role that abiotic factors can have by shaping the effect of parasitic challenges on honey bees.

KEY WORDS: Stress factors, *Apis mellifera*, *Varroa destructor*, low temperatures.

ORAL PRESENTATION

Effect of *Vespa velutina* queen trapping on honey bee colony development

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The Asian yellow-legged hornet *Vespa velutina* was accidentally introduced from China to other parts of the world: South Korea in 2003, Europe in 2004, and Japan in 2012. *V. velutina* represents a serious threat to honey bees, wild bees, native pollinators, and native wasps, causing several impacts on beekeeping, especially in high-density colonized areas where its massive predation towards honey bees contributes to colony losses. In some European areas, the beekeepers declared losses up to 50% of their honey bee colonies. To reduce this impact, many beekeepers adopted different trapping methods for *V. velutina* queens, in particular during spring when the founder queens begin the construction of their nests. The objective of trapping *V. velutina* queens during spring is to limit the foundation of hornet colonies in proximity to apiaries and consequently reduce hornet population and its impact in summer and autumn, when the predation of *V. velutina* is normally rather intensive. To test this hypothesis, the development of honey bee colonies in areas where the capture of *V. velutina* queens had been performed in spring was compared with honey bee colony development in areas where the spring trapping activity had not performed. The experiment was carried out in Liguria Region (Italy) within the LIFE STOPVESPA project, during the years 2017 and 2018 in an area with high density of *V. velutina* nests. In both years, three couples of similar apiaries where selected and 40 bottle traps where placed within a radius of 700 m from an apiary of each couple, while the other was used as an untreated control. Colony development was periodically checked till the next spring. Data analysis highlighted that honey bee colonies developed better in the apiaries where the spring queen trapping had been carried out compared to the control apiaries where the capture of *V. velutina* queens had not been performed. In the apiaries where no countermeasures were undertaken to limit the impacts of *V. velutina*, heavier honey bee colony losses where observed. The impact of spring trapping on not target insects can be considered acceptable in comparison to the widespread use of insecticides for *V. velutina* control.

The harmonic radar for tracking insects: the case of the invasive hornet *Vespa velutina*

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Tracking technologies are commonly used for studying movements of several animal species. Although on large animals the weight and size of the equipment is not a relevant issue, insects tracking necessarily requires the use of light and not-invasive approaches. Therefore, harmonic radars may represent one of the methods that can be used for tracking insects, due to the small dimension and weight of the transponder. In recent years, the Polytechnic and the University of Turin have developed an innovative harmonic radar technology, which is capable of following insects even in complex environments. This technology has been applied with success for tracking and locating nests of the Asian yellow-legged hornet *Vespa velutina*, an alien species that is invading several European countries and that is able to produce serious impacts on beekeeping and biodiversity. Hornets have been tracked through passive tags that were glued to their thorax. The developed tags, with a weight of no more than 15 mg, did not affected the hornets in their usual colonial activities, such as flying and preying honey bees for feeding the brood.
The analysis of 389 tracks from nine localities of Liguria region (Italy) allowed detecting the position of several nests and gathering important biological and ecological information on *V. velutina*. In the study areas, hornets have been tracked for an overall length of 37 km (mean single tracking length of 96 ± 62 m). Detected nests were located at a mean distance of 395 ± 208 m (min = 72 m; max = 786 m) from the apiary where hornets had been caught while hunting honey bees. Tagged hornets were flying at a mean speed of 6.7 ± 2.3 m/s, but the flying speed was reduced to 4.1 ± 1.3 m/s when carrying a prey to the nest. Furthermore, the harmonic radar was capable of tracking hornets either in natural and urban landscapes without any particular issue, showing difficulties only in environments with steep slopes. The harmonic radar developed for tracking the flying activity of *V. velutina* could be used for tracking other insects that are able to carry transponders with a weight of 10-15 mg. This technology is particularly useful for the management of invasive colonial insects that requires an early detection, but could find research application also on species that cannot be tracked with conventional methods.

KEY WORDS: harmonic radar, *Vespa velutina*, asian hornet.

ORAL PRESENTATION

Detecting colonies of social insect species through thermography

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Vespa velutina is an invasive alien species, introduced in France in 2004, that is spreading in Italy and in many other European countries. The hornet is able to generate negative impacts on beekeeping and entomological biodiversity, due to the intensive predatory activity towards Apis mellifera and other native insect species. Control strategies for *V. velutina* in Europe are mainly based on the timely detection and destruction of colonial nests. The effectiveness of the control strategy could be increased if the colonies are removed before the birth of the gynes, which generally occurs from the month of September. However the colonies, built in most cases on treetops, are often not detectable until late autumn, since covered by the tree canopy. Nests of several social insect species tend to maintain a temperature that, for at least part of the day, differs from the temperature of the surrounding environment, a phenomenon particularly evident with societies formed by a large number of individuals. Thermography may therefore represent a tool that can be used for detecting this thermal difference, and find a potential application in control strategies for *V. velutina*. Therefore, the effectiveness of thermography was tested under controlled conditions on some V. velutina nests that were located in Liguria region (Italy). The adopted infrared (IR) camera was able to detect nests in the morning before sunrise, when there is a temperature difference between the nest and the tree canopy, and in absence of branches that could mask the nests. The colonies were detectable up to a distance of 30 m by simply observing the display of the IR camera, even without a subsequent electronic post processing of the images. This experiment was performed during the month of August, which is one of the hottest month of the year in Italy. Consequently, event better results can be achieved in other periods of the year, during which the vegetation undergoes a lower heating, or with IR cameras with higher resolutions. This technology can also find application for other social species, whose nest is characterised by a thermal difference from the surrounding environment.

KEY WORDS: V*espa velutina*, nest detection, invasive species, early warning, thermography.

POSTER

Phytosociology, preferences and floristic mapping of the most appreciate species by *Apis mellifera* and other Apoidea

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The alteration of natural and rural landscapes of northern Italy, caused by the broad urbanization and the intensive use of agricultural lands, has led to a reduction in the nectariferous species with the depletion of the food resources available and necessary for the survival of the bees. The existence of architectural barriers determined by human settlements, and the reduction in cultivated areas of nesting sites for wild bee species often alters and obstacles bee movements in the areas in search of the food source. The impact of these factors is extremely variable in relation to the bee-species studied, *Apis mellifera* L. or other social or solitary bees. In the case of *A. mellifera*, numerous factors should be considered and they imply both the survival of the colony and the productive capacity in terms of honey produced. The analysis of the territory is therefore fundamental, supported by phytosociological studies, use of databases related to land use and apiaries position as a proxy indicator of territorial suitability for the creation of maps aimed at verifying the potential beekeeping of the territories surrounding the hives. It is evident how these maps, obtained with the support of Web & Mobile App and GIS, become a scientific tool for the study and design of environmental improvement interventions and for crop management in support of the honey bees. With regard to this last aspect, the results of some specific studies on nectariferous species, potentially introduced in association with other crops to support bees in times of difficulty, are reported. Furthermore, in the panorama of highly anthropic environments, botanical gardens and urban parks, rich in numerous species that alternate blossoming period to improve the aesthetic purpose, can be used as open-air laboratories for the study of the relationships among bee-species, environment, floral species, and flowering. Through the study of the flower-bee network, it is possible to identify the most suitable species to support both *A. mellifera* and/or other species of Apoidea, both generalists and specialists. It is also possible to deepen the relationships that correlate the flower traits to the different species of bee present considering the interaction between the morphology/bioethology of the bee and the botanical characteristics.

KEY WORDS: flower-bee interaction, network, GIS.

The influence of *Phacelia tanacetifolia* on early-spring and late-summer foraging behaviour of *Apis mellifera* and other bees

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In Lombardy (Northern Italy), where the environment is fragmented and mostly anthropic, the availability of food for bees is strongly limited during some periods of the year. Bees are stressed by this and other factors and often face the winter with a relatively low amount of stocks. This turns out in a high mortality rate, detectable when the activity starts again. To contrast this trend, and therefore increase bee survival, and as an alternative to artificial food supplements, researches are recently focusing on nectariferous species, especially on the ones which flowering time matches the period of highest food stress. Among these, *Phacelia tanacetifolia* is spreading. It is a plant native to North America, generally used for forage and silage but recently rediscovered as an excellent annual honey plant. This work assessed the potential of this species for pollinators, in particular honeybee and other Hymenoptera, through two types of sampling: stored pollen and nectar into three hives, positioned at the edge of the experimental fields at the beginning of flowering; and the monitoring within the flowering field, in plots of one square meter, proximal and distal to the hives, from the beginning until the end of flowering. The experimental phase started from sowing in two different periods of the year as nutritional support in spring and late summer. The hives were checked three times (at the beginning, peak and end of flowering) during the monitoring period and the stocks evaluated. The observational plots were repeated twice per week (with favourable weather conditions) from morning to afternoon, at given temporal sessions. During each session, the following parameters were recorded: number of inflorescences per square meter; environmental condition; number of *Apis mellifera* visiting the flowers within each plot in five minutes, and which resource (pollen or nectar) was collected. The same data were also collected for other Hymenoptera which entered the plot. The monitoring verified that nectar and pollen of *P. tanacetifolia* are appreciated by bees belonging to the following genera: *Bombus, Andrena* and *Halictus*. *Apis mellifera* was more interested in nectar, perhaps in connection with the need to carry out a particular manipulation of the flower during the collection of pollen. As expected, a correlation was observed between the visits of *A. mellifera*, for the collection of nectar and pollen, and the temperature, which influences the number of flowers visited by each individual bee especially in autumn. The use of this exotic species is now under debate, considering the risk of its switch to an invasive species and the possible disturbance to pre-existing relationships of native plant and pollinators. Further studies are needed to evaluate the costs and benefits related to its use.

KEY WORDS: pollinators, stocks, pollen collection, nectar collection, exotic species.

POSTER

Some insights on honey bee immune response to Deformed Wing Virus, Black Queen Cell Virus and Sacbrood Virus infection

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Colony Collapse Disorder (CCD) of honey bee has been widely studied in the past decade and it is nowadays well recognized as a syndrome with multifactorial origin that leads to sudden depopulation of the hives. Among factors leading to CCD, pathogens and parasites may play a major role. According to the literature, the chronic exposure of bee colonies to biotic stressors, such as viruses and parasites, acts in an indirect way by deregulating the immune pathways linked to the antimicrobial response (Toll and Imd pathways). Nevertheless, a better comprehension of the impact of every actor involved is necessary to elucidate the mechanisms that underlie these interactions. In the frame of the Interreg-Alcotra project "Innov'api", which was monitoring the health status of several honey bee colonies managed with two alternative methods for varroa mite control, we selected a subset of samples collected from Piedmontese apiaries in July and September 2019 (before and after the summer acaricide treatment). The selected colonies were characterized by a wide range of viral loads, previously assessed by qPCR analyses carried out on pooled adult bee samples. We then investigated the variability of infection level among individual bees belonging to the same colonies, by focusing on the three most widespread ssRNA+ viruses (Deformed Wing virus, Black Queen Cell virus, Sacbrood virus). The microsporidian Nosema ceranae, another major stressor involved in CCD, was also included in the analysis. On the same samples, we quantified the expression levels of five genes belonging to the Toll and Imd pathways (three antimicrobial peptides, a peptidoglycan receptor and a NF-kB factor) and of vitellogenin, a protein involved in the immunocompetence, in order to investigate their correlations with biotic stress level. Viral loads as well as gene expression values showed very different levels among individuals within the same colony, allowing us to study how these pathogens may affect the individual immune responses. Our multivariate analysis suggests that DWV and BQCV loads positively correlated with the production of antimicrobial peptides such as apidaecin and hymenoptaecin and with the expression level of the NF-kB factor dorsal-1A. Moreover, the type of varroa control treatment had an impact on viral loads and gene expression levels, whereas vitellogenin transcript levels were mostly affected by the sampling time. As the microsporidian N. ceranae was seldom detected, we were not able to evaluate the impact of this pathogen on the regulation of the investigated pathways.

KEY WORDS: *Apis mellifera*, virus, DWV, BQCV, SBV, immunity, honey bee health.

POSTER

Seasonal variations of viral load in Piedmontese apiaries managed with brood removal or chemical varroa control

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Varroa destructor mite is regarded as a major parasite of *Apis mellifera*, as it is responsible for many colony losses both directly and indirectly, by virus transmission. The current control strategies are based on the use of synthetic acaricides, which can compromise the quality of the hive products due to the presence of chemical residues, and whose activity is limited by the selection of resistant strains over time. Through a collaboration with the INRAE-PACA researchers from Avignon and with the Italian and French national associations of beekeepers UNAAPI and ADA France (Interreg-Alcotra project "Innov'api"), we tested a sustainable control technique against varroa. To this aim the innovative technique of brood removal combined with oxalic acid treatment was compared to conventional control with a synthetic acaricide (amitraz). In Piedmont, more than 200 colonies from both migratory and stationary apiaries managed by local beekeepers were monitored: the project took into account several parameters, such as population of adult workers and brood amount, productivity and health status of the hives. The latter was described by the assessment of varroa infestation levels (obtained by counting the phoretic mites) and infection levels of five viruses (deformed wing virus, DWV; acute and chronic bee paralysis virus, ABPV and CBPV; black queen cell virus, BQCV; sacbrood virus, SBV). Quantification of the viral loads was carried out through Real-time qPCR analyses, after reverse transcription of the RNA extracted from two pools of 40 nurse bees per colony. The "Innov'api" experimental design, by including at least six sampling dates per year and having covered three beekeeping seasons (2017-2020), has allowed us to describe the seasonal trends of the main honey bee viruses in relation to the population dynamics of the parasitic mite and host colonies. In Piedmont, a high prevalence of DWV, BQCV and SBV was found, with virus-specific fluctuations, whereas ABPV and CBPV were detected sporadically. However, considering the viral loads of the infected samples, we highlighted positive correlations between ABPV and DWV, as well as between BQCV and SBV. Showing higher levels in spring and summer, BQCV and SBV shared similar dynamics to those of the hives and positively correlated with the number of adult bees and brood. By reaching maximum peaks in autumn, DWV followed the trend of varroa population. Moreover, DWV load was positively correlated with the infestation levels of the hives. The overall results show that the brood removal technique is as effective, and sometimes quicker, in lowering the varroa population, as well as the associated viral load, as the conventional technique.

KEY WORDS: *Apis mellifera*, *Varroa destructor*, viruses, ABPV, CBPV, BQCV, DWV, SBV, Innov'api.

ORAL PRESENTATION

Sub-lethal effects of Spinosad on the learning ability of *Apis mellifera*

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The role played by honeybees through pollination services and the direct supply of different products is well known. Unfortunately, during the past 15 years bee populations in many parts of the world have faced a rapid decline. Possible causes include lethal and sub-lethal effects due to exposure to pesticides. Although this aspect has been widely studied, there are still few data on the effects of sublethal doses of some natural insecticides on the behaviour of *A. mellifera*. The aim of this research was to verify the effects of sublethal doses of a natural insecticide based on Spinosad on the learning ability of *Apis mellifera ligustica* Spinola foragers. This was evaluated in the laboratory using the proboscis extension reflex method. Direct exposure (contact) of foragers to increasing doses of the insecticide allowed defining a dose-response relationship, from which the concentrations of DL0.2 and DL20 were derived. These sublethal doses were used in the behavioural bioassays. Linalool was used as an odorous stimulus. In our preliminary results, bees exposed to DL20 showed a reduction of associative learning ability compared to bees exposed to control. This is consistent with the effects of other insecticides and indicates that exposure in the open field to natural insecticides based on Spinosad could negatively affect the foraging behaviour of *A. mellifera* foragers.

