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## **Final Report**

**Covering the project activities from 01/07/2013 to 30/11/2018**

Reporting Date

**<26/02/2019>**

LIFE+ PROJECT Acronym

**<LIFE CONOPS>**

### Data Project

<b>Project location</b>	Greece – Italy
<b>Project start date:</b>	<01/07/2013>
<b>Project end date:</b>	<30/11/2018>
<b>Total budget</b>	2.989.314 €
<b>EC contribution:</b>	1.480.656 €
<b>(%) of eligible costs</b>	50%

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## 2. Executive Summary (maximum 5 pages)

Invasive mosquito species (IMS) have raised great concern over their establishment in European countries mainly in respect to public health issues, particularly affecting the “entrance gate” countries. Main goal of LIFE CONOPS project was the management of these species with focus on Italy and Greece in respect to Climate Change consequences and parameters, dealing with population monitoring and socioeconomic factors of both countries. The project implementation started on July 1, 2013 and its actions were completed on November 30, 2018 (including a prolongation of twelve months).

LIFE CONOPS project mainly consists of three distinct phases. The first includes **preparatory actions**, that aim to identify current situation [**Action A.1**], the environmental parameters that affect the establishment of IMS [**Action A.2**] and finally, the assessment of current situation in terms of socio-economic impacts [**Action A.3**]. All Preparatory Actions (Actions A.1 until A.3) were completed and the respective deliverables have been submitted. More specifically, in the terms of Action A.1 several results (deliverables) were obtained presenting the current situation. Furthermore, a series of bioassays was conducted, in both Italy and Greece, to assess the resistance status of *Aedes albopictus*, Skuse, (the Asian tiger mosquito) populations against the currently used common insecticides (and registered in EU). Furthermore, we evaluated, under laboratory conditions, the vector competence of the Greek strain of *Ae. albopictus* (an established population from Athens). The results confirmed that *Ae. albopictus* is more efficient vector for chikungunya (CHIKV) than dengue-2 viruses (DENV 2). Within Action A.2, the environmental parameters influencing the invasive mosquitoes' entry, spread and establishment were assessed, resulting in the development of spatial databases and maps of the areas presenting an IMS suitability factor. Furthermore, during Action A.2 a multi-criteria analysis was performed aiming to identify the most suitable areas/places for Action B.2 implementation. In addition, during Action A.3., the socioeconomic aspects of IMS management costs were determined for public bodies (paid by municipalities, regions, etc.) and private prevention in both Greece and Italy. At this point, it should be noted that for Greece, LIFE CONOPS initiated the systematic entomological surveillance for *Ae. albopictus*. Before 2014, only sporadic data have been collected in relation with imported human cases of mosquito borne diseases.

The second phase referred to the **implementation actions**. The first action [Action B.1] was the development of an innovative prototype mosquitoes monitoring device [MD], while the second one [Action B.2] consists of the implementation of a pilot surveillance plan in

selected high-risk areas (selected points of IMS entry). Other actions of the second phase included, the development of biodegradable substances to control IMS based on origanum essential oil enriched with carvacrol [Action B.3], a pilot surveillance plan in selected high risk areas, which included both the prototype monitoring devices and extended network of ovitraps [Action B.2] and the determination of relevant climatic and environmental data in order to estimate the impact of climate changes (suitability maps [Action B.4]). Moreover, Action B.5 concerned the design of management plans to control IMS while Action B.6 deals with their pilot implementation. Finally, the development of Integrated Management Plans to control IMS was the subject of Action B.7.

In **Action B.1**, regarding the development and optimization of the Prototype Monitoring Device (MD), a major delay was faced as the whole process of constructing and testing the MD proved to be much more complex, labor demanding and time consuming than expected. Thus, instead of the foreseen completion date of September 2014, we were able to have completed and installed by the end of June 2015 a 3<sup>rd</sup> generation of the MD operated in the premises of BPI for field testing. This 9-month delay was unforeseen but impossible to avoid and consequently led to delays in the upcoming Actions (mainly B.2 and C.1). Thus, we asked for a prolongation of the project duration, which was granted by the European Commission on December 2017.

