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ORIGINALBEITRAG

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Entwicklung nachhaltiger Pflanzenschutzstrategien zur Bekämpfung von Schadschmetterlingen im Olivenanbau

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Zusammenfassung Traditionelle, extensiv bearbeitete Olivenhaine, aber auch moderne Intensiv-Plantagen mit künstlicher Bewässerung und hohem Einsatz von Düngern bzw. chemischen Pflanzenschutzmitteln kennzeichnen die derzeitigen verschiedenen Anbauformen der Olive im Mittelmeerraum. Schadlepidopteren wie die Olivenmotte (*Prays oleae*, Lep.: Yponomeutidae) und die Jasminmotte (*Palpita unionalis*, Lep.: Pyralidae) werden durch regelmässigen Insektizideinsatz bekämpft. Das von der EU geförderte internationale Forschungsprojekt TRIPHELIO zielte auf die Entwicklung insektizidfreier Alternativmethoden durch (1) die Optimierung der pheromongestützten Überwachung und Verwirrtechnik, (2) der Anwendung von Habitatmanagement-Strategien zur Förderung natürlicher Gegenspieler, und (3) dem Einsatz von *Trichogramma*-Schlupfwespen. Zusätzlich wurden Module für eine optimale Anwendung biotechnischer und biologischer Methoden bezüglich der Phänologie der Schadinsekten und möglicher Nebenwirkungen von Pestiziden erarbeitet. Die intensive Kooperation zwischen Wissenschaftlern und Praktikern aus mehreren Ländern Europas und Nordafrikas erlaubte den Entwurf möglicher Lösungsansätze für verschiedene Anbaubedingungen und klimatische Regionen des Mittelmeerraumes. Die wichtigsten Ergebnisse und Ausblicke für eine zukünftige praktische Umsetzung werden in dieser Veröffentlichung beschrieben.

Schlüsselwörter Olivenanbau · Biologischer Pflanzenschutz · Verwirrtechnik · Pheromonapplikation · *Trichogramma* · Habitat Management · Nachhaltige Landwirtschaft · *Prays oleae* · *Palpita unionalis* · Reduktion des Pestizideinsatzes

Towards sustainable control of Lepidopterous pests in olive cultivation

Abstract Current olive growing practices range from the traditional Mediterranean olive grove to intensively managed olive plantations. Insecticides against major olive pests, like the olive moth (*Prays oleae*, Lep.: Yponomeutidae) and the jasmine moth, (*Palpita unionalis*, Lep.: Pyralidae) are still applied frequently. The European Union-funded international research project TRIPHELIO is aimed at the development of economically

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feasible and sustainable insecticide-free control methods for key Lepidopterous pests of olive. Main research activities focus on (1) the use of pheromones for mating disruption and improved monitoring of target pests, (2) habitat management strategies to enhance the activity of natural enemies in the olive grove, and (3) the use of inundative releases of mass-reared egg parasitoids of the genus *Trichogramma*. Moreover, tools for successful integration of the methods developed into an integrated pest management (IPM) strategy for olive pests were explored by optimising techniques for surveillance as well as considering potential side-effects of common pesticides on beneficial organisms. An intense exchange of scientific information and technology between European and North African countries was undertaken to create solutions for a wide range of olive growing regions. Key results and recommendations for further essential steps towards practical implementation are presented in this publication.

Keywords Olive cultivation · Biological control · Mating disruption · Pheromone application · *Trichogramma* · Habitat management · Sustainable agriculture · *Prays oleae* · *Palpita unionalis* · Pesticide reduction

Introduction

Olive growing is of great economic and socio-cultural significance for the Mediterranean region, where 98% of the world's olive production is located (Civantos 2001). A companion of mankind since ancient times, the olive tree, *Olea europea* L., and its cultivation have been subjected to distinct changes during recent decades. High market pressures, as well as the implementation of specific agricultural policy measures (including production subsidies in the EU), have resulted in intensified crop management being introduced into traditional olive growing systems (López-Villalta 1996). New practices include the use of irrigation, application of artificial fertilisers, repeated short-term planting of early-fruiting cultivars at high densities, and intensive use of pesticides in order to push up olive yields as well as to increase the returns on the cash crop.

The olive tree is attacked by various insect pests, which can cause considerable yield losses, and current olive cultivation involves regular use of plant protection products. However, frequent insecticide applications pose the risk of environmental pollution and contamination of the olive products (Jardak and Ksantini 1996; Calbras et al. 1997; Cirio 1997). Moreover, increasing pesticide resistance of target pests led to the use of higher dosages, which is detrimental to the natural enemy complex, resulting in the subsequent appearance of secondary pests like the black scale *Saissetia oleae* (Oliv.) (Katsoyannos 1985). These developments have also steadily raised input costs for olive growing (Jardak and Ksantini 1996). The establishment of producer groups, such as ATRIAs (Agrupaciones para Tratamientos Integrados en Agricultura) in Spain, has certainly improved the situation by imposing strict application of integrated pest control, which lowers the risk of a negative environmental impact of plant protection (López-Villalta 1999). Recently, the International Organization for Biological Control (IOBC/wprs) has published „*Guidelines for Integrated Production of Olives*" to give further recommendations for more sustainable olive production (Malavolta et al. 2002). These guidelines are based on comprehensive joint research activities in olive plant protection during the past few decades (Cavalloro and Crovetti 1985; Delrio 1992). Nevertheless, the use of insecticides in olive cultivation in order to maintain high output levels of the crop is still frequent and widespread. Advisory plant protection services that can help the olive farmer to implement integrated pest management (IPM) strategies properly are still not well developed in the various olive-producing regions. This is especially true for those countries where the transition from traditional low-input orchards to intensified olive plantation systems is currently taking place.