KEY WORDS: honeybees, insecticides, sublethal doses.

POSTER

Management of the invasive alien species *Vespa velutina*: the LIFE STOPVESPA project

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Vespa velutina is an invasive alien species native to the southern part of China, that arrived in France in 2004 with few hibernating queens, and that was identified in Italy for the first time in 2012. With its intensive predation activity, the species is able to cause serious impacts on beekeeping and the environment in newly invaded areas. The LIFE14 NAT/IT/001128 STOPVESPA project "Spatial containment of *Vespa velutina* in Italy and establishment of an Early Warning and Rapid Response System", financed by the European Commission, operated in Italy from the 1st of August 2015 to the 31st of July 2019. The project developed a management strategy for the containment of *V. velutina* through a rapid detection and destruction of the colonies besides to expanding the monitoring network, assessing the impacts on beekeeping and the environment, involving and sensitizing stakeholders, citizens and administrators, and developing an harmonic radar capable of tracking the flight of the hornets and locate the nests. The control strategy and the early warning and rapid response system that were established allowed to increase the number of nests destroyed within the month of September, thus before the dispersion of gynes. About 2,000 nests were located and inactivated thanks to the activity of 2 monitoring teams and 3 destroyer teams of the project and 5 Civil Defence teams. During the period of activity of the project, the expansion rate of the species in Liguria decreased from about 20 to 3.2 km per year, and the colonised area, which in the period 2013-2015 reached 930 km2, in the years 2016-2018 increased with an annual rate of only 110 km2. Thanks to the work and skills of the Department of Electronics and Telecommunications of the Polytechnic of Turin, two entomological harmonic radar prototypes were developed. They allowed to track the flight of hornets, equipped with passive transponders, and locate their nests. Studies conducted in 2017 and 2018 on 6 experimental apiaries allowed to quantify the impacts of *V. velutina* on honey bee colonies; where no countermeasures were undertaken, an average loss of 18% of honey bees was assessed. Observations conducted to evaluate the effect of *V. velutina* queen spring trapping showed that honey bee colonies in apiaries where the trapping activity was performed reaches a greater development than colonies in areas without a trapping activity. Studies conducted to evaluate the impact of *V. velutina* on biodiversity highlighted that the species could ascertain an impact on the biodiversity of native insects. A negative impact has been assessed on some wild bees and Lepidoptera species, especially in the period July-October. This effect is particularly serious as it is synergistic with other causes that are leading to a generalized decrease of pollinators. The experience acquired with the LIFE project was made available to ISPRA for the preparation of the National Action Plan against *V. velutina*.

KEY WORDS: *Vespa velutina*, alien species, harmonic radar, early warning, impacts, management.

POSTER

Use of propolis against the mite *Varroa desructor* in *Apis mellifera*: prevention or curative social behavior?

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Self-medication, defined as a specific therapeutic behavior adopted in response to pathogens and parasites, plays a main role among the variety of behavioral defense mechanisms evolved by animals. According to the most recent definitions, adaptive behavior should meet the following conditions: 1. to involve the ingestion or external application of a third species or chemical compound; 2. to be initiated by parasite infection; 3. to increase the fitness of the infected individual or its genetic kin; 4. to be costly to uninfected individuals; 5. to be relevant in the natural environment of the host. Therefore, a clear example of behavioral defense in social insects is the collection of tree resin and its use in the hive as propolis, with bioactive properties. In *Apis mellifera*, an increased propolis content in the hive may correspond to variations in the microbial load of the colony and to a downregulation of the individual immune response of the bees. In the last five years, we conducted laboratory and apiary experiments to verify the hypothesis that the use of propolis in *A. mellifera* colonies may represent a case of social medication against the *Varroa destructor* mite. Through behavioral observations we showed that *Varroa*-infested colonies increased the resin foraging rate in comparison to non-infested ones. Afterwards, bioassays were performed to test the effects of propolis on the fitness of experimentally infested bees kept in metal cages in the presence or absence of raw propolis. In addition, toxicological tests were conducted to evaluate the acaricidal or narcoleptic effect of crude propolis on *V. destructor*. Our results demonstrated the positive effects of propolis on the lifespan of *Varroa*-infested adult bees. A low narcoleptic effect (19–22%) of raw propolis on phoretic mites after 5 hours of exposition was also observed. Considering these results and the fact that worker bees prepare brood cells for oviposition by applying propolis on their inner surface, we tested the hypothesis that propolis can interfere with *Varroa* reproduction phase by conducting laboratory trials using artificial brood cells. Propolis extracts caused an increase in *Varroa* mortality and a decrease in mite fertility in treated brood cells as compared to control cells. In conclusion, our findings seem to confirm the hypothesis that increased resin collection represents an example of social medication to mitigate the detrimental effects of parasitism.

KEY WORDS: propolis, social Immunity, self-medication, medication of kin, social medication.

ORAL PRESENTATION

Can propolis consumption reduce *Nosema ceranae* infection in *Apis mellifera*?

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Nosema ceranae is a widespread obligate intracellular parasite of the ventriculus of many *Apis* species, including *A. mellifera*, potentially leading to colony death. In the past the antibiotic fumagillin was largely used to control this disease. Because this product has now been banned in many countries, there is a need for alternative and safe products effective against *N. ceranae*. It is known that propolis, produced by bees mainly from resinous substances collected on woody plants, can decrease the microbial load of the hive. We hypothesized that propolis might also reduce *N. ceranae* infection of individual bees and that bees might consume propolis as a form of self-medication. To verify these hypotheses, we evaluated the effects of an ethanolic extract of propolis administered orally on the longevity and spore load of experimentally N. ceranae-infected worker bees. In separate experiments we also tested whether infected bees were more attracted to a diet containing propolis, and if they consumed a greater proportion of propolis in comparison to uninfected bees. Our propolis extract contained primarily derivatives of caffeic acid, ferulic acid, ellagic acid and quercetin. The results showed that propolis extract increased the lifespan of infected bees and also significantly reduced the *N. ceranae* spore load. Choice, scan sampling and food consumption tests did not reveal any preference of *N. ceranae*-infected bees for commercial candy containing propolis. In conclusion, our research supports the hypothesis that propolis represents an effective and safe product to control *N. ceranae* but worker bees seem not to use it for self-medication when infected with this pathogen. Further investigations on the possibility of including propolis in the diet of bees as a realistic perspective of controlling nosemosis type C in apiary are necessary.

KEY WORDS: ethanol extract, infection, Microsporidia, nosemosis, self-medication.

POSTER

FOR[m]AGE, BEES & FRUITS: bee-fruit synergies with forage farming systems in rainfed Mediterranean environment

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The Italian Ministry of Agricultural, Alimentary and Forestry Policies has funded the project "FOR[m]AGE, BEES & FRUITS": bee-fruit synergies with forage farming systems in rainfed Mediterranean environment", acronym 4APIFRUT. The hypothesis that neglected natural resources such as the potential benefits from native legume species, extensive cultivation of local fruit varieties and beekeeping might integrate the traditional productions (milk, cheese, meat) of agro-silvo-pastoral farms will be tested, in order to increase their incomes and competitiveness. The scheduled workpackages would promote agro-forestry practices within organic farming, increasing the energy efficiency and providing ecosystem services. In this frame, the entomological factors, regarding particularly the pollinators, will have an essential role. Four Operating Units, each with a different expertise, compose the research team of the project. They belong to the University of Sassari (Entomological section of the Department of Agricultural Sciences) and the National Research Council (ISPA; ISPAAM; IRET). The project will last three years (2020-2022) and involves four representative private farms located in Northern Sardinia. All farms practice extensive animal husbandry – dairy sheep, meat cattle, and donkeys – in a typical Mediterranean environment with a remarkable presence of wild pear trees. In this context, project activities will be aimed to: a) increase legume forage and grain yields; b) produce organic fruit from local varieties by grafting wild pear rootstocks; c) supply high quality honey by increasing and stabilizing pollen and nectar availability. Two Operating Units will deal with agronomic aspects. ISPAAM will attend the multifunctional exploitation of forage and grain legume species (sulla, sainfoin and white lupine) in new cultivation environments monitoring legume performances, ground covering, phenological stages, grain and forage yields, and N-fixation ability. ISPA will create extensive organic pear groves from unproductive native wild pear rootstocks (*Pyrus sylvestris*, *P. amygdaliformis*) grafted, in a planned way, with Sardinian varieties that hold high resilience to the climatic change and the environmental conditions. The two remaining Operating Units will follow the entomological project side. Even if their cooperation will be continuous and synergistic, the Sassari University will work prevailingly on beekeeping, IRET on biodiversity and pest control. Beekeeping is the main tool in enhancement of the farm incomes. Honey and pollen productions, obtained by increasing the surface area of leguminous plants and by grafting wild pears, will be studied. The major insect pests, their damages and their natural enemies will be monitored in order to implement possible pest control measures.

KEY WORDS: agroforestry, bees, cover crops, fruit crops, natural enemies, organic agriculture, insect pests.

POSTER

Detoxification as a key to understand the interaction between pollen and toxic compounds in honey bees

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Honey bees *(Apis mellifera* L.) play a vital role in ecosystems as plant pollinators and are essential for both plant biodiversity and agricultural production; indeed, one third of world crop production relies on animal pollination. Recent research highlighted the role of multiple interactions among several stress factors as a major cause of widespread colony losses threatening those essential ecosystem services. Pollen is the only source of proteins for honey bees and contains other substances that are necessary for normal growth and development. On the other hand, pollen may also contain toxic compounds, or it could be contaminated by widespread insecticides. Therefore, in many cases, to exploit the beneficial effects of pollen, metabolic detoxification (which is often based on Cytochrome P450 monooxygenases) must be activated with a costly process in terms of energy; for this reason, the interaction between pollen and toxic compounds deserves a close scrutiny. To gain insight into the possible interactions between pollen and toxic compounds that can be associated to it, we carried out dedicated lab experiments using nicotine as a toxic compound, both because it is common in some nectars and pollens and for its affinity with some insecticides. To this purpose honey bees were fed with nicotine, pollen or both and treated or not with a common inhibitor of detoxification (i.e. the insecticide synergist piperonyl butoxide (PBO), a P450 inhibitor, enhancing the toxicity of pyrethroid and neonicotinoid insecticides). The experiment was replicated three times in the early season and in the late season, to evaluate possible differences related to the presence Deformed Wing Virus (DWV): a common pathogen that is rare in Spring and widespread in late Summer. Preliminary results suggest that nicotine negatively affects bee survival only early in the season, when viral infection is low; under this condition, pollen seems to counteract the negative effect exerted by nicotine. Late in the season, when viral infection is higher, nicotine alone doesn't seem to be similarly harmful whereas the presence of pollen and nicotine together significantly reduce the survival. Finally, PBO appears to be active only in presence of both factors.

KEY WORDS: honey bees, detoxification, pollen, nicotine, Cytochrome P450.

Chronic oral exposure to field-realistic pesticide combinations via pollen and nectar affects thermogenesis in a solitary bee

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Wild and managed bees play an essential role in maintaining food production and plant biodiversity. However, bees are declining worldwide and pesticides are often signaled as one of the main causes of these declines. Generally, pesticide risk assessment schemes are based on testing lethal effects of single compounds following acute exposure. Yet, in agricultural environments, bees are exposed to combinations of pesticides for long periods of time. In addition, sublethal effects are rarely considered in current pesticide regulation, which increases the uncertainty around the safety of pesticides released in the market. In this study, we assessed the effects of field-realistic concentrations of three pesticides on the solitary bee *Osmia bicornis* L. (Hymenoptera: Megachilidae) in the laboratory. We measured pesticide residues in the pollen and nectar of melon flowers of 5 commercial fields in central Spain. We detected 19 pesticides, of which 11 had allegedly not been applied during the crop cycle. This result shows that agroecosystems are often much more contaminated with pesticides than it would expect. One of the most frequent combinations found included two neonicotinoid insecticides (acetamiprid, imidacloprid) and a triazole fungicide (myclobutanil). Throughout their life span, we exposed newly-emerged O. bicornis females orally via pollen and sugar syrup to these three compounds, alone and in combination (8 treatments including a control), at the observed concentrations. We measured pollen and syrup consumption, longevity, ovary maturation and thermogenesis. Although pesticide concentration was higher in pollen than nectar, pesticide uptake occurred mostly via syrup. At the tested concentrations, we did not find any effect on longevity and ovary maturation. Therefore, under current risk assessment schemes, we would have wrongly concluded that at field-realistic concentrations, these compounds and their mixtures were safe to *Osmia* bees. However, in all treatments containing imidacloprid bees showed lower syrup consumption and disrupted thermogenic capacity that impaired flight activity. Our results have important implications for pesticide regulation. The incorporation of tests specifically intended to detect sublethal effects into risk assessment schemes is essential to evaluate the consequences of pesticide exposure on the dynamics of bee populations in agroecosystems.

KEY WORDS: *Osmia bicornis*, sublethal effects, imidacloprid, melon.

POSTER

Synergistic effects of insecticide-fungicide combinations on bees: a comparative approach

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Bees play a key role in providing an essential ecosystem service in the form of pollination for crops and wild plants. Yet, this service is at risk because bees are declining worldwide and agricultural intensification is recognized as one of the main drivers. In agricultural environments, managed and wild bees are routinely exposed to a plethora of toxic plant protection products. Among the different possible pesticide combinations, neonicotinoids are known to interact synergistically with fungicides. In a previous study we found strong synergistic effects when bees were orally exposed to field-realistic doses of a neonicotinoid (clothianidin) with a triazole fungicide (propiconazole). The study was performed with three bee species with contrasting life history traits, *Apis mellifera*, *Bombus terrestris* and *Osmia bicornis*. In the current study we have used a similar methodology to assess the combined toxicity of sulfoxaflor, a new competitive modulator of nicotinic acetylcholine receptors that is being proposed a substitute for neonicotinoids, and fluxapyroxad, a succinate dehydrogenase inhibitor fungicide. We have found weaker synergistic effects restricted to early assessment times and intermediate sulfoxaflor doses in two of the three species. Our study confirms that bee species differ in their sensitivity to pesticides and, more importantly, in their response to pesticide combinations. Our results highlight the need to consider both different bee species and test compound combinations in environmental risk assessment schemes.

KEY WORDS: *Apis mellifera*, *Bombus terrestris*, *Osmia bicornis*, ecotoxicology, risk assessment.

ORAL PRESENTATION

Study of the apoidea in urban systems through the support of mobile technologies and GIS

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Numerous researches show that the richness in species of our planet is progressively rarefying. This is more evident in the industrialized countries were species are subjected to many anthropic perturbations. The loss of biodiversity weakens all the components of the ecosystem and, in detail, the organisms more "fragile" like the Apoidea. The alteration of natural environments with the consequent decrease in foraging and nesting sites is well known in Lombardy region where the Po Valley is characterized by a continuum of areas with monoculture or well-defined crop successions (winter cereals - maize) and where the natural areas are increasingly limited. This zone is also fragmented by numerous urban environments where the city of Milan and its hinterland occupy a predominant space. The cities, if on the one hand are unequivocally sources of stress for insects, to the point that in the past they were considered the main responsibles for biodiversity losses, they have recently been reevaluated, as there are species that seem to find, in the numerous parks and gardens present, conditions favouring their settlement. In particular, the floral essences, introduced in the green areas because of their high aesthetic and recreational value during the flowering period, provide important sources of nourishment for many Apoidea, either solitary either social, as well as honey bees. This work reports the preliminary results of the first year of monitoring carried out on the city of Milan used as a model to evaluate, through a multidisciplinary approach, the impact of the components of an urban environment on the presence/absence/spread of the Apoidea. The aim is to create an updated photograph of the species of bees, mapping their presence/abundance in relation to the environmental variables and the floral species present in the various city areas through the support of the Web & Mobile App and GIS. The application of data-driven models for the analysis of the relationships between the presence/absence of bees and environmental variables will allow the identification of model areas where environmental indicators recall a high probability of hosting bees. Specifically, innovative communication, education and dissemination strategies of the project results, based on information and communication technologies, will be used in order to make the data collection phase and the communication of the results more effective.

