

IOBC/wprs Bulletin Vol. 53, 2010

Working Group „Integrated Protection of Olive Crops”. Proceedings of the Meeting at Bragança (Portugal), 10-12 October, 2007. Edited by: Argyro Kalaitzaki.
(ISBN 978-92-9067-227-2) [xiv + 152 pp.]

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Bactrocera oleae: Genetics, Physiology, Control

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Genetic improvements to SIT for fruit fly control

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Abstract: The Sterile Insect Technique (SIT) is an effective, species-specific and environmentally friendly method for controlling pest populations. SIT involves releasing millions of sterile insects over a wide area to mate with the native insects that are present. Native females that mate with the sterile males produce non viable offspring, leading to a decline in the target pest population. SIT has been used successfully for control of several tephritid fruit fly species in programmes targeting local eradication, suppression below economic thresholds, and prevention of establishment. Modern genetic methods hold out the prospect of significant operational and cost-effectiveness improvements to the SIT, and for extension of the SIT to a broader range of pest species. Genetic improvements include: more reliable identification of released individuals with a fluorescent genetic marker; removing the need for radiation-sterilisation (“genetic sterilisation”); reducing the hazard posed by non-irradiated accidental releases from the mass-rearing facility by arranging that the insects need an artificially-provided condition, for example a dietary supplement, in order to survive or reproduce; providing automated sex-separation prior to release to eliminate females from the release population (“genetic sexing”). Oxitec has developed the RIDL[®] suite of genetic technologies and has demonstrated all of these properties in Mediterranean fruit fly (*Ceratitidis capitata*) and Mexican fruit fly (*Anastrepha ludens*), as well as in moths and mosquitoes. SIT would be a useful tool for the Integrated Pest Management of olive fly (*Bactrocerea oleae*) but implementation has been constrained by the difficulty of mass-rearing, including the lack of a cheap and consistent

artificial diet and changes in behaviour such as time of mating. Research into these issues is ongoing in Greece and at the IAEA. The cost-effectiveness of an SIT approach would be greatly improved by the ability to release only males. Each female olive fly lays 50-400 eggs at one per fruit, causing significant damage to the crop, regardless of whether she has been sterilised or not. In addition, if males and females are released together, the males may court the sterile females, and consequently not seek out the wild females as effectively as if they had been released alone. This distraction effect of sterile females on sterile males has been shown to have a major impact on the male effectiveness of Mediterranean fruit fly in the field. Oxitec's RIDL technology could be applied to olive fly to rear populations of male insects which produce no female progeny in the absence of a specific diet supplement. In addition, they could be reared safely in olive growing areas without the need for irradiation to sterilise them.

Susceptibility of Sardinian olive cultivars to *Bactrocera oleae* infestations

G. Delrio, S. Deliperi & A. Lentini 17-21

Abstract: The susceptibility of the main olive cultivars grown in Sardinia (Italy) to olive fly attack was studied in order to improve pest management techniques. Observations were carried out during 2004-2005 in an organic olive grove where the three following cultivars were randomly distributed: Manna, an early ripening cultivar with large drupes; Bosana, a medium-late ripening cultivar with small drupes; Semidana, a late ripening cultivar with medium sized drupes. The first generation of olive fly females preferred ovipositing in Manna olive fruits, which are characterised by larger size and lower hardness, whereas drupe size did not influence significantly female fruit preference in autumn. In this period, olive fruit susceptibility was likely to be more related to different amounts of epicuticular waxes on fruits from the three diverse cultivars. The olive fruit resistance to *Bactrocera oleae* development was evaluated estimating the mortality of eggs and young larvae. The highest percentages of mortality, which were reached in August, differed significantly among cultivars in both years (92% for Manna, 68% for Bosana and 79% for Semidana in 2004, and 72%, 42% and 44% in 2005, respectively). At harvest time, the percentages of wormy olives were 24 and 27% for Manna, 65 and 32% for Bosana, and 75 and 55% for Semidana, in 2004 and 2005, respectively. The susceptibility of the three cultivars to the olive fly seems to be strictly related to the over-time-changing oviposition preference and to the cultivar response to the mortality factors, such as high summer temperature.

