LIFE13 ENV/GR/000414

FINAL Report version for the website: www.biodelear.gr
Covering the project activities from 01/06/2014 to 25/10/2019
3/11/2020
LIFE BIODELEAR

Project Data

Project location	GREECE
Project start date:	01/06/2014
Project end date:	01/06/2019 **Extension date:** 25/10/2019
Total Project duration (in months)	64.9 months (including **Extension of** 4.9 **months**)
Total budget	2,205,454 €
Total eligible budget	1,102,727 €
EU contribution:	1,102,727 €
(%) of total costs	100%
(%) of eligible costs	50%

Beneficiary Data

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1. List of keywords and abbreviations

1. AUTH: Aristotle University of Thessaloniki
2. BGLC: Beneficiary Group Leaders Committee
3. BPI: Benaki Phytopathological Institute
4. BPR: Business Planning Report
5. CAGR: Compound Annual Growth Rate
6. DC: Dissemination Committee
7. EAR: Economic Analysis Report
8. FC Financial Committee
9. FR: Final Report
10. GDP: Gross Domestic Product
11. IR: Inception Report
12. IMS: Integrated Mediterranean Strategy
13. IPMT: International Pheromone McPhail Trap
14. IS: Integrated Strategy
15. MTR: Mid Term Report
16. MTD: Mass Trapping Density
17. PR: Progress Report
18. SC: Steering Committee
19. SSDA: Soil Science Department of Athens i.e. the new name of Soil Science Institute of Athens (SSIA)
20. T-MC: Technical-Monitoring
21. UTH: University of Thessaly

2. Executive Summary

The Mediterranean fruit fly (medfly), *Ceratitis capitata*, is considered one of the most destructive pests of fresh fruit worldwide, because of its broad distribution, the long list of hosts and its high multivoltinism (completion of many generations per year). Currently, pest management for the medfly is mainly conducted with the application of synthetic insecticides, which have adverse effects to both the environment and human health. Alternatives to chemical control approaches include the application of Sterile Insect Releases (SIT) and the implementation of mass trapping, which can both be applied over large areas and gradually suppress medfly population at densities lower than the economic injury level. Most of the various trapping devises that are often employed in medfly mass-trapping effort include food lures that attract both males and females. A male specific trapping system also exists. In recent years, a powerful lure consisting of ammonium acetate, trimethylamine and putrescine has been extensively used for both population monitoring and mass trapping purposes. The above "luring" system is rather expensive, especially for low-income countries, and has been accused of including compounds with toxic properties for humans. To accomplish its goals, mass trapping should be implemented based on a knowledge intensive strategic plan that considers not only aspects of the crop but also of the environment and growers' income.

The main objective of the "LIFE Biodelear project" was to develop an Integrated Mediterranean Strategy (IMS) for the control of the Mediterranean fruit fly with the use of the innovative, patented, environment-friendly and low-cost food attractant, Biodelear. Biodelear is non-toxic to humans, attracts more female than male medflies, has minor effects on non-target and beneficial insects and can be applied for a sustainable management of medfly. The IMS that was developed in the course of the LIFE Biodelear project is based on biological data regarding trends of the year round Mediterranean fruit fly population and includes several components that assure (a) a timely application of the mass trapping technique, (b) elimination of insecticide use, (c) employment of sanitation actions to reduce levels of medfly population and (d) constant monitoring of environmental aspects such as pesticide residues, biodiversity, and soil and irrigation water quality.

The project was initiated according to the timetable on the 1st of June 2014 and completed within 65 months (25-10-2019). All actions (A1, A2, B1, B2, C1, C2, E1, E2, and D1&D2) were successfully executed resulting in a wealth of valuable data. Amendments to the original plan have been considered to address emerging issues which, however, assured the scientifically sound execution of all trials. The coordinating beneficiary performed all necessary management activities, to ensure the synchronized function and cooperation of the four scientific teams (SSDA, AUTH, UTH and BPI). The LIFE Biodelear project included the following phases:

A1. Production of traps and attractants: The optimum dose of Biodelear was determined. Large quantities of Biodelear were synthesized and traps and other consumables purchased.

A2. Preparatory monitoring of the demonstration areas: Preparatory monitoring actions were conducted prior to establishing pilot testing sites to evaluate medfly population dynamics, fruit infestation rates, pesticide residues, arboreal and ground biodiversity and soil quality. Preparatory monitoring also preceded the real scale application of mass trapping with Biodelear.

B1. Implementation of innovative Med traps at pilot scale: The performance of mass trapping with Biodelear to control medfly was compared to mass trapping with the common attractant Biolure and the conventional management that involves insecticide application.

B2. Implementation of innovative Med traps at real scale: The performance of mass trapping with Biodelear to control medfly in a large area (10.3 ha) was contrasted to non-managed control plots (1.7 ha).

Over the last phase of field operations, the developed IS was applied to 10.3 ha citrus orchards. The results of this operation were quite positive since a complete elimination of insecticide use was accomplished, the infestation of citrus fruit was maintained at low level, pesticide residues were below detection levels and arthropod diversity was similar to unmanaged control plots.

In brief, the project LIFE Biodelear **succeeded to**:

1) develop a sound, integrated, and environmentally sustainable approach to control the medfly in Mediterranean citrus orchards, based on the IMS tool that has been elaborated following an extensive testing at real/large scale,

2) reduce citrus fruit infestation rates down to 1%,

3) eliminate insecticide use to control medfly and contribute towards rendering Mediterranean farming less dependent on pesticides, in compliance with the EU policy (Dir 92/43 EEC and Reg 2005/396/EC),

4) demonstrate that mass trapping with Biodelear is an economically viable approach to control medfly without adversely affecting human health and the environment. A thorough Business Planning report (BPR) (as advised by the Technical Officer, Ref. Ares (2018)1644471 - 26/03/2018) revealed that the attractant Biodelear is a viable product ready to enter the market,

5) assess the socio-economic and environmental impact of mass trapping with Biodelear on reliable indicators according to the GA,

6) establish processes that may lead to remediation of the ecosystem of the citrus orchards when the IMS tool is considered,

7) establish appropriate methodologies that can be adopted in other related projects for monitoring (a) soil and arboreal biodiversity in citrus orchards, (b) medfly population in mass trapping control projects, c) pesticide residues in citrus fruits, and (d) soil and water quality

8) provide the farmers with brochures and advise them on how to protect their citrus crops from the medfly pest without the use of pesticides, so that citrus fruit production becomes a profitable venture for them.

The LIFE Biodelear project designed and implemented an aggressive **dissemination strategy** in Mediterranean countries, targeting farmers and their associations, as well as local and regional agricultural authorities. Following a series of informative actions, events and workshops, the project gained the acceptance of the targeted stakeholders. The successful implementation of field actions that resulted in the control of the Mediterranean fruit fly in the area of Kampos Chios with the innovative attractant Biodelear and the development of a sound IMS have contributed to a positive reception of the project outcomes by stakeholders and the scientific community. There is a list of producers' organizations that have expressed interest to

use Biodelear and the IMS. After the positive effect to the local community and wide acceptance of Biodelear, we broadly disseminated our findings to both Greek and Mediterranean stakeholders. To further disseminate our findings, we developed informative guidebooks for policy makers that include a thorough assessment and evaluation of the project's results.

LIFE Biodelear established a network among research centers, universities, farmers and their associations in Greece and other Mediterranean countries, regional and national authorities, and representatives of the fruit trading industry aiming at promoting the project's achievements.

The "After LIFE Communication Plan" of the project achievements in combination with the continuous engagement of all partners, ensures the best possible dissemination and communication of project results in the following years enabling transferability within Greece and other Mediterranean countries.

3. Introduction

Background, problems, objectives: Agriculture represents over 15% of the GDP in Mediterranean countries. The Mediterranean fruit fly, (medfly), which infests more than 300 different plant species (mainly fresh fruits, but also vegetables and nuts) is considered one of the most notorious agricultural pests in the world. In Greece, high populations build up from July to November. Depending on the area and local climatic conditions, adult medfly activity, as is estimated by adult captures, may persist throughout the year in the southern parts of the country and cease for several months in winter and spring in cooler areas. Medfly causes major yield losses in fruit production and severe economic concerns in the agricultural sector. Females oviposit on ripe or ripening fruit, causing extensive fruit drop, while larvae develop in fruit flesh rendering them unsuitable for human consumption and trading. In addition, the presence of an established medfly population may elicit a range of quarantine regulation restricting fresh fruit trading and exports.

Environmental problem/issue addressed: Management of medfly populations, in conventional farming depends almost exclusively on insecticide applications (cover and bait sprays) that, as it is scientifically proven, have serious shortcomings, such as the development of pest resistance to insecticides, decrease of the biological diversity, and negative effects on soil properties and the ecosystem. Moreover, the use of pesticides has detrimental effects on the health of farmers and the consumers of agricultural products.

Alternative control methods for medfly include the application of the Sterile Insect Technique (SIT), fruit sanitation, biological control, lure and kill technologies (including mass trapping). Lure and kill are considered an environment-friendly technique; however, depending on the adopted killing system it may negatively affect biodiversity and beneficial fauna by attracting and killing large number of non-target insects. This is because several lure and kill devices use insect attractants, such as yellow color and food baits that besides medfly attract non target insect species including beneficials. In addition, most of the commercially available products are considered expensive for commercial farms, especially in developing countries and poor areas. Integration of the different methods in a broader strategy is expected to provide a sustainable and environmentally friendly management of medfly and that of other fruit flies. The implementation of such a strategy and its acceptance by fruit growers is challenging, especially when mild intervention such as mass trapping is included.

Outline of concepts and approaches: LIFE Biodelear is an ambitious project that addresses medfly control in Mediterranean citrus orchards, with an integrated and environmentally sustainable strategy based on a novel, non-toxic attractant. The project developed a new, environmentally safe, mass trapping technology, an integrated mass trapping strategy for individual farms (IS) and an Integrated Management Strategy (IMS) for larger Mediterranean citrus orchards. During the 5-year course of the LIFE Biodelear project we adopted mass trapping with the attractant Biodelear as the main tool to manage medfly populations in traditional citrus orchards in the island of Chios. The ultimate aim included the development of an integrated strategy based on the above mass trapping system and the elimination of any synthetic insecticide use. The production of the attractant Biodelear is based on the Maillard reaction. Biodelear is a mixture of volatile compounds derived from the Maillard reaction between urea and fructose in the presence of water, entrapped in the amorphous nitrogen polymers called melanoidins and liberated slowly by oxidation of these polymers working synergistically in attracting mainly medfly females. This renders Biodelear fundamentally different from all current commercial attractants available on the market for controlling medflies, which are based on compounds that are often considered toxic for humans (e.g. trimethylamine). It should be noted that in this trapping system no insecticide or other toxic elements is used.

Almost all field experiments were conducted in the traditional citrus farms in the Kampos area in the island of Chios, which is well regarded as a unique and protected environment. The preparatory monitoring of the pilot area revealed that the initially selected farms differed more than expected in the variety of citrus species and cultivars, the age, height and canopy size of the trees, the tree planting distances, and the number of species and density of non-host plants. To address the above issues, the experimental plan of the pilot phase had to be revised to allow a proper scientific evaluation. The new experimental plan increased the analytical power for the statistical analyses and facilitated extraction of solid conclusions and sound interpretation of the results regarding the validity of mass trapping with Biodelear. Several technical and methodological issues have been revisited during the preparatory phase as well. The optimal Biodelear doses have been determined, the sampling plan for monitoring the arthropod diversity within the orchards was developed, the sampling plan for assessing soil quality and insecticide residues in fruits have been worked out. The Pilot implementation included the application of our experimental plan in commercial farms and the evaluation of the mass trapping technique using Biodelear against the conventional control approach and the most promising commercial mass trapping devices available. Unmanaged control farms have been included as controls as well. During the two years of the pilot phase (B1) we have been working on developing a new integrated strategy (IS) for the management of medfly in citrus orchards, and on establishing well-appreciated indexes and analyses to assess the environmental impact. The implementation of the new integrated strategy was applied to the demonstration area of 10.3 ha, which was the next step for the control of the medfly insects in the citrus orchards (B2) for another two years of duration using such as the IS, the well -appreciated indexes and analyses to assess the environmental impact and all the methodologies (ground and arboreal biodiversity, soil quality, pesticide residues, medfly population) evaluated indicators to develop the IMS to control medfly without negatively affecting the environment and the human health. Economic aspects and risk analyses have been developed covering all the actions (A2, B1 & B2).

Results and environmental benefits in a nutshell:

- The results demonstrated reduction of infestation of the citrus fruits to acceptable levels even close or below 1% compared to the preparatory monitoring of the demonstration area before the initiation of the project. Moreover, these levels were very similar or slightly better compared to conventional practices (use of pesticides and insecticides) and with the use of the attractant Biolure for controlling the medfly, which was one of the most ambitious targeted goals of the initial proposal.
- In addition, the extensive use of mass trapping with Biodelear was proven to be neutral for the ecosystem with trends to remediate the distracted citrus farms compared to the previous conventional practices that have been applied (before the establishment of Life Biodelear trials) for the control of the Mediterranean fruit fly. Also, the Biodelear attractant was found to be a viable commercial product for both the local and international markets.
- To eliminate insecticide, use to control medfly and contribute towards rendering Mediterranean farming less dependent on pesticides, in compliance with the EU policy (Dir 92/43 EEC and Reg 2005/396/EC).

4. Administrative part

The structure of the project management is given in Figure 1.

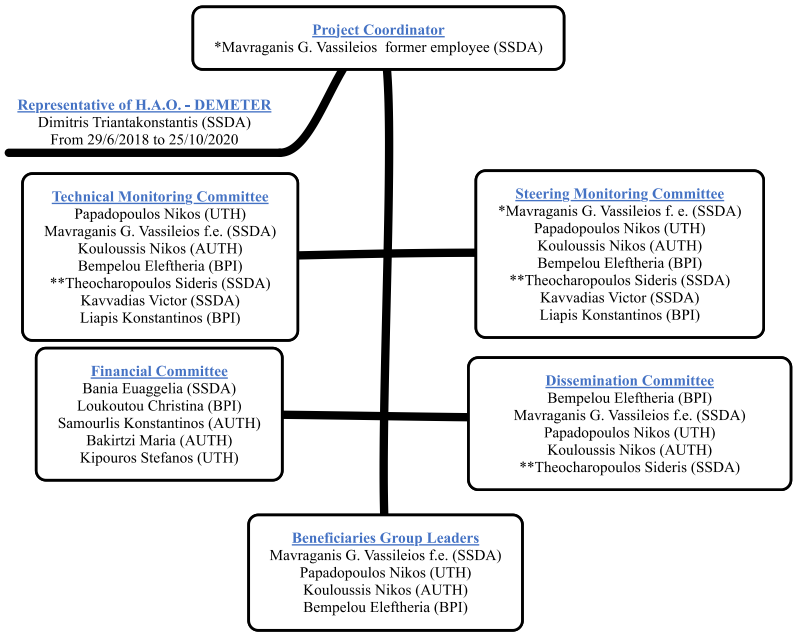


Figure 1.

*Dr. Vassilis G. Mavraganis changed his employment status to personnel contracted from 9/10/2018 to 25/10/2019, **Dr. Theocharopoulos Sideris (SSDA) retired in the year 2018.

5. Technical part

This chapter describes the work carried out during the project, per action. All the produced deliverables and outputs of the project are included as Annexes of the current Final report.

5.1. Technical progress, per Action

5.1.1 Action A1: Production of traps and attractants

Foreseen start date 1/6/2014 *Actual start date 1/6/2014*
Foreseen end date 30/11/2017 *Actual end date 30/11/2018*

Objectives: To test the optimum dose of the Biodelear attractant, to produce the required quantities of the attractant Biodelear and, to purchase attractants, traps and other consumables.

Biodelear Production (SSDA): Biodelear was produced with a one pot reaction between fructose and urea in the presence of water in a ratio of 3:1:1 w./w./w. fructose/water/urea (i.e. with 900 g of fructose / 300 ml of water / and 300 g of urea), resulting in an amount of 1500 g of Biodelear. Urea was added in small amounts in the refluxed solution of the dissolved fructose to achieve the Maillard reaction product named Biodelear patented by V.G. Mavraganis et al. 2011.

Biodelear Efficacy tests: The efficacy tests, which aimed to determine the optimum amount of Biodelear that would be used during the demonstration actions B1 and B2 in the pilot areas, were performed in field sites close to University of Thessaly (UTH) and in the University farm of the Aristotle University of Thessaloniki (AUTH), from September 2014 to April 2015.

Methodology: The three different doses of Biodelear tested were 17, 51 and 85 g impregnated in custom-made sponge dispensers (wettex). Wet (filled with 10% of propylene glycol in a 250 ml water solution) Plastic McPhail-type traps were used. They were checked every week and all captured flies were counted, identified, sexed and removed.

Results and conclusions: The results revealed that in both trials the lowest tested dose of Biodelear (17 g) was the best for both male and female medflies. Interestingly, all Biodelear doses attracted low numbers of non-target and beneficial insects.

Problems

The efficacy tests were conducted in two orchards located in Central and North Greece (Lechonia, and Thessaloniki, respectively) and not in the island of Chios as originally planned. This change was dictated because the process of selecting and renting appropriate farms in Chios took longer than expected. Therefore, we decided to run two instead of one experiment in the area of UTH and AUTH respectively to avoid possible delays and increase the credibility of our experimentation.

Evaluation

Action A1 has been successfully completed. It was concluded in October of 2018 instead of 2017 as foreseen in the proposal due to additional demands for the production of the attractant Biodelear.

Indicators of progress

The indicators of progress foreseen in the proposal were:

- *Synthesis of 100 kg to 200kg of attractant Biodelear depending on the results from the efficacy tests (5.5% of the total Biodelear from June to August of 2014) (17.2% of the total Biodelear produced until the December of 2014) (77.7% of the total Biodelear produced until the March of 2017).*

The production of Biodelear started from the beginning of the project (June 2014) and finished in October 2018. A total of approximately 400 kg of Biodelear (for the two efficacy tests, Action B1 and Action B2) were produced by SSDA.

- *The result of the efficacy test will be known and estimated by the end of December of 2014.*

The results were delivered on time.

- *Purchasing of the following items: 1200 McPhail traps, 100 pieces of commercial Biolure attractant, 200 pieces commercial Capilure for conducting male annihilation studies, appropriate reagents quantities to synthesize the attractant Biodelear.*

There was a substantial increase on the number of the traps and attractants for the two efficacy tests (see above problems), monitoring and mass trapping technique used during the implementation of the project for the protection from the medfly insects due to the change of the planning of the action B1, to meet certain specifications necessary for the citrus orchards in the island of Chios, nevertheless the expenses were within the limits of the initial proposal, for the detail.