KEY WORDS: *Anthidium*, biodiversity, model, Web & Mobile App.

POSTER

Exposure to the insecticide sulfoxaflor and fungicide azoxystrobin reduces foraging activity, pollen deposition and colony growth in bumblebees

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The intense use of pesticides to maximize crop production is considered one of the most important drivers of global wild bee decline. Sulfoximine-based insecticides, potential alternative to neonicotinoids, have been shown to negatively affect the reproductive success of bumblebee colonies. Nevertheless, their effects on bumblebee activity under realistic field conditions remain largely unknown. Moreover, very little is known about effects of other pesticides largely used in agriculture such as fungicides. Here, we investigated the direct and combined effects of the insecticide sulfoxaflor and the fungicide azoxystrobin on the foraging activity, pollen deposition and colony growth of *Bombus terrestris* under semi-field conditions. Colonies were kept in large cages of *Phacelia tanacetifolia*. Pesticides were applied directly to the plants. We found both sulfoxaflor and azoxystrobin to reduce individual foraging performance and final colony weight. Moreover, the fungicide application negatively impacted visitation rate, pollen deposition and the number of flights per day. No interactive effect was detected. Our results show that the use of both systemic insecticides and fungicides can reduce bumblebee foraging activity, negatively affecting colony growth. The use of fungicides during bloom has the potential to severely impact bumblebee populations foraging in agroecosystems compromising the pollination services they provide.

KEY WORDS: bees, pesticides, pollination services, pollinators, sulfoximine.

ORAL PRESENTATION

Bee health: the impact of sublethal and interaction effects of pesticides

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Bees are indispensable pollinators, vital for global food production and wild plant biodiversity. Consequently, bee health decline has broad environmental implications. Multiple stressors are driving bee health, a major one being pesticides. While single pesticides can cause lethal effects, bees are frequently exposed to sublethal levels of pesticides, which do not cause death but behavioural or physiological alterations that can finally affect bee health. Furthermore, bees are not simply exposed to a single pesticide at the time but are more frequently exposed to multiple ones simultaneously (pesticide "cocktails"). This raises concerns, given that pesticide cocktails can cause "synergistic effects", leading to a particularly amplified toxicity: pesticide doses that would not harm bees alone become harmful in combination. Our recent work demonstrated that these synergistic effects can pose harm to bees via both lethal and sublethal effects. In addition, we recently discovered that pesticides can cause adverse synergistic effects even when combined with common environmental stressors such as poor nutrition, causing both lethal and sublethal effects. Nonetheless, sublethal and interaction (e.g. cocktail) effects of pesticides are only marginally considered in research and pesticide risk assessment schemes, thus possibly leading to limited and/or inaccurate conclusions.
Here, we address some of the current major challenges research and environmental risk assessment are facing: the assessment of sublethal and interaction (synergistic) effects of pesticides in bees. We describe this broad issue from multiple perspectives, presenting our current research that involves the investigation of pesticide exposure, toxicity, and risk. We underline the complexity of pesticide effects on bees, the consequent difficulty of performing an accurate assessment of their risk, and we propose refined approaches. Our international project aims at laying the foundation of future integrations and result interpretations of bee health surveillance initiatives, intending to produce harmonized scientific methods and knowledge that could also guide policy makers in the improvement of pollinator's health and environmental sustainability.

KEY WORDS: Bees, behaviour, sublethal, interaction, *Apis mellifera*, bee health, pollinators, pesticides.

ORAL PRESENTATION

Beekeeping and climate change: perceptions of beekeepers and changes in the management of the beekeeping farms

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Climate change occurred in recent years has given rise to serious threats impacting on the most varied sectors and in particular on the environment. One of the pressing problems is the decline of pollinators, whose action is essential for pollination services, agricultural productivity, biodiversity conservation and for the production of honey, this latter dependent exclusively on the honey bee. *Apis mellifera* is considered as the most important pollinator, a plastic species bred all over the world, able to make up for deficiencies in pollination following the decrease in wild pollinators. The effects of climate change on the bee are the subject of many studies, and policy measures have been implemented, nationally and internationally, in order to balance the protection of the environment, the economic importance of agricultural yield and bee products. In this context, the Interreg Alcotra research project "CClimaTT – Cambiamenti Climatici nel Territorio Transfrontaliero" was launched, based on three foundations to know, to communicate, and to act, in order to deal with the issue of climate change, and aimed at investigating its effects on honey bees and beekeeping. To carry out this research, several actions have been performed in the Cuneo province (NW Italy), in two different environmental areas (river park and intensive viticulture area), including a survey, through focus group discussions, to point out i) the perception of beekeepers regarding the effects of climate change on the honey bee and on the beekeeping sector; ii) the possible changes in the management of beekeeping activity; iii) the current positive and negative aspects of the sector. The last question was addressed to elicit the strengths, weaknesses, opportunities, and threats of beekeeping, in order to complete a SWOT matrix based on the participants' perceptions. The greatest importance of this analysis consisted in putting at the center the beekeeper, who personally faces up the emerging problems due to the climate, thus allowing to detect the main effects, direct and indirect, of climate change on the bee and consequently on the business economics: scarcity of nectar resources, pollen and honeydew in the environment; weakening or loss of honey bee colonies; reduction or absence of production; greater infestation of the *Varroa destructor* mite; new threats. To cope with these emergencies, beekeepers had to take the following measures: supplemental sugar feeding to help colonies; intensive transhumance practice; brood interruption methods in winter period for the varroa control; increased production of nuclei to maintain profits. The main strengths of the beekeeping activity that emerged are the strong motivation and satisfaction of the beekeepers and the growing visibility of the bee in public opinion, while the negative aspects concern the low average production of the farm, the reduced strength of the colonies, the lack of institutional support and competition with poor quality bee products. On the basis of the direct responses of the beekeepers it will be possible to implement the actions aimed at limiting the sector problems caused by uncontrollable elements, such as climate change.

KEY WORDS: honey bee, climate change, beekeepers, focus group, SWOT.

POSTER

Infection routes of deformed wing virus (DWV) in honey bees and impact on pathogen's virulence

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Honey bees (*Apis mellifera* L.) suffer from several stress factors among which the deformed wing virus (DWV) has a particularly deleterious impact to the colonies. Normally present in the hive as a benign covert infection, in late summer, DWV can give rise to overt dangerous symptomatic infections, becoming a main driver of colony collapse.
Virus transfer among bees can follow a number of complementary routes. The transmission mediated by the ectoparasitic mite *Varroa destructor* is particularly deleterious since the mite is linked with DWV in a mutualistic symbiosis. The mite-mediated viral transmission can take place both during the mite's reproductive phase, within the brood cells, and during the phoretic phase, on adult bees. Furthermore, since DWV is found both in the nurse's hypopharyngeal glands and in the hives' products, such as honey and pollen, DWV transmission can also occur through larval and adult food. Despite this rich and diverse set of transmission routes of the pathogen within the hive, little is known about the relative influence on the infection outcome, although the clear importance under the epidemiological point of view. To gain insight into the influence of different infection routes on the pathogen virulence, virus-free bees were infected with DWV by means of artificial mite infestation or by administration of food (i.e. larval food or sugar syrup) containing 10^3 DWV viral copies. Both pre-imaginal stages and adults were infected. At the emergence, symptoms of DWV infection were noted and viral levels quantified by means of RT qPCR. Then, bees were maintained under controlled lab conditions and daily monitored to assess the survival. Individual 5-day-old worker bees exposed to the different DWV infection routes at different developmental stages were also used for a transcriptomic analysis to investigate stress response and the expression of immune related genes and pathways. As expected, both infection routes (i.e. food and mite mediated) negatively affected the survival of bees but they had a stronger impact on the pre imaginal stage than on the adult bees. Overall, the mite mediated viral transmission appeared to be the most detrimental in terms of bees' mortality and viral load. The transcriptomic analysis of bees contributed to unravel the molecular basis of the observed response. Despite the fundamental role of DWV with respect to honey bee health and the importance of the transmission route of the pathogen on the fate of the infection, little was known up to now on this aspect. The integration of these information into a convenient epidemiological framework will substantially contribute to a better understanding of the pathogens' dynamics within the hive.

KEY WORDS: Honey bees, DWV, infection route, epidemiology, RNAseq.

POSTER

SESSION XIII

Anagyrus vladimiri: an effective parasitoid of *Pseudococcus comstocki*?

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*Pseudococcus comstock*i (Kuwana) (Hemiptera: Pseudococcidae), coming from Eastern Asia, is characterized by a marked polyphagia toward several species of ornamental (*Morus* spp., *Prunus laurocerasus*) and fruit (*Malus domestica*, *Pyrus* spp., *Prunus persica*) plants. Its presence on the European continent was firstly recorded in 2004 in Italy and France. High infestation levels of this pest, due to the trophic activity of nymphs and mature females on buds, sprouts, leaves and fruits, strongly inhibited plant development, with major economic losses. Further indirect damages are due to abundant honeydew production, followed by the development of sooty mold. Since 2006, outbreaks of this pest have been reported in the north-east of Italy, with special reference in organic apple orchards and vineyards in the Veneto and Emilia Romagna regions. This highlighted the need of an effective and sustainable pest management strategy. Some species of hymenopteran parasitoids were identified as potential biocontrol agents (e.g. *Clausenia purpurea* (Ishi) (Encyrtidae)). One of them is *Anagyrus vladimiri* Triapitsyn, an encyrtid species already employed with success to manage *Planococcus ficus* (Signoret). In this study, the effectiveness of *A. vladimiri* as biocontrol agent against *P. comstocki* was assessed. Its host searching behaviour as well as the parasitization and superparasitization levels were quantified. No-choice and choice tests were performed comparing the parasitoid efficacy toward *P. comstocki* and *P. ficus*, selected as an optimal host for the development of this encyrtid. Lastly, the fitness of *A. vladimiri* wasps developed on *P. comstocki* and *P. ficus* was analysed, comparing duration of pre-imaginal development, emergence rates, sex ratio and mean body size. Overall, our results showed fully comparable performances of this parasitoid on the two hosts, allowing us to consider *A. vladimiri* as a potential effective biocontrol agent to manage *P. comstocki* populations.

KEY WORDS: biological control; integrated pest management; mealybug; Hymenoptera; invasive species.

ORAL PRESENTATION

Biological insecticides: a possible use against *Halyomorpha halys*

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Halyomorpha halys is an invasive Asiatic pentatomid accidentally introduced in North America in the nineties of the last century and successively in Europe in the first decade of 2000s. More than 300 wild and cultivated plants are suitable hosts for this insect that feeds on and damages mainly fruits and seeds. Poor results have been obtained from the use of chemicals against *H. halys* so researches have switched to biological alternatives aimed at the control of the different developmental stages of the insect, including early stages like the egg masses. Application of biopesticides and/or biological active ingredients has been evaluated in this study on 120 single *H. halys* egg masses under laboratory conditions. Products have been selected based on the possibility of their actual use in organic farming, and their effect has been evaluated calculating the percentage of egg hatching and nymphs mortality after 15 days following direct spraying on the egg masses. Two commercial products containing the entomopathogenic fungi *Beauveria bassiana* (strain ATCC 74040, Naturalis®) and *Paecilomyces fumosoroseus* (strain FE9901, NoFlyTM Wp) were used, together with three more wild entomopathogenic fungal strains from the CREA-Research Centre for Plant Protection and Certification collection: one *B. bassiana* (Bba18/T03), and two *Metarhizium brunneum* (Mbr17/T101 and Mbr17/T105) strains. One azadirachtin A-based product (NeemAzal®T/S) was also tested, and two more trials were performed using a *Bacillus thuringiensis* commercial formulation (Rapax® AS, *Bacillus thuringiensis kurstaki* strain EG 2348) and the same pure *B. thuringiensis* strain (EG 2348) isolated from the commercial product. Two control trials were included in the experiment, one with no treatment at all (CTRL AS) and one treated with a 0.02% water dilution of Tween®80 (CTRL TW). After treatment, egg masses have been isolated in single Petri dishes containing food and water (a slice of carrot and wet cotton) for first instar nymphs. The lowest hatching percentage (10%) was recorded in the NoFlyTM WP treatment while in control treatments it was as high as 95% and 71% for no-treatment and Tween®80 water dilution respectively. In remaining treatments hatching percentages were always lower than 40%. In fungi treatments, the effects on the newborn individuals were evidenced by dead nymphs showing symptoms of mycosis: they were 71% and 94% in Naturalis® and NoFlyTM Wp treatments, respectively. Mycosed dead nimphs were also 89%, 64% and 100% in Bba18/T03, Mbr17/T101 and Mbr17/T105 treatments, respectively.

KEY WORDS: *Beauveria bassiana*, *Metarhizium brunneum*, entomopathogenic fungi, egg masses, biological control.

POSTER

Lethal and sublethal effects of synthetic insecticides and plant essential oils toward the omnivorous mirid *Nesidiocoris tenuis*

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Omnivorous mirid bugs are a key component of solanaceous crop protection. They are able to increase crop resilience against arthropod pests by preying on a variety of pests and, in prey absence, by stimulating the plant defences. *Nesidiocoris tenuis* (Reuter) (Hemiptera: Miridae) is widely employed in protected tomato crops in the Mediterranean basin, via artificial releases or conservation strategies. Among these, the use of selective and alternative pesticides is of paramount importance in Integrated Pest Management framework. In this context, we aimed at characterizing the lethal and sublethal impacts of three insecticides (i.e., chlorpyrifos, lambda-cyhalothrin and spinosad) and four nanoemulsioned plant essential oils (i.e., anise, fennel, garlic and lavender) on *N. tenuis* adults. Through topical exposure, we first conducted concentration-mortality bioassays on the mirid predator, then we assessed the sublethal effects on the reproduction and the behavior (orientation towards volatiles of a host plant) of two low lethal (LC10 and LC30) and a sublethal (LC1) concentrations. The tested insecticides and essential oils caused different toxicity towards *N. tenuis* adversely affecting its reproduction and behavior. The results thus suggest that these substances should be carefully used in areas where the predator is released or conserved, and that these could be taken into consideration when aiming at limiting the crop damage caused by this species.

KEY WORDS: integrated pest management, organic farming, botanicals, orientation behavior, reproduction.