Consumers are concerned also in non-producing, but importing countries like Germany (Öko-Test-Magazin 2004) where the consumption of olive products is rising steadily, reaching levels comparable to those of producer countries (Fig. 1). Olive oil, especially, is perceived as a valuable product not only for its flavour in cooking, but also due to its particular value for health maintenance (Wahlqvist and Kouris-Blazos 1996), e.g. as an important component in the Mediterranean diet (Fankhänel 2002) recommended by the World Health Organisation (WHO). Demand for organically grown olive products is rising and consumers are willing to pay higher prices for olive oil of excellent quality and integrity (Öko-Test-Magazin 2004). This situation has opened up new opportunities for olive-producing and exporting countries to offer certified organically grown products (Jardak and Ksantini 2003). Food safety control regulations prevent access to the market of olive products of inferior quality or with critical pesticide levels [Botitsi et al. 2004; Codex Alimentarius FAO/WHO (http://www.codexalimentarius.net); Food monitoring of the German Federal Office of Consumer Protection and Food Safety (BVL) (http://www.bvl.bund.de/lebensmittel/monitoring.htm]. This urges the producer to have a reliable interest in pesticide-residue-safe crop production systems.

All these factors have led to an urgent requirement for alternative, insecticide-free—or at least insecticide-reduced—strategies in olive plant protection. In recent years, much progress has been made with respect to control of the olive fruit fly, *Bactrocera oleae* Gmel. (Diptera, Tephritidae). By using IPM strategies, total cover sprays of insecticides have been replaced by bait spraying treatments; these are applied mainly from the ground and only when a particular level of economic threshold was exceeded (López-Villalta 1999). Other new methods consider mass trapping, or the attract and kill techniques involving pheromone and food baits (Broumas et al. 2002; Mazomenos et al. 2002; Ragoussis 2002).

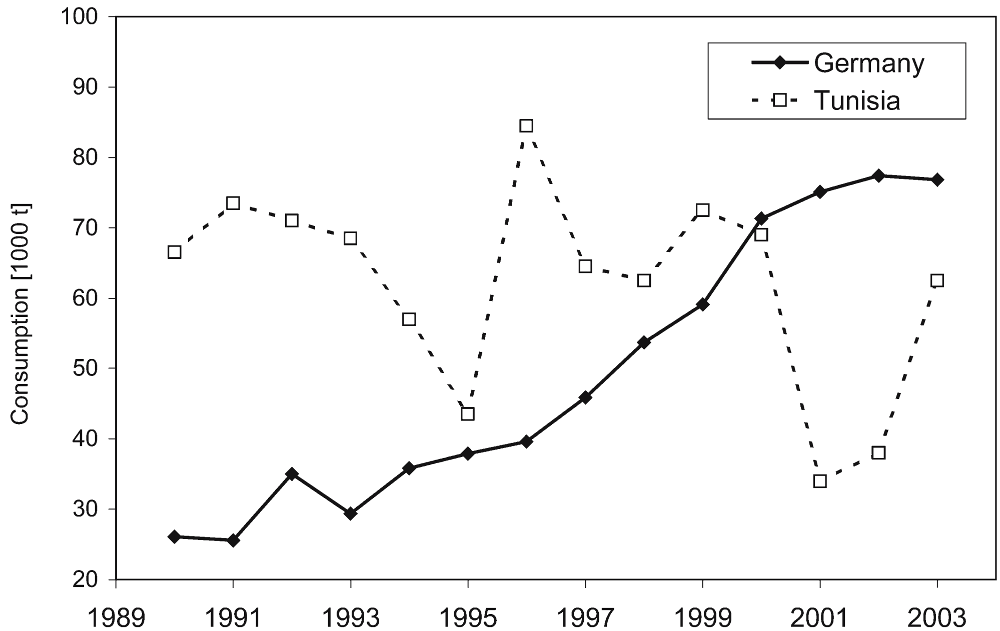

Fig. 1 Consumption of olive products (olive oil and table olives) since 1990 in Tunisia (main producer country) and Germany. Consumption per capita 2003: 0.9 kg in Germany and 6.3 kg in Tunisia (Data source: © IOOC statistics 2004)

Occasionally, similar destructive pests belonging to the order Lepidoptera occur. Among them, the olive moth, *Prays oleae* Bern. (Lep., Yponomeutidae) appears regularly in a wide range of olive growing areas, causing considerable yield losses (Ramos et al. 1989). Most recently, the first individuals of this moth have been discovered also in the Californian olive growing regions (http://www.oliveoilsource.com/olivenews5-3.htm).
Other Lepidopterous species, such as the jasmine moth, *Palpita unionalis* Hübn. (Lep., Pyralidae), are still considered as secondary pests, but under particular olive growing regimes, such as olive nurseries or irrigated young olive plantations, their importance can increase (López-Villalta 1999; Antonelli and Rossi 2004).