- *Preparation of the traps (5.5% prepared from June to August of 2014) (17.2% prepared until the December of 2014) (77.7% prepared until the March of 2017).*

The preparation of the traps was completed by 11/4/2016. Few additional traps were purchased for the needs of action B2 at 9/11/2016. The extension of the duration of the action A1 until October 2018 is justified because of the increased needs of the Biodelear attractant for the action B2.

Milestone

- *"Determination of the optimum amount of the innovative attractant to be used during the demonstration (Report)"*

The optimum dose was determined to be 17 g Biodelear per trap.

5.1.2 Action A2: Preparatory monitoring of the piloting and demonstration areas

Foreseen start date 1/6/2014 *Actual start date 1/6/2014*
Foreseen end date 30/3/2017 *Actual end date 31/3/2017*

Objectives: Selection of the demonstration areas and execution of preparatory monitoring. Recording of the practices that farmers follow to address medfly threat in the demonstration areas and assess their impact on medfly population, fruit infestation rates, farm environment (soil, quality, biological diversity, pesticide use and management) as well as on aspects of fruit production (associated cost, quality of fruit, productivity of trees). Perform preparatory monitoring action before the initiation of pilot (B1) and real scale (B2) testing.

Medfly population monitoring (AUTH)

A2 preparatory monitoring for the actions A1 and B1.

Introduction: The population density of the Mediterranean fruit fly in the experimental orchards was assessed for 2014 and 2015.

Methodology: In 2014, we started the preparatory monitoring on 30th of October with 9 McPhail traps baited with Biolure scattered in the experimental area. In 2015, five trapping stations, consisting of one McPhail trap baited with the three-component Biolure dispensers and one Jackson trap baited with the male specific attractant trimedlure, each placed ca 15m apart, were established in each experimental orchard. Captures of medflies, as well of non-target insects were recorded and removed from traps weekly. Replacement of trimedlure and Biolure dispensers was conducted every 3 and 4 weeks respectively

Results: The average number of captures during the season of 2014 was 6.45 Flies per Trap per day (FTD) most of which were females. The population of females was higher during November but in December it dropped to very low levels. In addition, the number of non-target insects captured was relatively low throughout the experimental period. In 2015, although traps were deployed in January, the first captures were recorded late in June and were considerably higher compared to 2014. Captures peaked in July until mid-late August and reached an average number of 62.9 FTD. Medfly population in the area was rather low from mid-September until October.

Conclusions: During the period of medfly activity, population densities were high throughout the period of the preparatory monitoring for the pilot scale and particularly during August 2015 reaching an average of 175.5 flies/trap. These results are aligned with earlier studies in the area of Chios [Byron I. Katsoyannos et al. 1998] and justify the implementation of the experimental actions of the research program (i.e., actions B1 and B2).

A2 for the time period of the action B2 (real scale)
Introduction: We used data from the B1 action collected in 2016 and data collected from January 2017 until late March 2017, before the real scale mass trapping phase started. *Methodology:* We followed the same procedure as in 2015 and the same experimental citrus orchards One farmer abandoned the project, so instead of four plots we used the three experimental orchards used in 2015

Results: The first extensive captures in 2016 were recorded in June. Male medfly captures peaked for a second and third time in mid-August and mid-September, respectively. Similar patterns were observed also in female medflies. In 2017, populations were similar at all plots and at near zero levels during the observation period, which was continuous all year long and lasted from the beginning of 2016 until late March 2017.

Conclusions: Medfly population levels were high in the experimental orchards during the observation period of 2016, which was continuous all year long. A considerable number of medflies was caught during summer season (high peaks in July and August) at a critical time point of the growth season, which is regarded as the first period of citrus susceptibility, while a second medfly population peak was observed in autumn, which coincides with the second period of citrus growth. From the beginning of 2017 until late March captures were near zero as expected and in accordance with previous years.

Assessment of Biological Biodiversity and of Citrus fruit infestation rates (UTH)

Ground and arboreal biodiversity

A2 action for the pilot scale: In each of the four selected / rented experimental orchards we established a grid of five pitfall traps (20 in total each time). All trapped arthropods were counted and identified, and beetles (Coleoptera) were further identified and analyzed as they are considered important for estimates of ground biodiversity. Furthermore, using appropriate dichotomous keys we managed to identify most species of Carabidae and that of other Coleoptera families. Three samplings were performed at three different dates i.e., 23/6, 19/7 and 9/9 of 2015.
In total, 1,557 arthropods were retrieved from the pitfall traps during the three samplings. Data analysis revealed that neither citrus orchards nor different sampling dates had a significant impact on abundance. Nevertheless, Orchards 3 & 4 had slightly higher abundance of arthropods relative to the other two. The Insecta class represented 58.76 to 86.02% of all arthropods collected, followed by Diplopoda (9.32-37.53%), Arachnida (2.13-4.04%) and Chilopoda (0.00-0.62%). The number of captured insects was not affected by the citrus orchards or the sampling dates. Sampling date had a significant effect on the abundance of ground beetles with the number of captured individuals being significantly lower in the first sampling (23/6) relative to the other two. Finally, the relative abundance of ground Coleopteran families varied (though not significantly) among citrus orchards. Overall, the above information provides a good background information for the biodiversity of the ground arthropods in the area of Kampos Chios. As it was expected, season (capture date) may affect the abundance and diversity of specific insect taxa (the date of capture was a significant predictor for the abundance of ground beetles only but not for that of other arthropods); however, the four different farms considered, have rather similar profiles in terms of abundance of arthropod, insect and Coleoptera taxa. This information was considered for the development of the experimental design that has been applied in B1.

A2 for the real scale: Background information was collected before the commencement of Biodelear evaluation at real scale, since new plots were added in our experiments (for details see B2 action below). Data from the pilot phase were considered for all plots included in B2.
Methodology: By applying the protocol, we established a grid of 70 pitfall traps in total. Sixty (60) traps were placed in the Biodelear treatment and ten (10) in Controls citrus orchard. We performed one sampling at 2/5/2017 soon before the establishment of the Biodelear mass trapping. Data from the pilot phase were also used as background information in B2 since all farms used in B1 were included in B2 providing collectively adequate information for ground arthropod biodiversity.
Besides ground biodiversity, our initial plan included the assessment of the arboreal biodiversity as well. However, the extended flowering period of citrus trees during March-May 2017 resulted in the presence of a high honey-bees population in the experimental orchards. In order to avoid any concerns of beekeepers regarding potential harmful effects of "fogging" with an insecticide on the honey-bees we decided to skip the arboreal preparatory monitoring. Preliminary data collected in June and August 2016 in the frameworks of B1 were considered as the only available background information for the arboreal biodiversity.
Results: In total 1,064 arthropods were retrieved from the pitfall traps during the preparatory sampling. The average number of arthropods was higher in the Control orchards relative to Biodelear selected ones.

Conclusions: The preparatory monitoring of the ground biodiversity between the real scale Biodelear selected citrus orchards and the Controls suggest a higher abundance in the second case though not significant.

Citrus infestation rates

A2 for the Pilot scale: To estimate fruit infestation rates mandarin oranges were collected from different farms in Kampos Chios during autumn 2015. Fruits were transferred to the laboratory and the proportion of infestation was determined.

Results: The infestation rates ranged from 25 to 12% for clementine and the local Chios mandarins respectively. The average number of pupae per fruit (considering infested and non-infested fruit) was estimated to 3.8 and to 0.5 for the two cultivars respectively.

Conclusions: The infestation rates of mandarin oranges in the area of Kampos Chios (both clementine and local cultivar) were high (well above the economic damage which is range in most years between 5-8 % fruit infestation depending on the current prices of citrus species). Based on these findings we proceeded to establish the B1 action in the area of Kampos Chios.

A2 for the Real scale: Since the estimation of citrus infestation rates coincides with their ripening period late in autumn, it was not possible to perform preparatory monitoring in the real scale Biodelear selected plots and the respective Controls because renting of farms concluded in January 2017. Instead, we used the data collected in November of 2016 during B1 action to perform this comparison. These data regard both the mass trapping treated and the control plots of all farms that were included in B2. The protocol regarding the sampling method of citrus fruits is described in action B1.

Results: The total percentage of infested sweet oranges (22.38 %) in the Control orchards was higher than that of the Biodelear treated plots (6.40 %). Likewise, infestation rates of mandarin oranges (12.40 %) in Control were higher than that in Biodelear treated plots (0.90 %)

Conclusions: Citrus fruit infestation rates were significantly higher in the Control orchards relative to the Biodelear ones (fruit sampling of 2016). Although medfly populations may vary from year to year, this tendency is expected to remain during the real scale implementation since data from B1 action revealed that the application of mass trapping with Biodelear provides similar "protection" levels against medfly infestation compared to Biolure and the conventional (cover sprayings) methods.

Assessment of the Pesticides Residues (BPI)

A2 for pilot scale areas: The first fruit sampling was conducted in December 2014, following the protocol described in the Grant Agreement. The number of samples taken was 133, higher than those envisioned in the proposal, due to the larger area of the farms. The analysis revealed no detectable residues of plant protection products or plant growth regulators. This first sampling was considered as a preliminary screening of the pilot area of Kampos Chios, providing an estimation of the potential contamination of the cultivated areas during the specific sampling period.

In order to elucidate the chemical profile of the pilot area, a second citrus fruit sampling was carried out in June 2015. Sixty-seven samples of combined (oranges and mandarins) citrus fruits were collected and analyzed as reported above. Based on the obtained results, the insecticides chlorpyrifos (58) and deltamethrin (22) were determined, as well as the fungicide propamocarb (11).

The results of the above samplings provided information on the current status of pesticide residues in the experimental orchards of Kampos.

A2 for real scale areas: The third sampling regarding action A2 was carried out in January and March 2017, after the rental of the experimental orchards for the real scale. A total of 341 samples of citrus fruits were collected, prepared and analyzed as reported above, in order to provide data for the preparatory monitoring of B2. As has already been reported, the initial farms in which the preparatory monitoring took place were included in the large scale (10 ha) experimental orchards. Therefore, in this action (A2) the preparatory monitoring of real scale regarding pesticide residues was focused on the new orchards that participate in the project, since the rest of farms had already been examined in previous samplings, and therefore the number of samples is lower that the corresponding in the approved proposal.
According to the obtained results, the pesticides **spirotetramat (44)**, **dimethomorph (11)**, **flumioxazine (6)**, **mepanypirim (1)**, **azoxystrobin (4)**, **propamocarb (3)** and the **synergist piperonyl butoxide (76)** were determined.
In total, residues of plant protection products were detected at the 41% of the examined samples, whereas, the 59% of the collected samples were free from pesticides (No Detected), as shown in Figure A2-4. In conclusion, before the use of Biodelear mass trapping, detectable residues were observed in the 15% of the samples collected from the control orchards and in the 32.3% of the relative to Biodelear selected ones.

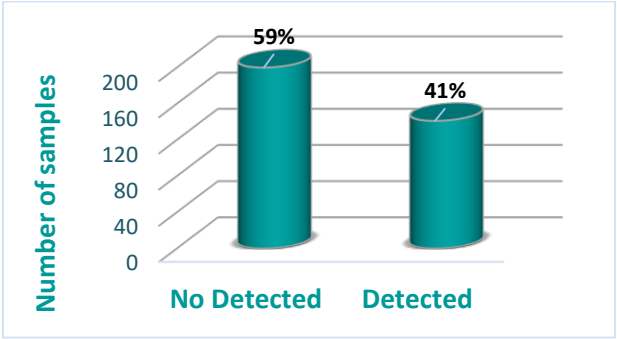


Figure A2-4. *Detected (n= 140) and not detected (n=201) citrus fruit samples at third sampling of Action A2 (preparatory monitoring of large scale).*

Assessment of soil quality (SSDA)

A2 for the Pilot scale: Fifty-two soil samples were taken from the four farms used in pilot testing and in different soil depths 0-30 and 30-60 cm (1st sampling; November 30th – December 3rd, 2014). Another 64 soil samples were taken in different soil depths 0-30 and 30-60 cm (2nd sampling; August 5th-7th, 2015) and 26 samples were taken in different soil depths 0-30 and 30-60 cm (3rd sampling; March 28th-29th, 2016) for the three control farms. Samples were carefully stored, preserved and transported to the laboratory.
SSDA, which is accredited according to ISO 17025, undertakes all the analyses, i.e., texture, pH, electrical conductivity, total salts, moisture content, saturation percentage, exchangeable cations/anions (Ca, Mg, Na), nutrient content (N, P, K), organic matter, available Fe, Mn, Cu, Zn, Cl^{-1}, SO_4^{-2}, NO_3^{-1}, NH_4^{+1}. Chemical and physical analyses were carried out following standard soil analysis methods.
Results: For the *1st sampling period*, and *2nd sampling period*, high concentration of water-soluble ions (Cl^{-1}, NO_3^{-1}, PO_4^{-3}, SO_4^{-2}) were determined in soils especially Cl^{-1} which may be

attributed to the low quality of irrigation water (intrusion of sea in water). Also, in both the two sampling periods high concentrations of Cu were reported. For the *3rd sampling period*, soils were rich in inorganic nitrogen, exchangeable Mg and K. They also presented high percentages of Cu, Mn, Fe and available Ca and P.
Conclusions: The results of the soil samples that have been collected during this Action indicated that soil quality was negatively affected by previous cultivation practices. The contamination of agricultural soils with Cu/Zn-based pesticides was attributed to the regular use of fungicides, which can potentially pose a risk to the environment, particularly if residues persist in the soil or migrate off-site and enter waterways. The high levels of soluble anions in soil of the citrus groves were attributed to both inappropriate nutrient fertilization practices taken place in the past as well as to bad quality of irrigation water mainly in coastal regions. Regarding soil nutrient availability, the results showed that, in general, there were no deficiencies of chemical elements.

A2 for the Real scale: Data on soil quality that were collected at the end of the action B1 were considered for the preparatory monitoring of B2. In addition, 52 soil samples were taken in two different soil depths 0-30 and 30-60 cm from 7 new orchards that have been added (5th sampling; 29-31 March 2017). All the above samples from the newly added orchards were treated only with Biodelear. The preparatory sampling for the large-scale overlaps with the completion of the action B1 due to common citrus areas used in action B2
Results: The soil analysis results indicated that available Cu, Zn, water soluble Cl^{-1}, SO_4^{-2} and exchangeable Mg were well above threshold levels. For the *5^{th} sampling periods*, soils presented high percentages of Cu, Mn, Fe and available Ca and P.

Problems

1) Preparatory monitoring for the action B2 was accomplished by collecting data from the new farms rented in January 2017 and using additional data of the action B1 for those farms that were included in B2. Depending on the monitoring action specific adjustments were made as it is explained above.
2) In the action A2 for the real scale the assessment of the arboreal biodiversity was not possible to attain due to the extended flowering period of citrus trees during March-May 2017 that resulted in the presence of numerous honeybees in the experimental orchards. In order to avoid any concerns regarding potential harmful effects of "fogging" on the honeybees we decided to skip the arboreal preparatory monitoring. As we reported in page 20, the two arboreal samplings that were performed during 2016 (June and August) in B1 were used as background information (preparatory monitoring) for B2. The fact that we included unmanaged plots (controls) in our experimentation provided an additional element to assess effects of mass trapping with Biodelear on arboreal biodiversity.

Evaluation

The implementation of the action A2 (Preparatory monitoring of the piloting and demonstration areas) was successful for both the actions B1 and B2.

Indicators of progress

The indicators of progress foreseen in the proposal were:

- *Collection and analysis of 40 + 40 + 60 to 70 soil samples –A total of 2800 to 3000 analyses (20 soil parameters of each soil sample). (Collection 40+40 soil samples will be completed*

from June of 2014 to August of 2015 for the two (2x1 ha) pilot citrus orchards). (Collection 60 to 70 soil samples from June of 2016 to August of 2017 for the 10-ha citrus orchard.

Completion of the collection and analysis of 142 soil samples –A total of 2840 analyses (20 soil parameters of each soil sample) for the four pilot farms and the three control farms were conducted. Completion of the collection and analysis of 106 soil samples and 52 soil samples in the March of the year 2017 before the implementation of the control of the medfly for the 10-ha citrus orchard a total of 3040 analyses for the action A2 for the B2 action.

- *Collection and analysis of 720 fruit samples –A total of 1440 analyses (checking 334 active compounds plus 19 acidic plant protective and plant regulating growth compounds). (Collection from August of 2014 to the end of December of 2014 for the two 2x ha citrus orchards) (Collection from August of 2016 to the end of December of 2016 for the 10-ha citrus orchard).*

Completion of the collection and analysis of 541 fruit samples in total, derived from the orchards involved in each phase (pilot and real scale) of the preparatory monitoring. –A total of 1082 analyses for pesticide residue detection in the citrus fruits. The reason for the lower number of fruit samples was due to the fact that there was an overlap between the Action B1 and preparatory Action A2 (because some of the citrus orchards in the Action B1 were used also in the Action B2) for the Action B2.

- *Completion of the pilot areas inventory (web available). (For the (2x 1ha) two citrus orchards results the web available information will be informed from December of 2017 to January of 2018) (For the 10-ha citrus orchard results the web available information will be informed from December of 2018 to January of 2019).*

The inventory was completed and made available on 2017. All data collected regarding both the pilot and real scale have been uploaded and are freely available to LIFE Biodelear partners and EC.

- *Completion of risk analysis of the pilot areas. (From December of 2016 to August of 2017 for the two (2x ha) citrus orchards) (From December of 2018 to June of 2019 for the 10-ha citrus orchard).*

The risk analysis was completed on 25 October 2019. Data collection and first attempts to perform the risk analysis were conducted in 2017. However, a first internal revision revealed that additional elements and new structure should be incorporated. The additional elaboration resulted in a delay in the delivery of the risk assessment.

- *Acquisition of a permit for the implementation of the protection of the 12 ha citrus orchards from the Med fly from the Ministry of Agricultural and Food Development (from June to August of 2014 the permit will be obtained).*

Two permits (each permit has a duration of 3 years) were acquired from the Ministry of Rural Development and Food covering the whole duration of the program.

Deliverables

- *"Web available inventory of the pilot area"*

The Inventory of the pilot area can be accessed at https://www.biodelear.gr/index.php/en/the-project/results, after logged in with username and password given in the Dissemination Action.