**Action B.2** concerned the pilot implementation of a surveillance plan in the points of IMS entry. This surveillance plan included the installation of the MD in 8 points of entry in Greece and 4 points of entry in Italy and the establishment of an extended network of ovitraps in both Greece and Italy at selected sites where the MDs were installed or other areas that are considered as high risk areas (e.g. based on suitability of entrance and establishment of new IMS). Although the action referred to three monitoring periods (mosquito periods), due to the delay in the construction of the MD, the results of the 1<sup>st</sup> monitoring period (2015) were only based on the oviposition network and not on MDs. The project's prolongation enabled the LIFE CONOPS consortium to not only acquire data during the 2017 mosquito period but also to acquire mosquito surveillance data from the MDs' operation during the 2018 mosquito period (Deliverable B.2.10). Thus, three mosquito periods are now fully covered, as foreseen. The respective Deliverables of Action B.2 were the databases generated from the monitoring surveillance network established in the points of entry. In this respect, the databases from MDs and ovitraps' network of Greece and Italy were submitted for 2017 and 2018 mosquito periods (Deliverables B.2.10 and B.2.11) revealing that in areas with medium or high densities of mosquitoes MDs collection efficacy was very good. Samples collected from the MDs were

tested afterwards for their CHIKV and DENV status. All tested samples were found negative to the viruses.

**Action B.3**, included the selection of the most favorable Essential Oils (EOs) and their large-scale production for further evaluation in pilot scale. Among numerous EOs tested, the following three were selected as the most suitable: I) the EO of *Origanum* (V 12), which displayed simultaneously the best repellent and larvicidal properties, II) the EO obtained from the summer collection of unripe *Juniperus phoenicea* berries (J 17), which was selected for the implementation of large scale repellency tests, and III) the EO C 15 originated from the processed industrial by-products of *Citrus sinensis*, which was selected for the larvicidal pilot scale tests. Finally, after laboratory bioassays for both larvicidal and repellent efficacy, the EO of *Origanum* was selected for further field trials and evaluation both as spatial repellent and larvicidal (pilot field tests in a park and in catch basins, respectively).

In **Action B.4**, the spatial risk databases for the establishment of IMS in Greece and Italy were produced, taking into account several environmental and socioeconomic parameters. The Action's results were very useful for the development of management plans against IMS because they present the areas that will be most affected by IMS and thus these management plans will be more intense in these areas in comparison to others that are not affected to such a degree.

**Action B.5**, included the development of the management plans for the IMS control, which already have been submitted in the previous reports. It must be noted however, that the developed management plans are already **legally in force in Greece since September 2016**, according to a Ministerial Circular issued by the Hellenic Ministry of Health adopting the LIFE CONOPS' Management Plans as a means for the confrontation of CHIKV, DENV and Zika virus in Greece. This action was of high importance since the LIFE CONOPS team was able to test, evaluate and improve the produced management plans by using data from real cases, concerning all CHIKV, DENV and Zika virus infections imported in Greece. For Italy, in 2016, after the adoption of LIFE CONOPS management plans for the control of IMS *Ae. albopictus*, the Emilia-Romagna Region has increased surveillance and vector control by creating a task force consisting of expert entomologists that supports the Local Public Health Units and Municipalities to evaluate the effectiveness of the extraordinary measures and the possible health risk remaining downstream of the mosquito-control treatments.