POSTER

First multi-target application of exclusion net in nectarine orchards: effectiveness against pests and impact on beneficial arthropods

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In the past, the fruit orchard protection was mainly based on the use of synthetic pesticides to prevent or limit pest damage. However, the indiscriminate and excessive use of insecticides has often induced resistance in pests in addition to increasing hazards for workers, consumers and the environment. In peach and nectarine orchards, the management of main pests such as *Grapholita molesta* (Busck) has influenced the crop protection guidelines for years. Despite the efficacy of well-established IPM procedures against this pest, insecticide applications to supplement this strategy under high pest densities and to control new invasive pests, such as *Halyomorpha halys* (Stål), nullify them. Physical exclusion strategies have been evaluated as an alternative for the control of insect pests. In 2016 and 2017, the effect of anti-insect nets on pest populations and fruit damage as well as the possible impact on predators were monitored in Piedmont (NW Italy) in two nectarine orchards during the growing season and at harvest. In each orchard, trials were sorted in a randomized complete block design with three replicates for each of the following treatments: netted plots, un-netted control plots, and un-netted plots treated with insecticides. The netted plots were covered with a pearl anti-hail photoselective net placed at the petal fall and removed after harvesting. During this time, no insecticides were applied in netted plots and un-netted control plots. Moreover, every 10 days, pest [*G. molesta*, *H. halys*, *Drosophila suzukii* (Matsumura)] and predator populations were monitored using traps and visual inspection in netted and un-netted control plots. Predator populations were also assessed after harvesting thanks to a knockdown treatment applied in all the three treatments. At harvest, samples of fruits were checked for damage caused by *G. molesta* and *H. halys*. The pearl anti-hail photoselective net significantly reduced *G. molesta*, *H. halys* and *D. suzukii* and populations in comparison with un-netted control plots. At harvest, fruit damage caused by *G. molesta* and *H. halys* in netted plot was reduced up to 90% and to 78%, respectively, compared with insecticide-treated plots. During the growing season, higher captures of predators were recorded with yellow sticky traps in un-netted control plots in comparison with netted plots; however, at the end of the trials, no differences were recorded between the netted and the un-netted plots treated with insecticides following the final knockdown treatment. The exclusion net proved to be effective in controlling *G. molesta* and *H. halys* damage preserving the production of healthier fruits with a strong reduction of insecticide treatments (up to seven less) and of their related costs. Moreover, the exclusion net can be considered as a ready to use protection system against invasive alien species, a severe and growing threat due to globalization and climate change.

KEY WORDS: *Grapholita molesta*, *Halyomorpha halys*, *Drosophila suzukii*, predators, photoselective net.

Evaluation of seed predation by carabid beetles in a wheat agroecosystem of Central Italy using PCR-based gut-content analysis

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As generalist predators, carabid beetles can play a key role in biological control of weed seeds in agroecosystems. Weed seeds are an important issue in agricultural fields, e.g. wheat, requesting high costs and efforts for their management. Therefore, improving our knowledges on the feeding behaviour and seasonal effectiveness in seed predation would be useful to enhance biological control.
We designed and evaluated species specific primers, targeting chloroplast genes fragments, to detect predation upon three economically relevant weeds: Lolium multiflorum, Papaver rhoeas and Sinapis arvensis. Amplicon sizes ranged from 244 to 287 bp, allowing the detection of recent feeding events.
From May to October 2018, we collected carabid beetles using pitfall traps filled with EtOH, in a wheat field at the experimental station of the Department. Samples of Pseudoophonus rufipes and Pterostichus melanarius were dissected and the gut processed for DNA extraction. PCR reactions were conducted to evaluate the entity of field predation upon weed seeds. The observed overall predation by P. melanarius towards the three species was higher than that of P. rufipes. In particular, overall predation by P. melanarius upon L. multiflorum was 27.2% compared to 2.9% by P. rufipes.
Our results highlight a positive overall predation towards weeds, confirming the importance of the two carabid species in reducing the amount of seeds in the soil. The enhancement of soil functions and ecosystem services aiming at promoting carabid beetle diversity and foraging activity might contribute to reduce herbicide use maintaining a reasonable crop yield.

KEY WORDS: PCR, molecular gut-content analysis, biological control.

POSTER

Integrated control of *Planococcus ficus* by mating disruption and augmentative release of *Anagyrus vladimiri*

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The vine mealybug, *Planococcus ficus* (Signoret), is one of the grape pests most difficult to control effectively, also in view of its cryptic behavior that reduce the efficacy of the chemical control. The cryptic behavior and the increasing restrictions on the use of insecticides promoted the development of alternative and sustainable control practices, such as classical biological control and pheromone-based control strategies. These techniques have been tested together in a field trial in a highly *P. ficus*-infested organic vineyard in Sardinia in 2019. The treatments compared were: mating disruption, through the release of 500 dispensers/ha for a total sex pheromone load of 90 g synthetic sex pheromone; augmentative biological control, through the release in mid-late June of 2,000 *Anagyrus vladimiri* Triapitsyn/ha; joint application of mating disruption and biological control; untreated control. Each treatment was repeated three times in plots of 5-7 ha. The effectiveness of the tested treatments was compared by counting the number of captured males in monitoring pheromone traps, and assessing the mealybug density per plant, the parasitism rate by *A. vladimiri* and the crop damage. At the experimental densities of *P. ficus* and without chemical applications, both sustainable control techniques were ineffective, as mating disruption did not suppress male captures in pheromone traps, and the parasitoid release did not increase the parasitism rates. Mating disruption applied jointly with *A. vladimiri* release did not affect the foraging behavior of parasitoids. Overall, mating disruption, parasitoid release and both techniques, applied together, did not reduce the density of *P. ficus* nor the crop damage at harvest compared to control plots. Possible causes could be a mealybug density too high for an effective disruption of male-female sexual communication, the release of an undersized number of parasitoids or a delayed release. Our results highlight the importance of defining protocols of integrated management in which the control efforts are adapted to the pest infestation level. Further studies are required in order to determine; the mealybug density above which mating disruption is ineffective; the optimal release period of parasitoids; the number of parasitoids to be released with regard to the mealybug density; the potential integration with compounds allowed in organic agriculture; and the economic sustainability of the proposed solutions.

KEY WORDS: biological control, pheromone-mediated control, vine mealybug, organic viticulture.

ORAL PRESENTATION

Pheromones of the fig weevil *Aclees taiwanensis*: evaluation of their attractiveness in the laboratory and in the open field

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The fig weevil Aclees taiwanensis Kôno (Coleoptera: Curculionidae) is an invasive species of Asian origin, accidentally imported in Italy (Tuscany) in 2005 through the trade of ornamental plants and it is currently present in seven regions of Central-Northern Italy. The insect attacks many species of the genus *Ficus* including the common fig, *Ficus carica*. The damage to plants is caused both by adults, which feed on buds, leaves, and ripening fruits and by the xylophagous larvae that dig tunnels at the base of the trunk. The control strategies implemented so far, with synthetic chemicals or biological agents, have not given satisfactory results. However, in other species of xylophagous beetles, it has been shown that the capture of adults through aggregation or sex pheromone can be applied to monitoring and/or control. This work aimed to verify whether reconstituted mixtures of Volatile Organic Compounds (VOCs), emitted by adults of both sexes of *A. taiwanensis*, are involved in the recognition and attraction of conspecifics and if it is possible to use them to control the fig tree weevil. The compounds contained in the VOCs were sampled through solid-phase microextraction (SPME) from insects collected in late spring-early summer 2019 and identified by gas chromatography coupled to mass spectrometry. The identified VOCs, terpenes with limonene as the main compound, were used both in the laboratory and in preliminary field tests to evaluate their bioactivity. In the behavioral laboratory tests, in a two-way olfactometer, the reconstituted VOCs containing the main compounds were found to attract the opposite sex. Reconstituted VOCs of both sexes containing also minor components attracted only females. These latter mixtures were also tested between June and July 2020 in a fig orchard. The reconstituted VOCs applied on salivary rollers placed under a textile sleeve (Rhynchotrap) surrounding the base of the tree trunk, resulted more attractive than control.The results obtained, although preliminary, suggest an attractive effect of reconstituted VOCs and their possible application for monitoring or mass trapping. However, further investigations will be necessary to determine the most appropriate modality to release the reconstituted VOCs in the field, the different activity of enantiomers of the chiral compounds, and the differences in the profile of the VOCs of adults collected in different periods of the year.

KEY WORDS: pheromone traps, olfactometer, invasive species, *Ficus* spp., monitoring.

POSTER

New findings on parasitoids of *Cryptoblabes gnidiella* in Italy

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Native to the Mediterranean basin, the honeydew moth, *Cryptoblabes gnidiella* (Milliére), represents a primary fruit pest in Uruguay and Brazil. Beyond South America and southern Europe, this moth has currently spread throughout North, West and South Africa, India, Pakistan and South-East Asia, New Zealand, USA, Bermuda and Hawaii. It is a polyphagous species, feeding on more than 60 species belonging to 30 plant families, including the spurge flax, *Daphne gnidium* (Thymelaeaceae), its most common wild host plant. Though *C. gnidiella* was never deemed as a major pest of grapevine in the Mediterranean area, an observed increase in populations in recent years led to reconsider its importance in Europe as a primary carpophagous moth. Larval feeding on mature grapes, especially on late ripening varieties, accounts for a progressive decay and drying of the bunch, and eventually led, in high humidity rate conditions, to the proliferation of pathogenic rotting molds and saprophagous insects. Despite its increasing importance, very little is known about the parasitoid community associated with this moth in both vineyards and wild environments. We provide here an updated list of parasitoids of *C. gnidiella*, with particular reference to the Italian fauna. So far, 29 taxa have been associated with the honeydew moth worldwide, including doubtful records and unidentified species. They belong to eight families of Hymenoptera (Bethylidae, Braconidae, Chalcididae, Eulophidae, Ichneumonidae, Perilampidae, Pteromalidae, Trichogrammatidae) and one of Diptera (Tachinidae). Only six taxa have been recorded in Italy: two braconids, *Phanerotoma* sp. and *Bracon hebetor*, three ichneumonids, *Itoplectis* sp., *I. alternans, I. tunetana*, and the bethylid *Goniozus gallicola* (the latter doubtful). Thanks to our investigations, carried out in recent years in Italian vineyards (Tuscany, Apulia) and on *D. gnidium* in Tuscany, five previously unrecorded parasitoids have been found on *C. gnidiella*: two braconids and three ichneumonids. These new findings may shed light on potential biological control agents of what can be considered a full-fledged threat for Mediterranean viticulture.

KEY WORDS: grapevine, biological control, Hymenoptera, Diptera, honeydew moth.

POSTER

A study on the parasitoidsof *Halyomorpha halys* for the development of biological control strategies in Trentino, Italy

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The brown marmorated stink bug (BMSB), *Halyomorpha halys* Stål, is an invasive, herbivorous insect species that was detected in Italy in 2012, in the Emilia Romagna region. It is considered a major threat to European agriculture because it damages many crops. Due to its high mobility, chemical control is considered inefficient. Thus, the biocontrol could be a valid alternative. This research aimed to investigate the occurrence of parasitoids' activity in the province of Trento, to assess which species can parasitize the BMSB eggs, and to evaluate their potential parasitization rates. The study was conducted from April to August 2019 covering the area from Mezzocorona to Besenello, comparing two approaches. 1) Fourteen sentinel egg traps, baited with egg masses produced by BMSB individuals in laboratory conditions, were positioned in seven sites. 2) Egg masses were directly attached to tree leaves in other four sites. The total was 255 exposed egg masses. We found that the second method was more efficient to attract parasitoids. Moreover, besides finding the indigenous generalist parasitoid *Anastatus bifasciatus*, we detected, first-ever in Trentino, the alien wasps *Trissolcus mitsukurii* and *T. japonicus*, highly specialized parasitoids of BMSB, native to Asia. They showed remarkable parasitism rates (73 – 89%), suggesting that their employment for classical biocontrol on BMSB has the potential to reduce the latter's damages.

KEY WORDS: *Trissolcus japonicus, Trissolcus mitsukurii,* brown marmorated stink bug, parasitoids, biocontrol.

POSTER

Chestnut ecotypes resistant to *Dryocosmus kuriphilus*. New assessments for integrated pest management

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The Asian Chestnut Gall Wasp (ACGW) *Dryocosmus kuriphilus* Yasumatsu (Hymenoptera: Cynipidae), reported in Italy since 2002, is a major threat to chestnut orchards. ACGW lays eggs into chestnut buds and induces the development of galls that cause severe reduction of plant photosynthetic activity. This damage results in an intense productive decline in the attacked plants. Currently, the most effective method to control ACGW is the use of its natural enemy *Torymus sinensis* Kamijo (Hymenoptera: Torymidae), introduced in Italy since 2006. However, despite the good results achieved, the parasitoid is still unable to completely control the pest in some areas and with particular climatic conditions. Therefore, besides the evaluation of the parasitoid control action, one aim of the Chestnut Special Project, coordinated by the Institute for Sustainable Plant Protection (IPSP) of the Italian National Research Council (CNR), is to assess resistance or tolerance to ACGW attack in local chestnut ecotypes. Through sampling and based on agronomic and morphological plant descriptors, some ecotypes, showing resistance/tolerance to ACGW, have been identified in the Campania region. To highlight the potential phenomenon of cross-resistance the ACGW resistant ecotypes have also been evaluated against *Gnomoniopsis castaneae* Tamietti (syn. *G. smithogilvyi*), the main agent involved in nut rot in chestnut. During the study, monitoring activities were carried out in Campanian chestnut orchards where both susceptible and resistant ecotypes were present. The aim of this monitoring was to compare the damage levels caused by *D. kuriphilus* and *G. castaneae* on different ecotypes and to characterize the type of resistance. Two samplings per year were performed: the first between June and July, to evaluate the eggs laid by ACGW or repellence phenomena expression; the second one in spring, to observe the presence of ACGW loculi and larvae, and their development stage. Several variables have been considered: galls per linear meter, healthy and infested shoots, healthy leaves, the mean number of loculi and larvae per gall, the percentage of *T. sinensis* parasitism. The totality of the considered parameters highlights that the resistant ecotypes showed a significant difference compared to the susceptible ones. In particular, shoots from resistant trees resulted mostly healthy and free of damage caused by *D. kuriphilus*. A single ecotype showed almost complete absence of attacks and damages. Conversely, the susceptible ecotypes show a higher number of galls, both on shoots and leaves, a lower number of healthy and well-developed leaves, and lower parasitization levels.
Similar results were observed about the presence of *G. castaneae*: even in this case the resistant ecotypes displayed a lower incidence of damages ascribable to the pathogen. This result shows how the hypothesis of cross-resistance is a real phenomenon and an interesting field for future investigation.

KEY WORDS: *Castanea sativa*, cross-resistance, invasive species, parasitization.