- *"Risk analysis of the conventional practices to address med fly" (Report)".*

Risk analysis of the pilot area was completed 30/06/2019

5.1.3 Action B1: Implementation of innovative Med traps at pilot scale

Foreseen start date: 01/06/2015 *Actual start date: 15/06/2015*
Foreseen end date: 31/12/2016 *Actual end date: 31/12/2016*

Objectives: To control medfly with mass trapping using the innovative attractant Biodelear at pilot scale and the comparison of the proposed system with mass trapping with the commercial attractant Biolure and conventional control (cover sprayings) methods. To evaluate whether infestation of citrus fruits, farms biological diversity, pesticide residues and the soil quality – are affected by mass trapping using Biodelear. To develop an integrated strategy for managing medfly at local scale (Integrated Strategy, IS)

Experimental farms and protocols

The ultimate target of the current action was to develop a sound Integrated Strategy for medfly that would preserve environmental quality and benefit both the consumer and the farmer. Following the original proposal, we selected and rented farms and started monitoring the medfly population with traps and sampling of fruits. However, we soon realized that the farms differed more than we expected in the variety and age (height and canopy size) of the trees they contained as well as in the tree planting distances. There were also other differences, such as in the number of species and density of non-host plants in each orchard. Based on that we decided to amend the original experimental plan to generate replicates and enhance our ability to statistically analyze and better interpret the results. The new experimental design includes 4 farms of 1 – 2 ha each. Each farm was divided in 2-3 plots of different treatments, i.e. plots with traps baited with Biolure, plots with traps baited with Biodelear, and plots managed conventionally with the use of pesticides. We have also included, as controls, three smaller farms (≈0.5 ha) in the area that received no treatment at all. All these farms were included in the second phase of the project where Biodelear has been applied in a real scale (B2). Hence, the following treatments were established in September 7, 2015: (a) Biodelear (100 traps baited with 17g Biodelear per ha), (b) Conventional (treated with cover insecticide applications of deltamethrin on October 7, 10 and 31 by the farmers), (c) Biolure (100 traps per ha baited with Biolure dispensers), and (d) Control (unmanaged plots receiving no insecticide application or any other medfly intervention method). Three of the farms included plots with treatments a, b, and c and one with treatments a and b only. Details of the seven farms used in the current action are given in Figure B1-1. All Mass Trapping Devices (MTD) were checked every 2 weeks and the aqueous solution of 10% propylene glycol was renewed. Biodelear and Biolure dispensers were renewed every one and three months respectively. There were 14 experimental plots in total. Following an initial analysis regarding the cost of Biolure and considering also the recommended deployment density of Biolure dispensers for mass trapping purposes by commercial agents, trap density was revisited in 2016. The density of the mass trapping devices was adjusted to 100 and 70 per ha for the Biodelear and Biolure treatments, respectively. UTH was actively involved in designing the general experimental plan and those protocols regarding

adult trapping; however, AUTH was responsible for reporting and implementing the current protocols.

Briefly, the following assessments were conducted: (a) medfly population monitoring (AUTH), (b) infestation rates on sweet and mandarin oranges (UTH), (c) ground and arboreal arthropod diversity (UTH), (d) pesticides residues (BPI), and (e) soil quality – chemical and physical properties of the soil- (SSDA).

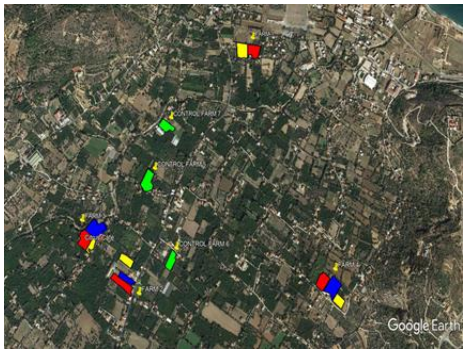


Figure B1-1. *Google earth view of the seven farms used in our experiments. The red areas in farms 1, 2, 3 & 4 are the Biodelear. The blue areas in farms 2,3 & 4 are the Biolure. The yellow areas in farms 1,2,3 & 4 are the conventional (use of insecticides). The green areas 5,6 & 7 are the control*

Medfly population monitoring (AUTH)

Methodology: The population density of the Mediterranean fruit fly was assessed with McPhail type traps (baited with Biolure dispensers) and Jackson traps (baited with trimedlure). We established four experimental citrus orchards Captures of medflies, as well as those of non-target insects were recorded and removed from traps every week.
Results: During both years, 2015 and 2016, the medfly population was high at the beginning of the trapping season in the orchards treated with conventional methods but decreased as time advanced. Medfly population trends were similar between Biodelear and Biolure citrus orchard, which showed that population suppression was comparable between the two-trapping methods. The average female medflies captured per trap per day (FTD) throughout the season was higher in the control farms during 2015 compared to Biodelear mass trapping farms. In 2016 there were no do not point to large differences among treatments.
Conclusions: Population trends were quite similar in the Biodelear and Biolure citrus orchards and the ensuing population suppression was comparable.

Assessment of citrus fruit infestation rates and of the biological diversity in the pilot scale area field trials (UTH)

Citrus fruit infestation rates
Methodology: Infestation rates of citrus fruit were used for measuring the efficiency of mass trapping with Biodelear. Given that the susceptibility of citrus fruit to medfly attacks differ among citrus varieties, fruit infestation rates were measured individually for sweet oranges and mandarin oranges. In this context, 50 sweet orange and 100 mandarin orange fruits were randomly collected from each experimental plot (4 Biodelear-treated plots, 4 Conventional plots, 3 Biolure-treated plots, 3 control plots), taking into consideration to include all available cultivars at approximately equal proportions. Recently fallen fruits were also included in the sampling besides those collected from the trees. All fruit samples were placed in paper bags, treated with 5% boric acid to eliminate possible fungal infection and transferred to the laboratory of Entomology and Agricultural Zoology at the University of Thessaly to be

inspected for medfly infestation. Fruits from each plot were visually inspected for the presence of medfly oviposition stings. Fruits with at least one oviposition sting were considered as infested. During the Pilot scale, fruit sampling took place on November 23th and 24th, 2015 (sampling of 1st year) and December 6th, 2016 (sampling of 2nd year). A total of ≈2.250 and 2.000 fruits were collected during the first and second sampling period, respectively. Infestation rates of sweet oranges and mandarin oranges were recorded for each treatment (Biodelear-treated, Biolure-treated, conventional orchards and control (unmanaged orchards).

Results:
First year sampling (2015): The mean percentage of sweet oranges with oviposition stings ranged from 10.8% in the Biolure plots to 34.4% in the controls. The mean percentage of mandarin oranges with oviposition stings ranged from 4.33 % in Biolure plots to 20.48 % in controls.
Second year sampling (2016): The mean percentage of sweet oranges with oviposition stings ranged from 7.22% in Biodelear to 25% in controls plots. The mean percentage of mandarin oranges with oviposition stings ranged from 0.99% in Biodelear to 10.68% in control plots.
Conclusions: The application of mass trapping with Biodelear provided similar "protection" levels against medfly infestation compared to Biolure and the conventional (cover sprayings) methods. Moreover, in the case of mandarin oranges, infestation rates in 2016 were slightly below 1% reaching the target level set in the proposal.

Biodiversity

Ground arthropods sampling
Methodology: A grid of five pitfall traps in each experimental plot (4 Biodelear-treated plots, 4 Conventional plots, 3 Biolure-treated plots, 3 control plots) was established. In total there were 14 plots and each time 70 pitfall traps were placed. Pitfall traps' positions during samplings were selected so that they covered the orchards' area in a uniform way. Each trap included a white plastic container covered by an elevated white plastic dish as a roof, and it was filled with approximately 300 ml of a 1:1 water solution with antifreeze liquid 30% w/w in ethylene glycol and a few drops of dishwashing liquid. Traps remained for 48 h in the experimental plots. Then, their content was sieved, placed into sealed plastic bags and transferred to the laboratory for inspection. We performed six samplings during B1 action on 11/10 and 21/11/2015 (1st year sampling period) and on 20/4, 18/6, 23/8 and 17/10/2016 (2nd year sampling period).
Results: In total 25,750 arthropods were retrieved from the pitfall traps during the six sampling dates. Data analysis revealed that both treatment (medfly control method) and sampling date as well as their interaction affected captures in pitfall traps. Arthropod captures were higher in control followed by Biodelear and Biolure plots. The lowest number of arthropods was found in the conventional plots.
Conclusions: The implementation of mass trapping with both Biodelear and Biolure increased ground arthropod abundance relative to conventional plots (full cover sprayings). However, untreated orchards (controls) still had higher abundance relative to Biodelear and Biolure treated ones, suggesting that the recovery of ground biodiversity is a long-lasting procedure.

Arboreal arthropods sampling
Methodology: Fogging method was used for sampling arboreal biodiversity. Our sampling protocol involved the selection of one, similarly-sized, sweet orange tree in the middle of each experimental plot. A SWINFOG SN50 (Germany) thermal fogger provided with PHOBI E (etofenprox 30 %) insecticide was used. We performed two applications, one preliminary to evaluate the performance of our protocol by fogging 4 trees (each one in a different treated plot) and one full scale by fogging 14 trees (one tree in each of the 14 experimental plots). Foggings took place between 07:00 and 09:00 when the air temperature was relatively low and therefore arthropods activity was minimal. Before each fogging, the ground beneath the trees was lined down with white nylon sheets 4x4 m to ensure collection of arthropods from both the trunk and the fronds. Each tree was fogged for approximately 2 min, making sure that the whole trunk and foliage area received insecticide. Half an hour after the applications, specimens from each sheet were collected and placed into sealed plastic bags and transferred to the laboratory for identification. We performed two sampling during B1 action on 27/7 and 26/8/2016.
Results: In total 882 arthropods were collected from the two samplings performed. The average arthropod abundance ranged from 91.8 arthropods (control) to 27.6 arthropods (conventional) among treatments. Data analysis revealed that treatment had no significant impact on the mean number of tree arthropods.
Conclusions: Medfly control treatment had no effect on arboreal biodiversity of sweet orange trees in Kampos Chios.

Assessment of the Pesticides Residues (BPI)

B1 action for the pilot scale area: The first sampling regarding the B1 action was conducted in December 2015.
Methodology: Sampling was carried out separately for each medfly management treatment (Biodelear, Biolure, conventional) and analyzed as it is described in A2.
Results: In total 92 samples were collected, comprising 42 samples from conventional and 22 and 26 from Biolure and Biodelear, respectively. Based on the obtained results, 39.1% of the analyzed samples gave positive determination for chlorpyrifos (Conventional, Biolure and Biodelear), 32.6% for deltamethrin (Conventional and Biolure).
Conclusions: The insecticides chlorpyrifos ethyl and deltamethrin were detected not only in the conventional plots as expected, but also in the Biodelear (chlorpyrifos ethyl) and Biolure plots. The detection of the insecticides in Biodelear and Biolure plots was attributed to previous use, considering that the experimental plan was established three months only before the sampling. It should be noted that the fungicide propamocarb, which was detected during the preparatory monitoring, was not found.

B1 action for the pilot scale area - confirmatory sampling: Following the first sampling regarding the B1 action in December 2015, a confirmatory sampling took place in January and March 2017.
Methodology: The samples were collected separately for each medfly management treatment, Biodelear, Biolure and Conventional.
Results: A total of 219 samples of citrus fruits were collected, prepared and analysed as reported above. 13.6% of the analyzed samples gave positive determination for **spirotetramat**, 12.3% for **deltamethrin** 5%, for **dimethomorph**, while **piperonyl butoxide** was detected in only one sample.

Figure B1- 4. *Pesticides detected (number of positive samples) in a total of 219 citrus fruits collected.*

Conclusions: The completion of sample preparation and analysis showed measurable residues of 4 active compounds in citrus fruits. Furthermore, no detectable residues of plant protection products or plant growth regulators were determined in the Biodelear/ Biolure treated plots. It is worth noting that chlorpyrifos was not detected at the confirmatory sampling, despite the fact that it had been detected at the first sampling.

Assessment of Soil quality (SSDA)

Methodology: One hundred and six (106) soil samples were taken from seven different orchards and in two different soil depths 0-30 and 30-60 cm (4th sampling; 1-3 March, 2017). The above-mentioned soil samples were taken from citrus orchard plots that were used as Biodelear, Biolure, Conventional and Control treatments. Late in 2017 and early 2018, as the results of all previous soil chemical and physical properties analysis were available, SSDA decided to conduct an additional sampling in the orchards in order to cross-check the high values of concentrations above limits, especially in Cu, Zn, Cl^{-1}, SO_4^{-2} and Mg. For the additional sampling 98 soil samples were taken from seven different orchards and in two different soil depths 0-30 and 30-60 cm (6th sampling; 29 January - 1 February, 2018).
Samples were carefully stored, preserved and transported to the laboratory. Chemical and physical analyses were carried out following standard soil analysis methods.
Results: For the 4^{th} and 6^{th} sampling periods, it can be concluded that the soil samples were far below the threshold level for salinity, exchangeable Ca ranged within normal range and concentrations of exchangeable K in experimental sites were below threshold limits. Exchangeable Mg in all sites was well above the threshold value. This can be attributed to high concentrations of Mg ions in irrigation water indicating intrusion of seawater in aquifer. High P levels were recorded in one orchard site mainly due to Olive Mill Waste (OMW) that was disposed of regularly for many years, as well as the placement of poultry manure. Total N was lower than the threshold value. High surface soil concentrations of total N were registered in the same orchard mentioned above probably due to OMW disposal and poultry manure as well. Soils in experimental sites and in particular surface soils were rich in organic matter. Concentrations of NH_4^{+1} in pilot fields were below threshold values. NO_3^{-1} concentrations were well above the threshold levels in all sites and cannot be attributed to medfly control practices. Probably nitrates are relatively higher in the areas that agricultural activities (nitrogen fertilization and manures) are taking place. Sulfate concentrations were well above the threshold levels may be due to the use of sulfate base fertilizers. In fact, soil pH in study sites is characterized as alkaline and the use of sulfate fertilizers is a common practice. Therefore, water soluble Cl^{-1} in some orchards was found at high levels probably due to the bad quality of irrigation water that was used in these pilot fields. The presence of Cl^{-1} usually reveals seawater

intrusion into the aquifer. Values of extractable Mn were below threshold value of 50 mg kg^{-1}. Extractable Cu in most pilot fields varies in high levels indicating the use of copper-containing fungicidal sprays. The contamination of agricultural soils with Cu-based pesticides presents a major environmental concern. The extensive use of fungicides can potentially pose a risk to the environment, particularly if residues persist in the soil or migrate off-site and enter waterways. Differences among management practices within each pilot site may have been masked by the high contents of soil Cu because of the extensive use of copper-based fungicides. Data in most of the experimental sites have values of latent Fe below threshold values. Concentrations of extractable Zn were close or above the upper thresholds indicating pollution effects caused by fertilizers and pesticides.

Conclusions: The results of the soil samples that have been collected during this Action confirmed that the orchards have concentrations above limits in Cu, Zn, Cl^{-1}, SO_4^{-2} and Mg, a fact that is affecting negatively the soil quality. In order to examine the reasons for these high concentrations, we decided to evaluate the quality of irrigation water in the pilot fields.

Problems

1. The performance of ANEL McPhail-type traps during 2015 was not satisfactory. A different trap type (the International Pheromone McPhail Traps [IPMT]), was used in 2016 to address this issue.
2. To cross-check the high values of concentrations above limits, especially in Cu, Zn, Cl^{-1}, SO_4^{-2} and Mg, SSDA conducted an extra sampling (6^{th} sampling) in the orchards of the 4^{th} sampling, in order to cross-check the high values of concentrations above limits.

Evaluation

All objectives of the action B1 have been achieved. Citrus fruit infestation rates in plots treated with mass trapping with Biodelear were similar to conventionally treated plots and those of plots treated with mass-trapping with Biolure and much lower than control non-treated plots. Hence, mass trapping with Biodelear can reduce medfly population densities and achieve rates of medfly management similar to well established practices. In addition, an IS based on mass trapping with Biodelear was developed. This was an important element for the implementation of the action B2. During the course of B1, we gathered a wealth of data (substantially more than envisioned in the Grant Agreement) demonstrating that mass trapping with Biodelear has no negative effect on ground arthropod diversity and abundance. The thorough pesticide residues analysis revealed that mass trapping treated plots were free from insecticide in contrast to conventionally treated ones. Soil and water analysis demonstrated general issues (high concentrations of Cu, Mg, Zn Cl^{-1} and SO_4^{-2} in all plots) for the area of Kampos Chios, which however, are not affected by the medfly management practices tested. The above data support the adoption of mass trapping with Biodelear at local scale.

Indicators of progress

The indicators of progress foreseen in the proposal were:

- *Checking and counting on the male/female population of the med flies as well as counting and identifying any non-target insects attracted by the innovative attractant Biodelear and by commercial products (Biolure and Capilure) on all the traps (90 IP McPhail traps for*

Biodelear, 90 IP McPhail traps for Biolure and 18 traps for Capilure) (For the year 2015 during the end of the December of 2015 to January of 2016 and for the year 2016 during the end of the December of 2016 to January of 2017 will be assessed the efficacy of the Biodelear food attractant in comparison to the commercial Biolure food attractant and to the previous conventional ways of applications (bait cover sprays).

Following the revised experimental plan given above, the original medfly fly population monitoring was modified. The new monitoring system was substantially more demanding and labor intensive, however, it provides scientifically sound data. Seventy trapping stations randomly distributed within the experimental plot, consisting of one McPhail and one Jackson trap each and placed ca 15m apart, were established in all 14 experimental plots for monitoring medfly population throughout the action B1. The above trapping stations provided a robust mean for a thorough analysis of medfly population trends in the four plots established. The experimental design adopted allowed the statistical evaluation of the results. The obtained data are given as graphs and tables. In total close to 5,040 checks of traps have been conducted during this action and a total of 102,933 male and 35,606 female medflies have been recorded and identified in the trap stations. Likewise, about 26,995 non-target insects were recorded.

- *Assessment of citrus crops infestation (For the year 2015 the end of the December of 2015 and January of 2016 will be assessed the citrus crops infestation using the Biodelear food attractant in comparison to the commercial Biolure food attractant.*

The infestation of citrus was assessed in December 2015 and November 2016 in the plots treated with mass trapping with Biodelear, mass trapping with Biolure, conventional control approaches and non-treated plots. Both sweet oranges and mandarin oranges have been collected, transferred to the laboratory and thoroughly examined.

- *Collection and analysis of 50+50 soil samples (2000 analyses) Collections of soil samples for the year 2015 from December of 2015 to April of 2016 and for the year 2016 from December of 2016 to April of 2017.*

One hundred and six (106) soil samples were taken from seven different orchards and in two different soil depths 0-30 and 30-60 cm (4th sampling; 1-3 March 2017). An extra sampling of 98 soil samples was conducted 29 January - 1 February 2018).

- *Checking and measuring the biological diversity for the whole fruiting seasons for the two years of the implementation of the protection of the citrus orchards from the med fly insects.*

A total of 70 pitfall traps have been established and checked 2 times in 2015 and 4 times in 2016. A total of 25,750 arthropods have been collected, transferred to laboratory and identified. Sampling in 2016 covered the whole season, while in 2015 focused in the ripening period of citrus fruits (October - December). It should be noted that the establishment of B1 action took place in the middle of June 2015. Two measurements of arboreal arthropod diversity were taken during 2016 including a preliminary trial, since some methodological issues were encountered.