The **Action B.6** concerns the pilot implementation of the LIFE CONOPS management plans. In this Action, in 2015 and 2016, the Greek team, except from the entomological surveillance activities, knowledge was transferred from the Italian partners in order to create

the required capacity for further actions which were implemented for the first time in Greece. So, several actions were implemented, among which we underline the door-to-door activity performed for the first time in Greece in the Municipalities of Palaio Faliro (2017) and Vravrona (2018) while in Italy the implementation continued for another two seasons (2017 and 2018). Moreover, the application of the Sterile Insect Technique (SIT) was accomplished in Vravrona in collaboration with the International Atomic Energy Agency (IAEA) and University of Thessaly. The initial plan, as described in the last progress report, was to implement the SIT in Chania. However, after several meetings with IAEA, the LIFE CONOPS team realized that the distance of Chania from Attica could jeopardize the success of SIT. Thus, a suitable area near Athens was identified and the pilot implementation of SIT was conducted in Vravrona (Attica) which is close to Athens airport (El. Venizelos) and Benaki Phytopathological Institute (entomological laboratory facilities). Finally, for the first time, a holistic approach for the development of mosquito control agents of natural origin, was developed and applied. Within the frame of Action B3, as discussed above, a large number of Essential Oils (EOs), retrieved from Mediterranean plants and Agro-industrial wastes, were tested as potential means for the control of mosquitoes (larvicidal and/or repellent efficacy). So, in Action B.6 two rounds of field tests (2017 and 2018) were implemented allowing a better evaluation of the efficacy of the larvicidal and repellent activity.

**Action B.7** concerned the development of integrated management plans for the control of IMS. In this context, the LIFE CONOPS MPs (action B5), after their pilot implementation for almost 4 consecutive years. Thus, during Actions B6 and B7 it was possible to collect extra useful data because of the implementation of the management plans in other areas different from the pilot areas (see below more details in the description of Action B.7).

The third phase of the project's implementation includes Actions concerning the monitoring of the project's impact. In particular, Action C.1 refers to the monitoring of the pilot implementations performance, Actions C.2 and C.3 concern respectively the assessment of the environmental and socio-economic impacts of the management plans.

The **Action C.1** results concern the monitoring of MD performance and the surveillance for mosquito and mosquito borne diseases. In particular, in Deliverable C.1.1 the comparison results among MDs, BGs and ovitraps (in PMAs where it was possible) are included. This activity was performed for 9 (out of 12) PMAs installed in Greece and Italy. In the following PMAs, namely Thessaloniki (Greece), Patra (Greece) and Ancona (Italy) we were not able to establish a continuous data flow, as it was not possible, despite the repetitive efforts of the LIFE CONOPS team, to solicit a local company or expert to collect the required samples and maintain

the surveillance network. The results presented in Deliverables C.1.1 and C.1.2 are indicative that the operation outcome of MDs is comparable to other adult traps (BG-sentinel), and in some cases their catches exceed those of traditional methods. This observation was very obvious in the case of Chania, where the mosquito population was bigger, resulting in a larger number of mosquito catches. On the other hand, in regions with low mosquito populations both BG and MD presented very low catches as expected. The Deliverable C.1.3 refers to the evaluation of LIFE CONOPS management plans implementation with respect to the entomological surveillance, resistance prevention and annoyance level of residents due to adult mosquitoes. Finally, the Deliverable C.1.4 includes the evaluation of the LIFE CONOPS management plans effectiveness when implemented for the imported cases of Dengue, Zika and Chikungunya.

In **Action C.2**, first of all an investigation regarding the main characteristics of the PMAs in Greece and Italy that enhances the establishment of IMS as well as the natural means that are expected to be affected by the implementation of IMS management plans was conducted. The collected data include land uses, meteorological data (temperature, relative humidity, wind speed and direction, rainfall), a set of risk areas (based on the current land uses) and the main water bodies identified in each area, since water is the main natural mean that can be potentially affected by the use of mosquito population control substances. These data were mapped for each PMA and the main IMS pathways within each area were identified and illustrated in these maps. This analysis was focused on *Ae. albopictus*, the IMS with the highest presence in both countries. Furthermore, for each PMA, microclimatic data were collected from the meteorological stations of the MDs. These microclimatic data include temperature, relative humidity, wind speed and direction, and rainfall. The results of the above investigation, data collection and analysis were presented in the respective deliverables.

Furthermore, calculation and analysis of the environmental footprint of the designed and implemented IMS Management Plans was performed for the entire LIFE CONOPS' pilot implementation period.