POSTER

Host-location behavior of the generalist parasitoid *Theocolax elegans*: role of VOCs emitted by *Rhyzopertha dominica* larval faeces

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Integrated Pest Management in post-harvest can use a limited range of authorized techniques and products. In this context, the use of classical biological control agents is controversial, due to concerns about possible contamination attributable to the release of natural enemies. *Theocolax elegans* (Westwood) (Hymenoptera: Pteromalidae) is a generalist ecto-parasitoid, able to parasitize the larvae of several beetle species (*Sitophilus* spp., *Stegobium paniceum* L., *Rhyzopertha dominica* F., *Callosobruchus* spp.), as well as to attack the larvae of the Lepidoptera *Sitotroga cerealella* (Oliver). *Theocolax elegans* is a cosmopolitan insect and can be found naturally within infested grains. Field studies shown that the employment of this parasitoid can be useful for the control stored-product pests. Nevertheless, few studies focused on the behavior and the preferences of this wasps, and this information can be helpful to understand the outcomes in field conditions. The aim of this study was to evaluate the olfactory preferences of *T. elegans* females towards different potentially attractive substrates. In detail, responses to wheat (var. Tito Flavio) uninfested and infested by *R. dominica* larvae and the faeces resulting from trophic activity of the host larvae were evaluated. Lastly, gas chromatography coupled to mass spectrometry was used to identify the volatile components emitted by the substrates to determine the presence of any kairomones for parasitoid females.
The olfactory preferences were tested in bioassays in still-air arena, in which one stimulus (no-choice) or two stimuli simultaneously (choice) were presented to every single female. During the observation (5 minutes), the residence time spent on each stimulus was also recorded. Bioassays showed that females of *T. elegans* preferred *R. dominica* infested wheat to healthy wheat and spent significantly more time in both choice and no-choice contexts. In addition, when a small amount (0.05 g) of feces produced by *R. dominica* larvae was added to healthy kernels, these became more attractive than healthy wheat and were as attractive as the infested kernels. This result was supported by the analysis of the residence times, highlighting that the parasitoids spent the same time inspecting the healthy wheat added with faeces and the infested wheat. Therefore, the larval faeces seem to be the key cue to attract the parasitoids, since, even in absence of kernels, the females of *T. elegans* were attracted by faeces. The analysis of volatile emissions showed a limited number of molecules produced by the substrates. However, dodecanal was emitted exclusively by healthy kernels, while fenchone, methyl decanoate and 1-pentadecene just from the infested ones. Among these compounds, only methyl decanoate and 1-pentadecene were also identified in the volatile profile of larval faeces, suggesting that these molecules can be responsible for the attractiveness toward the parasitoid and that fenchone could be directly related to the presence of *R. dominica* larvae.

KEY WORDS: biological control, semiochemicals, kairomones, choice-test, GC-MS.

ORAL PRESENTATION

Comparison between sustainable strategies for controlling the European corn borer: potential impacts on beneficial fauna and aflatoxin perspective

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The European corn borer (ECB), *Ostrinia nubilalis* Hübner (Lepidoptera: Crambidae), is a major pest in maize fields. Furthermore, the ECB infestations is considered as predisposing factor for fungal colonization, including the toxigenic strains of *Aspergillus flavus* and *Aspergillus parasiticus*, well known producer of aflatoxin B. During the two years project (2017-2018), the efficacy of a biological strategy based on *Thrichogramma brassicae* and *Bacillus thuringiensis* and a conventional chemical approach (chlorantraniliprole) were compared with an untreated control. The aims of this study were to asses: i) the efficacy of the strategies on ECB infestation; ii) the impact of the strategies on beneficial fauna; iii) the role of ECB in promoting aflatoxin B1 kernels contamination. Conventional chemical approach based on chlorantraniliprole has been detected as the best strategy to reduce pest outbreak followed by biological control and the untreated control. However, no significant differences in production were found among all the strategies. The role of ECB in promoting aflatoxin B1 was confirmed only when insect damages occurred simultaneously with strong climate conditions, such as happened in 2017, when the high ECB infestation, together with the dry and hot meteorological conditions resulted in high level of aflatoxin B1 in pre-harvest maize. On the other hand, in the low risk condition of aflatoxin contamination, like 2018 season, the concentration of this contaminant was significantly lower than the limit in all strategies. Moreover, any significant differences in the abundance and activity density of ground dwelling arthropods and canopy insects were found among pre and post treatment, demonstrating a short time selectivity of chlorantraniliprole on natural enemies. In conclusion, our study provides a practical contribution for sustainable maize production in Italy, demonstrating that biological strategy, although less effective than chemical control, can be a feasible strategy in organic system to obtain a partial control of the second larval generation, without any increment of AFB1 in grains and yield loss.

KEY WORDS: European corn borer, *Ostrinia nubilalis*, biological control, integrated pest management, aflatoxin B1.

POSTER

Molecular identification of koinobiont parasitoids in galling insects, the case study of *Dasineura oleae*

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Dasineura oleae was usually considered as a minor pest in olive orchards, but in the last years, several outbreaks have been registered in some Mediterranean countries, including Italy. In order to actuate conservation biological control programmes, the identity and abundance of its parasitoids must be assessed. In Italy *Mesopolous mediterraneus*, *Mesopolobus aspilus*, *Platygaster demades* and *Platygaster oleae* were recorded as parasitoids of *D. oleae*. While parasitization may be evident in some groups (e.g aphids), in galling insects will not be evident if the gall is not dissected. Furthermore, all *D. oleae* parasitoids detected in Italy are koinobionts, therefore parasitization will not be evident until development of third instar larvae. Additionally, the relative abundance of each parasitoid species can't be assessed until adult emergence, but the available techniques do not allow an accurate estimation due to the high parasitoid mortality in laboratory conditions. The aim of the present work is to develop a molecular technique that would enable both an early detection of the parasitism rate and the assessment of relative abundance of each group of parasitoids, independently from the development rate. For this purpose, leaves showing typically galls of *D. oleae* were collected from organically managed olive orchards located in the Grosseto district (Italy) nearby the core of the outbreak area. The sampling was performed in February, when most of the population of *D. oleae* turns in to a third instar larvae. 979 galls were dissected under sterile conditions, visually inspected and checked for vitality movements, signs of parasitization or other abnormalities, then individually stored in ethanol at -20°C. Moreover, some infested branches were laboratory-reared to obtain adults of parasitoids and D. oleae. Several adults of *Mesopolobus* spp. and *P. demades* were identified while no individuals of *P. oleae* were obtained. Sequences of a cytochrome oxidase subunit I (COI) gene region from reared parasitoids were obtained and aligned with those available in GenBank of *Mesopolobus* spp. and *Platygaster* spp. Three forwards and one reverse primer were designed for each target parasitoid to anneal to specific conserved COI region. Primer sets were individually tested for their specificity in order to select the most suitable primer pairs for the detection of parasitoids. Primers were also tested in multiplex PCR to prevent interferences and interactions among primers. The results have demonstrated that the molecular approach is more sensible than visual inspection. Furthermore, the rapidity of this method allows to screen a large number of samples providing an important information about parasitization efficacy of *Mesopolobus* spp. and *Platygaster* spp. parasitoids. However, similar studies showed that molecular method may provide parasitization estimation similar to traditional methods, highlighting how the magnitude of difference between the methods presumably depends on the biology of the studied species.

KEY WORDS: *Dasineura oleae*, *Platygaster* spp., *Mesopolobus* spp., koinobionts parasitoids, biological control, multiplex real time PCR.

POSTER

Classic biological control of the brown marmorated stink bug through releases of *Trissolcus japonicus*: detailed investigations on the impact and parasitization efficacy in Emilia-Romagna region

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Following the decree "Emergency measures for the prevention, control and contrast of the brown marmorated stink bug" of 29 April 2020, during summer 2020, a concerted work of classical biological control which involved a large area of Northern Italy took place by means of inoculative releases of the exotic parasitoid *Trissolcus japonicus* (Hymenoptera: Scelionidae). This work is part of the releases carried out by the Emilia-Romagna region and it has been performed with the aim of studying the efficacy and diffusion of the parasitoid and its impact on non-target species providing important results for planning future biological control actions for the sustainable management of the brown marmorated stink bug *Halyomorpha halys* (Heteroptera: Pentatomidae). The study was conducted in two selected fields cultivated with *Paulownia tomentosa* in the provinces of Modena and Reggio Emilia. To obtain information on the ability of the parasitoid to spread from the release point, each site was divided into three concentric areas with radius of 10, 30 and 50 m respectively, whose center corresponded to the release point. The number of plants to monitor was calculated based on the size of each concentric area and on a total inspection time of 6 hours / man. The monitoring included the inspection of trees and herbaceous species, collecting naturally laid eggs of *H. halys* and other pentatomids. In addition, in only one of the two sites, sentinel egg masses were exposed on non-monitored plants in a number that was proportional to the size of each concentric area. In each site, the number of female fertile parasitoids released ranged between a minimum of 100 up to a maximum of 450. The releases were carried out at the end of June and July 2020. Collected eggs were classified based on their distance from the release point and were kept under controlled condition to verify the number and species of emerged parasitoids. The unhatched eggs were dissected with the purpose to include the number of fully developed parasitoid adults that failed to emerge among the total number of parasitoids within the parasitization rate.
Considering the target species, the results showed a parasitization of 5% by *Trissolcus mitsukurii* (Hymenoptera: Scelionidae), of 1.5% by *Anastatus bifasciatus* (Hymenoptera: Eupelmidae) and of 0.5% by the hyperparasitoid *Acroclisoides sinicus* (Hymenoptera: Pteromalidae). *Trissolcus japonicus* has shown a parasitization rate close to 1% against the target specie and has never been found in eggmasses of non-target species. Egg masses parasitized by *T. japonicus* were found at a distance of 50 m from the release point. Molecular analyses are underway to verify whether the parasitoids belonged to the released populations or to adventive populations already present in the area.

This work is part of the project Haly.Bio, funded by the Emilia Romagna region within the Rural Development Plan 2014-2020 Op. 16.1.01 – GO EIP-Agri - FA 4B, Pr. "Haly.Bio" and coordinated by CRPV

KEY WORDS: Biocontrol, parasitoids, invasive species, Pentatomidae, Scelionidae.

POSTER

Repellency of four essential oils against *Halyomorpha halys* adults in different physiological-behavioural phases

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Halyomorpha halys (Stål) (Heteroptera: Pentatomidae) is a highly polyphagous pest native to East Asia, accidentally introduced in several countries. It causes severe damage to numerous agricultural crops during its summer feeding phase and becomes a dwelling nuisance in autumn, due to the aggregation of overwintering adults inside buildings. The attempts to counteract *H. halys* outbreaks with chemical control led to a massive increase in broad-spectrum insecticide use, disrupting previous IPM programs, increasing costs for the growers and causing negative consequences to the environment and consumers. In the view to develop sustainable strategies to manage this pest, an approach is to consider the use of natural substances, such as essential oils (EOs), which are safe, biodegradable, and can be used also in combination with other techniques. The repellent potential of four EOs (ginger, clove, vetiver and turmeric) was tested on *H. halys* adults of three different physiological-behavioural phases: exiting from overwintering (EXOV), active during summer (SUMM), entering in overwintering (ENOV). In a two-choice apparatus, fresh food was used as attractant in both sides, together with three males when testing ENOV individuals. A filter paper with different concentrations of the EOs (50%, 25%, 12%, 6%, 3% and 1%) was the treatment side, and a filter paper with ethanol acted as control. The position of the individually tested bugs was recorded after 1, 6, 24 hours. Results showed that all tested EOs were repellent at concentrations higher than 3%, independently of sex or time of exposure. Turmeric and clove were the most repellent EOs, whereas ginger and vetiver showed on average a medium-low repellency. Significant differences emerged among the physiological-behavioural phases, with SUMM individuals showing a greater repellency to many of the tested concentrations, and EXOV individuals being overall the least susceptible to these substances. The response to vetiver oil was ambiguous, as at 25% it elicited both a strong repellency in SUMM and a strong attraction in EXOV. Turmeric and clove EOs are promising candidates in integrated pest management strategies to reduce attacks by *H. halys* to susceptible crops especially during summer, as well as to prevent the entrance of overwintering bugs in buildings in autumn.

KEY WORDS: Invasive species, agricultural pest, nuisance pest, eco-friendly strategies, natural substances, integrated pest management, Pentatomidae.

ORAL PRESENTATION

The use of long-lasting insecticide-treated nets to control the Japanese beetle *Popillia japonica*: a laboratory and field evaluation

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The Japanese beetle *Popillia japonica* Newman is a quarantine insect pest that recently invaded northern Italy, between Piedmont and Lombardy regions. This was the first record of this pest in mainland Europe. From its discovery onwards, the invaded territory is getting wider, reaching in 2019 more than 7,500 square Km in Italy. In the meanwhile, findings of adults of *P. japonica* occurred also in Switzerland in 2017. Due to its high polyphagy and the ability to rapidly spread into new areas, this insect is considered a serious threat to agriculture. Confirming the foregoing, it is noteworthy that the new EU regulations have inserted the Japanese beetle within the EU list of priority pests (Reg UE 2019/1702). In the last decades, several trials on biological control of the Japanese beetle by entomopathogenic nematodes and fungi have been carried out with variable efficacy. However, the necessity of an integrated pest management approach to improve its control has arisen. Long-lasting insecticide-treated nets (LLINs) are a technology recently evaluated for the control of other agricultural pests with an attract-and-kill strategy. Here, we present results from laboratory evaluation of two different LLINs, Storanet (BASFTM) and ZeroFly (VestergaardTM), against *P. japonica* adults. Both were effective in killing the beetles; however, some differences emerged if different exposure times were compared: ZeroFly always gave 100% mortality in tests from 5-sec to 30-min exposure; Storanet showed 100% mortality only with 30-min exposure and going down to 99%–89% mortality for 15-min to 5-sec exposure. Furthermore, in order to define the modality of the field use of such LLINs, we present the result of field experimentation in which attract-and-kill devices were left exposed outside for the entire flight period of *P. japonica* , which is roughly from June to September. Results showed that the full effectiveness of the LLIN was maintained for the first-month exposure. After that time, the effectiveness appeared to progressively decline.

KEY WORDS: alien pest, integrated pest management, scarab beetle, pyrethroid.

POSTER

Application of agroecological strategies at farm scale: effects of cover crop termination on the ground-dwelling arthropods

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Cover crops are one of the most implemented practices to increase diversity and improve sustainability of agricultural systems. The inclusion of cover crops in rotations, instead of long bare fallow periods, may positively cascade up on multiple ecosystem services. A number of studies have documented positive effects on yield, nutrient availability, nitrogen fixation, weed control and arthropod dynamics.
A key aspect of cover crop management is the method of termination before cash crop is planted. The impact of cover crop termination methods on arthropods has been poorly investigated and the aim of this study was to assess the effects of termination methods on ground-dwelling arthropods. The conventional mechanical termination method in integrated and organic farming - i.e. incorporation in the soil as green manure by means of a disc harrow - was compared to the flattening by a roller crimper. This two-year study was carried out from September 2015 to August 2017 in the experimental organic farm of CRA-SCA in Metaponto (MT), Southern Italy. Two different crop systems were investigated: i) cauliflower (*Brassica oleracea* L. var. *botrytis*) was grown in autumn after the termination of a mixture of *Vigna unguiculata* (L.) Walp., *Pennisetum glaucum* (L.) R. Br. and *Raphanus sativus* L.; ii) tomato (*Solanum lycopersicum* L.) was cropped in spring and summer after the termination of a mixture of *Hordeum vulgare* L. and *Vicia sativa* L. Ground beetles (Coleoptera: Carabidae), rove beetles (Coleoptera: Staphylinidae) and spiders (Araneae) were sampled by means of standard pitfall traps throughout the growing season of both cash crops. All beetles were identified to species, except for individuals belonging to the subfamily Aleocharinae (Staphylinidae); spiders were identified to family level. Overall, the termination techniques of cover crops strongly affected the abundances of the arthropods, with specific responses influenced by taxa and sampling dates. Ground beetles (4380 individuals sampled) showed inconsistent responses between sampling seasons. Roller crimper overall increased the abundance of these beetles in the first growing season of both cash corps, whereas in the second year no significant effect could be attributed to the termination technique. Rove beetles (1675 individuals sampled) were more abundant in plots where the cover crops were terminated by roller crimper. The only exception was in the second growing season of tomato, when the scarcity of captures did not allow any sound analysis. Finally, the green manuring increased the abundance of spiders (4468 individuals overall caught), especially in the first sampling date after cover crop termination. Albeit different taxa showed different response, the termination of cover corps by roller crimper, which leaves plant residues on soil so obtaining a natural mulching, seems to generally increase the abundance of ground dwelling arthropods. Given that most of sampled species belongs to generalist predators, their increased abundance could possibly lead to augmented levels of biological control. This topic should be investigated with experiments specifically planned to evaluate predation rate.