- *Completion of the IS before the beginning of B2 action.*

The Integrated Strategy was developed at the end of B1. The strategy was based on obtained results and included a spatial component that analyzed the dispersion of alternative, key medfly hosts. Auxiliary methods to reduce levels of medfly infestation such as fruit sanitation were included in IS. The IS was considered appropriate for the implementation of B2

- *Confirmatory tests to verify the absence of pesticides in the demonstration areas of the two citrus orchards (1 ha each) (at the end of the fruit season of 2016 Collection of 40+40 samples).*

The confirmatory test was applied in B1 for all the plots and no detectable pesticide residues were found in the Biodelear/Biolure treated plots. For the confirmatory test 158 fruit samples were analyzed.

Deliverables

- *Development of an Integrated Mediterranean Strategy (IMS) against med fly" and "Scientific, technical and financial analysis of the obtained results against anticipated environmental Benefits. Strengths, Weaknesses, Opportunities and Threats (SWOT) and Cost Benefit Analysis" for the action B1. The Deliverable was requested from the EU Commission by question 8 from the letter Ref. Ares(2016) 187117-13/01/2016"*
  The above deliverable was completed considering both the actions B1 & B2.

Milestones

- *"Identification of an appropriate Integrated Strategy for the protection against med fly at local level".*
  An Integrated Strategy was developed for the protection against med fly at local level, as was foreseen)

5.1.4 Action B2: Implementation of Innovative Med Traps at Real Scale

Foreseen start date: 1/06/2017 Actual start date:1/01/2017
Foreseen end date: 1/12/2018 Actual end date: 31/12/2018

Objectives: To implement the IS (developed during the pilot phase, B1) in a large ("real scale") area and the development of an Integrated Management Strategy (IMS) considering additional adjustments and modifications. To control medfly and reduce infestation rates of citrus fruit in a large area. To protect and/or restore the environment in the area of Kampos Chios. Emphasis has been placed (beside reducing medfly infestation on citrus fruit), on environmental quality indices, such as arthropod abundance and diversity, soil and water properties and pesticide residues in fruit. To conduct financial and technical evaluation of the IMS and identify whether restrictions in its adoption exist. To widely disseminate in all European Mediterranean countries the IMS and the outcome of the trials. To consider all different factors that affect the efficacy of the proposed methodology across the Mediterranean countries, i.e. climatic conditions, law restrictions, social acceptability. To develop scientific, technical and financial analyses of the obtained results against anticipated environmental benefits. To conduct Strengths, Weaknesses, Opportunities and Threats (SWOT) and Cost Benefits analysis. To extend the implemented actions from local scale to regional/national/Mediterranean scale.

Experimental farms and protocol

A total area of 12 ha citrus orchards was selected. All experimental orchards of the B1 action were included in B2 with addition of new ones (conventionally treated) in an attempt to establish a more robust experimental arena. By applying the same protocols developed in B1, during 2017 and 2018, we estimated both the ground and the arboreal arthropod abundance and

diversity as well as citrus fruit infestation rates in both mass trapping treated and non-treated orchards (Controls) (UTH). The medfly population (AUTH), the pesticides residues (BPI) and the soil quality (SSDA) were monitored throughout the two years of the implementation. Mass trapping was achieved by deploying 985 IPMT traps baited with Biodelear at a density of approximately 100 traps per ha. Mass trapping devices were set and functional throughout this two-year period, with regular checking and servicing. In early spring 2017 there was a thorough checking and servicing of the mass trapping devices, just before the commencement of B2 phase.

Medfly population monitoring (AUTH)

Methodology: To determine medfly population trends, three trapping systems were employed. IPMTs baited with Biodelear, IPMTs baited with Biolure and Jackson traps baited with trimedlure. To catch and retain flies, in both IPMTs a 10% propylene glycol in water solution was added. Trap check and service was performed weekly throughout the year. The number of male and female medflies and that of non-target insects captured were recorded. In addition, a sample of adult female medflies captured alive were maintained in wooden cages until they were dissected for counting the number and size of their ovaries in order to determine the level of reproductive maturity in adult female medflies captured in Biodelear treated orchards compared to the ones captured in the control orchards.

Results: Mass trapping with Biodelear during 2017 resulted in high suppression of medfly population, especially during the first period (23/05-05/09) which is critical in citrus fruit development. During the first period of citrus fruit development, from late May to early September, a significantly higher number of medflies was captured in the control farms compared to the treatment plots. During the second period (14/9-05/12), the average FTDs in control and treatment farms were at similar levels.

Mass trapping with Biodelear during 2018 suppressed the medfly population especially during the first period (16/04-08/08). FTDs throughout the first period were higher in the control compared to mass trapping plots. During the second period (16/08-15/11), FTDs captured in the control and treatment plots were significantly different in the McPhail traps. However, captures in Jackson traps were similar between control and mass trapping plots.

In both years (2017 and 2018) the number of non-target and beneficial insects (pinned and preserved for identification) were similar in control and treatment plots. The proportion of reproductively mature females captured were similar between control and mass trapping plots.

Conclusions: During B2, more than 10,000 checks of traps have been conducted and a total of 296,614 individual medflies have been recorded and identified in the trap stations. Likewise, about 148,135 non-target insects were recorded.
Mass trapping with Biodelear resulted in a notable population suppression of medflies, especially in the first period extending from late spring until the end of summer, when citrus fruit development is at a critical stage. Moreover, the number of non-target and beneficial

insects was at similar levels in the control and treatment orchards, which shows that Biodelear does not have a negative impact on non-target and beneficial insects.

Assessment of citrus fruit infestation rates and arthropod abundance and diversity (UTH)

Methodology: To assess the efficacy of mass trapping with Biodelear at real scale on the infestation rates of citrus fruits, we performed two fruit samplings at 29/11/2017 and 25/11/2018 when most of sweet orange and mandarin orange cultivars were ripe or ripening and therefore susceptible to medfly infestation. Details regarding fruit sampling and handling are given in Annex B1 of the Mid-term report.
Results: Infestation rates of sweet oranges in 2017 was significantly higher (24.58 %) in control orchards relative to mass trapping treated ones (9.84 %). Likewise, the total percentage of infested mandarin oranges was significantly higher in control orchards compared to mass trapping treated ones (21.26 and 2.32% for control and mass trapping orchards respectively).
Infestation rates of sweet oranges in 2018 was significantly higher (12.50%) in control orchards relative to mass trapping treated ones (5.48%). On the other hand, the total percentage of infested mandarin oranges was similar in control orchards compared to mass trapping treated ones (1.38 and 1.24% for control and mass trapping orchards respectively). It should be noted that infestation rates in mandarins for an unknown reason were very low in the area in 2018.
Conclusions: The implementation of the Biodelear Integrated Strategy (IS) at real scale resulted in very similar "protection" levels against medfly infestation rates. Therefore, we consider that the adoption and application of the mass trapping technique using the novel attractant Biodelear by means of the Integrated Strategy developed, can be as effective to control medfly as other methods including conventional sprayings.

Ground arthropod diversity and abundance

Methodology: Following the preparatory monitoring of the ground biodiversity, by utilizing the same pitfall trap grid (60 traps in the Biodelear treated orchards and 10 in the controls) we performed three samplings, at 21/6, 28/8 and 20/11 of 2017 using the methodology described above (Ground Biodiversity-Preparatory). Five additional samplings were also performed during 2018 at 12/3, 19/5, 25/7, 24/9 and 26/11. During the second sampling of 2018 (19/5) we added 19 and 8 pitfall traps into the Biodelear treated and control orchards respectively, in order to increase the data collection accuracy. This was done in an attempt to collect some rare ground beetles found in back 2017 in order to calculate more accurately the related biodiversity indexes.
Results: In total 13,583 arthropods were retrieved from the pitfall traps during the eight sampling dates. Data analysis revealed that only sampling date had a significant effect on captured arthropods, while medfly control approaches had not. The number of arthropods captured in control plots were similar to that in mass trapping treated plots.
Conclusions: The implementation of the Biodelear Integrated Strategy (IS) at real scale over two consecutive growing seasons in citrus orchards, appeared to restore the ground arthropod biodiversity up to the levels of the untreated orchards (controls). These results are very encouraging considering the limited time frame (2 years) within which this restoration attempt took place.

Arboreal arthropods sampling

Methodology: Fogging method was used for arboreal arthropod sampling, as described in B1 action. We performed three samplings in 2017 (20/6, 29/8 and 21/10) and three in 2018 (20/5, 24/7 and 25/9) respectively. Our sampling protocol involved the selection of one sweet orange tree of similar size in the middle of each experimental orchard. In the Biodelear treated orchards, we performed fogging applications in 9 out of the 10 selected orchards since one of our experimental plots was part of a resort property. As far as control orchards are concerned, we decided to perform two instead one fogging each time in the largest orchard (0.9 ha) since it was almost double in size relative to the other two.

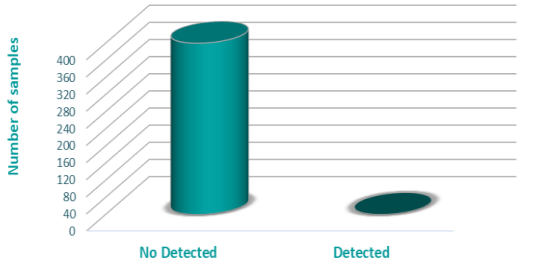
Results: In total 5,112 arthropods were collected during 2017 & 2018 samplings. Sampling date had a significant impact on the arboreal arthropod collections. The interaction between sampling date and treatment was also significant. On the other hand, treatment did not affect arthropods' abundance.

Conclusions: As in the case of the ground biodiversity, the implementation of the Biodelear Integrated Strategy (IS) at real scale over two consecutive growing seasons in citrus orchards resulted in almost identical levels of arboreal arthropod biodiversity relative to untreated orchards (controls).

Assessment of the Pesticide Residues after the implementation of the Real Scale areas (BPI).

B2 action for real scale areas: Following the preparatory monitoring of the real scale experimental orchards, confirmatory samplings were conducted in all citrus farms in December 2018 and February 2019. In total 400 samples were collected from both mass trapping and control plots. The completion of sample preparation and analysis (as already described), showed that no detectable residues of plant protection products or plant growth regulators were determined.

Figure B2-6. *Not detected pesticide residues in citrus fruit samples (n=400).*



Assessment of soil quality in mass trapping treated and control plots (SSDA)

Methodology: Seventy-six soil samples were taken from thirteen different orchards and in two different soil depths 0-30 and 30-60 cm (7th sampling; 25-29 June, 2018). Another 76 soil samples were taken in two different soil depths 0-30 and 30-60 cm from the same orchards (8th sampling; 28-31 January 2019). Chemical and physical analyses were carried out following standard soil analysis methods.

Results: For the 7th and 8th sampling periods, it can be concluded that the soil samples results were similar to the sampling periods 4 and 6 (See Action A2 pages 20-21). Concentrations were above threshold limits for Cu (>3 mg kg^{-1}), Zn (> 8,1 mg kg^{-1}), Cl^{-1} (>50 ppm), SO_4^{-2} (>30 ppm) and Mg (>2 cmol+ kg^{-1}) at the most samples.
Irrigation water quality: Although irrigation water analysis was not foreseen in the proposal it was decided to evaluate the quality of irrigation water in the pilot fields since excess accumulation of Cu, Zn, Mg and soluble ions were registered in soil. Salinity, specific ion toxicity, may have potential negative impacts or may cause hazards on soil quality and crop yield. Concentrations of NO_3^{-1} over 45-50 ppm are above threshold limits (FPS, 1999; FPTC 2008; EU Directive 98/83/EK), while concentrations of Cl^{-1} over 250-500 ppm are above threshold limits (FPS, 1999; FPTC 2008; EU Directive 98/83/EK; WHO 2008). High levels of soluble anions (NO_3^{-1}, Cl^{-1}, and SO_4^{-2}) were also registered in soil of citrus groves due to both inappropriate nutrient fertilization practices taken place in the past as well as to bad quality of irrigation water mainly in coastal regions.

Conclusions: Regarding the soil analysis results, the concentrations were above threshold limits for Cu, Zn, Cl^{-1}, SO_4^{-2} and Mg at the most samples and for this Action as well. The irrigation water analysis confirmed that the quality of the water used for the citrus orchards is low, as it has very high conductivity, especially high concentration of Cl^{-1} and high values of Mg. Concerning the high SO_4^{-2} values, can be explained from the over application of sulfur-based fertilizers of the local farmers. As for the excess Cu and Zn contents, they are attributed to the excessive use of fungicidal plant protection products in citrus plantations, highlighting the importance of appropriate applications of pest control products. Moreover, excess soil copper can lead to reduction in plant-available iron, resulting in chlorosis of citrus leaves and alteration of soil pH. Therefore, the use of Biodelear attractant will contribute to the control of medfly and in combination with a minimum data set of soil quality indicators will lead to the recovery of citrus ecosystem services.

Problems

1. No data on arboreal arthropod diversity were collected in spring because of late blooming and the presence of high bee population. However, three such samplings were conducted later in the season providing adequate data to evaluate possible effects of mass trapping on arboreal arthropod diversity and abundance.
2. High values of concentrations above limits, especially in Cu, Zn, Cl^{-1}, SO_4^{-2} and Mg were detected in both control and mass trapping treated plots. To cross-check this result, an extra sampling involving irrigation water was conducted.

Evaluation

All objectives of the action B2 have been achieved. Citrus fruit infestation rates in plots treated with mass trapping with Biodelear were lower than that of the non-managed orchards served as controls. The control of the medfly with the Biodelear attractant reduced the fruit infestation rates down to acceptable levels for both sweet and mandarin oranges. It seems that recovery of the ecosystem in the citrus orchards to natural levels (ground and arboreal biodiversity) has been accomplished (see also the Economic Analysis). Hence, mass trapping with Biodelear can reduce medfly population densities and achieve acceptable levels of medfly control in large

areas as well. It is expected that the longer, thorough application of the mass trapping strategy that we developed will result in progressive reduction of infestation rates and the medfly populations in the treated area. During the course of B2, we gathered a wealth of data (substantially more than envisioned in the proposal) demonstrating that application of the IS does not have any negative effects on ground arthropod diversity and abundance. Likewise, the no effects on arboreal arthropod diversity has been detected. The thorough pesticides residues analysis revealed that IS treated plots were free from insecticides. Soil and water analysis demonstrated general quality issues (high concentrations of Cu, Mg, Zn Cl^{-1} and SO_4^{-2} in all plots) for the area of Kampos Chios, which are not however related with the practices tested for the control of medfly, neither the conventional ones used. Based on the results obtained in B2 and IMS strategy was developed. An area wide adoption of the IMS strategy that will be supported by local producers' organizations will maximize its efficacy. The IMS that has been developed can be used in other areas and other countries. The emphasis is placed on citrus fruits; however, the IMS might be equally suitable for other crops especially for early ripening ones such as apricots and peaches. The above data support the adoption of the mass trapping with Biodelear at regional and international level in Mediterranean countries.
The nontoxic innovative Biodelear attractant can be a viable product ready to enter the market as was reported in the Business planning report and the Economic Analysis report of the action B2.

Indicators of progress

The indicators of progress foreseen in the proposal were:

- *Checking and counting on the male/female population of the med flies as well as counting and identifying any non-target insects attracted by the innovative attractant Biodelear (900 (IP) McPhail traps) and the Jackson traps with the Capilure attractant (90 Jackson traps) will be sufficient to control and monitor the med fly population in action B2 for the two years tests. (Depending on the population of the med fly from 24 services of the traps to 72 services of the traps – for 6 months of the fruiting season). For the year 2017 during the end of the December of 2017 to January of 2018 and for the year 2018 during the end of the December of 2018 to January of 2019 will be assessed the efficacy of the Biodelear food attractant in the large scale of the 10 ha of citrus orchard implementation from the protection of the med fly insects in comparison with the previous implementations on the 1 ha citrus orchard and to the previous conventional ways of applications (bait cover sprays).*

In 2017 and 2018 the detailed and thorough monitoring started on 4/6/2017 and 16/3/2018 respectively). For medfly population monitoring we established 83 trapping stations consisting of one IPMT with the attractant Biodelear, one IPMT with Biolure and one Jackson trap with trimedlure. Hence, a total of 249 traps were serviced at weekly intervals (except for winter season when checks were conducted every two weeks). The above sampling plan was the most appropriate for generating data reflecting medfly population dynamics in control and mass trapping treated plots. A total of 985 IPMT baited with Biodelear were deployed in 10 ha for mass trapping medfly adults. These traps were serviced regularly. The orchards used in B1 were included for comparison purposes. An economic analysis has also been concluded.

- *Assessment of citrus crops infestation in comparison with the previous implementations on the 1 ha citrus orchard and the conventional way of protection of the citrus crops (2, one for each year).*

Assessment of citrus crop infestation in action B2 in comparison to B1 was conducted. The Economic Analysis report provided information about the actions B1 and B2 in their implementation for the control of the medfly insects, in comparison with conventional practices of controlling the medfly insects and the commercial attractant Biolure.

- *Collection and analysis of 60 to 70 soil samples – 1600 analyses for the orchard in consideration. The collection of soil samples will take place at the end of the fruit season of the large scale at the end of December of 2018.*

For the action B2, 76 soil samples were taken from thirteen different orchards at 25-29 June, 2018 and another 76 soil samples were taken from the same orchards at 28-31 January 2019.

- *Assessment of the biological diversity on the soil and in the canopy of the citrus trees in the pilot field. For the whole fruiting seasons for the two years.*

The biological diversity on the soil and in the canopy of the citrus trees was assessed at real scale over two consecutive growing seasons in citrus orchards and the recovery of the ecosystem reached natural levels and the report was completed in January of 2019. Fourteen number of samples (sampling dates) were conducted including both mass trapping treated and control orchards. A total of 668 (ground biodiversity) and 78 (arboreal biodiversity) sampling units were collected transferred to the laboratory and all captured arthropods were identified.

- *Confirmatory tests to verify the absence of pesticides in the pilot field (At the end of the fruiting season of 2018 (December of 2018)).*

The confirmatory samplings were conducted in all citrus farms in December 2018 and February 2019 and the absence of pesticide residues was verified at the end of the real-scale implementation.

- *Completion of the scientific-technical – financial study of the proposed methodology. Extension to cover other Med countries (2018 December to January of 2019).*

The Scientific, technical-financial study of the proposed methodology was completed at the end of the year 2019 (report) and also the extension to cover other Med countries was completed at the end of the year 2019. The Scientific, technical and financial analysis of the obtained results against anticipated environmental Benefits. Strengths, Weaknesses, Opportunities and Threats (SWOT) and Cost Benefits Analysis from the implementation of the actions B1 and B2 was developed based on the results on the Business plan report and on the Economic Analysis report on the performance in the action B2, the Biodelear attractant and the IMS tool against the other methods of protection from the medfly insects such as the conventional way (pesticides and insecticides) and the use of the commercial attractant Biolure. The extension of the implemented actions from local scale to regional/national/Mediterranean scale based on the results from the above deliverable and the economic assessment of the attractant Biodelear from the BPR & EAR.