Finally, an assessment of the potential environmental impacts derived by the conventional way to control mosquito larvae (use of Diflubenzuron [DFB]) as well as by the use of the alternative larvicidal essential oil of *Origanum*, that was developed in the context of Action B.3 and tested in the context of Action B.6, was performed. In fact, the results of the 1<sup>st</sup> Report (Deliverable C.2.3) guided the further implementation of Action B.3 towards the development of an *Origanum* Essential Oil (CREO), the composition of which would ensure



the most effective control of IMS while simultaneously it would present minimized aquatic ecotoxicity.

In **Action C.3**, the scientific team of LIFE CONOPS selected a variety of methods for a holistic estimation of the socioeconomic aspects of the problem both from a citizens' and an experts' point of view. The aim of Action C.3 was to evaluate various categories of costs and benefits induced by the implementation of management plans targeted to the control and prevention of invasive mosquito species in Greece and Italy. The analysis was based on a mixture of methodological approaches, including the choice experiment method for the elicitation of benefit levels, citizen's surveys on the overall socioeconomic impact of the problem and a survey targeted to stakeholders' and experts' for the identification of qualitative parameters associated with the prevention and control of invasive mosquitoes. The analysis evaluated in monetary terms the potential benefits of improved mosquito control programs and attempted to compare them with various cost categories associated with the mosquito problem from both a citizens' and an experts' point of view. The empirical findings form a basis for a cost-benefit evaluation of the current prevention programs and the design of improved mosquito management plans, as well as for the suggestion of policy proposals based on various future scenarios related to the spread of invasive mosquito species and related epidemics. Findings show a higher preference for improved programs targeted at health aversion over nuisance aversion in Greece, while Italian citizens and experts emphasize on the overall harmfulness of mosquitoes. Greek citizens seem more prone to pay against possible health consequences and specifically against the spread of unknown to them diseases, implying a risk averting behavior against invasive mosquito species and appear more willing to accept a higher cost (for an improved control program) at the present eliminating possible effects in the future. The overall socioeconomic analysis indicated that the application of improved management plans at a national level in Greece (like those already applied in Emilia Romagna, Italy) is economically justified.

In **Action D.1**, the official logo of the project is already provided, while in **Action D.2** the official website of the project was established in September 2013 and has been regularly updated with the results of the project but also with news and information about recent guidelines on mosquito control (an updated deliverable is attached – Deliverable D.2). **Action D.3** aims at the implementation of activities that ensure the dissemination of the project objectives, actions and results to relevant stakeholders, scientific community and public in general. Throughout the whole duration of the project, an extensive informative campaign was

implemented. It is noteworthy, that the dissemination activities in almost all cases surpassed the target numbers foreseen in the proposal. For example, 13 scientific articles in peer review journals were published during LIFE CONOPS when at least 4 were foreseen in the proposal. A total number of 20 TV or Radio interviews were broadcasted while at least 10 were foreseen in the proposal. Please see action D.3 below for more details. In **Action D.4**, 13 project noticeboards were produced and installed in the PMAs that accepted their installment. The problem with their installation was that the PMAs are located by definition in Points of Entry and mainly ports and airports. For this reason, the administration of most of the airports was very reluctant in placing the notice boards at their premises in the fear of negative impact on their own public perception. So, we decided to install the rest of them (3) in selected stakeholders' premises, in areas that could be visible after it is installed (Metropolitan Park Antonis Tritsis, Partner's premises, Regional Unit of Crete, etc). In **Action D.5**, the Layman's report was designed and produced with information about the objectives of LIFE CONOPS, the actions implemented and the results (Deliverable D.5.1 and D.5.2).

BPI was responsible for the effective management of the project, the operational internal communication, the effective administrative and technical control of the project and the successful implementation of all Actions through decision-making, consultation and guidance to the Associated Beneficiaries. 143 formal meetings were carried out during the whole project (Deliverable E.1.2). The project progress has been monitored continuously, and the results of this monitoring activity are presented in the QA/QC reports (**Action E.2**). The 6<sup>th</sup> and 7<sup>th</sup> QA/QC report, are presented in the Deliverables E.2.7 and E.2.8.