Entomopathogenic fungi show promise for biological control of olive fly puparia in the soil

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Effectiveness of mass trapping by bottle traps baited with salt sardines to control *Bactrocera oleae* (Gmelin)

V. Caleca & M. Maltese 23-24

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Spintor® Isco, an innovative system for the control of the olive fruit fly *Bactrocera oleae* in IPM

Maria Torné, Doris Paroonagian, Costas Mavrotas & Mark Miles 25-36

Abstract: SPINTOR® ISCO, GF-120®, is a newly developed pre-mixed concentrated fruit fly bait containing a reduced risk toxicant to both mammals and non-target insects. GF-120® attracts and controls multiple species of tephritid fruit flies and contains an optimized blend of feeding attractants and the active ingredient, spinosad; an insect control product derived from a naturally occurring soil bacterium, *Saccharopolyspora spinosa*. It provides improved consistency, attractiveness, selectivity, pre-harvest intervals and overall efficacy when compared to current bait-toxicant mixes. GF-120® is approved for use in organic (OMRI™ Listed) and conventional production systems in many countries. Application is by ultra low volume with large droplets (4-6 mm of diameter) that help the product to remain viable in the field for extended periods of time when compared to other baits. GF-120® has an excellent environmental profile and has

been demonstrated under laboratory, and field conditions to be an ideal product for use in IPM programs. In laboratory studies which mimic realistic exposure, GF-120[®] was reported to be non-toxic to two important beneficial in olive, *Cryptolaemus montrouzieri* and *Rodolia cardinalis*. Effects on the highly sensitive Hymenoptera parasitoid, *Psytalia concolor* were investigated in two trials under field conditions in Tunisia. Olive fruit from trees treated with GF-120[®] at 1L/ha (0.24 g a.i./ha) were taken to the laboratory where emergence of fruit fly larvae was observed. Parasitism rates were recorded and GF-120[®] was shown to have no adverse effects on *O. concolor* (IOBC class 1). The effect on beneficials from a single application of GF-120[®] at 1.25 l/ha (0.3 g a.i./ha) was investigated in two field trials conducted in Greece. Dead arthropods were collected from treated trees for up to 20 days after treatment. GF-120[®] was found to be harmless (IOBC class 1) to *Chilocorus* sp., *Coccinella dipunctata*, *Chrysopa* sp., and demonstrably safer to Hymenoptera parasitoids and Syrphidae predators than Fenthion and alpha-cypermethrin. In Spain as part of the aerial efficacy trials carried out by “Departamento de Sanidad Vegetal de Málaga” an area of commercial scale was treated with GF-120[®] (500 hectares). The impact on beneficial arthropods was assessed and GF-120[®] was found to have no adverse impact on a wide range of beneficials species. Transient effects were noted on Hymenoptera parasitoids (Chalcidoidea) but with recovery observed 20 days after application. GF-120[®] applied twice at 1 l/ha within a 9 day interval showed no negative impact on honeybees, *Apis mellifera* in a trial covering 800 hectares of olive trees in Spain. In conclusion, GF-120[®] is highly selective to beneficials and pollinators whilst delivering outstanding control of tephritid fruit flies. These attributes make it an ideal fruit fly control product for use within IPM programs in olives.

Study of the effectiveness of various insecticides used for the control of *Bactrocera oleae* (Gmelin) (Diptera: Tephritidae) in Crete olive groves.

Argyro Kalaitzaki, Kyriaki Varikou & Venizelos Alexandrakis 37-46

Abstract: The control of olive fruit fly *Bactrocera oleae* (Gmelin) (Diptera: Tephritidae) in Greece has been based mostly on bait sprays with organophosphate insecticides (usually fenthion and dimethoate) for more than 30 years. Extensive research has been carried out the last ten years by the Institute of Olive Tree and Subtropical Plants of Chania in order to study the effectiveness of various insecticides, against *B. oleae*, in olive groves of Chania. The results of last two years study concerning the effectiveness of various insecticides, applied by bait from ground sprays, are represented. In 2005, the tested insecticides were lambda-cyhalothrin 10% and a-cypermethrin 10% which were compared to the reference product fenthion 50%. The mean number of *B. oleae* per trap was significantly higher in the plots that were sprayed with fenthion compared to those that were sprayed with lambda-cyhalothrin and a-cypermethrin. In 2006, the tested insecticides were lambda-cyhalothrin 10%, a-cypermethrin 10%, deltamethrin 2.5%, deltamethrin 6.25%, deltamethrin + thiacloprid and Spinosad 0.24% which were compared to fenthion 50%. No significant differences were found in the olive fly captures among the tested insecticides. In both years, the percentage of the olive fruit fly infestation remained generally in very low levels and was not observed significant difference among the tested insecticides.