- *Completion of the Integrated Mediterranean Strategy. (2018 December to January of 2019).*

The Integrated Mediterranean Strategy was completed at the beginning of the year 2019. The IMS development was possible because the implemented IS in the action B1 provided enough information so in turn the IMS was implemented in the action B2 successfully.

Deliverables

- *"Development of an Integrated Mediterranean Strategy (IMS) against med fly".*

The report was delivered on 22/10/2019.

- *"Scientific, technical and financial analysis of the obtained results against anticipated environmental Benefits. Strengths, Weaknesses, Opportunities and Threats (SWOT) and Cost Benefits Analysis".*

The report was delivered at 25/10/2019. SWOT analysis was based on a thorough economic analysis of the project's achievements (integrated technology of pest management with the use of nontoxic means for the control of the medfly pest in large scale area).

- *"Extension of the implemented actions from local scale to regional/national/Mediterranean scale."*

The report was delivered at 25/10/2019.

Milestone

- *"Validation of the Integrated Strategy against med fly and extension from local to regional/national/Mediterranean scale."*

The developments of the IMS and the extension from local to regional/national/Mediterranean scale were achieved by the validation of the Integrated Strategy against medfly coupled with dissemination activities (see action D1 for details).

5.1.5 Action C1: Monitoring of project's environmental impact

Foreseen start date: 1/6/2014 *Actual start date: 1/6/2014*
Foreseen end date: 25/10/2019 *Actual end date: 25/10/2019*

Objectives: Monitoring of project's environmental impact by the assessment of biological diversity and of citrus fruit infestation rates in the demonstration areas (UTH) and by monitoring of the medfly population in the demonstration areas (AUTH), the pesticide residues assessments (BPI), plus the soil quality – chemical and physical properties (SSDA).

Fruit infestation rates at the end of Action B1

Based on the fruit infestation results, the project's aim for the Pilot Phase was dual: (a) the efficiency of mass trapping with Biodelear to be equal or better than that of the conventional way of medfly management, and (b) the fruit infestation percentage to be lower than 1% by using mass trapping with Biodelear.
Results: At the end of B1 (Dec. 2016), fruit sampling in Biodelear-treated orchards revealed that the infestation rates of sweet oranges and mandarin oranges were 7.22 ± 3.21% and 0.99 ± 0.99 %, respectively (see Action B1).
Conclusions: Our results suggest that mass trapping with Biodelear reached the threshold infestation level (<1%) only for mandarin oranges. However, the efficiency of mass trapping with Biodelear against medfly in citrus orchards was found to be similar to the conventional

methods (cover spays) and the mass trapping with Biolure for both sweet (7.23±4.1 and 11.70±5.8 % respectively) and mandarin oranges (4.94±3.5 and 1.85±1.8 % respectively) (see Action B1).

Fruit infestation rates at the end of Action B2

Including Control – non treated plots during the execution of large-scale test (B2) we managed to generate robust results regarding effects of the IS strategy on fruit infestation rates.
Results: The implementation of the IS strategy employing mass trapping with Biodelear as the main tool against adult medflies reduced fruit infestation rates by approximately 15 and 7% in sweet oranges in 2017 and 2018 respectively. Reduction of infestation rates was high in mandarin oranges in 2017 (approximately 20%). In 2018 infestation rates of mandarin oranges in both treated and control plots were very low and did allow the extraction of solid conclusions. The target of reducing fruit infestation below 1% was approached only in the case of mandarin oranges in 2017.
Conclusions: Overall the implementation of the Biodelear Integrated Strategy (IS) dramatically reduces citrus infestation rates. However, to reach negligible infestation levels longer term and an areas wide implementation might be required.

Biological diversity at the end of Action B1

Ground biodiversity
Methodology justification: Ground-dwelling beetles (Coleoptera: Carabidae), have been frequently used as bio-indicators for studying the effects of agricultural practices in agroecosystems, including Mediterranean perennial crops (Pizzolotto et al., 2018). The carabid beetles are used as bio-indicators in farming systems because:

a) they occur in all temperate agroecosystems due to the high number of geographically wide-ranging generalist (ubiquitous) species, and those occupying a wide range of habitats (eurytopic) (Rainio and Niemela et al., 2003),
b) they are predators of many pests, including aphids, lepidopterous larvae, and slugs,
c) they are easily captured and identified,
d) they respond relatively quickly to a wide variety of husbandry practices (Holland and Luff, 2000),
e) their diversity and species richness are expected to be greater where agrochemical inputs were reduced (Pearsall and Walde, 1995; Ellsbury et al., 1998; Labrie et al., 2003; Simon et al., 2010)

Consequently, we used Carabidae species as bio-indicators of ground diversity for assessing environmental impact of different medfly management methods in both pilot and real scale implementation. Accordingly, the Species Richness index and the other related diversity indices were calculated based on Carabidae species data.
According to the project aim, mass trapping with Biodelear is expected to have an increase of 10-20% in Species Richness (S) in the Biodelear-treated orchards compared to the conventional ones. To address the project aims we compared the ground biodiversity and ground abundance in Biodelear-treated orchards with that in conventional and Biolure-treated ones. To determine potential increase in Species Richness (S) after mass trapping with Biodelear compared to the conventional way of plant protection, we calculated the S only for Carabidae species for the two alternative medfly management methods. Besides Species Richness (S), biodiversity was

estimated by the soil arthropod abundance and the following diversity indices: Shannon-Wiener diversity index (H), Shimpson's index of dominance (D) and Shannon's evenness diversity index (E) (see details in Mid-term report). Arthropod abundance was estimated as the mean number of arthropods per trap per treatment.
Results: Ground-dwelling arthropod abundance and diversity indices estimated for Carabidae species in Biodelear-treated, Biolure-treated and conventional orchards. Species Richness index was estimated at 6.25 and 5.75 species for Biodelear-treated orchards and Conventional orchards respectively, indicating an increase of 8% in Biodelear-treated orchards. Data analysis revealed that soil arthropod abundance was significantly increased in Biodelear-treated orchards compared to conventional ones. In contrast, there was no significant difference in soil arthropod abundance between the two mass trapping methods. Diversity indices did not differ significantly between Biodelear-treated orchards and Biolure-treated or Conventional orchards.

Arboreal biodiversity
To estimate arboreal arthropod abundance in the Pilot phase of the program, arthropods specimens were collected after fogging one sweet orange tree.
Results: The mean number of arthropods/sweet orange tree in Biodelear-, Biolure- and Conventional-treated orchards was 49.0 ± 14.1, 31.3 ±14.7 and 27.6 ± 7.1 respectively. Even though arboreal arthropod abundance in Biodelear-treated orchards was substantially higher than that in conventional or Biolure-treated citrus orchards, there was no statistical difference among treatments, implying that mass trapping with Biodelear had a negligible impact on the arboreal abundance compared to conventional way of plant protection (incl. mass trapping with Biolure).
Conclusions: Mass trapping with Biodelear was found to enhance soil arthropod abundance in citrus orchards, compared to conventional methods. In addition, there was no difference in arthropod abundance between Biodelear-treated and Biolure-treated orchards. Mass trapping with Biodelear resulted also in an increase of 8% in Species Richness (S), compared to conventionally treated orchards. However, the basic dominant carabid fauna in the study area was found to be represented by generalist species, which are usually less sensitive to environmental factors than specialists (Rainio and Niemela et al., 2003). Moreover, because most Carabidae are univoltine it may take many years before consistent noticeable changes can be detected (Holland and Luff, 2000). Taken together, it is assumed that there was an increase in species richness during that short period of two years but the project target (10-20%) was not achieved. Overall, it is suggested that mass trapping with Biodelear did not have a negative impact on the soil arthropod community.

Biological diversity at the end of Action B2
At the end of B2 (Real scale), the project goal was the recovery of biological diversity in the ecosystem to the accepted natural levels that refer to those ones expected to have in the absence of pesticides and agrochemical in the citrus orchards. Given that organic orchards provide a diverse range of environmental conditions suitable for a wider diversity of soil and arboreal arthropod species (Holland and Luff, 2000; Miliczky et al., 2000), untreated orchards can serve as control orchards for assessing recovery process of the ecosystem biodiversity to the natural levels. In this context, we compared a) arboreal arthropod abundance, b) ground biodiversity and c) ground abundance in Biodelear-treated orchards and untreated citrus orchards in the Kampos area.

Moreover, it is well appreciated that arthropod population dynamics, particularly for carabid beetles, is strongly influenced by climatic conditions (Thiele 1977; Pajač Živković et al., 2016). To this end, the projected comparisons of the arthropod community status (diversity and abundance) between the zero-time of the project (i.e., initial conditions, 2014-2015) and at the end of the project (Action B2) were not conducted, because results were expected to be affected by the annual variation in temperature and humidity conditions. Comparisons were conducted for treatment and control throughout the same year (2016 for B1 and 2018 for B2). Instead, comparisons between Biodelear-treated and untreated (control) orchards were conducted i) at the end of the Pilot Scale implementation (Action B1, 2015-2016) and ii) at the end of the Real Scale implementation (Action B2, 2017-2018).

Arboreal biodiversity

Methodology: For details see Action B2. Arboreal arthropod abundance was measured as the mean number of arthropods/sweet orange trees in each treatment.

Results: At the end of Action B1, the arboreal arthropod abundance was estimated at 49.0 ± 14.1 and 91.8 ± 37.8 arthropods in Biodelear-treated and control (untreated) orchards, respectively. After applying mass trapping with Biodelear on a large scale, the arboreal arthropod abundance in citrus orchards increased at 65.1 ± 8.6 arthropods, being very close to that in controls (66.5 ± 7.8 arthropods). Data analysis revealed that arboreal arthropod abundance did not differ significantly between treatments both at the end of pilot phase and real scale.

Conclusions: At the end of B1, even though arboreal arthropod abundance for Biodelear-treated orchards was found to be lower than that of controls (untreated orchards), there was no statistical difference between them. At the end of the Real Scale implementation (B2 action), the natural balance of the ecosystem was not only maintained after mass trapping with Biodelear but was enhanced. Overall mass trapping with Biodelear had no negative effect on arboreal arthropod abundance and diversity.

Ground biodiversity

Methodology: For details see Action B2. Biodiversity was estimated by the soil arthropod abundance and the following diversity indices for Carabidae species: 1) Species Richness (S), Shannon-Wiener diversity index (H), Shimpson's index of dominance (D) and Shannon's evenness diversity index (E).

Results: At the end of Action B1, the ground-dwelling arthropod abundance was estimated at 80.6 ±7.3 and 60.5 ± 5.6 arthropods for control and Biodelear-treated orchards, respectively. Arthropod abundance in Control citrus orchards was significantly higher than that in Biodelear-treated ones. At the end of B2, soil arthropod abundance was estimated at approx. 20 arthropods/trap/sampling. No differences between Biodelear-treated and control orchards were detected.

Diversity indices for both treatments (untreated and Biodelear treated citrus orchards) at the end of Action B1 and B2 were estimated. Species richness index, based on the number of Carabidae species, was approximately 6.5 species for both Biodelear-treated and untreated citrus orchards. Shannon-Wiener diversity index ranged from 0.58 ± 0.1 to 1.22 ± 0.1 for Biodelear-treated orchards, while it was approx. 1 for the untreated (control) ones. Shimpson's index of dominance was about 0.65 in both treatments during the pilot trials while it ranged from 0.25 ± 0.1 to 0.40 ± 0.1 in the real scale experiments. Shannon's evenness diversity index

was estimated at about 0.5 for the control orchards and it ranged from 0.30 ± 0.1 to 0.67 ± 0.1 in Biodelear-treated ones. Data analysis revealed that there were no differences in values of diversity indices between control and Biodelear-treated orchards for both the pilot (B1) and real scale (B2) implementation.
Conclusions: Real scale mass trapping with Biodelear in citrus orchards decreases the differences in ground-dwelling arthropod abundance between Biodelear-treated and control orchards, implying that the recovery of biological diversity in the ecosystem to the accepted natural levels has been accomplished. The decrease in soil arthropod abundance in both control and Biodelear-treated orchards in Real Scale trials compared to those during the pilot phase can be explained by the annual variation in climatic conditions.
On the other hand, there were no differences in diversity indices between treatments (regardless of experimental set up). Even though the Shannon diversity and dominance indices as well as the Simpson diversity index are commonly used as biodiversity indicators, previous studies revealed that they are only weakly correlated with local species diversity in cultivated areas (Duelli and Obrist, 1998; Rainio and Niemelä, 2003).

Medfly population monitoring

Real scale mass trapping with Biodelear in citrus orchards decreases the pest population levels in experimental orchards compared to control. In all cases, adult medfly captures during the citrus growth season at the end of pilot and real scale implementations were higher in the control compared to the citrus orchards treated with Biodelear. Thus, mass trapping with Biodelear resulted in high population suppression of medflies especially during the critical first period for citrus production both in 2017 and 2018.
Moreover, the number of non-target and beneficial insects were at similar levels in the control and treatment orchards. Therefore, treatment with Biodelear does not have any negative effect on the populations of beneficial insects.

Pesticide residues in citrus fruits

During the whole duration of the project (A2, B1 and B2), more than 2000 citrus fruit samples were sampled in accordance with Directive 91972/2003/EC. All samples were analyzed with two multiresidue analytical methods capable of analyzing 334 pesticides and 18 plant growth regulators. As regards *action A2 for pilot scale areas*, 133 samples were collected (1st sampling: December 2014) and analyzed for a wide variety of pesticides and their metabolites and plant growth regulators. Pesticide residues were not found in any of the samples above the limit of quantification (LOQ). In July 2015, a total of 67 samples were collected from the same orchards. The samples were collected during the cultivation season in which insecticide application was conducted. Pesticide residues, such as the chlorpyrifos, deltamethrin and propamocarb, were found in many farms revealing an irregular practice of conventional control management against the medfly.
Additionally, during the monitoring of pesticide residues for *action A2 for real scale areas*, a total of 341 samples were collected at January – March 2017, from conventional and orchards (in which Biodelear or Biolure was used). Residues of pesticides, spirotetramat (74 samples), dimethomorph (22 samples), deltamethrin (27 samples), propamocarb (3 samples),

mepanypirim (1 samples), flumioxazine (6 samples) and azoxystrobin (4 samples). A synergistic to pesticide active substances compound, piperonyl butoxide was also detected (45 samples) shown that chemical pollution is not limited to active substances.
*During the action B1 action for the pilot scale area, t*he first sampling was conducted in December 2015. In total 92 samples were collected, comprising 42 samples from conventional and 22 and 26 from Biolure and Biodelear, respectively. Based on the obtained results, 39.1% of the analysed samples gave positive determination for chlorpyrifos (Conventional, Biolure and Biodelear), 32.6% for deltamethrin (Conventional and Biolure). An additional confirmatory sampling of 219 fruit samples took place in January and March 2017 and residues of spirotetramat, deltamethrin dimethomorph and piperonyl butoxide was observed.
Finally, in the implementation of action B2, a total 400 samples were collected in December 2018 and February 2019, from both mass trapping and control plots. The completion of sample preparation and analysis (as already described) showed that no detectable residues of plant protection products or plant growth regulators were determined.

Overall evaluation of soil quality
During the five-year period of the study (A2, B1 and B2), 550 soil samples were taken from the experimental orchards, in different soil depths 0-30 and 30-60 cm. Soil samples were analyzed for the following properties: texture, pH, total salts, saturation percentage, electrical conductivity, organic matter, total N, available P, exchangeable cations (K^+ Ca^{++}, Mg^{++}, Na^+), available Fe, Mn, Cu, Zn, water soluble ions Cl^{-1}, SO_4^{-2}, NO_3^{-1}, and NH_4^+.
Soil analysis during initial action A2 revealed that available Cu, Zn, water soluble Cl^{-1}, and SO_4^{-2} and exchangeable Mg were well above threshold levels and therefore it was decided statistical analysis to focus on the above soil parameters throughout the whole soil campaign. The high-water soluble SO_4^{-2} may be due to use of sulfate base fertilizers, while the Cl^{-1}, and exchangeable Mg concentrations may be attributed to bad quality of irrigation water that was used in these pilot fields. Concerning Cu and Zn, the high concentrations above threshold levels probably are related to the extensive use of fungicides in the citrus orchards of the area for years.

More specifically, soils in pilot fields and in particular surface soils (0-30 cm) were rich in organic matter (>3.4%). Unfortunately, this was mainly attributed to large quantities of fallen fruits the last years mainly due to inappropriate cultivation practices and low prices of the fruit.

Soil quality is negatively affected when Cu>3 mg kg^{-1}, Zn> 8.1 mg kg^{-1}, Cl^{-1}>50 mg kg^{-1}, SO_4^{-2}>30 mg kg^{-1} and Mg^{++}>2 $cmol_+$ kg^{-1} (Marx et al., 1999; MAFF, 1988; ILACO, 1985, Horneck et al., 2011). The results from the experimental orchards of Kampos Chios indicated that the values on these 5 parameters were well above threshold concentrations in soil surface (0-30cm) throughout the soil campaign and this was the main reason that it was decided to focus on them for the analysis of soil quality. Moreover, these 5 parameters have been evaluated at the deliverable "Risk Analysis Report". Soil samples with values above threshold levels in 4 or 5 of these parameters are characterized by severe risk, regarding the possibility that Biodelear or/and current cultivation practices induced residual effects or negative trends in soil quality. When we have 2-3 parameters above these levels, the risk is moderate and when we have 0-1 the risk is **mild**. The majority of the samples collected had values above these levels in 4 or 5

of these parameters, and the rest had values above these levels in 2 or 3 of these parameters (see deliverable "A Simplified Report for policy makers regarding the results of the environmental monitoring and the obtained results").

High concentrations of Cl^{-1}, SO_4^{-2} and Mg^{++} in soils were probably attributed to high concentrations of these ions in irrigation water and the use of sulfate base fertilizers due to soil pH alkalinity. The variability of soil pH in the pilot fields was high. Excess levels of DTPA-Zn and DTPA-Cu were also registered in surface soils (0-30cm) highlighting a possible intensive use of copper-containing fungicidal. In fact, the regular use of fungicides can potentially pose a risk to the environment, particularly if residues persist in the soil or migrate off-site and enter waterways. On the other hand, concentrations of the above metals in 30-60 cm were lower than in 0-30 cm, especially in DTPA-Zn results. This trend is closely related to soil texture and to high organic matter content. The soil texture of the orchards used for the study was CL (clay loam) or C (clay). The presence of clays and organic matter in the studied soils reduces substantially the toxic load during infiltration in deeper soil horizons avoiding therefore serious contamination of ground waters. However, if conventional crop protection practices continue to take place where citrus crops are considered among the main cultivations and soils are generally "light" in texture and poor in organic matter then surface and ground water contamination will occur.