Regarding **Action E.3**, networking activities with other relevant EU projects were established. This effort was reinforced by the dissemination activities and resulted in an extensive After-LIFE Communication Plan (Deliverable E.4.1). In **Action E.5**, the 5<sup>th</sup>, 6<sup>th</sup> and 7<sup>th</sup> Carbon footprint Reports are given with the Deliverables E.5.3, E.5.4, E.5.5. In **Action E.6**, two Independent Auditors, one for the Italian beneficiaries and one for the Greek beneficiaries, delivered their Audit reports to BPI, which are submitted to the European Commission with Deliverable E.6.1.

### 3. Introduction (1 page)

**Description of background, problems and objectives:** Globalization of trade and travel has facilitated the spread of non-native species across the earth. A proportion of these species become established and cause serious environmental, economic and human health impacts. These species are referred to as invasive. As a long-time centre for trade, Europe has seen the introduction and subsequent establishment of more than 11,000 alien species, at least 15 % of which are invasive. Insects are the dominant group among non-native terrestrial invertebrates in Europe: of 1,522 established species, 1,306 (86%) are insects.

Regarding mosquitoes, several invasive mosquito species (IMS) have been inadvertently introduced in Europe, where they meet favourable environmental and climatic conditions enhanced by Climate Change, to establish permanent populations. In fact, it is estimated that due to the foreseen Climate Change trends, the IMS problem will be more intense in the immediate future. Consequently, new sanitary and environmental risks are faced, including the (re)-appearance of Mosquito Borne Diseases such as CHIKV, DENV and West Nile virus (WNV) which are currently emerging in different Member States, requiring the adoption of specific measures.

For this reason, LIFE CONOPS project has developed a network of 12 prototype devices for the monitoring (surveillance) of the IMS populations in selected high-risk areas in Greece and Italy, in order to contribute to the development of the integrated management plans for the confrontation of the IMS problem both in the current period as well as in the future, based on climatic and environmental projections. Management plans aim at the control of already established IMS and at the prevention of the establishment of newly introduced IMS in Europe, in a sound environmental way. Data related to invasive mosquitoes' distribution, seasonal variation and possibility of developing resistance to registered biocides, are always required for the selection and proper implementation of the correct responses and measures.

In a nutshell, **the expected results from the LIFE CONOPS project** implementation were:

- a network of 12 prototype devices for the monitoring (surveillance) of the IMS population in selected Points of Entry in Greece and Italy;
- the development of integrated management plans for the confrontation of the IMS problem both in the current period as well as in the future, based on climatic and environmental projections.

## 4. Administrative part (maximum 3 pages)

### 4.1. Description of the management system

Benaki Phytopathological Institute (BPI) was the Coordinating Beneficiary of the project and the project management-coordination was performed by Dr Antonios Michaelakis. BPI was responsible for the effective management of the project, the operational internal communication, the effective administrative and technical control of the project and the successful implementation of all Actions by providing decision-making, consultation and guidance to the Associated Beneficiaries. Other members of BPI staff with coordinating duties were Dr Panagiotis Milonas, Dr Dimitrios Papachristos, Dr Dimitrios Kontodimas and Dr Kyriaki Machera.

Three (3) Management Committees (Scientific, Financial and Technical) were formed in order to provide successful and effective project management in the respective sectors. Each Committee consisted of one representative from each LIFE CONOPS beneficiary, while all three were coordinated by Dr Antonios Michaelakis.

More specifically, the Scientific Committee decided on the implementation of the Actions, monitored the scientific rationality and Action performance and evaluated the results of each Action.

The Financial Committee monitored the economic figures of the project and performed the necessary contacts with beneficiaries' financial departments. Each partner maintained up-to-date books of account, in accordance with the accounting rules imposed by the relative legislation of each country.