Funding of collaborative research within the field of olive plant protection by the European Commission has a long history, resulting in several significant research projects during the last 20 years (C.E.C. Programme on Integrated and Biological Control in olive growing, 1979–1983; ECLAIR-project, Contract AGRE.0013: „*The development of environmentally safe pest control systems for European olives*“, 1990–1994). In 2000, the EU announced a specific call to develop „*Tools for sustainable development concerning cash crops, including olive*“ within the programme „*INCO2: Confirming the international role of community research*“. The topic was based on agreements made by the Euro-Mediterranean Forum in Naples (8–9 July 1998) on future co-operation in agriculture and agro-industries between European and Mediterranean countries. Following this call, a consortium of scientific experts in olive cultivation, biological control and pheromone research from Europe and North Africa was built up to elaborate biological and biotechnological control methods for the control of Lepidopterous pests of the olive tree in a joint research effort („TRIPHELIO“, „*Sustainable control of Lepidopterous pests in olive groves—integration of egg parasitoids and pheromones*“, Contract ICA4-CT2001-10004). The project was initiated and coordinated by the Federal Biological Research Centre for Agriculture and Forestry (BBA) in Germany for a period of 42 months (2001–2005).

TRIPHELIO aimed to integrate three complementary approaches: (1) the optimisation of available pheromone-based techniques for monitoring and for control by mating disruption, (2) the exploration of various strategies to protect and enhance key natural enemies in the olive grove habitat, and (3) the development of biosafe inundative releases of mass-reared egg parasitoids of the genus *Trichogramma* (Hym., Chalcidoidea) as efficient control tools for olive cultivation. The present paper summarises information on the pest problems, research activities and results achieved by TRIPHELIO as well as conclusions and recommendations for further research and development.

Biology of target pests and current control practices

Olive moth

The olive moth (*Prays oleae*; Fig. 2) develops three generations per year and is highly adapted to the phenology of the olive. Larvae of the phyllophagous generation overwinter as leaf-miners. The last instars are external leaf feeders, but may also occasionally damage buds of the olive shoots. Adults emerge in early spring and eggs are laid almost exclusively on olive flowers just before opening at phenological stage „D“ (Arambourg and Pralavorio 1986). This anthophagous generation develops on flower buds, flowers and, at later instars, on whole flower clusters. Larvae of the subsequent carpophagous generation penetrate the developing fruit and feed inside

Fig. 2 Adult of the olive moth, *Prays oleae*

Fig. 3 Adult of the jasmine moth, *Palpita unionalis*

the endocarp. The emergence of the full-grown last instar for pupation cause the fruit to detach from the stalk and to fall. As a consequence, an attack by *P. oleae* usually causes two series of premature fruit fall: one in early summer, directly after penetration of the larva into the fruit (this loss of fruit can usually be compensated by the tree, except in years with low fruit-set), and a second pre-harvest fall, incurring serious crop losses. Control treatments are applied against the anthophagous as well as the carpophagous generation. Action thresholds have been established for particular olive growing regions. For instance, in IPM-managed olive groves in Spain, the decision to treat the anthophagous generation depends on the level of infestation, but also on the general flowering and fertility index of the olive tree in the particular season. Insecticide applications against the fruit generation are usually advised when early fruit infestation by viable eggs exceeds 30–40% in Spain (López-Villalta 1999) or 20% in Tunisia (Jardak and Ksantini 1996). To control the carpophagous generation, the insecticide has to penetrate the fruit in order to eliminate the larvae, with organophosphates (e.g. dimethoate) currently the products of choice. To date, *Bacillus thuringiensis* var. *kurstaki* (or *B. thuringiensis* var. *kurstaki* × var. *aizawai*) is the only available biological product to control this pest. If properly applied, efficacy is good, but the most critical drawback is that only externally feeding larval stages will be affected, i.e. the last instars of the phyllophagous and the young instars of the anthophagous generation (at the beginning of flowering). As a consequence, the level of control obtained is not always sufficient to prevent serious attack by subsequent generations on olive fruits, especially in years of high population density (Jardak 1994) or with general low fruit-setting of trees (Yamvrias and Young 1977).

Jasmine moth

Palpita unionalis (Fig. 3) is a multivoltine species with several overlapping generations per year. Larvae overwinter and first adults occur in early spring. Later, the development of the larvae is not well synchronised, and is temperature-dependent, resulting in the simultaneous presence of eggs, different larval stages, pupae and adults throughout the season (López-Villalta 1999). Usually, population densities are highest from July to October (Arambourg 1986). Feeding of larvae has been observed on several plant species belonging to the Oleaceae (Genus *Olea*, *Ligustrum*, *Fraxinus*, *Jasminum*, *Phillyrea*). In olive, both leaves and fruits are damaged. In nurseries or young orchards, feeding damage can reach up to 90% of the leaf area, thereby seriously affecting the development of the plant shoots. High infestations in late summer and autumn during fruit ripening may also reduce the fruit yield by 30% (López-Villalta 1999). Damages caused by *Palpita* have been reported from Greece (Vassilaina-Alexopoulou and Santorini 1973), Italy (Triggiani 1971, Fodale and Mule 1990, Antonelli and Rossi 2004), Israel (Avidov and Rosen 1961) and Egypt (El-Kifl et al. 1974). Ornamentals also suffer from larval attacks on leaves and flower buds (Lo Pinto and Salerno 1994). According to Gargani (1999), a heavy outbreak in a jasmine plantation, cultivated for the production of perfume, was observed in France. Insecticides like dimethoate, deltamethrin and cypermethrin can be used for the chemical control of the jasmine moth in olive cultivation (López-Villalta 1999). Arambourg (1986) recommended the use of *B. thuringiensis* var. *kurstaki*, but no other biological or biotechnological control means are currently available.