Risk Analysis from the beginning of the project and at the end of the project

In the risk analysis from the beginning of the project and at the end of the project was evaluated by the estimated risk in all the indicators (action C1) by their assessment and was monitored in several steps of the project as to take the appropriate measures either for changing the scientific strategy or experimental design.

Efficacy

Risks: 1) *Medfly infestation of sweet oranges in untreated (control) orchards is low, and* 2) *Medfly infestation of mandarin oranges in untreated (control) orchards is low*

Risk assessment: The infestation rates of sweet oranges and mandarin oranges in untreated (organic) orchards in Kampos area remained above 1% during the Project, implying that medfly damage in the study area during the project (2015-2018) is quite high enough for assessing the efficacy of mass trapping with Biodelear.

Risks: 1) *The effectiveness of mass trapping with Biodelear is less than that of conventional sweet orange orchards,* 2) *The effectiveness of mass trapping with Biodelear is less than that of conventional mandarin orange orchards,* 3) *The effectiveness of mass trapping with Biodelear is less than that of mass trapping with Biolure in sweet orange orchards* and 4) *The effectiveness of mass trapping with Biodelear is less than that of mass trapping with Biolure in mandarin orange orchards* (Pilot Phase, B1)
Risk's assessment: No statistical differences in fruit infestation rates were found between medfly management treatments. However, sweet oranges are more sensitive to medfly infestation than mandarin oranges, irrespective of the management treatment. Therefore, the

risk of high infestation rates in Biodelear-treated orchards compared to conventional orchards or Biolure-treated orchards was low at the end of Pilot Phase (B1) for both sweet oranges and mandarin oranges. Therefore, the efficiency of mass trapping with Biodelear is equal than that of the conventional way of fruit fly control, including mass trapping with Biolure.

Risks: 1) *The effectiveness of mass trapping with Biodelear is low for sweet orange orchards*, and 2) *The effectiveness of mass trapping with Biodelear is low for mandarin orange orchards*. Risk's assessment: At the end of B1, the percentage of infestation was higher than 1% for sweet oranges, implying that the risk remains high. In contrast, infestation rates of mandarin oranges were approximately 1%, suggesting that the risk is "seldom likely" to occur. Besides the Project Scope, both risks were monitored during the Real Scale operation (B2) because of the high priority of the risks. Results revealed that the risks of low effectiveness of mass trapping with Biodelear in sweet orange and mandarin orange in Kampos is seldom or reasonably likely to occur. Reduction of fruit infestation to levels lower than 1% (commercial accepted level) after mass trapping with Biodelear has not achieved. Nevertheless, a substantial reduction in fruit infestation especially in mandarin oranges has been reported, highlighting the potential of Biodelear to control medfly.

Biodiversity

Arboreal arthropod

Risks: 1) Arboreal arthropod abundance for Biodelear-treated orchards is lower than that for conventional citrus orchards. 2) Arboreal arthropod abundance for Biodelear-treated orchards is lower than that for Biolure-treated citrus orchards (Pilot Phase, B1)

Risk assessment: At the end of B1, arboreal arthropod abundance in Biodelear-treated orchards is substantially higher than that in conventional orchards or Biolure-treated citrus orchards. Therefore, the impact although positive is rather negligible regarding effects of mass trapping with Biodelear on arthropod abundance of citrus trees, compared to conventional way of protection (incl. mass trapping with Biolure).

Risk: Arboreal arthropod abundance for Biodelear-treated orchards is lower than that for untreated (control) orchards (Real Scale, B2)

Risk assessment: At the end of B2, even though arboreal arthropod abundance for Biodelear-treated orchards found to be lower than that for untreated (control) orchards, there was no statistical differences between treatments. Similarly, the risk remains low at the end of project, implying that Biodelear traps are not expected to have a negative impact on arboreal arthropod abundance of citrus trees.

Ground arthropods

Risk*: Fruit fly control has not a negative impact on ground biodiversity of conventional citrus orchards.* (Preparatory Phase, A2)

Risk assessment: During the Preparatory Phase, as far as ground biodiversity in the study area is regarded, Species Richness was low in citrus orchards (1.75 Carabidae species). Therefore, the risk is negligible.

Risks: 1) *Ground biodiversity for Biodelear-treated orchards is lower than that for conventional orchards,* 2) *Ground biodiversity for Biodelear-treated orchards is lower than that for Biolure-treated orchards, and* 3) *There is not a satisfactory increase of Species richness index for Biodelear-treated orchards compared to conventional orchards* (Pilot Phase, B1)
Risk assessment: Based on Species Richness (S) index, the risk of a relative low ground biodiversity in Biodelear-treated orchards compared to conventional orchards or Biolure-treated orchards is low at the end of Pilot Phase. On the other hand, mass trapping with Biodelear results in an increase of 8% in Species Richness, compared to conventional orchards. That means that the risk of not achieving Project Scope (an increase of 10-20% in S) at the end of B2 is reasonably likely to occur. However, we accept that risk because our results revealed that the basic dominant carabid fauna in study area is represented by generalist species However, generalist Carabidae species are usually less sensitive to environmental factors than specialist species (Rainio and Niemela et al., 2003). Moreover, because Carabidae are univoltine it may take many years before consistent noticeable changes can be detected (Hollanf and Luff, 2000). Long-term evaluations of contiguous fields are needed to determine the effect of mass trapping with Biodelear on ground biodiversity.

Risks: 1) *Ground biodiversity for Biodelear-treated orchards is lower than that for untreated (control) orchards,* and 2) *Ground abundance in Biodelear-treated orchards is lower than that in untreated (control) orchards* (Real Scale, B2)
Risk assessment: During the Project, species richness, based on the number of Carabidae species, ranged from 4.5 to 6.25 and from 5.33 to 7.33 for Biodelear-treated orchards and control orchards, respectively. However, there was no statistical differences between treatments. Therefore, the risk is low at the end of the project. In addition, the soil arthropod abundance was higher in control orchards compared to Biodelear-treated orchards at the end of B1, implying that risk is likely to occur. However, there was no statistical differences between treatments at the end of B2, suggesting that the risk is low.

Pesticide residues
Although pesticide residues are possible to occur due the use of PPP according to their already studies residue behavior as estimated during the MRL setting procedures under Regulation 396/2005, based on the specific results obtained from the screening of the specific areas in Chios, the magnitude of harm in the specific areas from previous uses or environmental pollution in negligible, thus the risk of pesticide residues to reach a magnitude that will cause harm in near zero. The risk was significant during the cultivation period (July 2015 and January – March 2017) where residues were identified. However, form the last sampling and after the use of Biodelear in the specific orchards no residues were identified minimizing the risk.

Soil quality
In September 2014, during action A2, risk is maintained due to intensive cultivation practices in the past. The risk will be minimized in the coming years when considering adoption of a sustainable fertilization and crop protection strategy. It is of vital importance to adopt an integrated soil management system which can effectively improve soil quality in Mediterranean citrus groves and protect soil from further degradation. It is urgent to adapt to a design and

adjust the local farming systems for promoting soil conservation, as well as increase its fertility, while controlling the environmental impact.

Problems

A delay in completion of Risk Analysis was reported, due to the fact that some deliverables could not be completed on time, but the problem was rectified by the 5-month extension given from the EU Commission.

Evaluation

The methodologies were defined and the developed indicators were recorded and the results were very useful and interesting.

Indicators of progress

The indicators of progress foreseen in the proposal were:

- *Definition of monitoring methodology.*

The monitoring methodology was defined for the infestation of citrus fruits, the Biodiversity of ground and arboreal and the medfly population in the implementation areas, the pesticide residues and the soil quality – chemical and physical soil properties at the beginning of 2015

- *Implementation of indicators monitoring methodology and periodical recording of their values.*

The implementation of the indicators monitoring methodology and periodical recording of their values were delivered in 2015, 2016, 2017 and 2018.

- *Completion of two Risk Analyses (the first Risk analysis will be obtained at the end of the 2017 and the second at the end of the 2018).*

The completion of the risk analysis was completed and delivered for actions A2, B1, B2 at 25/10/2019.

Deliverables

- *"A simplified report for policy makers regarding the results of the environmental monitoring and the obtained results (in Greek and English)"*

It was created and distributed to stakeholders completed at 25/10/2019.

- *"Risk analysis in the beginning and by the completion of the project".*

Completed at 25/10/2019.

5.1.6 Action C2: Monitoring of project's social-economic impact

Foreseen start date: 1/4/2014 *Actual start date: 2/4/2014*
Foreseen end date: 30/4/2019 *Actual end date: 30/9/2019*

Objectives: To monitor the social and economic impact of the project across all main target groups (i.e., Greek and Mediterranean citrus producers and stakeholders). To assess the economic viability of the IMS. To determine whether the novel attractant Biodelear can become a marketable product.

Methodology:
Establishment of a network: In accordance with Action D1, a network was established between research centers, universities, farmers and their associations, regional and national authorities, representatives of the industry/market community, scientists from other, already existing, European networks and also with other LIFE projects related to pest and agricultural production management. To this goal two texts/ invitations were created in Greek and English language.
The invitation for participation to the network of LIFE- Biodelear was sent to farmers, citrus producers, stakeholders and regional/local Authorities. All the participants of the network were recorded accordingly (*excel file attached in the deliverable of the establishment of the network)* and continually received feedback for the dissemination actions (workshops, publications etc.), the Newsletters, the creation of the project's animation films and the survey with questionnaires.
Survey: A survey was conducted focusing on the social-economic impact of the project in an effort to monitor the indicators of C2. Three different questionnaires were created, for the farmers who participated in the project and farmers from Chios island (Q1), farmers in general (Q2) and stakeholders (Q3). All questionnaires consisted of semi -closed questions. Data analysis was based on descriptive statistics with the use of the Excel. A special platform was developed in the website of LIFE-Biodelear (https://www.biodelear.gr/index.php/el/to-ergo/erotimatologia, https://www.biodelear.gr/index.php/en/the-project/questionnaires) containing all three questionnaires. When a questionnaire was answered, all data were recorded automatically. After the completion of the survey all data were exported in an excel sheet. Simultaneously, each questionnaire was sent to the members of our network via e-mail and by post and was also sent via e-mail to all beneficiaries of the project who further forwarded it. In addition, questionnaires were distributed (hard copy) by the members of the project in all dissemination actions that took place during the survey (e.g. 18th Panhellenic Entomological Congress).
In total more than 600 questionnaires were distributed and 463 questionnaires were finally filled in. In particular and as regards farmers who participated in the project and farmers from Chios, out of 50 questionnaires distributed, 36 were answered. Similarly, 100 questionnaires were distributed in farmers in general and 76 of them were filled in, while regarding stakeholders 351 questionnaires were answered out of the 400 initially distributed.

Questionnaire 1 aimed at updating the knowledge and the satisfaction of farmers after the application of the mass trapping technique with the attractant Biodelear and its social/ environmental and economic importance, comparing with a similar questionnaire carried out in 2016 (preliminary survey already submitted). As observed from the data collected, 36 farmers completed the questionnaire, covering the whole range of age groups, with the majority (75%) of the respondents to be males and all permanent residents of Chios. 39% farmers have small citrus orchards (up to 0.5 ha) and that they have positive opinion about organic farming (58%). Their majority (75%) stated that they all cultivate citrus under conventional condition, with an exception of 4 farmers who cultivate organically. *Ceratitis capitata* is considered as the most (49%) important insect that damage citrus orchards. Additionally, 47% of the farmers stated that they do not apply plant protection products, while among those who apply sprays the 22% and 25% uses pyrethroids or organophosphates insecticides, respectively. Regarding insect mass trapping 83% of the farmers answered that they are aware of the technique, with 55% of

them to believe that it is an effective technique for the control of *Ceratitis capitata*. Regarding the attractant Biodelear, the respondents believe that it is a friendly and non-harmful method and also, they consider that using mass trapping technique will contribute to the reduction of producing cost in citrus orchards. Eighty-nine % of the farmers stated that they have noticed an increase in the fruit production following the application of Biodelear in their orchards simultaneously with an improvement in the quality of the citrus fruits produced. All farmers showed a positive perspective on the application of agricultural practices not harmful to the environment in order to control Medfly, while 75% of them believe that through the application of mass trapping technique with the attractant Biodelear, there will be an improvement in the living standards of their area. Conclusively, the majority of the respondents believe that mass trapping with attractant Biodelear will help to develop a better agriculture non-toxic to humans and the environment and all farmers (100%) stated that they will purchase Biodelear if it was commercially available, providing very promising results for the future steps of the attractant Biodelear.

Questionnaire 2 was distributed in farmers, members of our network or not, and aimed in the recording of their awareness regarding mass trapping with Biodelear, its social/ environmental and economic importance and finally their will to adopt the developed Integrated Mediterranean Strategy (IMS). It was filled in by 76 farmers, belonging to all group stages and educational levels and 70% of the respondents were male. From the total of the respondents, 57% said that they were not residents of Chios, while 43% said they were residents of Chios being employed as farmers (67%), as citrus producers (25%) and as agronomists (6%). In total 55% of the respondents stated a positive opinion regarding organic farming and 80% of the farmers agreed with the use of an organic method for pest control at their orchard avoiding environmental contamination. The majority of the farmers (40%) consider *Ceratitis capitata* (Medfly) as the major pest that damages citrus fruits, while 89% of the total answered that they are aware of mass trapping technique and that they have positive opinion (82%) about the control of *Ceratitis capitata* with the mass trapping technique but only a 35% apply mass trapping for control of medfly up to now. Regarding the attractant Biodelear, farmers believe that is an environmentally friendly and non-harmful method (61%). In addition, most respondents (67%) stated that they would be positive in using the attractant Biodelear and that they will adopt mass trapping with attractant Biodelear (96%). Farmers agreed that the use of Biodelear in citrus orchards will contribute to the establishment of a healthier environment and they also believe that the quality of citrus fruits will be improved. In other words, farmers consider that they will claim better commercial prices of their citrus fruits after the use of Biodelear. With the use attractant Biodelear they hope that it contributes to the improvement of their standards of living. In conclusion, the vast majority consider that the use attractant Biodelear will contribute to a healthy environment.

Questionnaire 3 was developed in Greek and English language and was distributed in stakeholders, policy makers, regional/ local authorities and citizens in general. It was filled in by a total of 351 participants comprising of 50% women and 50% men, of all age groups and with their majority (78%) of them to be of higher educational level. Only 42 stakeholders said that are permanent resident of Chios island. Furthermore, 27% of the respondents are employed at a research institute, 19% are agronomists while 20% said that they have other professional activities. Lower percentages were observed for employees in private sector or state/

government organizations (12% and 11%, respectively) while only 11% said that have self-employed. In general, stakeholders know about mass trapping and are favorable in the application of organic farming. They believe that the use of organic farming in neighboring fields will benefit the environment and their quality of life. Answering that mass trapping with the attractant Biodelear is an environmentally friendly and human-friendly method, they are positive in using the attractant Biodelear. Moreover, stakeholders believe that there will be an improvement in the quality and yield for orchards using it and a better commercial price will be achieved for the citrus products. This approach is believed to improve the overall quality of life in a rural area due to the reduction of the use of plant protection products after their replacement by the attractant. Above all, stakeholders have excellent or good opinions about the implementation of the project. It is considered very optimistic that in total 55% expressed a supportive view for the innovative attractant Biodelear, and that 59% expressed a positive point of view in the results of the project LIFE-Biodelear.

Results: This section is divided into two parts: (a) stakeholder's indicators & (b) economic analysis.

Indicators:

1. **Number of farmers from Greece that will become members of the network of action D1.** *Accepted value of participation: 100 members, Satisfactory: 150 members, Target: 250 members*

A total of 131 Greek farmers became member of the LIFE Biodelear network. This is a an accepted to satisfactory response.

2. **Number of Greek farmers that are willing to adopt the developed IMS.** *Considering that at the moment there are no farmers who are participating in such management scenarios a minimum participation of 20% of the farmers belonging to the above network (indicator 1), is considered accepted, a participation of 30% is considered satisfactory and the participation of 50% could be set as the target value.*

The analysis of the questionnaire revealed that 67% out of the 76 farmers participated in the survey are willing to adopt the IMS developed.

3. **Number of farmers from other Med countries that will become members of the network of action D1.** *Accepted value of participation: 150 members, Satisfactory: 200 members, Target: 350 members*

Only 3 farmers from other Mediterranean countries joined the LIFE Biodelear network. This was not a satisfactory outcome, despite the continuous effort (i.e. addressing personalized letters etc.) to reach farmers in Spain and Cyprus after the targeted events organized in both these countries. In an effort to expand this limited number and based on our "After LIFE Plan", publication of articles in magazines and newspapers of rural and environmental interest will continue to be published, while its representatives will present and distribute project material at events, exhibitions, conferences and meetings at EU level.

4. **Number of farmers from other Med countries that are willing to adopt the developed IMS.** *Considering that at the moment there are no farmers who are participating in such*

management scenarios a minimum participation of 20% of the farmers belonging to the above network (indicator 3), is considered accepted, a participation of 30% is considered satisfactory and the participation of 50% could be set as the target value.

This indicator is directly related to the previous indicator and therefore likewise not attained. However, it has to be mentioned that the manager of the largest citrus farm in Cyprus (5000 hectares) was very positive to use the IMS approach developed by the LIFE Biodelear project. He has been registered as a member of our network and is/was continuously informed about the news and the obtained results of the project.

5. **Numbers of stakeholders, policy makers and regional/local authorities from Med countries that will become members of the network of action D1.** *Accepted value of participation: 40 members, Satisfactory: 50 members, Target: 80 members*

A total of 130 stakeholders, policy makes, and regional local authorities officials became members of the LIFE Biodelear network. Their countries of origin were Greece, Cyprus, Tunisia, Spain, Portugal, Italy, Algeria, Morocco, Turkey and Israel, while there was a network member from Belgium as well. This is a very satisfactory response. Fifty-seven of them were agronomists and plant protection agents, 45 researchers and 29 officials regional, local authorities.

6. **Reduction of expenses at field level due to the reduction of pesticides use against med fly. Because of the adoption of the proposed IMS.** *It is anticipated that IMS could result to total expenses reduction by a minimum of 90% from the conventional way of protection (meaning using the cover spray technique with insecticides) if the application of the Biodelear in the same way is consistent for many years the reduction of expenses annually will be achievable therefore to reach the above target.*

In a nutshell the target of reaching the 90% reduction for the cost of protection from the medfly insects can be achieved by a combination of using the IMS tool, the Biodelear attractant in the mass trapping technique and with the added value of money of selling prices by the producers as organic citrus fruit products.

The assessments of the reduction of expenses at field level due to the reduction of pesticides used against medfly insects are based in the Economic Analysis report, and in that report was used the value of **0.6** euro per piece including all the accessories related to the ready use formulated Biodelear attractant which was estimated in the BP report. In the figure C2-1 is presented the example for the mandarins in the second year of implementation on the action B1 in the citrus orchards, provided us with three cases of protection from the medfly insects competing against each other with the Biodelear attractant to exhibit a compatible cost against the use of insecticides and much cheaper approach than the Biolure attractant. The values below are based on the following:

a) the loss of the cost production (euro per 0.1 ha per year) due to the infestation by the medfly insects of the mandarin fruits and
b) the cost of protection (euro per 0.1 ha per year) from the medfly insects.