The Technical Committee was responsible for all technical aspects of the project (definition of the technical specifications for acquired equipment, market research, evaluation of technical offers etc.) and its demonstrative operation.

One hundred forty-three (143) formal meetings (e.g. meetings in partners premises, skype meetings, phone discussions, see Annex E.1.2 for more details) were carried out during the whole project implementation. Several informal and internal meetings as well as skype meetings were also implemented between the Greek and Italian beneficiaries. The associated beneficiaries submitted to the coordinating beneficiary monthly progress reports which are incorporated in the integrated monthly reports of the project as well as financial reports (every 3 months).

To sum up, during the whole project duration, LIFE CONOPS received seven visits from the monitoring team (Dr A. Koutsolioutsou) and EU representatives, apart from the presentation of the project at the LIFE NEEMO meeting at Thessaloniki (21.04.2016).

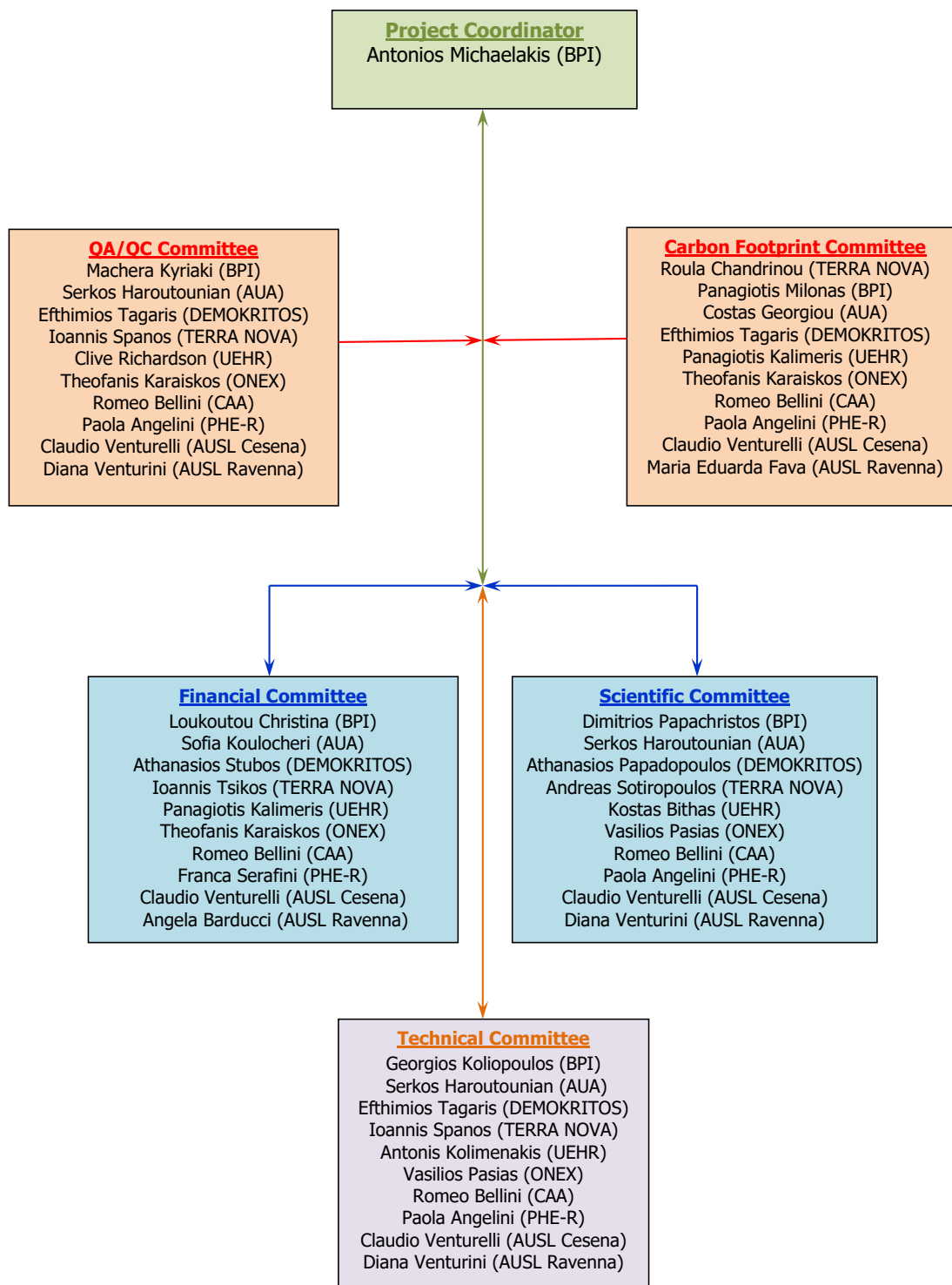
It should be noted that communication and interaction between the coordinating beneficiary and the associated beneficiaries (telephone, emails and Skype meetings) were conducted daily in order to ensure the successful implementation of actions and plans of the project. Furthermore, monthly reports were submitted to the monitoring team regarding the progress of the project (monthly reports). These reports are given attached (Deliverable E.1.8).