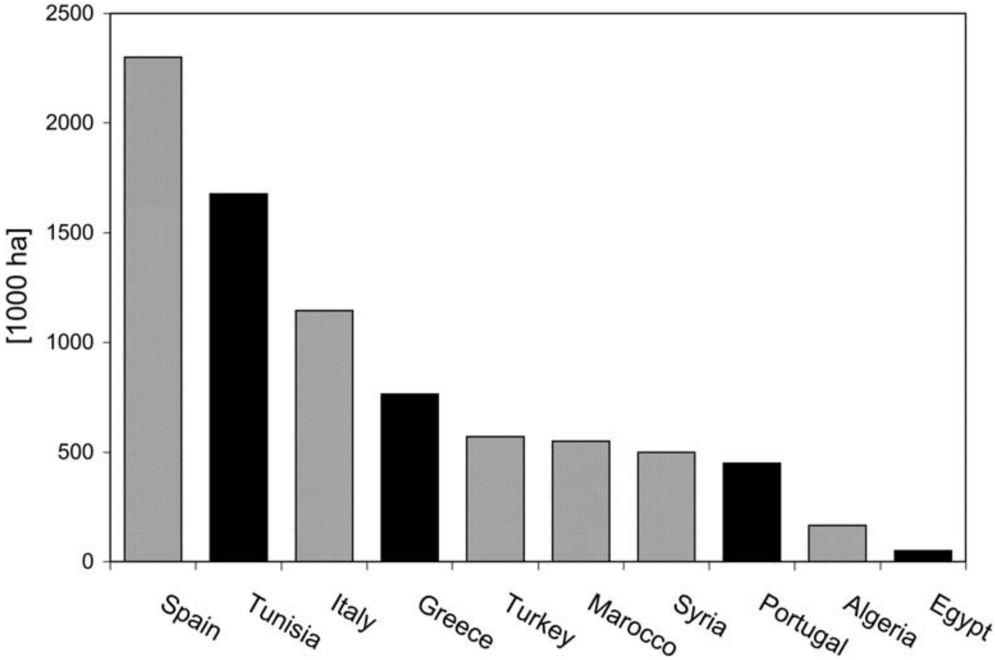

Fig. 4 Olive production area in the Mediterranean region in 2002, including the TRIPHELIO-partner countries Tunisia, Greece, Portugal and Egypt (black columns) (Data source: © FAOSTAT 2003)

Olive growing under present conditions

The main goal of TRIPHELIO was to provide environmentally sound and economically feasible technologies for the control of Lepidopterous pests for a wide range of olive-producing countries in the Mediterranean basin. Environmental conditions (climate, rain, soil, sea-level) vary considerably between the southwest/southeast Europe and North-Africa, and influence olive cultivation and pest problems. However, general management strategies also deeply affect the status of plant protection in this crop system. Olive farming is far from homogenous and comprises different concepts that can be roughly assigned to the following categories (Beaufoy 2000): (1) low-input traditional orchards with little or no chemical input, but with a high labour input, (2) intensified traditional plantations with systematic use of chemical products, weed control and soil management, (3) intensive modern plantations of smaller tree varieties with intensive use of artificial fertilisers, pesticides, and irrigation.

The countries participating in the project were Portugal, Tunisia, Greece and Egypt, thus representing a considerable diversity of olive-growing regions from both an economical and an ecological point of view. Tunisia and Greece belong to the main olive-producing countries in the world (Tunisia: 243,000 t/annum, Greece: 459,000 t/annum in 2003/2004; IOOC-Statistics 2004) and olive cultivation is characterised by a high level of management. The Tunisian olive-growing area is the second largest in the world (Fig. 4), and about 65% of the production is exported (Fig. 5). In contrast, olive production in Portugal and Egypt is less extensive (Portugal: 46,500 t/annum, Egypt: 97.000 t/annum in 2003/2004; IOOC-Statistics 2004), and is carried out in a more or less traditional way, although there has been an increasing trend towards intensification in the last few years. This refers especially to table olive production (98% of total production) in Egypt, where table olive varieties were recently planted in desert areas due to the governmental activity of the „General Organisation for the Development of the Desert" (IOOC 1996), thereby increasing exportation of table olives more than 10-fold during the last 10 years (1993/1994: 5,000 t/annum, 2003/2004: 55,000 t/annum, IOOC-Statistics 2004).

The main experimental approach in the TRIPHELIO-project was therefore to work out and conduct comparable field trials as case studies under different olive growing conditions. In Portugal, the main experimental field sites were located in traditional, low-input managed olive groves of organic production (Fig. 6). In Tunisia and Greece, the test fields were groves created several decades ago, and which were under IPM control and soil management (Fig. 7). In Egypt, main trials were conducted in an intensively managed olive plantation established in 1996 in the desert near Cairo (Fig. 8).