KEY WORDS: predators, Carabidae, Staphylinidae, Araneae, pitfall traps.

ORAL PRESENTATION

Entomopathogenic nematodes as a potential biocontrol agent against *Bactrocera oleae*

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The olive fruit fly *Bactrocera oleae* is one of the most serious and economically damaging insects worldwide, affecting the quality and quantity of both olive oil and table olives. Until now, control measures for this pest have been based mainly on the use of organophosphate insecticides, but in the last decade, the European Community (EC) has defined new approaches on their usage, introducing restricted limits for the registration of new pesticides and the elimination of conventional products characterized by a high environmental impact and human and animal health risks. At the same time, the EC strongly encourages non-chemical alternatives and the use of natural products in plant pest control. Third instar larvae and pupae of several tephritid flies were reported to be susceptible to entomopathogenic nematodes (EPNs), but few studies have been carried out on the olive fruit fly. To evaluate the susceptibility of *B. oleae* larvae and pupae to EPNs, laboratory bioassays were performed in 2018-2019. Two commercial EPN species, *Steinernema feltiae* and *Heterorhabditis bacteriophora* and two indigenous Italian strains of *H. becteriophora* and *Steinernema carpocapsae* were used. Moreover, a semi-field bioassay, using olive fruits in the soil was performed to evaluate if EPNs were able to enter inside the olives and kill *B. oleae* during the pupal stage or the emergence of adults. All bioassays with *B. oleae* were performed in well plates, filled with sterile soil (n = 30 for each EPN strain and insect stage). Adult emergence and mortality were recorded daily for 15 days. Dead pupae and adults were dissected to assess nematode infection. Our results indicate that all tested life stages of *B. oleae* are susceptible to EPNs. Although larvae were more susceptible to EPNs infection than pupae, they spent a relatively short time in the soil before developing into pupae. The most noteworthy result was obtained with *S. feltiae* which was able to infect more than 80% of larvae and it killed the pupae inside olive fruits and the adults during their emergence with the same efficacy. Since this tephritid fly spent several months in the soil, the use of EPNs, in particular *S. feltiae* could be a promising method to control this pest.

KEY WORDS: *Heterorhabditis bacteriophora*, olive fruit fly, *Steinernema* spp.

POSTER

Preliminar assessment of commercial and experimental insecticide toxicity on *Chilocorus bipustulatus*

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The complex of natural enemies of the black scale *Saissetia oleae* (Olivier), in the Sicilian olive groves consists of several indigenous species, and among them the ladybeetle *Chilocorus bipustulatus* (L.) is one of the most common. Insecticides used to control the insect pests such as the olive fruit fly *Bactrocera oleae* (Rossi) and the black scale itself, can affect the survival of such biocontrol agents.
In the framework of a research project (DI.OL.) funded by Ministry of Agricultural, Food and Forestry Policies, laboratory ecotoxicological bioassays were carried out to assess the toxicity of several agrochemicals commonly used in the olive grove to control the two insect pests (i.e., pyriproxifen, dimethoate, spinosad, paraffinic mineral oil, sweet orange essential oil), two experimental clay-based formulation (bentonite and n-hexanal-containing bentonite), and a commercial formulation of mycorrhizal fungi. Residual tests were carried out by spraying twigs about 10 cm long and keeping predator adults into the experimental arena. Mortality was assessed every 24 h for a week.
The two bentonite materials induced extremely low and not statistically significant mortality compared to dimethoate and mycorrhizal fungi. Further tests were performed to evaluate the lethal effect of a different bentonite formulation and two zeolites (n-heptanal-containing bentionite; clinoptilolite and copper(II)-exchanged clinoptilolite) in addition to dimetoate, the product containing mycorrhizal fungi and a product containing *Beauveria bassiana* where zeolites and bentonite induced very low mortalities significantly different from those induced by dimethoate and mycorrhizal fungi. To date, there are few studies on side effects on *C. bipustulatus* and the data obtained in this research represent a first contribution to the knowledge of the toxicity on this predator not only of conventional products, but also of new formulations and experimental eco-friendly promising products to be used in the IPM strategies in olive grove. Funding was provided by the Ministry of Agricultural, Food and Forestry Policies in the framework of the research project DI.OL." - Defense from harmful organisms in conventional and intensive olive crops" (CUP C31I16000010001).

KEY WORDS: *Chilocorus bipustulatus*, insecticides, porous mineral powders, entomopathogenic fungi, toxicity.

POSTER

Applied Biotremology: a strongly expanding new discipline

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Biotremology is the science that studies mechanical communication using surface-borne waves in animals. Among the numerous animal classes that use vibrations, arthropods make the largest use of them to mediate different interactions (e.g. predation, parasitism, mating). In many insect species, mating behavior relies on vibrational signals and the establishment of a stable vibrational duet between a male and a female is a prerequisite to accomplish mating. The exchange of vibrational signals provides relevant information, among which are included identity, position, distance, but also quality (e.g. size, age, health status) of the communicating individuals. In this way, vibrational signals fulfill a fundamental role in the process of pair formation of many insects whose behavior, on the other hand, could be potentially manipulated through the transmission of artificial disrupting signals into the substrate (i.e. plants). This is the concept basic to the "applied biotremology", a new discipline in strong expansion considering the number of new specialized labs worldwide. This research involves several topics and species, including bark beetles, social insects, bugs and hoppers that are either vectors of plant diseases or responsible for massive yield losses. Among the possible approaches of applied biotremology we can find techniques of trapping, detection, deterrence and mating disruption. The latter has been applied as an open field experiment since 2017 in Trentino for the control of two grapevine leafhoppers, *Empoasca vitis* and *Scaphoideus titanus*. Thanks to the common efforts of engineers and entomologists, new prototypes of disturbance vibrations emitters have been developed and installed in an organic "vibrational vineyard". After 3 consecutive years of application, without any use of insecticides, we measured a significant reduction (< 50%) of the leafhoppers populations and related direct damage in the treated area, compared to the control. On the contrary, no impact has been detected on the populations of predators that commonly use vibrational signals (i.e., spiders and lacewings). The validation of the vibrational mating disruption can be assessed through a direct method, using manual sampling, and an indirect method, using yellow sticky traps. Surprisingly, we found that the sticky traps placed inside the treated area captured many more leafhoppers and with a significantly higher sex rate (% males, constantly around 90%) than those inside the control. We hypothesize that this is due to the reduced chance of males meeting females which forces them to stay in a constant stay of call-fly, thus making them more susceptible to be caught by the sticky traps. In this way, the sticky traps could be used in the future as a tool to assess the actual functioning of the vibrational mating disruption in the field. In conclusion, this and other experiences currently at work in different research centers, clearly indicate that applied biotremology is a promising novel approach for use in crop protection. Methods of mating disruption, as well as trapping and monitoring are potentially achievable for all pests that rely on vibrational signals for intraspecific communication, provided the deep knowledge of the behaviors associated with the vibrational communication is available. For all these reasons, we foresee that innovations developed thanks to the applied biotremology, as new eco-compatible tools, will soon be available for IPM implementation in the field.

KEY WORDS: biotremology, mating disruption, vibrational communication, IPM.

ORAL PRESENTATION

Can indigenous scelionid parasitoids exploit eggs of the exotic *Halyomorpha halys*?

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The establishment of *Halyomorpha halys* (Stål) (Hemiptera: Pentatomidae) in North America and Europe has caused severe economic damage to many crops, including fruit and hazelnut. Due to the difficult management of this exotic pest, research aimed at investigating the presence and effectiveness of natural enemies, both in native and recently invaded areas. In the native range, the main natural enemies are egg parasitoids, among which the most effective are *Trissolcus japonicus* (Ashmead) and *Trissolcus mitsukurii* (Ashmead) (Hymenoptera: Scelionidae). In Europe, little is known about the host range and the impact of most indigenous scelionid species so far. Therefore, the presence of scelionid species developing on native hosts (Hemiptera: Pentatomidae, Scutelleridae) in Piedmont (NW Italy) and their ability to develop in *H. halys* eggs were investigated. During 4-year field surveys carried out from 2016 to 2019, egg masses of 10 indigenous hosts were collected and reared, and 10 egg parasitoid taxa were obtained in laboratory. Among them, *Telenomus* spp., *Trissolcus belenus* (Walker) (Hymenoptera: Scelionidae) and *Anastatus bifasciatus* (Geoffroy) (Hymenoptera: Eupelmidae) were the most abundant ones. To evaluate their ability to develop on the exotic host, scelionid females emerged in the laboratory were exposed to *H. halys* eggs. *Trissolcus kozlovi* Rjachovskij, a species also emerged from field-collected *H. halys* eggs, was the only species able to emerge in significant numbers from *H. halys* eggs; however, all the tested *Trissolcus* species significantly increased the mortality of *H. halys* eggs. Based on the achieved results and since little information is available on *T. kozlovi*, its host range was assessed in the laboratory, and its impact on *H. halys* was evaluated in the field with augmentative releases in two hazelnut orchards. Among the 12 species tested in the laboratory (Hemiptera: Pentatomidae, Scutelleridae), *Arma custos* (F.), *Pentatoma rufipes* (L.) and *Peribalus strictus* (F.), with the highest parasitism rates, were the most suitable hosts, while only *Nezara viridula* (L.) proved to be an unsuitable host. In the field, *T. kozlovi* emerged from *H. halys* eggs in both hazelnut orchards after three releases of 400 females and 100 males each, but it was never found in the next two years. This study provided knowledge on the scelionid species associated with indigenous hosts and their adaptation to the exotic host, and represents a starting point to investigate the interactions between native and exotic parasitoids and hosts, following the arrival of *H. halys*, *T. japonicus* and *T. mitsukurii* in northern Italy.

KEY WORDS: Scelionidae, Pentatomidae, Scutelleridae, *Trissolcus kozlovi*, parasitism rate.

POSTER

Biological control and ant management against mealybugs in vineyards of Emilia

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Mealybugs (Hemiptera: Pseudococcidae) represent one of the most detrimental pests in Italian vineyards. Besides *Planococcus ficus* (Signoret), a species native to Eastern Asia, *Pseudococcus comstocki* (Kuwana), is causing several damages on vine plants, resulting in important economic losses for growers. The partial inefficacy of chemical applications in controlling mealybug infestation as well as their impact on environment have led to the research for the adoption of environmentally-friendly alternatives, as biological control. Natural enemy activity may be hindered by tending ants, which are known to create a mutualistic relationship with mealybugs, resulting extremely aggressive against beneficial insects. The aim of this work was to evaluate the combined release of two important natural enemies, *Anagyrus vladimiri* Triapitsyn (Hymenoptera: Encyrtidae) and *Cryptolaemus montrouzieri* Mulsant (Coleoptera: Coccinellidae), for the control of mealybugs. Beneficial releases were carried out both in pesticides-free trial plots, using exclusion cage method, and in commercial vineyards using an integrated approach including the use of insecticides against mealybugs. Field trials were also carried out to mitigate ant attendance by means of sugar dispensers in order to improve parasitization and predation. Both *P. ficus* and *P. comstocki* were parasitized by *A. vladimiri*, even if parasitization showed to be higher on *P. ficus*. Also the predator demonstrated to be effective in controlling Pseudococcidae infestation, showing a strong density dependent activity. Overall, natural enemy releases have shown a huge potential in controlling mealybugs and represent a basic tool to implement integrated pest management of these pests. The use of sugar dispensers reduced ant activity and enhanced ecosystem services, showing great potential in boosting biological control against mealybugs. High variability among vineyards occurred, and a multi-year evaluation of the strategy is recommended.

KEY WORDS: *Planococcus ficus*, *Pseudococcus comstocki*, *Anagyrus vladimiri*, *Cryptolaemus montrouzieri*, biological control, ant-mealybug association, sugar dispensers, vineyard.

The olive leaf gall midge and its parasitoids: the monitoring network in Tuscany

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The olive leaf midge *Dasineura oleae* (Angelini) (Diptera; Cecydomyiidae) is a secondary pest in olive orchards that generally causes low-level infestation and negligible damage to the plant. The feeding activity of the larvae on the leaves induce the formation of galls and the alteration of the physiological vegetative processes. In cases of serious infestation, with the formation of galls on the inflorescence, negative effects on production can also occur. According to the dictates of IPM (Integrated Pest Management), the identification of a control strategy for a specific pest, on a large territorial scale, involves some fundamental steps such as the knowledge of its population and infestation dynamics. Furthermore, it is necessary to collect data that come from a monitoring network that takes into account the main environmental variables regulating the biological cycle of the pest, at different levels. In 2019, we developed the *D. oleae* monitoring network in the province of Grosseto with the aim of assessing the progress of the infestation and the presence of indigenous antagonists, essential in the conservation biological control strategy.
The work started with the identification of 24 monitoring points (olive groves of about one ha) distributed throughout the province and affected by infestation of *D. oleae*. Subsequently, on each of them, using the transept methodology, the work carried out involved the removal of portions of vegetation (4 shoots / plant out of 6 plants) to be analysed in the laboratory. A methodology for assessing the infestation was therefore developed with the definition of risk classes of production damage, assessing both the severity of the infestation and the extent of biological control. In fact, we quantified the parasitization by parasitoid koinobiont Hymenoptera of the genera *Platygaster* and *Mesopolobus* which, already reported in previous studies, are known for their relevant role in the control of the phytophagous. Subsequently, the data obtained in the monitored olive groves were used to perform a landscape-scale study of both olive leaf gall midge and its parasitoids at four different territorial scales (buffers of 250, 500, 750 and 1000 meters respectively). In particular, the effect of the amount of olive crops and semi-natural habitats was investigated, as well as the diversity of land use through the Shannon index, using generalized linear models and cluster analysis. It has been observed that parasitization is positively influenced by landscape diversity measured at the 250 m buffer. At higher buffers (750 m and 1000 m), instead, it was observed that the diversity of the landscape seems to positively influence the abundance of the olive leaf gall midge. The obtained results provide useful information on the strategy for the control of this pest and at the same time are fundamental to refine the design and management of the monitoring network for *D. oleae*.

KEY WORDS: *Dasineura oleae*, *Platygaster*, *Mesopolobus*, IPM.

ORAL PRESENTATION

Biological control of *Erasmoneura vulnerata* using generalist predators

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Outbreaks of the Nearctic leafhopper *Erasmoneura vulnerata* are increasingly frequent in north-eastern Italy. Naturally occurring predator and parasitoids are not effective in the control of the leafhopper populations. In organic vineyards, the control of this pest is critical because of the reduced effect of organic-certified insecticides. In this context, an augmentative biological control strategy based on predator releases could represent a potential option. Here we investigated the effect of the use of two generalist predators *Chrysoperla carnea* and *Orius majusculus* in vineyards infested by *E. vulnerata.* As a preliminary step, laboratory and semi-field tests were performed to evaluate the predatory activity of the two species against the juvenile forms of *E. vulnerata*. Both species exhibited a strong voracity under laboratory conditions and significantly reduced the density of the leafhopper in semi-field experiments. Releases of the two predators were performed during the summer in an infested vineyard. The two predators significantly reduced leafhopper densities. However, the impact of both predators in the field was lower than in the laboratory and semi-field conditions. Further investigations are planned in order to test the effect of timing and release densities.

KEY WORDS: biological control, leafhoppers, grapevine, integrated pest management.