Comparison between current compounds for bait spray against olive fruit fly, *Bactrocera oleae* Gmelin in Iran (Qazvin-Tarom Sofla)

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Lure-and-kill formulations used in olive culture have an adverse effect on important species of the natural enemy complex of arthropod pests

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Introduction of the African parasitoid *Psytalia lounsburyi* in south of France for the classical biological control of *Bactrocera oleae*: will hybridization affect establishment and population growth?

Jean-Claude Malausa, Arnaud Blanchet, Marie-Claude Bon, Sandrine Cheyppé-Buchmann, Géraldine Groussier-Bout, Walker Jones, Charles Pickett, Nicolas Ris, Marie Roche, Marcel Thaon & Xavier Fauvergue 49-55

Abstract: Classical biological control can be considered as a particular type of biological invasion in which experimental approaches are possible. The scientific objective of our research programme is to test *in natura* the influence of intra-specific hybridization on the success of biological invasions in general, and of biological control introductions in particular. The model organism is the endoparasitoid *Psytalia lounsburyi* Silvestri (Hymenoptera: Braconidae). Parental populations originating from Kenya, South Africa, and their hybrids will be introduced in France to control *Bactrocera oleae* (Rossi) (Diptera: Tephritidae), the most important pest of olives in southern France. Experiments conducted during 2007 have improved our basic knowledge of *P. lounsburyi* at the genetic level (microsatellite markers and geographic structure) and at the phenotypic level (life-history traits) allowing us to design efficient rearing methods. Intensive field surveys have also been carried out to identify 60 experimental sites where these exotic parasitoids will be introduced in 2008 in order to study the genetics and demography of introduced populations.

Susceptibility of *Bactrocera oleae* Gmelin (Dipt.: Tephritidae) to entomopathogenic fungi

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Field studies on *Psytalia concolor* (Hymenoptera: Braconidae) and the caper fly, alternative host to the olive fruit fly, in south Sardinia

Giovanni Marongiu, Francesco Foddi, Riccardo Pesci & Federico Corda 57-61

Abstract: During 2004 and 2005 a series of investigations was conducted on populations of *Psytalia concolor* (Szépl.) (Hymenoptera: Braconidae) an endophagous parasitoid of tephritid fruit flies, of its main host in the Mediterranean basin, the olive fruit fly *Bactrocera oleae* (Gmelin) (Diptera: Tephritidae) and of what is thought to be an alternative host reported in Tunisia, *Capparimyia savastani* (Martelli) (Diptera: Tephritidae).

The purpose of the investigations was to determine whether the latter host, whose larvae infest caper flower buds, could also be parasitized by *Psytalia* in Italy, whether it could contribute substantially to the population growth of the parasitoid or whether it could in any case enable it to survive for long periods of the year in the absence of olives and hence of *B. oleae*.

The findings have shown that the parasitoid is capable of parasitizing *C. savastani* in Italy too and of being hosted by the latter during that period when parasitizable stages of the main host *B. oleae* are absent. The parasite load of *Capparimyia* is not however considered sufficient to allow it to multiply abundantly.

Assessing the effect of soil treatments with the entomopathogenic fungus *Metarhizium anisopliae* (Metchnikoff) Sorokin against puparia of *Bactrocera oleae* (Diptera: Tephritidae) on soil dwelling non target arthropods

Inmaculada Garrido-Jurado, Cándido Santiago-Álvarez, Mercedes Campos & Enrique Quesada-Moraga 62-66

Abstract: The objective of this study was to determine the persistence of the autochthonous *Metarhizium anisopliae* EAMa 01/58-Su isolate in the soil when applied beneath olive trees for controlling olive fly puparia and to elucidate its possible effect on non-target soil dwelling arthropod communities. For that, we selected 200 olive trees in an organic olive orchard at the province of Málaga (Spain) to be sprayed either with a 2.5×10^7 conidia m^{-2} suspension of the fungus on the ground beneath the tree canopy (100 trees) or with the blank formulation as controls (100 trees). Before fungal treatments, we selected 10 trees from the treated ones for evaluating both the possible presence of indigenous entomopathogenic fungi in the soil by using the Galleria Bait Method and the evolution of the conidial densities in the soil after spraying.