Results and discussion

Improvement of monitoring systems for target pests

Monitoring by sex pheromone traps was applied in all TRIPHELIO-countries using similar trap systems (delta-trap, funnel-trap). Thus, comparative data sets on the flight phenology of the olive moth and the jasmine moth were obtained during the project. Nowadays, monitoring olive moth flight by use of these traps is a standard procedure in olive-producing countries applying IPM (Jardak et al. 1985; Ramos et al. 1989; López-Villalta 1999), but this is not the case everywhere. For instance, in Egypt we gained valuable data on the flight phenology of *P. oleae* in different olive agroecosystems by this method for the first time.

Concerning the jasmine moth, new knowledge on the distribution and abundance of this secondary pest was obtained. During the project period, the formulation of the principal pheromone components (E)-11-hexadecenal and (E)-11-hexadecenyl acetate (Mazomenos et al. 1994) and a suitable trap system were optimised (Athanassiou et al. 2004). The moth was caught sporadically in Portugal, Tunisia and Greece during all three years, but was collected frequently in Egypt, where continuous captures with several peaks suggested an overall seasonal activity of this insect. Higher numbers, exceeding 100 ma-

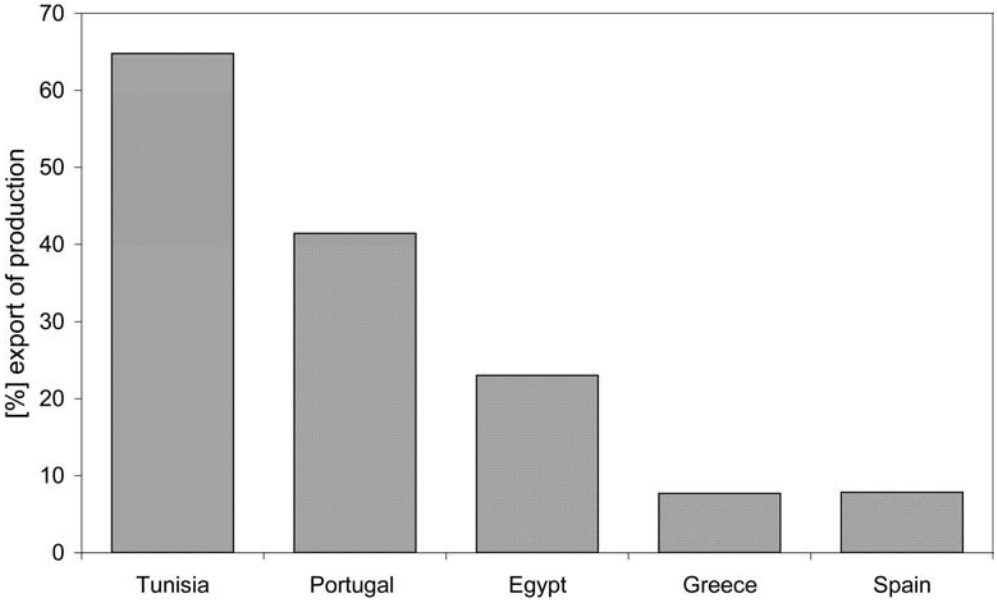

Fig. 5 Export range (in percent) of total production (olive oil and table olives) between the years 1992 and 2004 in several olive producing countries. In Egypt, 98% of total production is table olives (Data source: © IOOC Statistics 2004)

Fig. 6 Olive grove under organic production in Northern Portugal (Tras-os-Montes region). Olive cultivars (oil): Cobrançosa, Madural and Verdeal Transmontana, age of trees about 45 years, tree density 124 trees/ha

Fig. 7 Integrated pest management (IPM)-managed olive grove in Tunisia in the vicinity of Sfax (southwest coast). Olive cultivar: Chemlali (oil). Tree age: 80–90 years, tree density 17/ha

les trap^{-1} week^{-1}, were found in the young plantation near Cairo. Here, also damage on leaves and fruits was noted, indicating that this type of orchard (irrigated, and with hot climatic conditions) represents a preferred, and therefore susceptible, biotope for potential outbreaks of the jasmine moth.

Our findings demonstrated the need for regular monitoring of olive moth and jasmine moth in all olive-growing areas using a sensitive detection system. The distribution and regular application of these pheromone trap systems is highly recommended also in those countries that do not yet possess the state-of-the-art technology required to use them. Decision makers in plant protection should act accordingly.

New data on the developmental biology of both pests were obtained during our project, and studies were conducted on the morphology of the antennal structures as principal organs of intraspecific pheromone-steered communication (Shehata et al. 2003; Stefanos 2003).

Mating disruption with sex pheromones

The principal technique for mating disruption of the olive moth had been already explored before TRIPHELIO started (Mazomenos et al. 1999a, 1999b). However, experience was available only from field trials in Greece and therefore the first challenge was to optimise pheromone application for use in a wider range of climatic conditions. The formulation of the main pheromone

Fig. 8 Olive plantation in Egypt in the desert area near Cairo (established 1996). Main cultivars: Toffahy and Shamy (table olives), tree density 330 trees/ha. The plantation is drip-irrigated. Fertilizers, chemical weed control and pesticides are regularly applied

component (Z)-7-tetradecenal (Campion et al. 1979) into a β-cyclodextrin-complex (Mazomenos and Moustakali-Mavridou 1997), and its application in polypropylene tubes at a rate of 40–50 g (a.i.) per hectare, resulted in comparable stability and sufficient release rates of the pheromone for up to 6 weeks field exposure, even in the hot and dry climatic conditions in Tunisia and Egypt. A new procedure was successfully developed for formulating the jasmine moth pheromone components for a field trial in Egypt (B.E. Mazomenos et al. unpublished data).