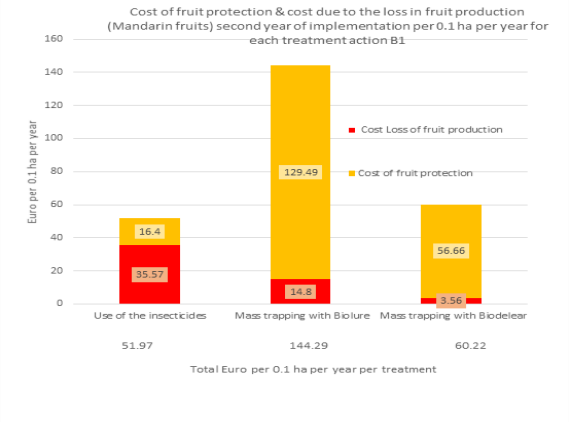
Under these circumstances and considering the fact crop yield of the mandarins was estimated to be about 1.8 tons per 0.1 ha per year

a) the loss of the cost production was estimated to be €35.57, €14.8 and €3.56 in respect to the insecticide, Biolure and Biodelear method of protections respectively,
b) the cost of protection was estimated to be €16.4, €129.49 and €56.66 for the insecticide, Biolure and Biodelear method of protections respectively.

In a similar way as above the crop yield of the oranges is estimated to be about 2.4 tons per 0.1 ha per year in the implemented area of the project and in both citrus fruit orchards the Biodelear attractant method exhibited a compatible overall cost against the use of insecticides and of course a much cheaper approach than the Biolure attractant method of protection. The implementation of the protection with the Biodelear method had demonstrated that the reduction of expenses annually to be consistent as was assessed in the EA report from the first year to the second year for the actions B1(pilot-small scale) and B2 (demonstration-large scale scale) in their two-year duration each. For both actions the second year had the better performance of lowering the infestation from the medfly insects meaning that the recovery of the ecosystem together with the IMS tool and the Biodelear attractant with the mass trapping technique worked synergistically to control the above insects. The extra prices of the organic citrus fruits in comparison to the nonorganic citrus fruits can be used as a profit benefit for the producers, thus i.e. with a relative small increase of 2% and 6% in the prices corresponding to the nonorganic mandarins and oranges respectively for the Biodelear attractant method of protection, are enough of reaching the total revenue as the production for the nonorganic citrus fruits (protection from medfly insects with insecticides), while the needed increases for the Biolure attractant method of protection from the medfly insects for the same reason are up to 13% and 23% for mandarins and oranges respectively. For the same reason, if there was an increase of 10% and 15% for the mandarins and the oranges respectively, the created net benefit profit would be used to compensate completely the cost of protection with Biodelear and the IMS tool to almost 100%, while for the Biolure attractant the proper increase would be up to 31.5% and 43.5% to reach the same target.

Conclusion: It is clear that in Integrated Mass Trapping Strategy for controlling the med-fly with the Biodelear attractant productivity increases significantly and, although we have relatively increased costs, the final net economic benefit is similar to the use of the insecticides but differs significantly from the use of Biolure. In addition, in both alternative methods (Biolure & Biodelear), the farmer is able, if he meets other requirements, to receive a certification for organic products, which can increase the selling price by 30-40%. However, the great advantage of the Biodelear attractant lies in the non-financial benefits created such as the recovery of the ecosystem and that has not any toxic effect to nature or to human health with satisfactory control of the medfly insects.
In addition, the analysis of the questionnaire revealed that 89% out of the Chios farmers who were actively involved in the project stated that, as they actually observed by the end of the project, a drastic reduction in the production cost is expected. Other Greek farmers (about 46%), also answered that they anticipated a reduction of expenses. Finally, stakeholders and citizens (about 61%) also expressed their positive view regarding the reduction of production costs.

Figure C2-1. *Comparison of the cost of citrus orchard protection methods for the medfly insects and the cost due to the loss in fruit production (Mandarin fruits) in the second year of implementation in the action B1*



In addition, the analysis of the questionnaire revealed that 89% out of the Chios farmers who were actively involved in the project stated that, as they actually observed by the end of the project, a drastic reduction in the production cost is expected. Other Greek farmers (about 46%), also answered that they anticipated a reduction of expenses. Finally, stakeholders and citizens (about 61%) also expressed their positive view regarding the reduction of production costs.

7. **Number of farmers, stakeholders, policy makers and regional/local authorities satisfied from the project outcomes.** *Accepted percentage of satisfied stakeholders: 80%; satisfactory: 90%; target: 100%.*

The analysis of the questionnaire revealed that 100% of the farmers who participated in the project were very satisfied with its results and that they would purchase the attractant Biodelear if it was commercially available. Farmers in general (67%) answered that they were positive in using the attractant Biodelear, while stakeholders and citizens when they were asked to rate the application of the project, at a percentage of 55% expressed a supportive view for the innovative attractant Biodelear.

Additionally, two of the farmers involved in the project, when interviewed by the agro-newspaper "Ypaithros Chora" stated that:

Mr. Vasileios Drosinos: "*As far as harvest is concerned, things are much better, just to look at the flies caught in traps"* and

Mr. Ioannis Anastasakis: *"I cultivate 1 ha of oranges and mandarins in which I've placed traps as well as 0.8 ha in which I have not placed traps at all. The difference in the infestation of the fruits was very great".*

Finally, the research team of LIFE-Biodelear has accepted e-mails from farmers who wanted to be provided with the attractant Biodelear in order to use it.

8. **Improvement of crop yield and quality at the three pilot farms.** *An increase in yield really will depend from what the kind of citrus orchard will be selected so an expected value*

greater than the yield obtained with the use of the conventional way of protection (meaning using the cover spray technique with insecticides) will be considered satisfactory.

The farmers who participated in the program answered questions so that this indicator could be evaluated. The results in the four years implementation of the control of the medfly with the Biodelear attractant and the IMS tool resulted in reduction in the infestation with respect to the conventional approach (cover sprays with insecticides) and of course recovery of the ecosystem to natural levels as the evaluation of the indicators for arboreal and ground biodiversity was assessed. The crop yield in both the mandarins and the oranges with the Biodelear attractant method and the IMS tool was substantially increased by decreasing the crop infestation against the Biolure attractant method and insecticide use as was demonstrated in the EA report and mentioned above. Therefore, the protection from the medfly by Biodelear is considered adequate and satisfactory. Eighty nine percent of the farmers involved in the project stated that they have noticed an increase in their fruit production following the application of Biodelear in their orchards while 78% of the farmers expect an increase in the price of their citrus production in the market.

An increase in yield was possible for all the citrus orchards more or less with respect to the control of the medfly with the Biodelear attractant compared to the conventional way of protection, and this was assessed by the evaluation of the infestation rates between the treatments control (citrus orchards without any control actions B1 & B2), protection from the medfly insects by insecticides action B1, protection from the medfly insects by the Biolure attractant action B1, and finally protection from the medfly insects by the Biodelear attractants actions B1 & B2.

Therefore, the protection from the medfly by Biodelear is considered adequate. Eighty nine percent of the farmers involved in the project stated that they have noticed an increase in their fruit production following the application of Biodelear in their orchards while 78% of the farmers expect an increase in the price of their citrus production in the market.

9. **Number of farmers/citizens who, because of the project, will become aware of environmental issues associated with pesticides use.** *A minimum number of 500 farmers/citizens is considered satisfactory. For this indicator, an initial recording of the people awareness on the specific environmental issue at the pilot area will be performed by distributing specific questionnaires. Such questionnaires will be distributed at least two more times during the project lifetime*

A survey was conducted focusing on the social-economic impact of the project in an effort to monitor the indicators related to Action C.2, following the methodology described above. The survey was carried out from March 2019 until October 2019. A similar survey with questionnaires was carried out in 2016 having only to farmers of Chios as the target group. The responses to the questionnaires were unique and anonymous. A total of 463 questionnaires were filled in (36 from farmers who participated in the project and farmers from Chios, 76 from farmers in general and 351 from stakeholders).

Results of the survey: The detailed results of the survey are reported.

The analysis of the questionnaires (Q1, Q2 and Q3) showed that farmers are who are ready to implement mass trapping with the new innovative attractant Biodelear to manage medfly, It seems that they have understood that the attractant Biodelear contributes to the reduction of infections rates, assists in the increase of production due to the minimization of infections and contributes to the increase of biodiversity. Finally, it was observed that there is a trend of farmers to adopt practices, which are friendly to the environment and human health. This shall result in plant commodities free of pesticide residues.

Problems

In general, no problems were encountered during the implementation of this action, nevertheless, in the Indicator number 3 'Number of farmers from other Med countries that will become members of the network of action D1' although actions were taken to attract farmers from other Mediterranean countries (attending conferences, organizing a workshop in Cyprus, etc.), it was not possible to obtain additional members.

Evaluation

Monitoring of socio-economic impacts on main target groups, such as farmers and local authorities was initiated in 2016. Suitable indicators have been specifically identified and methodologies for data collection have been discussed. In general, were obtained reliable information from the implementation of the project targets of its socio-economic impact.

Indicators of progress

The indicators of progress foreseen in the proposal were:

- Establishment of suitable socio-economic indicators.)

Initial suitable socio-economic indicators have been established

- Definition of monitoring methodology.

Monitoring methodology has been fully defined.

- Implementation of indicators monitoring methodology and periodical recording of their values.

Implementation of indicators monitoring methodology and periodical recording of their values were fully carried out as planned.

- Development and distribution of questionnaires.

Three questionnaires have been developed by taking into account the response of different target audiences and distributed in a series covering the period 2016-2019.

- Number of web site visitors. Satisfactory 3000 visits/year; target 5000 visits/year.

The number of web site visitors surpassed the target of the 5000 visits/year. Specifically, year 2014-2015, 30589 visits; year 2016, 11820 visits; year 2017, 14527 visits; year 2018, 11622 and year 2019, 4620. A total of 73178 visits in the five years duration of the project.

Deliverables

- "A Simplified Report for policy makers (in Greek and in English)"

It was delivered at 25/10/2019.

- "Effect of IMS on the socio-economic status of Mediterranean countries (Report)"

It was delivered at 25/10/2019.

- "The Business planning for the Biodelear attractant".

This deliverable was requested by EU Ref. Ares (2018) 1644471-26/03/2018.

5.2 Dissemination Actions

5.2.1 Objectives

The Dissemination strategy began early on the project and lasted until the entire project lifetime. The dissemination strategy included a significant set of actions to disseminate the results of the project so that the knowledge gained will be actively communicated to those stakeholders in Mediterranean area that may best use of it and apply the lessons learnt from the project.

5.2.2 Dissemination: overview per activity

- *Benaki Phytopathological Institute (BPI) was responsible for the dissemination of the project.* All beneficiaries under the guidance of the Dissemination Committee performed significant dissemination actions.
- The dissemination strategy followed was as planned and the objectives were reached, gaining a positive and promising reaction feedback from all the target groups involved (farmers, students, agronomists, policy makers, stakeholders).

- The dissemination strategy was built upon three basic pillars:

1. Development of a functional and attractive project web site,
2. Establishment of a network between research centers, universities, farmers and their associations, regional and national authorities, representative of industry/market community etc.
3. Dissemination of project's achievements to stakeholders, scientists, policy makers and mainly to producers of citrus orchards and other cultivated crops affected by the medfly insects in Greece and other Mediterranean countries.

- LIFE Biodelear is presented through a modern, attractive and very rich **web-site** (**http://www.biodelear.gr**). The construction of the website was undertaken by the team of SSDA-BPI and was delivered according to that timetable (in January 2015).

All beneficiaries participated in website enrichment through the coordinator SSDA and BPI by sending data and information to be uploaded. Two versions of the website are available, i.e. English and Greek. There are many pages that a visitor can visit and be informed regarding the project progress, aims, activities and expected results (news page, newsletters, photo galleries studies, research articles, maps, statistical data, and many others). Inside its menu a **project diary** was created, in which all beneficiaries reported their activities and progress. Additionally, in the homepage of the website there is also a link to its corresponding Facebook page.

- The website has also an **web inventory** which contains data and information regarding control of the medfly with innovative attractant Biodelear with the mass trapping technique in the citrus orchards in the island of Chios in comparison with conventional techniques of controlling the medfly insects, the impact to the ecosystem, soil status of the citrus orchards and pesticide residues in the citrus fruits before and after the implementation of mass trapping technique and related issues, and others.

During the five years of the project, there were many visitors from all over the world as it can be seen in annexes. The total number of visitors at project's end is up to 73178 visitors from Greece, Spain, Cyprus and Mexico.

- A **network** was established between research centers, universities, farmers and their associations, regional and national authorities, representatives of industry/market community, scientists from other, already existing European networks and also with other relative LIFE projects relative to pest and agricultural production management. At a first approach, all stakeholders and growers who participated in the two workshops organized in Chios, the participants in the 3rd Workshop of LIFE-Biodelear in Nafplio-Greece (April 2017) and in the "IOBC WPRS Meeting on Citrus Pests, Diseases and Weeds" in Valencia-Spain were included in our network. Secondly, and to the goal of broadening the project's network, a text/ invitation was created in Greek and English language and sent to all that maybe interested. The members of the network were recorded and had feedback of the project's achievements throughout its duration.

In accordance with the Grant Agreement, an intense and fruitful dissemination strategy was accomplished throughout the duration of the project.

- Starting with the creation of the project's logo,
- The website of LIFE-Biodelear (www.biodelear.gr) was afterwards developed, launched and maintained.
- Seven information signs were placed in the citrus orchards involved in the project, displaying full information of beneficiaries, funding, duration and a short description of the project.

As regards the dissemination of project's progress and results, an extensive informative strategy was implemented, with

- Four workshops in Greece (3 in Chios and 1 in Nafplio),
- One two-day workshop in Cyprus
- LIFE-BIODELEAR participated with a dedicated section in the IOBC/WPRS Meeting on Citrus Pests, Diseases and Weeds in Spain.
- The project was presented in 6 scientific conferences (1 national, 2 European and 3 international),
- The implementation and some of the obtained results were published in scientific magazines (2 in the Integrated Protection of Fruit Crops IOBC-WPRS Bulletin and 1 in DEMETER magazine).
- The implementation and some of the obtained results were published in the press (2 in the e-newspaper "Ypaithros").
- LIFE-Biodelear developed the main project's leaflets (2200 copies) in November 2015

- LIFE-Biodelear circulated 7 Newsletters (#1 550 gr; #2 660 en and 660 gr; #3 550 en and 550 gr; #4 580 en and 580 gr; #5 450 en and 450 gr; #6 300 en and 300 gr and #7 200 en and 200 gr) which were distributed all the dissemination actions (workshops, conferences, exhibitions) in which the research team of Biodelear participated and 10% of them were distributed in students and visitors in the premises of all beneficiaries.
- LIFE-Biodelear circulated 3 informative banners.

All printed documents were generated in Greek and English languages and the LIFE logo, the logo of the project and the corresponding of the beneficiaries (coordinating and associated) were reported.

- Two additional leaflets were created specified in the use of the McPhail and Jackson trap and in the mass trapping with the attractant Biodelear. Those leaflets were printed in 50 copies each only in Greek language and were distributed to all the participants of the closing ceremony in Chios (2019).
- The coordinator of the project Dr V. Mavraganis as well as other members of the research team gave interviews in press (2 in radio stations, 3 in TV and 2 in newspapers), while
- The objectives and the development of the project were presented in the e-newspapers and websites of rural interest (in a total of 14 presentations).

The research team of LIFE-BIODELEAR visited

- The local authorities of Chios (at the beginning and in the end of the project, 4 visits).
- Research institutes (1 presentation).
- Stakeholders (1 presentation) dealing with mass trapping.
- Presented the project in schools (1 presentation) and universities (2 presentations).

Moreover,

- It was presented in exhibitions (Agrotica (1), Florist Exhibition in Kifissia (2), Kipotechnika (1))
- Info days (Green Fund's and Greek LIFE Task Force workshop (2)).
- Promotional material of hats (500 pieces), bags (450 pieces), notepads (300 pieces) and pens (800 pieces) were designed and created with the logo of BIODELEAR and LIFE was printed in all.
- The project's video was created in December 2019 in English and Greek languages and in two versions, a long one with duration of 2 minutes and 4 seconds and a short version of 30 seconds. It was sent via email to more than 400 participants of the Life-Biodelear network, posted in the home page of the website and Facebook page and presented in all beneficiaries' institutes, in stakeholders. Additionally, during the 18th Panhellenic Entomological Congress, a TV was occupied exclusively for the reproduction of our video all day long.

Finally, after the completion of the project,

- The Layman's Report was created in both Greek and English languages.
- After-LIFE Dissemination Plan was created in english.
- A simplified guidebook to farmers was prepared in English, Italian, Spanish, Greek and French, as well as

- The simplified reports for policy makers regarding the results of the environmental and social economic monitoring and the obtained results, in both Greek and English languages.

Problems

There were no particular problems during the design and implementation of the dissemination strategy, due to the well planned and concerted actions considered and the cooperation of project beneficiaries.

Evaluation

The dissemination activities were very satisfactory and effective. There was a significant interest in the project, also from other countries and many visitors to the website from all over the world.
The events organized in the island of Chios, in the Nafplio of Argolis a major citrus producing region in Greece, in the "IOBC WPRS Meeting on Citrus Pests, Diseases and Weeds" in Valencia-Spain a major citrus producing region in Spain and in Cyprus University of Technology in Limassol of Cyprus plus the personal contacts and the dissemination material distributed during the events, raised the awareness of many stakeholders. It is indicative that the visitors of the web site were more than 73,000 until October 2019.
In that framework the beneficiaries are working to apply for a new project implementing the control of the medfly to larger citrus areas in Greece.

Indicators of progress:

- Web site completion (design and first version will be completed on month 6, but it will be continuously updated until the end of the project)

The web site was completed and was continuously updated until the end of the project.

- Construction and erection of three metallic boards with LIFE logo at pilot areas (the first two metallic boards with LIFE logo will be placed on the site of the two pilot areas by the end of the 2014 and the third metallic board with LIFE logo by the end of 2016 for the large-scale area)

The construction and erection of seven metallic boards (3 large and 4 smaller) was implemented.

- Number of events organized during the project (7) (will be completed during the last two years of the project)

The project organized 6 events in total; four in Greece and two in other Mediterranean countries.

- Number, frequency geographic distribution of web site visitors (at least 3000 visitors/year).

More than 73,000 visitors during the five years of the project.

- Volume of the dissemination material that will be produced and distributed (as defined in the "expected results").