Grapholita molesta (Lepidoptera: Tortricidae) monitoring in stone fruit orchards treated with mating disruption using kairomonal lures

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Oriental fruit moth, *Grapholita molesta* Busck (Lepidoptera: Tortricidae), is a key world-wide pest of stone fruit (peach and nectarine). In Italy, these crops are usually managed with the Integrated Fruit Production (IFP) program, which includes the use of sex pheromones for mating disruption (MD) of oriental fruit moth and intensive monitoring. The use of MD for *G. molesta* is typically effective in managing this pest either alone or with supplemental insecticide applications. However, effective monitoring of this moth under MD to assess the need for additional control methods is problematic due to a general shut-down in male sexual behaviours and zero moth captures in traps. Unexpectedly, during the last few years, sporadic infestations of *G. molesta* has occurred with MD usage in Emilia-Romagna region (Italy). The use of sex pheromone-baited traps in these orchards has apparently not been effective in characterizing pest pressure and the potential for fruit injury. In response, new lures for *G. molesta* have been developed: a blend of a synergized-sex pheromones (*i.e.*, *G. mol*esta and *Cydia pomonella* L. sex pheromones) in combination with volatiles characteristic of fruit fermentation (*i.e.*, acetic acid and terpinyl acetate). Our study evaluated a commercial lure (Pherocon OFM Combo Dual Trécé Inc., Adair, OK, USA), developed from previous research in the USA, and compared moth catches in traps baited with a standard sex pheromone lure (Pherocon OFM L2, Trécé Inc.) in stone fruit orchards over the last two seasons. In 2018, 13 randomized lure trials with five replicates of each lure were performed in orchards treated with several types of MD (using either white or orange delta traps). Similarly, in 2019 11 and 9 trials were performed, respectively, in orchards treated and not treated with MD (white delta traps). The trap color and lure selectivity were also evaluated in terms of non-target insect catches (*i.e.*, *Apis mellifera* L.). We found that: 1) trap color did not affect *G. molesta* nor *A. mellifera* catches; 2) the type of lure did not affect *A. mellifera* catches; 3) a significantly higher *G. molesta* male and total moth number was caught in traps baited with the new combo lure compared with the standard sex pheromone lure in both MD and non-MD blocks; 4) females comprised < 10% of the total *G. molesta* catches with the combo lure; 5) the number of males and total *G. molesta* caught in orchards with or without MD was significantly different. Our studies suggest that the management of *G. molesta* can be improved using the new OFM Combo Dual lure due to more effective monitoring of pest pressures, especially under MD.

KEY WORDS: oriental fruit moth, sex pheromone, kairomone, trap, integrated pest management.

POSTER

Female removal of *Cydia pomonella* through kairomones: a new management strategy is now possible

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The use of sex pheromones to achieve mating disruption (MD) of codling moth, *Cydia pomonella* (L.), is widely adopted as a part of an integrated approach to manage this key pest of pome fruits and walnuts. However, in some management programs, such as certified organic production in North America, the integrated approach is unable to maintain this pest at economic levels, and in some cases, growers are forced to abandon their orchards or to revert to conventional spray practices to re-establish codling moth control. The concept of 'female removal' (FR) through intensive trapping has recently become a viable consideration following the discovery of a four-component non-pheromone blend (pear ester, nonatriene, linalool oxide, and acetic acid). This approach works in tandem with MD because it can reduce populations in targeted 'hot-spots' and it allows disruption to be more effective when pest populations are much lower. Female removal also is likely the most effective strategy to combat immigration of female moths into a management block. Thirty-four field experiments were performed during 2018, 2019 and 2020 in both apple and pear orchards in the USA and in pear orchards in Italy. In each orchard, a minimum surface of 0.5 ha was treated with FR using 60 traps per hectare and a similar area in the orchard had no traps added. All paired blocks were sprayed with the same grower program. Non-saturating bucket traps or delta traps with sticky liners were used for FR. Buckets were partially filled with mineral oil to retain the moths. Traps were baited with different multi-component blends comprised of sex pheromone, pear ester, nonatriene, linalool oxide, and acetic acid according to the experiment. At mid-season, the paired blocks were sampled for fruit injury and the captured moths were counted, sexed, and females dissected to determine their mating status. Prior to harvest, all paired blocks were sampled again for fruit injury and moths were counted from all traps. Levels of fruit injury in both pears and apples were significantly lower in the trapped than untrapped blocks (seventeen sites considered as replicates per each crop). At harvest in apple, thanks to the FR technique combined with MD, the level of fruit injury was significantly reduced by 50-75% across each moth flight in the USA. In pear, the average fruit protection was 65% in the USA studies and 27% in Italy. The lower control in the Italian orchards was likely due to the lower performance of these multi-component lures in Italy compared to the USA. These promising results should be confirmed in other geographical areas and further studies to determine the suitable trap density according to the infestation level are required to refine the FR approach for codling moth management.

KEY WORDS: codling moth, bucket trap, female removal, integrated pest management, organic farming.

ORAL PRESENTATION

Mating disruption: a new strategy to manage the honeydew moth, *Cryptoblabes gnidiella*?

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The honeydew moth, *Cryptoblabes gnidiella* (Millière) (Pyralidae: Phicitinae), is a key pest in the vineyards in South America (e.g. Brazil and Uruguay). Although it has always been considered a secondary pest of grapevine in the Mediterranean area, it has recently assumed an economic importance for the damage caused in the coastal areas of Italy and France. In these environments *C. gnidiella* develops mainly on the ripening grapes (particularly on medium-late or late ripening varieties such as Sangiovese, Montepulciano and Aglianico), causing a progressive bunch decay. This research offers a focus on the effectiveness of mating disruption (MD) against this moth, focusing on results obtained over three years (2017-2019) in vineyards in central (Tuscany) and southern (Puglia) Italy. MD tests were carried out using the Isonet® CGX111 (ShinEtsu Chemical) experimental dispenser tested at 500 units / ha. The effectiveness of MD against *C. gnidiella* was monitored weekly by carrying out field checks in the MD vineyards, as well as in untreated and insecticide treated plots. Periodically, groups of dispensers were collected and analyzed by gas chromatography in order to monitor the residual pheromonal content and consequently estimate the release of the synthetic blend in the field (mg/ha/day). In the first two years the dispensers were applied only once, in April. In both years the release of the synthetic pheromone has not been uniform, ending early in the season. In order to maintain an adequate level of pheromone until harvest, the MD strategy was modified in the third year, using two distinct approaches: (i) two dispenser applications (in April and July); (ii) only one application postponed to July. In the first two years of tests, the percentage of infested bunches in MD plots was about 50% lower compared to the untreated plots but higher of about 35% compared to insecticide-treated plots. In the third year, the infestation decreased by 78.9% in first approach (i) and of 91.7% in the second one (ii), if compared to untreated plots, confirming a greater MD effectiveness with a postponed dispensers application. Overall, MD can be considered an effective and environmentally sustainable approach for the management of *C. gnidiella*. Further studies are underway to validate this strategy by assessing the impact of different densities of pheromone dispensers in selected vineyards in central and southern Italy.

KEY WORDS: honeydew moth, Pyralidae, Phycitinae, Integrated Pest Management, sex pheromones.

Two on one: mating disruption of *Lobesia botrana* and *Planococcus ficus* with Isonet® L PFX246

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The European grapevine moth (EGVM), *Lobesia botrana* (Denis & Schiffermüller) (Lepidoptera: Tortricidae) and the vine mealybug (VMB), *Planococcus ficus* (Signoret) (Hemiptera: Pseudococcidae) are two key grape pests in the Mediterranean countries. In Italy, both perform 3-4 generations per year causing relevant damages leading to major economic losses. Currently, different strategies are used to manage EGVM and VMB, ranging from the repeated applications of synthetic pesticides to newest and safest options in the frame of biological control. Concerning the latter, one of the most promising biocontrol options validated on both pests is mating disruption (MD). Indeed, recent research highlighted the effective management of EGVM and VMB through the application of hand-applied sex pheromone dispensers (for both species) or aerosol devices (for EGVM), with little non-target effects. However, a dedicated dispenser releasing both pheromones at the same time is currently lacking from the market, though it would be extremely helpful to reduce labour cost (i.e. the time required for dispensers' field application) as well as plastic amount in the vineyards. This 2-year (2017-2018) study evaluated the field efficacy of an experimental dispenser, namely Isonet® L PFX246 (ShinEtsu Chemical), at various densities (400, 500 and 600 units/ha), for managing *L. botrana* and *P. ficus* through MD. Trials were conducted in vineyards of Southern (Sicily), Central (Tuscany) and Northern (Veneto) Italy. In both years, the MD approach relying to Isonet® L PFX246 allowed a satisfactory management of EGVM and VMB along the productive season. MD vineyards showed a significant reduction of the infested number of flower clusters/bunches, as well as of the number of EGVM nests and VMB individuals per flower cluster/bunch compared with no-MD plots. Dispenser performances were comparable, with no significant differences among the three tested densities in all the study sites. Overall, our results pointed out the possibility to boost MD by combining sex pheromones of different vineyard pests in a single releasing device. The dispenser tested in this study allowed the simultaneous management of *L. botrana* and *P. ficus* allowing a reduction of labour cost, as well as limiting plastic and insecticide use in the field.

KEY WORDS: Lepidoptera, Hemiptera, biological control, integrated pest management, vineyard pests.

POSTER

Induced plant defences by essential oils on sweet pepper plants and associated trophic networks

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Plant essential oils (EOs) are a promising tool for the sustainable control of agricultural pests, however, their impact on plants and the associated trophic networks has been merely investigated. In the laboratory, we assessed whether EO applications can induce defences on sweet pepper plants, *Capsicum annuum* (Solanaceae). Based on the olfactory response of the model parasitoid *Encarsia formosa* (Hymenoptera: Aphelinidae), garlic and peppermint EOs were selected among different plant extracts over four botanical families (i.e., Apiaceae, Asteraceae, Liliaceae and Lamiaceae). The behavioural responses of the herbivores *Bemisia tabaci* (Hemiptera: Aleyrodidae), *Frankliniella occidentalis* (Thysanoptera: Thripidae) and the predators *Orius laevigatus* (Hemiptera: Anthocoridae) and *Nesidiocoris tenuis* (Hemiptera: Miridae) to EO induced plant volatiles were thus investigated. The expression level of defence-related genes in plant tissues and the phytotoxic effect were measured in response to EO foliar applications in sweet pepper plant. Garlic and peppermint EO sprayed plants, tested at different concentrations, triggered different behavioural responses on both pests and natural enemies in olfactory trials. Gene expression analysis showed activated defence signalling pathways in sweet pepper EO sprayed plants, while a variable phytotoxic effect was recorded. Our results suggest that sweet pepper plants can significantly mediate EO effects on pests and natural enemies. These findings could serve as a basis for future development of plant protection strategies in the framework of Integrated Pest Management. a valutare l'attività in vivo del peptide carausicina in seguito a immunostimolazione degli insetti.

KEY WORDS: biological control, botanical, IPM, plant response, predatory bugs.

ORAL PRESENTATION

Effectiveness of mating disruption to control *Grapholita funebrana* in Sicilian plum orchards

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The development of ecofriendly pest management strategies is of great importance especially in organic cultivation. Among these, the mating disruption could represent an alternative tool for the control of *Grapholita funebrana* (Lepidoptera: Tortricidae), a key pest of the plum orchards. To date, little information is available on the use of mating disruption against the tortricid. In 2012 and 2014, a study was carried out in three different Sicilian plum orchards, in order to evaluate the effectiveness of mating disruption on *G. funebrana*. Each plum orchard was 6, 4 and 3 hectares, respectively, of which 4, 3 and 2 hectares, respectively, were treated by placing about 500 pheromone dispensers per hectare (ISOMATE® OFM red FLEX) before the adults of the wintering generation emerged. One dispenser per tree was placed in the two outer rows of each experimental plot and one every two trees within its perimeter. The untreated remaining surface represented the control plot. During the research, no insecticide treatments were carried out in any of the 6 plots. Three pheromone baited sticky traps were placed in each of the 6 plots and were checked weekly. Male genitalia extraction of a sample of insects caught at each date have been made to confirm the specific identification of the tortricid. Starting from the first catches and up to the harvest (early August 2012, late July 2014), samplings of the fruits were carried out every two weeks on three blocks of trees of the Angeleno, Friar, President and Stanley cultivars within each plot. At each sampling date, 10 fruits per trees (90 per cultivar) were collected on three trees of each cultivar from each block, to record the occurrence of infestation by the plum moth. In 2014, the pheromone release rate from dispensers collected in the field at each fruit sampling date and from new dispensers, was estimated through solid phase microextraction followed by GC-MS analysis. In the treated plots, catches of *G. funebrana* adults throughout the study period were lower than the control plots. The fruit infestation in the treated parcels was significantly less than that found in the control in almost all cultivars. Despite this, in the cultivar most susceptible to the plum moth attack (Angeleno), the percentage of fruit infestation at harvest was high (from 41% to 60%). The amount of pheromone released by the dispensers collected in the field recorded a significant decrease starting from the second sampling date, remaining unchanged until the last sampling. The results show that in the treated plots, the pheromone density in the field determined a lower number of male catches in the traps. However, tortricid mating was probably not limited enough to reduce the infestation to economically acceptable levels. Therefore, further investigations aimed at evaluating the factors that determined the scarce effectiveness of the mating disruption in the pedoclimatic conditions of the study areas would be necessary, in order to make its application economically sustainable.

KEY WORDS: plum moth, sustainable pest management, mating disruption.

POSTER

Trissolcus japonicus and *Trissolcus mitsukurii*, Asiatic egg parasitoids of *Halyomorpha halys* in northern Italy and Switzerland

Giuseppino Sabbatini Peverieri[1], Francesco Tortorici[2], Gianfranco Anfora[3], Simone Bardella[4], Massimo Bariselli[5], Luca Benvenuto[6], Iris Bernardinelli[6], Alda Butturini[5], Stefano Caruso[7], Elena Costi[8], Paolo Culatti[5], Emanuele Di Bella[8], Martina Falagiarda[10], Tim Haye[11], Lara Maistrello[8], Giorgio Malossini[6], Alberto Mele[12], Lorenza Michelon[13], Silvia Teresa Moraglio[2], Alberto Pozzebon[12], Michele Preti[14], Pio Federico Roversi[1], Martino Salvetti[15], Davide Scaccini[12], Silvia Schmidt[10], Luciana Tavella[2], Maria Grazia Tommasini[16], Giacomo Vaccari[7], Pietro Zandigiacomo[18], Livia Zapponi[17]

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Halyomorpha halys is an invasive pest of Asian origin and accidentally introduced in Italy, Switzerland and almost all European countries. This pest causes severe damages on agricultural crops in Italy, with more than 500 million euro in 2019. Proposed chemical solutions for pest control do not guarantee an acceptable level of efficacy in the long-term and have a high negative environmental impact. On the other hand, classical biological control using the Asian egg parasitoid *Trissolcus japonicus* seems most promising for a sustainable management of *H. halys*. Unexpectedly, adventive populations of *T. japonicus* have been discovered in southern Switzerland (2017) and in north-western Italy (2016). Moreover, a second Asian egg parasitoid, *Trissolcus mitsukurii*, was also detected on *H. halys* eggs in central and northeastern Italy in 2018. Considering the exceptional findings of both parasitoids and the perspectives for the pest control, in 2019 a large-scale field survey all over northern Italy and Switzerland was performed to define for both species the abundance, distribution and impact on pest populations. Monitoring focused on collecting naturally laid egg masses of *H. halys* and other pentatomids. Overall, more than 4,630 egg masses were collected, of these more than 4,340 belonged to *H. halys*. Results of the survey indicate wide occurrence of these parasitoids in Northern Italy and Switzerland. In details, *T. mitsukurii* appeared to be widely distributed in the north-central and north-eastern part of Italy, whereas *T. japonicus* was detected in the north and south Switzerland and in the north-western part of Italy. Minor findings of one of the two species occurred in the area where the other species was most represented. Parasitism of *H. halys* by *T. japonicus* ranged between 0.45% and 53.85% of the egg masses and between 0.29% and 50.88% of eggs. In *T. mitsukurii* such percentages ranged between 0.17% and 20.20% of egg masses and between 0.09 e il 14.49% of eggs. The parasitism rates among egg masses (excluding unhatched and/or predated eggs) was very high reaching in some cases 100% in *T. japonicus* and 97.40% for *T. mitsukurii*. Parasitization by these two parasitoids on egg masses other than *H. halys* was 6.76% in *T. japonicus*, and ranged between 1.96% and 9.09% in *T. mitsukurii*.