Large-scale field trials (5 ha/country) for mating disruption of the olive moth were performed in Portugal, Tunisia, Greece and Egypt in all three years of the project. After improving the formulation, excellent male disorientation (>90%) was noted in all countries during the second flight in 2003 and 2004, but the effect on crop protection was not satisfactory and—especially at high population densities—fruit damage could not be reduced to below the economic threshold using this method. In 2004, the field trial in the young plantation in Egypt led to better pest control than in the other countries. Population density (as indicated by egg samples) and also fruit fall were significantly reduced in treated areas (efficacy >60%) and yield/tree was much higher (>58%).

Although the principal mechanism of the mating disruption treatment—male disorientation—worked successfully with the pheromone formulation, and dose and dispensers used under different olive growing conditions, successful matings obviously still occurred and these mated females were sufficient to cause considerable infestation. Besides the risk of invasion by mated females from neighbouring areas, spatial heterogeneity in moth distribution, with „hot spots" of high moth densities within the treated olive grove, may also allow matings without previous „pheromone correspondence". The mating disruption trial of the jasmine moth in the olive plantation in Egypt had a similar outcome: high levels of male confusion were noted, but suppression of the pest population was insufficient. More knowledge of the mating behaviour, dispersal and egg-laying behaviour of these pests is still needed and studies in this area should be encouraged. To increase the efficacy of mating disruption, a strong reduction of population density before pheromone application is essential, and we would also expect better control by long-term application of the pheromone over extended areas (Mazomenos et al. 1999a) as has been observed for other pests (Louis et al. 1996). The method would probably be more suitable for isolated, densely planted olive plantations like the experimental farm in Egypt. It will certainly be possible to greatly reduce the current costs of pheromone application after further progress towards commercial production.

Key natural enemies in the olive grove ecosystem

The existence of a particularly rich parasitoid fauna of both the olive and the jasmine moths has been documented in the literature, at least in some olive-growing regions (Arambourg 1986; Arambourg and Pralavorio 1986). Coleopteran, Neuropteran or other invertebrate predators, which attack the various stages of Lepidopterous pests, also occur in the olive grove ecosystem (Campos and Ramos 1985; Morris et al. 1999). These natural enemy communities are especially susceptible to disruptive measures of modern olive cultivation such as insecticide application or the removal of secondary vegetation, which may harbour essential food/shelter and prey/host resources. TRIPHELIO compared general assemblages of key natural enemies in the various olive growing areas (Abou-Elkhair et al. 2002; Nasr et al. 2002; Bento et al. 2005). Similarities in the parasitoid communities of the olive moth were found between Portugal and Tunisia, with *Chelonus eleaphilus* Silv. identified as the dominant parasitoid of the olive moth. In Egypt, the most important species was the larval parasitoid *Apanteles xanthostigmus* Hal. (Hymenoptera, Braconidae) (Table 1). Egg predation was identified as an important mortality factor, especially for the carpophagous generation, destroying up to 67% of eggs deposited on the fruits. A careful study in Portugal examined the role of ants in egg predation, suggesting that egg predators belonging to Chrysopidae and Coccinellidae might be negatively influenced by the presence of ants, especially by the species *Tapinoma nigerimum* F. (see also Pereira et al. 2004). This observation confirmed earlier results published by Morris et al. (1998).

Habitat management strategies

Olive trees take in most of their water requirements from the soil in spring and summer. It is therefore essential that as much water as possible is stored in the soil by removing weeds, thus reducing water losses through evaporation, especially under olive dry farming systems (López-Villalta 1996). As a consequence, soil tillage, mowing, or weed removal is carried out regularly in intensively ma-

Table 1 Species diversity and relative abundance of parasitoids collected from regular samples of the leaf and flower generation of *Prays oleae* in different partner countries during the season 2004

	Portugal			Tunisia	Egypt	
Parasitoid guild	Leaf	Flower	Fruit	Leaf	Leaf	Flower
Ovo-larvalparasitoids						
Ageniaspis fuscicollis var. *praysincola* Silv.	+[a]	++	+	++	–	–
Chelonus eleaphilus Silv.	++++	+++	++++	++++	–	–
Larval-parasitoids						
Angitia armillata Grav.	0	0	0	++	–	–
Apanteles xanthostigmus Hal.	++	++	+	r	++++	++++
Elasmus flabellatus Fonsc.	0	+++	0	0	–	–
Pupal-parasitoids						
Habrobracon crassicornis Thoms.	0	0	r	0	+++	+++

[a] Relative abundance of parasitoids: ++++ >50%, +++ =21–50%, ++ =11–20%, + =2–10%, r =1% and less (single specimen), 0 = not recorded, but known from similar samples in the region, – = not recorded in 2002, 2003 and 2004

naged olive groves (Fig. 7). However, these cultivation practices lead to a loss in biodiversity of flora and insect populations (Beaufoy 2000, 2001) and may have a particularly disturbing effect on groups such as predators or parasitoids by removing essential food/nectar/shelter resources. Maintaining or creating „ecological infrastructures“ is generally seen as an effective concept for enhancing biodiversity, and possibly also natural biological control, in agroecosystems (Boller et al. 2004; Malavolta et al. 2002).