In addition, beneficiaries hired personnel under full or part time temporary contracts, with specialists to facilitate the implementation of specific Actions of the project. The temporary personnel hired by each beneficiary reached the number of 41 scientists (22 women and 19 men).

Contracts and specific issues are presented in Annex E.1.3.

#### **Organigramme of the project team and the project management structure**

Three (3) Management Committees (Scientific, Financial and Technical) and two (2) Subsidiary (QA/QC and Carbon Footprint) Committees were formed at the beginning of the project. The management organogram of the LIFE CONOPS is illustrated below: Detailed personnel lists of the beneficiaries are presented in Annex E.1.4 (Figure 1).



**Figure 1.** Organogram of the management structure

## 4.2. Evaluation of the management system

### 4.2.1. Assessment as to whether the project objectives and work plan are still viable

Given the approved requested prolongation of the project duration, all the activities were successfully implemented according to the agreed work plan and in compliance with the foreseen time schedule. Its major goal of producing management plans was fully met as a result of the fruitful exchange of views, the collection of examples of good practice from many European mosquito experts. As a result, the overall assessment of the project as seen by the stakeholders, workshops and dissemination activities was very successful. Not only communication and cooperation between partners was excellent throughout the project work, but furthermore the objectives set in the project submission and the results agreed on were fully met.

### 4.2.2. Problems encountered

The major delay was faced in Action B.1. Based on our initial estimation (approved LIFE CONOPS' Proposal) it was foreseen that the first Prototype MD (MD demonstrator) should have been constructed and tested until September 2014. Instead, the whole process proved to be much more complex, labor demanding and time consuming. Eventually at the end of June 2015 the 3<sup>rd</sup> MD generation (2 previous generations which preceded, were tested and gradually improved) was completed and installed at the premises of BPI to be field tested. As already reported in the submitted Mid-term Report, this 9-month delay was unforeseen and eventually impossible to be avoided, but it led to the optimized design and performance of the MD demonstrator.

Furthermore, it should be underlined that the unexpected Capital Controls, which were enforced in the Greek Bank System in the beginning of July 2015, led to the creation of new significant, unforeseen, obstacles regarding the development of the rest MDs.

To cover the gap from the delay in the construction of the MDs, a network of ovitraps was placed in selected PMAs (Pilot Monitoring Areas) to collect the required IMS' population data. By this way, we managed to cover the monitoring results of the 1<sup>st</sup> monitoring period (2015) as described in the approved Proposal. Since, there was no previous knowledge in IMS distribution and abundance in Greece, we established a large network of ovitraps. For example, in Crete where the first record of *Ae. albopictus* was only recently confirmed (a single specimen in September 2014 from KEELPNO), a very dense ovitraps' network was installed, which covered not only the airport of Chania (which is the relevant PMA in Crete) but almost the whole of

Crete from Chania (west) to Agios Nikolaos (east). This dense network led to the identification of the first established population in Crete by the LIFE CONOPS team. From April 2016 and onwards, the MDs were gradually installed in the selected PMAs and therefore the 2<sup>nd</sup> monitoring period (2016) was implemented according to the approved Proposal.