The entomopathogenic fungus *Beauveria bassiana* (Balsamo) Vuill. was the most common species, being found in all the samples, while *M. anisopliae* was found only in one sample. After spraying the 100 treated and 100 control trees, soil samples beneath the 10 selected trees from the top 10 cm were taken to calculate the number of conidial forming units per gram of soil at 1, 7, 14, 28, 35, 42, 49, 56 and 63 days after treatment respectively. Our preliminary data indicate that the soil ecosystem favours the persistence of this autochthonous isolate, which could allow long term protection of the crop against olive fly puparia. In order to assess the possible effect of the fungal treatment on soil arthropod populations, 40 pitfall traps (7.5 cm diameter by 10 cm deep) placed beneath the tree canopy of randomly selected 20 treated and 20 controls trees, were sampled every two weeks. Our preliminary data indicate that formicidae species are the most abundant arthropods trapped, but no infected insects have been found in field as a result of the treatment to now.

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Natural control of olive enemies – Other olive pests

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Abstract: Knowledge about the beneficial arthropod community occurring in agroecosystems is an essential prerequisite for the development of plant protection programmes within the context of a sustainable agriculture. In the olive ecosystem the value of these beneficials was discovered mainly as a consequence of their elimination by the use of insecticides, leading to outbreaks of secondary pests. The objective of this study was to gain insight on the beneficial arthropod fauna occurring in olive groves from Alentejo (South region of Portugal), which was at that time a relatively undisturbed ecosystem. The study was carried out during two consecutive years, 1999 and 2000, in four olive groves located near Évora. Arthropods were collected by the beating technique, using a modified japanese umbrella device. Sampling was done weekly between March and November in 1999, and between April and November in 2000. In each sampling period, two branches randomly selected, from each of 60 trees per grove, were beaten.
- Results showed that 29% from the 138 858 individuals collected were predators (27%) or parasitoids (2%). Among the predators, the most numerous families were, by decreasing order of abundance: Aranea (11.6% of the total captured arthropods), Coccinellidae (8.9% of the total) and Formicidae (5.3% of the total). Staphylinidae, Carabidae, Chrysopidae, Hemerobiidae, Raphidiidae, Coniopterygidae and Mantidae were also captured in the canopy but, in total, they represented only 1.9% of the total individuals obtained. Aranea, Coccinellidae and Formicidae were present during all the sampling period. However Aranea and Coccinellidae were captured in higher numbers during June and August, while Formicidae were obtained mainly during June and July.
- Parasitoids were represented mainly by Chalcidoidea, representing 1.5% of the total captured arthropods, and Ichneumonoidea, representing 0.2%. Chalcidoidea were captured during all the sampling period, while Ichneumonoidea were obtained mainly between April and May, and in September.
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- Preliminary study of the parasitic complex associated with *Dittrichia viscosa* in Andalusia
S. Franco-Micán, J. Castro & M. Campos 139-143
Abstract: The interaction of organisms in the organic management cultivation of olive trees is caused by the diversity of plant species growing nearby, which directly influences beneficial insects. The systematic and widespread elimination of these plant species increases populations of insects harmful to olive trees. Among the plant species historically associated with olive groves is the *Dittrichia viscosa* L. Aiton, a perennial plant that blossoms between September and October. Its flowers are infested with *Myopites stylata* Fabricius, dipteran that causes gall formation where the larvae are parasited by *Eupelmus urozonus*, one of the main parasitoids associated with the olive fruit-fly (*Bactrocera oleae*). The aim of this study is to analyse the parasitic complex of *Myopites stylata* associated with *Dittrichia viscosa* in three olive zones in Andalusia (southern Spain). Galls were collected and kept separately in glass vials under incubator room conditions until adults emerged (photoperiod of 14:10h (L:D) at a temperature of 25°C ± 3°C. We observed the presence of Hymenoptera from the Chalcidoidea superfamily belonging to the Eurytomidae, Eupelmidae, Pteromalidae and Torymidae families in a decreasing order of quantity. The number of families, as well as the number of adults present in galls, changed according to the geographical location of the galls.
- Detection of *Verticillium dahliae* Kleb. from olive trees with chronic decline and dieback of branches and shoots
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Abstract: A study has been carried out to evaluate the influence of integrated olive production on the quality of Galega virgin olive oil. On three consecutive years, olive samples from two different groves in integrated production were taken, and submitted to extraction in industrial mills. The analytical determinations in olive oil were acidity, peroxide index, K_{232} and K_{270} , sensory analysis, fatty acid composition, total phenol compounds, tocopherols, oxidation stability and organophosphate pesticides. Quality criteria were within the European Union limits for extra virgin olive oil, and the organophosphate pesticides residues were always undetectable.

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