The project developed the main project's leaflets in November 2015 circulated 7 Newsletters and 3 informative banners. Two additional leaflets were created specified in the use of the

McPhail and Jackson trap and in the mass trapping with the attractant Biodelear. The Layman's Report and the After-Life Dissemination Plan, a simplified guidebook to farmers (in English, Italian, Spanish, Greek and French) as well as the simplified reports for policy makers regarding the results of the environmental and social economic monitoring and the obtained results were also created. Quantitative details are given in annexes

- Number of publications and national and international journals and announcements in international conferences (as defined in the "expected results").
- Number of network' s members (as defined in C2 action)

Network' s members have been recorded as defined in C2 action.

- Completion of the layman' s report

The layman' s report was completed.

Deliverables

- "Web site of the project"

It was delivered at 2015. **web-site (http://www.biodelear.gr)**

"Establishment of a network"

It was delivered at the end of the project. See Dissemination page 62

- "A simplified guidebook to farmers in English, Italian, Spanish, Greek and French"

It was delivered at 25/10/2019.

- "Layman's report"

It was delivered at 25/10/2019.

- "After-LIFE dissemination plan"

It was delivered at 25/10/2019.

Milestones

- "An active network between producers and their associations; scientific publications; edition of the "Integrated Mediterranean Strategy" and distribution to policy maker."

5.3 Project management and monitoring of the project progress

5.3.1 Action E1: Project Management

Foreseen start date: 01/06/2014 *Actual start date: 01/06/2014*
Foreseen end date: 1/06/2019 *Actual end date: 25/10/2019*

Status: Completed

Objectives: To ensure timely implementation of the project, good cooperation among Beneficiaries, quality assurance of the deliverables.

Project Management by SSDA

SSDA as the Coordinating Beneficiary with the coordinator himself has been in charge of providing effective management of the project and being the main link between the other beneficiaries and the Commission and the LIFE External Monitor Team. The coordinator and the management group of SSDA was in touch very often and meetings are held at least once a

month. All details regarding implementation of this Action are described in the section 4.1. Description of the management system of the Administrative part. In action E1 the Economic Analysis report and the added deliverable Business Planning report were delivered.

Table E1-1. *Milestones of the project.*

Name of the Milestone	Action	Status	Foreseen deadline	Fulfilled on
Determination of the optimum amount of the innovative attractant to be used during the demonstration	A1	Fulfilled	31/12/2015	31/12/2015
Identification of an appropriate Integrated Strategy for the protection against med fly at local level	B1	Fulfilled	31/12/2016	31/12/2016
Validation of the Integrated Strategy against med fly and extension from local to regional/ national /Mediterranean scale	B2	Fulfilled	31/12/2018	25/10/2019
An active network between producers and their associations; scientific publications; edition of the "Integrated Mediterranean Strategy" or "Integrated Pest Management Strategy" and distribution to policy makers	D1	Fulfilled	31/05/2019	25/10/2019

Table E1-2. *LIFE Biodelear reporting obligations. Initially and proposed submission days.*

Type of report	Foreseen Deadline	Actual Deadline	Actual Status
Inception Report	01/03/2015	01/06/2015	Submitted
Progress report	01/12/2015		PR was skipped due to the proximity to the MTR
Mid-term report	01/12/2016	30/12/2016	Submitted
Progress report	01/12/2017	25/04/2018	Submitted
Final report	01/05/2017	3/11/2020	Submitted

Indicators of progress:

- Submission of Inception Report, Annual and bi-annual Progress Reports, Mid-term Report, and Final Report.
  The inception report was submitted at 01/06/2015, the progress report at 1/12/2015 was skipped due to the proximity to the Mid-term report, the Mid-term report was submitted at 30/12/2016, the progress report at 25/04/2018 and the final report was submitted 3/11/2020.
- Completion of several key components of the project and the respective reports
  The completion of several key components of the project and the respective reports were delivered.

5.3.2 Action E2: Project Monitoring

Foreseen start date: 01/06/2014; Actual end date: 01/06/2014
Foreseen end date: 1/06/2019; Actual end date due to amendment:25/10/2019

Status: Completed

Objectives: *To measure and document the effective implementation of the project actions, attainment of expected results and assessment of environmental impact.*

Project Monitoring by SSDA

5.3.3 Objectives of the project

1. Implementation in real scale of an innovative, patented, environment friendly and low-cost food attractant (Biodelear) for the control of med fly, the attractant is non-toxic to humans, attracts selectively females med fly and virtually none of the non-targeted insects.
The objective was accomplished because during the course of B1& B2, by using the attractant (Biodelear) for the control of med fly, we gathered a wealth of data demonstrating that mass trapping with Biodelear has no negative effect on ground and arboreal arthropod diversity and abundance, on the contrary contributed to the recovery of the ecosystem in the citrus orchards in a sustainable way so that the control of the medfly to be attainable in both actions.

2. Elimination of insecticides use, which are toxic to humans and to the environment, decrease biological diversity by creating imbalances in soil, and make a plant more attractive to insects' pest.
The elimination of insecticides was accomplished because at the end of each action B1 & B2 the pesticide residues was found to be to zero thus the prerequisites of the recovery of the ecosystem to natural level was created by the use of the attractant Biodelear.

4. Development of a sound, integrated and environmentally sustainable technology to address Med fly in Mediterranean region, using as example the cultivation of citrus.
The development of a sound, integrated and environmentally sustainable technology to address Med fly in Mediterranean region, using as example the cultivation of citrus with the actions B1 & B2 was developed the IMS.

5. To render Mediterranean farming less dependent on pesticides-conformation of the EU policy: enhanced biological diversity (92/43 EEC) and Elimination of the residue levels of pesticides in or food and feed of plant and animal 2005/396/EC.
The above objective with the use of the Biodelear attractant in our endeavor was fulfilled with the above directives of the EU policy

6. In our project the maximum quantity of the 12 ha citrus orchards would be used and the amount of reduction of the insecticides would be of 100%
The above objective was delivered.

5.4 Analysis of long-term benefits

5.4.1 Visibility of project results

LIFE+BIODELEAR distinguishes the following results that will become apparent after a certain time period

· Positive acceptance by famers who participated in the project. The main accomplishments of the project have already received an initial acceptance by the farmers and authorities. After the positive effect in the local community and wide acceptance of Biodelear, a focused scaling-up strategy at national and Mediterranean levels has been developed.

· Biodelear has reduced the medfly infestation levels of both mandarin and orange crops. The reduction in infestation rates to less than 1% in the second year of implementation in some cases, such as for the mandarins, is within the target foreseen in the proposal

· Biodelear attractant promoted a substantial recovery of the citrus ecosystem in the implementation areas of the citrus orchards, selectively attracted medflies, and did not disturb the non-target insect populations in areas where mass trapping method used.

· An integrated pest management strategy was developed on a pilot scale to control the Mediterranean fruit fly without the use of insecticides by employing the mass trapping technique with the novel attractant "Biodelear", which is non-toxic to the environment. The Biodelear project foresees the use of the integrated Mediterranean strategy (IMS) tool in citrus orchards in the Mediterranean region

• The application of Biodelear in citrus orchards had no adverse effect on the environment and resulted in the elimination of insecticide residues in citrus fruits, rendering Mediterranean farming less dependent on pesticides, in conformation with the EU policies on enhanced biological diversity (92/43 EEC) and elimination of the residue levels of pesticides in food and feed of plants and animals (2005/396/EC). In fact, an improvement in the environmental quality of the pilot areas due to the developed integrated Mediterranean strategy (IMS) was identified. Nevertheless, further decrease in the risk of environmental degradation and improvement in the state of the environment is anticipated in the following years when the results of the IMS will be more noticeable and more farmers must have adopted the mass trapping technique using the novel Biodelear attractant, in combination with sustainable management of water and nutrients in the citrus and other crop orchards affected by medflies. Although the IMS was implemented on a local scale, the efficiency of the proposed strategy (IMS tool, novel attractant Biodelear with the mass trapping technique, sustainable management of water and nutrients and check of pesticides residues from previous practices) needs the intervention of authorities/policy makers in order to be implemented at regional or national level.

• Excess concentrations of some ions in the soil surface and irrigation water were recorded since the beginning of the monitoring of the pilot and demonstration areas. This was ascribed to inappropriate cultivation practices in the previous years. The IMS tool with the novel attractant in combination with a sustainable water and nutrient management plan for citrus orchards is anticipated to minimize the environmental impact of intensive citrus fruit cultivation.

• The developed indicators for monitoring and reporting the socio-economic impact of medfly control in citrus orchards contribute to sustainable decision-making by targeted stakeholders and improve the sustainability of citrus fruit protection. The socio-economic impact of LIFE

BIODELEAR will become more apparent in the coming years, as more information on the outcomes and benefits of the project will be provided to stakeholders as indicated in the After-Life Communication Plan

- Finally, according to the business plan and to economic analysis reports, Biodelear is a viable product ready to enter the market.

5.4.2 Necessity of project amendment

As it was clearly mentioned in the request letter to the European Commission on 27/2/2019, the prolongation of the project was necessary mainly in order to meet specific objectives, since the SSDA could not perform some of the foreseen activities of Actions A2, B1, B2, C1, C2, D1 & D2 due to the absence of the coordinator Dr V.G. Mavraganis for three months. It is noteworthy that the specific tasks, for which the prolongation was requested, were very important i.e. Delay in the completion of the risk analysis, delay in the completion in the development of the Integrated Mediterranean Strategy (IMS) against med fly, which will be based on thorough evaluation of the obtained results from the actions B1 and B2 etc. since they meant to provide data and dissemination material for meeting the project objectives. The identified effects and practices were incorporated into the decision-making tool of the project and in of the Actions A2, B1, B2, C1, C2, D1 & D2, as it was initially foreseen in the proposal.

5.4.3 Effectiveness of the dissemination - Drawbacks

In order to assess the effectiveness of the dissemination action, a series of indicators were set with related values referring to the reporting period. The indicators can be seen in the table of dissemination activities, confirming the effectiveness of the dissemination activities of LIFE+BIODELEAR project.

The consortium has carried out different activities to guarantee the dissemination of the progress of the project amongst the most relevant stakeholders. During the five years of the project, there was a significant interest in the project from Greece and other countries, as indicated by the gradually greater worldwide visits to the project website (73,000 by October 2019). The dissemination strategy (workshops, conferences, press interviews, meeting with local authorities, exhibitions, round-table discussions, etc.) made BIODELEAR recognizable among citrus-producing areas in Greece and some other Mediterranean countries (Spain and Cyprus), as well as in the science community among other European countries, specifically the LIFE projects (AgroStrat, OliveClima, Conops). There was a lot of interest from farmers regarding when the attractant Biodelear would hit the market. Biodelear attractant is proved a viable product to enter the market with profitable outcome for the farmers-producers and the consumers. An indirect measure of the dissemination efficiency is the quantity of publications in conference proceedings and journals that will maintain the visibility of BIODELEAR for several years. For information about future plans for the project, please see the deliverable “After Life: plan. Finally, no drawbacks were recorded during the project duration.

5.4.4 Replicability, demonstration, transferability, cooperation

The LIFE Biodelear activities took place in the Kampos citrus areas in the island of Chios in Greece, with results obtained for the initial two years of implementation in a 4-ha area and then for another two years of implementation in a 10.3-ha area. In both implementations, the results obtained are characterized by a high degree of reproducibility. For the evaluation of the results, the characteristics and particularities of the Kampos citrus areas in the island of Chios were taken into account. Thus, all proposed measures, means and directives on how to control the medfly are applicable also to other parts of Greece and other Mediterranean countries. Moreover, the control

of the medfly of the citrus fruits and the development of the IMS approach provides a clear Mediterranean added value to the results of the project.

Thus, the innovative attractant Biodelear was evaluated according to its performance in the implementation areas. It successfully controlled the medflies by reducing their infestation rate, in some cases down to less than 1%. It was generally compatible with other methods of medfly control, ensuring the recovery of the ecosystem of the citrus areas and that non-target beneficial insects, which works synergistically with the recovered ecosystem, were not attracted. Because it uses the developed IMS approach to control medflies in an efficient way, this method can be adapted for other Mediterranean countries. As mentioned above, this provides a clear Mediterranean added value to the results of the project.

The innovative Biodelear attractant can be used for the protection of other crops as well, although this has not been tested yet. Nevertheless, the Biodelear attractant in the present project has provided us with efficient protection against the medfly that is compatible with other protection methods (such as pesticides). Therefore, the Biodelear attractant, in combination with the IMS approach, can contribute efficiently, as with the citrus fruits, to the protection of other fruits, such as peaches, apricots, pears, grapes, pomegranates, etc., against medflies.

Agricultural food production is expected to increase up to 70% by 2050 according to Eurostat). There is going to be an increase in consumer awareness regarding food safety and quality. Stricter regulations are expected to be applied by international regulatory standards for agricultural products, and there is a large tendency for increase of land area of organic farming in Europe. For example, there was a 36.05% increase in the total land area of organic farming in Europe from 2012 to 2018.

The global market for agriculture is projected to grow from $75 billion in 2017 to $90 billion by 2023. Organic pesticides (the mass trapping method with attractants to control pests is considered part of organic pesticides) in particular are one of the fastest growing markets with a growth factor (CAGR) of 16 - 17%. Specifically, the market for organic pesticides in the world was valued at $3 billion in 2017. Organic pesticides are forecast to reach a market value of $8,225 million by 2025, growing at an estimated CAGR of 8% in the period 2020 -2025.

(Source Internet- Industry Analytical Consulting)
https://www.industryarc.com/Research/Organic-Pesticides-Market-Research-501539
(Source Internet:- Eurostat)
https://ec.europa.eu/eurostat/databrowser/view/t2020_rn120/default/table?lang=en

There is a clear tendency in the global market for organic farming, and therefore the prospect of introducing the attractant Biodelear in the local market in Greece would be a profitable venture, as reported in the business plan. The formulated attractant Biodelear is ready to enter both the global and local market as a viable product, and can become a beneficial item for producers and consumers, especially because of the efficiency of the protection it promises against the medfly and its ability to recover the ecosystem by its non-toxic effects on humans and the environment. It has a relatively low cost, and in the long run, economies of scale can be created by further reduction in costs. For producers, this implies the prospect of an increase in production, and for consumers, low prices of citrus fruits free of pesticide residues.

5.4.5 Best practice lessons

The entire project aimed to demonstrate best practices approach to the use of the innovative and non-toxic Biodelear attractant by developing an integrated Mediterranean strategy for medfly control. This strategy does not involve the use of insecticides and, thus, it protects citrus fruits and boosts production in a sustainable way. Although this approach was developed for the citrus orchards in the Kampos of the island of Chios in Greece, guidelines have been given on how

interested stakeholders from other Mediterranean countries may adopt, conform to and implement it (Policymakers simplified guidebook, Policymaker's guidebook with environmental issues, Farmers' and Layman's report).

The project contributed to the development of best practices for the protection of citrus fruits against medflies, using the IMS with the innovative Biodelear attractant in the mass trapping technique. Therefore, this approach contributes towards the complete eradication of insecticides, leading to the recovery of the ecosystem. Moreover, consultancy is given by the dissemination materials in 5 different languages on how to implement the IMS with the innovative Biodelear attractant.

The reduction of environmental footprint by the elimination of pesticide use in citrus orchards and the resultant recovery of the ecosystem presuppose reduction in energy consumption and, therefore, CO_2.

Based on several key impact indicators that were periodically monitored, the perspectives of the farmers/citizens on the socio-economic impacts (see action C2) were assessed. We recorded their positive responses on the IMS, which is based on the sustainable use of the innovative non-toxic Biodelear attractant to control the medfly. This method promises many collateral benefits, such as improvements in human health and the recovery of the ecosystem.

5.4.6 Innovation and demonstration value

The Biodelear project has a significant innovation value due to the innovative results and tools developed by the use of the innovative non-toxic Biodelear attractant with the mass trapping technique. Most of the results and tools are unique, and have strong demonstration characters. The beneficiaries, based on already known best practices such as the mass trapping technique, moved a step forward and developed unique tools, methodologies and an integrated Mediterranean strategy, which could be immediately applied to address specific issues, for example, to control medflies on different Mediterranean regions. The innovative aspects of the project included:

1. Efficacy tests to establish the optimum amount for the innovative non-toxic Biodelear attractant, eventually found to be 17g.
2. Development of methodologies for the creation of the proper indicators for the monitoring of the ecosystem biodiversity, such as arboreal and ground biodiversity, before and after the implementation of the medfly control using the innovative non-toxic Biodelear attractant with the mass trapping technique.
3. Pesticide assessment before the implementation of medfly control using the innovative non-toxic Biodelear attractant in citrus orchards in the island of Chios, and confirmatory tests for pesticide residues at the end of the implementation.
4. Soil assessment for the citrus orchards before and after the implementation of medfly control with the innovative non-toxic Biodelear attractant.
5. Integrated pest management strategy to ensure the suitability of the innovative non-toxic Biodelear attractant for the control of the medfly in different Mediterranean regions.
6. Business plan report, which concluded by stating that the innovative non-toxic Biodelear attractant is a viable product ready to enter the market
7. Economic analysis of the action B2, with the conclusion that the innovative non-toxic Biodelear attractant can be a more affordable alternative protection against medflies to insecticides when it is used on a permanent basis, thus creating economies of scale that will further reduce costs. This will ultimately render Mediterranean farming less dependent on pesticides, as required by the EU policies on enhanced biological diversity (92/43 EEC) and elimination of residue levels of pesticides in food and feed of plants and animals (2005/396/EC).

5.4.7 Long-term indicators of the project success

All monitoring indicators (scientific, technical and dissemination) were satisfactory. It has to be mentioned that the dissemination activity of the project was very successful.

The following indicators could be monitored on a long-term basis:

1. The adoption of IMS tool with the innovative non-toxic Biodelear attractant (the mass trapping technique) by an increasing number of local farmers,
2. The number of regional producers in Greece and in other European countries.
3. The number of stakeholders who adopt the IMS with the innovative non-toxic Biodelear attractant.
4. The innovative non-toxic Biodelear attractant, according to the economic analysis and business plan reports, is a viable product ready to enter the market, and many stakeholders have expressed great interest in the attractant and are excited for the future when the productive must have penetrated the market and is then competing against the use of pesticides.
5. The long-term indicator of the project success would be the After-LIFE plan for the project.
6. The number of farmers who will receive a "green certificate" when they adopt the control of the medfly by non-toxic means.
7. The long-term indicator of the project success would be the implementation of the Biodelear attractant in other projects for the control of the medfly for other fruits, such as peaches, apricots, pears, grapes, pomegranates, etc.
8. The expansion of the implemented actions from a local scale to a regional/national/Mediterranean scale.
9. The innovative non-toxic Biodelear attractant is considered in the After-LIFE plan for application to other projects in greater land areas to protect citrus or other fruits, such as peaches, apricots, pears, grapes, pomegranates etc., against the medfly.

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