KEY WORDS: biological control, brown marmorated stink bug, parasitization.

POSTER

Trissolcus mitsukurii attraction toward *Halyomorpha halys* volatilechemical cues

Marica Scala[1], Jalal Melhem Fouani[1], Livia Zapponi[2], Valerio Mazzoni[2], Antonio Biondi[3], Nuray Baser[4], Gianfranco Anfora[1,2]

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Halyomorpha halys (Hemiptera: Pentatomidae) is an invasive pest native to Eastern Asia, particularly harmful because it can attack more than 100 host plants species. The long distance dispersal capacity and the absence of efficient biocontrol agents make its control very difficult. Due to a lack of effective insecticides, investigating parasitoid traits linked to the biological control potential is of paramount importance for developing sustainable *H. halys* management strategies. A probable candidate for this purpose is the microhymenopteran *Trissolcus mitsukurii* (Hymenoptera: Scelionidae). This species is the principal egg parasitoids of *Nezara viridula* (Hemiptera: Pentatomidae) in Japan, however it can successfully attack several other stink bugs. Recently, *T. mitsukurii* emerged from *H. halys* egg masses sampled in different areas of northern Italy. Currently, there is little information about the biology and the biocontrol potential provided by this parasitoid. Parasitic Hymenoptera often use different kinds of kairomones as chemical cues to detect environments in which the prey is present, and, to orient towards the oviposition substrate. In some other *Trissolcus* spp. it has been proven that they use volatile cues to find adults and, more specifically, they use it to locate suitable substrates for oviposition. Thus, the aim of this project is to study the behavioral responses of the parasitoids which are elicited by different host-borne chemical cues and that could play an important role in the detection of the proper oviposition substrate. The trials are conducted using single *T. mitsukurii* female (2-4 day-old) exposed to different stimuli (e.g. volatile cues emitted by *H. halys* females). The behavioral response is recorded with a “video tracking” analysis software (Ethovision). The variables taken into account are: residence time in specific areas (sec); mean linear velocity (mm/s); angular velocity (°/s); distance moved (mm). As demonstrated by previous studies, we expect that a parasitoid triggered by the chemical cues released by a suitable host, shows a slow walking velocity, higher angular velocity, covers long distances in addition to longer residence time. The obtained results are presented and discussed in the framework of potential H. halys biological control programs.

KEY WORDS: *Trissolcus mitsukurii, Halyomorpa halys*, behavioral trials, chemical cues, biological control.

POSTER

Development and field-testing of a Decision Support System to control the populations of *Ceratitis capitata* in multi-varietal fruit orchards

Andrea Sciarretta[1] , Maria Rosaria Tabilio[2], Marco Colacci[1] , Pasquale Trematerra[1]

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In the present work, a decision support system (DSS) to control infestations caused by the Mediterranean fruit fly, *Ceratitis capitata* (Wiedermann), in multi-varietal peach orchards has been developed and evaluated in the field. The DSS included a semi-automatic monitoring of adults and a precision approach for insecticide treatments. The DSS consists of three algorithms. The first, based on the calculation of degree days, defines when the traps must be deployed in the field to start monitoring the population of *C. capitata*. The second one defines the areas to be treated and the type of treatment, based on the number of adult catches, the harvesting period and the phenological stage of the host cultivar. The third algorithm defines the treatment procedure, taking into account the characteristics of the selected insecticide (e.g. withholding period and efficacy duration time) and the adverse weather conditions (wind, rain). The DSS was evaluated in a fruit farm near Rome, comparing DSS assisted with conventional management, through a randomized complete block experimental design with three repetitions. In the plots DSS-assisted, a semi-automatic system for adult monitoring was used, consisting of electronic traps equipped with a camera, connected to internet for the real-time data transmission. The DSS returns a prescription map showing the areas to be treated and the type of treatment (bait or cover spray applications). The farmer was free to follow or not the DSS indications. First catches of *C. capitata* were observed on 30 June, while the degree day threshold was reached on 3 July, when the DSS started operating. The field trials produced 29 maps from 3 July to 1 September and showed a substantial reduction in the number of insecticide applications, areas treated and volumes of pesticide applications in the plots with DSS-assisted management. No significant differences were observed in fruit infestation levels between DSS-assisted and conventional management. The level of DSS acceptance by the farmer was 78%. The results obtained confirmed the need to fully involve farmers in the evaluation process of DSS.

KEY WORDS: Mediterranean fruit fly, peach orchards, automated traps, precision agriculture, integrated pest management, smart agriculture.

ORAL PRESENTATION

Native and exotic egg parasitoids of *Halyomorpha halys* in Piedmont: a 5-year field survey

Sara Scovero, Delia Leone, Silvia T. Moraglio, Marco G. Pansa, Francesco Tortorici, Luciana Tavella

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

Since its introduction into North America and Europe, the brown marmorated stink bug *Halyomorpha halys* (Stål) has caused severe damage to crops with heavy economic losses. The pest management basically based on the repeated use of insecticides has not been entirely effective given the limited knock down effect and short residual activity. Consequently, the research has focused on natural enemies able to adapt and effectively suppress the exotic stink bug, and in particular on egg parasitoids of the genera *Anastatus* and *Trissolcus*, which in the native area contribute to the pest control. To assess the impact of indigenous egg parasitoids on *H. halys* in Piedmont (NW Italy), a 5-year field survey was carried out from 2016 to 2020; egg masses were collected in 33 sites and reared in the laboratory to evaluate the parasitism rate. From the 6,209 field collected egg masses of *H. halys* (in total 155,303 eggs), *Anastatus bifasciatus* (Geoffroy) (Hymenoptera: Eupelmidae) was the most widespread egg parasitoid, emerged in all sites and years, but generally with a low impact (8-13%). Recently, adventive populations of *Trissolcus japonicus* (Ashmead) and *T. mitsukurii* (Ashmead) have been observed in North Italy. In Piedmont, *T. japonicus* was found in one site in 2018, in further five sites in 2019, and throughout the region in 2020, also due to the releases carried out in the national biological control programme. *Trissolcus mitsukurii* was found in four sites in 2019 and in further 14 sites in 2020. In the site where they were observed for the first time, the exotic parasitoids showed increasing parasitism rates from 15.6% in 2018 to 26.5% in 2020, when the overall parasitism rate, also considering *A. bifasciatus,* was 36.5%. Furthermore, in 2019 and in 2020 in Piedmont, adventive populations of *Anastatus japonicus* (Ashmead) were found emerging from *H. halys* eggs. During the 5-year survey in Piedmont, the two exotic scelionid parasitoids proved to spread rapidly in environments where *H. halys* is present, with an additive effect to that performed by *A. bifasciatus*. In light of the discovery of *A. japonicus* and the recent systematic studies on the genus *Anastatus*, it will be appropriate to further analyse the *Anastatus* specimens emerged from *H. halys* eggs in the years prior to 2019, in order to accurately check their identifications.

KEY WORDS: Pentatomidae, Scelionidae, Eupelmidae, *Trissolcus japonicus, Trissolcus mitsukurii*.

POSTER

Kill Rate' to evaluate the predation of the phytoseiid *Neoseiulus californicus* mass reared on alternative foods

Sauro Simoni[1], Giovanni Burgio[2], Elena Gagnarli[1], Franca Tarchi[1], Silvia Guidi[1], Donatella Goggioli[1], Alberto Lanzoni[2]

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To increase the commercial production of predators and parasitoids to be employed in biological control of pests and to reduce production costs, the mass rearing of phytoseid mites has been carried on the use of alternative foods which guarantee: 1) the development of the phytoseiid populations and 2) the maintenance of their predatory efficiency, in particular with respect to the target phytophagous species. In this contribution, the number of red spider mites - the tetranichid mite *Tetranychus urticae* Koch - killed by the phytoseiid *Neoseiulus californicus* (McGregor), mass reared on different foods, was assessed. These values were used to calculate a predation rate (Kill Rate) km, obtained by modifying the intrinsic growth rate of a population (rm), of *N. californicus* after years of breeding on substitutive food sources such as astigmatid mites, *Glycyphagus domesticus* (De Geer), *Lepidoglyphus destructor* (Schrank), and *Quercus* sp. pollen. The calculated km values were slightly higher for the phytoseidae previously maintained on the astigmatid mite *G. domesticus* and on *Quercus* sp. pollen. Based on the km values calculated, the mass rearing of *N. californicus* on these foods is confirmed as adequate option to provide high level of phytoseiid populations and high predation efficiency against the tetranychid target *T. urticae*. The chance of using the kill rate as index to group phytoseiid species biological control agents in the application of control strategies, is discussed, based on their adaptation to different food/prey, and on actual amounto of killed prey.

KEY WORDS: phytoseiids, *Tetranychus urticae*, control strategies, predatory potential.

ORAL PRESENTATION

New associations of natural enemies from different niches to synergise biocontrol of a new glasshouse pest

Emiliano Rinaldo Veronesi[1], Stephen Wratten[2]

[1] Lincoln University, Lincoln, New Zealand; [2] Lincoln University, Lincoln, New Zealand

The tomato-potato psyllid (TPP) is a major pest of solanaceous crops such as tomatoes, potatoes, tamarillos and capsicums, it feeds on plant sap and is a vector for CLso (*Candidatus Liberibacter solanaceorum*), a bacterial pathogen of plants which can drastically compromise yields. This bacterium is also responsible for the zebra chip disease, which causes black stripes in potato tubers and alters the taste, rendering the product unsuitable for the processed potato market. Worldwide, the main strategy against TPP involves the prophylactic use of broad-spectrum pesticides, of which up to 15 applications are made per season.
Nowadays, TPP is showing increasing resistance to pesticides and biocontrol seems to be the only suitable long-term approach. The mirid bug *Engytatus nicotiane*, a true bug almost unknown in the scientific literature, is showing good potential and several large-scale experiments involving whole plants confirmed its efficiency in managing this pest. The scope of this work is to combine *Engytatus nicotianae* with the parasitic wasp *Tamarixia triozae*, two biocontrol agents that exhibit different traits and belong to different guilds. In fact, whereas *Engytatus* shows a clear preference for TPP eggs and the first 3 stages, *Tamarixia* prefers TPP's fourth and fifth stages as a stimulus for egg laying. The needs of these two biocontrol agents appear to be complementary, suggesting a good chance of synergy with no or little competition. The final aim of this work is to combine *Engytatus* with *Tamarixia* in order exploit the full potential of these two agents in terms of TPP pest management.

KEY WORDS: TPP, tomato potato psyllid, *Engytatus nicotianae*, *Tamarixia triozae*, biocontrol, greenhouse tomato.

BINAGHI AWARD

BINAGHI AWARD

Hoverfly and ecological integrity in the Merlino Wood Natural Reserve

Umberto Maritano[1], Elio Giuliano[2], Maria Chiara Magnano[2]

[1] Indipendent researcher, Italy; [2] Naturalist illustrator, Italy

The use of hoverflies as bioindicators, through the application of the Syrph the Net database, is a fundamental tool for gathering information on the ecological integrity of an ecosystem, especially in forest landscapes. Merlino Wood is subjected to massive human pressure, but still represents an isolated source of saproxylic organisms of high conservation interest. In order to preserve rare species, strict habitat conservation measures are required.

KEY WORDS: Diptera, Syrphidae, Piedmont, Syrph the Net, *Spilomyia manicata*.

OSELLA AWARD

OSELLA AWARD

Invasive insect pests in the 21st century: current state and future challenges

Davide Rassati

University of Padova, Italy

The constant increase in the volume of goods shipped internationally has caused an impressive number of exotic species introductions around the world. This number is expected to further increase due to expanding trade networks and incorporation of new regions that can act as source of new species. Thus, biological invasions represent one of the most problematic challenges existing nowadays. Due to the high number of international ports, the high diversity of ecosystems, and the suitable climate, Italy is particularly prone to insect invasions. In the last few years a number of exotic insects have been introduced in Italy, causing massive damage to agriculture and forest ecosystems. The most famous examples are the spotted wing *Drosophila*, *Drosophila suzukii*, the brown marmorated stink bug, *Halyomorpha halys*, the oriental chestnut gall wasp, *Dryocosmus kuriphilus*, and the Asian longhorn beetles, *Anoplophora glabripennis*, but many other exist. The availability of tools able to intercept exotic species before they become established in the invaded environment is essential to prevent damages and reduce overall costs. Massive resources have been invested to this end, and a wide set of phytosanitary measures, strategies and tools are now available for being used both prior and after exotic species introduction. These progresses have undoubtedly aid in biosecurity capacity worldwide, allowing the interception, eradication, and containment of several exotic species around the world. Nonetheless, the phenomenon of exotic insect introductions is still far to be stopped. The question is: how can we further improve existing surveillance programs? At this regard, there are two aspects that are still understudied but that can potentially impact the efficacy of surveillance activities. The first one regards the role of natural forests surrounding import/export sites as source of native species that are at high-risk to be introduced in other continents. The study of these native communities could help to target the most likely invaders, with direct implications on the efficacy of surveillance programs. The second one is the role of microorganisms in insect invasions. There is increasing evidence that insect invaders can acquire novel microorganisms during invasion. These microorganisms can potentially confer important adaptations to its novel host, allowing it to overcome limitations of its original microorganisms. The occurrence of a such mechanism opens a new scenario in biological invasions that can overturn the traditional approach used in biosecurity surveillance. Species distribution models, for example, are commonly used by governmental institutions for planning surveillance programs and decide where to concentrate efforts and resources. The acquisition of novel microorganisms in the invaded environment can completely alter predictions for the establishment and spread of certain exotic species in a particular environment. Unraveling ecological factors that influence the acquisition of novel microorganisms and incorporating their role into ecological theories is thus fundamental to clarify the mechanisms behind insect invasions and aid in biosecurity surveillance.

KEY WORDS: Biological invasions, exotic species, surveillance, microorganisms.

AUTHORS LIST

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Mercalli L.	WEBINAR
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Candian V.	POSTER
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D'Isita I.	POSTER
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Gagnarli E.	ORAL
Germinara G.	ORAL
Germinara G.	POSTER
Jucker C.	ORAL
Jucker C.	POSTER
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Laudani F.	POSTER
Manachini B.	POSTER
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Sabbatini	
Peverieri G.	POSTER
Scala M.	POSTER
Sciarretta A.	ORAL
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AWARDS

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Rassati D.	OSELLA AWARD - POSTER

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