We tested different habitat management strategies in case studies in Portugal (elevated, semi-arid area in southwest Europe) and Egypt (North-African desert area). Created vegetation islands, consisting of flowering vegetables (radish, roquette, turnip, squash) or natural vegetation (wild groundsel) in Egypt, and of flowering buckwheat/carrot or natural vegetation in Portugal, were shown to be highly attractive for important groups of natural enemies (Hymenopterous parasitoids, Coccinellidae, Syrphidae, Anthocoridae, Chrysopidae). The abundance and diversity of these taxa was increased in the treated areas in comparison to plots kept without vegetation (Jorge et al. 2005; Nasr et al. 2004). In an experimental olive grove in Portugal, the omission of soil tillage resulted in the development of natural soil vegetation, and modified the composition of the soil fauna as well as of important groups of natural enemies. All these measures lead to a significant attraction of key natural enemies to treated areas.

Nevertheless, offering islands of flowering plants in the olive grove or changing the soil management did not significantly improve pest control in this short-term 2-year study. Higher parasitism and predation rates of various development stages of the target pests occurred sometimes, but not regularly, in plots with increased secondary vegetation. Considerable fruit infestation and serious yield loss could not be prevented. The possibility that olive pests profit from offering flowering plants could not be excluded, although we found no direct indication for this in laboratory experiments. We believe that the potential biocontrol effect of increased biodiversity associated with secondary vegetation can be assessed only by „holistic“ monitoring of the population dynamics of pests and key natural enemies in a long-term study (Bostanian et al. 2004). In Egypt, the „intercropping“ of olive trees with flowering vegetables was well adopted and used by the workers of the experimental olive farm; however, in most olive farming situations the artificial introduction and management of secondary vegetation will probably be too costly or difficult to apply in semi-arid and arid conditions where water is scarce. Keeping spontaneous natural vegetation in alleyways or wildflower strips in order to promote populations of beneficial insects can have additional advantages for the olive grove ecosystem. This refers especially to the prevention of soil erosion, water run-off, or long-term loss of organic matter in the soil (Malavolta et al. 2002), as was observed after intensive clearance of vegetation over several years in Spanish orchards (Beaufoy 2000, 2001). Taking all these potential benefits into account, we suggest that this concept warrants further investigation.

Other tools to protect or enhance key natural enemies

Following earlier work carried out during the former ECLAIR-project (McEwen et al. 1994; McEwen and Morris 1998), we applied various food sprays for potential encouragement of natural enemies in specific field trials in Portugal. Food sprays containing protein sources like L-tryptophan especially attracted Hymenopteran parasitoids. The abundance of Coccinellidae and Chrysopidae also increased in comparison to the control by most of the spray applications. Predation on olive moth eggs deposited on fruits was enhanced in all treatments (57–61%) in comparison to the control (40%). Nevertheless, the effect on fruit infestation was negligible.

Egg parasitoids of the genus *Trichogramma* are well known from many agroecosystems and have also been found in olive groves (see below). They belong to an important group of beneficials regulating pest densities and can be considered as an indicator taxon concerning potential side-effects of plant protection measures. We explored the kairomonal effect of the olive and jasmine moths sex pheromones, olive plant volatiles, and whole-body extracts of the jasmine moth on the attraction and egg-laying behaviour of several species of this genus known to occur in the olive grove ecosystem (Milonas et al. 2003). Some components of the synthetic sex pheromones had a slight repellent effect, some plant volatiles evoked weak attraction, and whole-body extracts of the jasmine moth elicited obvious arrestment behaviour. In general, the various groups of compounds tested did not strongly affect or disturb activity of *Trichogramma*. In a separate study, insecticides used currently in olive cultivation were tested for potential side-effects on *Trichogramma cacoeciae* March., following IOBC-guidelines. This was necessary to provide a reasonable framework for timing of biocontrol applications with unavoidable insecticide treatments. Dimethoate, still the most widely used pesticide in olive cultivation, retained a harmful activity to *T. cacoeciae* on olive foliage for about 2 months after treatment (Youssef et al. 2004).

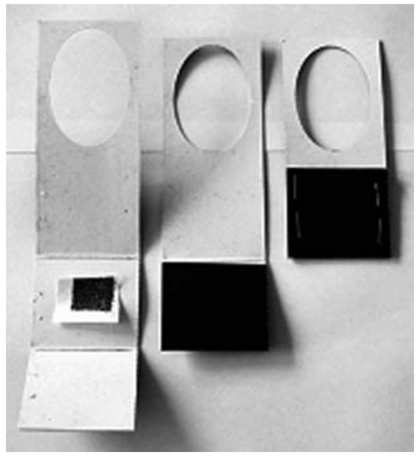

Fig. 9 Carton cards for releasing *Trichogramma*, used in 2004. The releasing material (left: parasitised eggs glued on paper) is completely enveloped after folding the card twice. Emerged wasps are able to leave the card

Releasing *Trichogramma* for the control of olive pests

At present, inundative releases of mass-reared egg parasitoids of the genus *Trichogramma* have become an essential part of the biological control of various lepidopterous pests (Smith 1996). These egg parasitoids can be applied with high selectivity and their mass-production on factitious hosts is economically advantageous. Successful application and pest control has been achieved even in complex habitats like forests and fruit orchards (Hassan et al. 1988; Li 1994; Smith 1996; Mills 2000), although heterogeneity in habitat structure might impede dispersal and activity of the parasitoids. The key to success relies mainly on careful selection of a suitable parasitoid species for the pest-crop system in question.

The first serious attempts to use *Trichogramma* in olive were undertaken by Stavraki (1977, 1985) in Greece, releasing the species *T. cacoeciae*, *T. dendrolimi*, *T. minutum*, *T. pretiosum*, *T. euproctidis* and two unidentified strains against the carpophagous generation of the olive moth in several field trials. These strains had not been collected from olive groves, but some of them were successfully used in other crop systems. Egg parasitism levels achieved were low (<10%) and, concerning pest suppression, releases were not unsuccessful. Jardak (1980) also studied a species (*T. oleae*; Voegele and Pointel 1979), isolated from eggs of *Prays oleae* in France, for its potential to control the olive moth.

More recently, small-scale field trials in Portugal to test control of the olive moth by inundative releases of *T. cacoeciae* showed promising effects, with a reduction in pest damage of up to 46% (Bento et al. 1999). This result, and the encouraging experience in similar crop systems, motivated us to investigate more systematically the potential of an application of *Trichogramma*-species for the control of Lepidopterous olive pests.

Within the first 2 years of TRIPHELIO, field application of mass-reared *Trichogramma* wasps (*T. cacoeciae* in Portugal and Greece, *T. oleae* in Tunisia and *T. evanescens* in Egypt) brought disappointing results. Observed parasitism of olive moth eggs on treated trees was below 30% and the achieved efficacy concerning pest control was far from sufficient in all countries. In Portugal, one factor partially responsible for the failure of releases was the occurrence of high predation by ants, which removed considerable amounts of released material shortly after installation in the field (Pereira et al. 2004). In the last year of the project, we were able to solve this problem by using cardboard devices that wrapped up the releasing material completely, so that ants could not contact it with their antennae (Fig. 9). These devices could also serve as prototypes for use in other crop systems with known high ant predation pressure.

In 2004, we also performed comparative field releases including local strains collected in olive groves by the project team in previous years. Releases were performed from early spring against both generations of the olive moth, and in Egypt also against the continuous presence of the jasmine moth. Despite some critical drawbacks concerning doses and timing of releases, much higher levels of egg parasitism were obtained by these local strains: 35% for a local strain of *T. oleae* in Tunisia; 59% for *T. cordubensis* in Portugal; up to 91% for *T. bourarachae* (on olive moth) and 83% for *T. cordubensis* (on jasmine moth) in Egypt. Significant effects of releases on pest densities and infestation levels were also noted. The best results were obtained by several releases of 3,000,000 wasps/ha (= three cards with 3,000 wasps each per tree) at 2-weekly intervals in the olive plantation in Egypt (Fig. 8). The amount applied is considerably higher relative to what has been used in other crop systems (corn: 150,000 wasps/ha, walnut: 500,000 wasps/ha, apple: 900,000 wasps/ha, pine: 1,500,000 wasps/ha), but we consider this result as a starting point for further optimisation of the method.

Conclusion and outlook

With respect to the outcome of former international research projects dealing with olive plant protection, significant progress was made by TRIPHELIO regarding the following aspects:

- Our research demonstrated that the optimised mating disruption technique for the olive moth worked well under different climatic conditions, resulting in high levels of male disorientation. Efficacy of this method can certainly be raised by strict suppression of the first generation, and extended spatial and temporal application.
- Important steps forward were made regarding monitoring and control of the jasmine moth.
- Our comparative investigation allowed us to differentiate between useful and less useful concepts for habitat management in different olive growing commodities.
- In comparison to previous experience on the use of *Trichogramma* in olive, a significant improvement was achieved by using locally collected, native strains, which demonstrated a promising field performance in their habitat of origin. Further research and development should focus on suitable application designs (releasing devices or other releasing techniques such as spray application, doses, timing) for these strains, as well as on optimisation of available mass production systems in order to maintain their particular qualities and to ensure sufficient output to treat larger areas.

All these approaches have to be considered as „soft“, preventive strategies and we have learnt that their true potential for plant protection will probably be apparent only after repeated application. Moreover, studies on their combined use with other environmentally safe methods are now necessary. For example, the systematic use of *B. thuringiensis* applications against external feeding stages of the target pests can help suppress their densities to a level where the other methods may work better.

All efforts should be made to bring our achievements towards successful commercialisation and practical implementation. We have already stressed the high interest of both consumer and producer parties in pesticide-reduced or even pesticide-free olive production. Therefore, it is not reasonable to wait another 10 years before further progress is made. Much of the experience gained right now will be lost if the work cannot continue, and a laborious re-invention of the wheel will be required. Only when the necessary funding is provided, can further progress in the development of sustainable plant protection in olive growing be expected.

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