Although the ovitraps network covered sufficiently the surveillance requirements of the 1<sup>st</sup> monitoring period (2015), the prolongation of the project's duration enabled the operation of the MDs for in-field surveillance purposes during 3 mosquito periods (from 2016 to 2018) as it was originally foreseen in the approved Proposal. Thus, the prolongation made feasible the acquisition of mosquito surveillance data as well as MDs' operation performance data, during the 2018 mosquito period. Consequently, the deliverable concerning the 3<sup>rd</sup> (final) monitoring period, includes the results of 2017 and 2018 mosquito period (Deliverables B.2.10, B.2.11, B.2.12). The importance of the approved prolongation lies on the fact that we were able to implement the surveillance for all MDs for a whole mosquito period which, for the pilot implementation areas, usually begins in March and ends in late October each year (or even November, depending on the climate conditions). It is reminded, that according to the proposal, five pilot areas would be selected, three of them in Greece and the other two in Italy, to host the MDs. In each area, two MDs would be installed and there was the provision to keep two MDs as a back-up. Nonetheless, due to the large number of points of entry and the larger than expected size of the MD, instead of the foreseen distribution of the MDs according to the proposal, it was decided to choose 12 Pilot Areas (instead of 5 foreseen), eight in Greece and four in Italy (more details in Action B.2). During the 1<sup>st</sup> reporting period (2015) the MDs were not yet finalized. During the 2<sup>nd</sup> reporting period (2016), 8 out of 12 MDs were installed and operated in the points of entry. During the 3<sup>rd</sup> reporting period (2017 and 2018) all MDs were fully operating. Thus, the prolongation of the project allowed us to cover the lost period of 2015 during 2018. For more details, please see Action B.2 annexes.

Moreover, it should be noted that despite the above-mentioned delays, the first year (2016) of the MDs' operation as well as the demonstration of the MDs that took place, revealed significant potentials for market exploitation of the developed MD. The LIFE CONOPS' team, that is engaged with the development of the MD, identified a potential pathway between the project and the entomological surveillance global market. The approved prolongation of the project's duration enabled LIFE CONOPS to further investigate these market potentials.



## 5. Technical part (maximum 50 pages)

The basic idea of the project was to implement the newly released European Centre for Diseases Prevention and Control (ECDC) “Guidelines for the surveillance of invasive mosquitoes in Europe” in both countries in order to improve the States’ capacity of active detection of invasive mosquito species (IMS) and then include these activities in the structural permanent activity of Public Health Services. The problem of IMS, such as *Aedes albopictus*, *Aedes aegypti*, *Aedes atropalpus*, *Aedes koreicus*, *Aedes japonicus*, *Aedes triseriatus*, is of paramount importance in the EU as has been proved by the increasing number of detections in different EU countries and because of the public health risk related to the vector capacity of these mosquito species. Among these IMS *Ae. albopictus* is the most famous “aggressive day-time biter”. Already it is established in many European and neighbour countries; arrived in Albania in 1979 and has since spread across Europe. Moreover, it has already been responsible for recent local cases and outbreaks of CHIKV and DENV. These diseases cause acute fever - arthralgia and there are no specific therapeutic agents to treat infected people. Although *Aedes aegypti* is not located in the mainland of Europe, is established in Madeira island (Portugal) and in Turkey (close to borders with Georgia). This mosquito species is considered as the primary vector of CHIKV, DENV and Zika virus.

### 5.1. Technical progress, per task

#### 5.1.1 Action A: Preparatory actions

##### **Action A.1: Current state of the problem targeted**

Foreseen start date: 01/07/2013      Actual start date: 01/07/2013

Foreseen end date: 30/06/2014      Actual end date: 30/06/2014

##### **Achievements and description of the progress:**

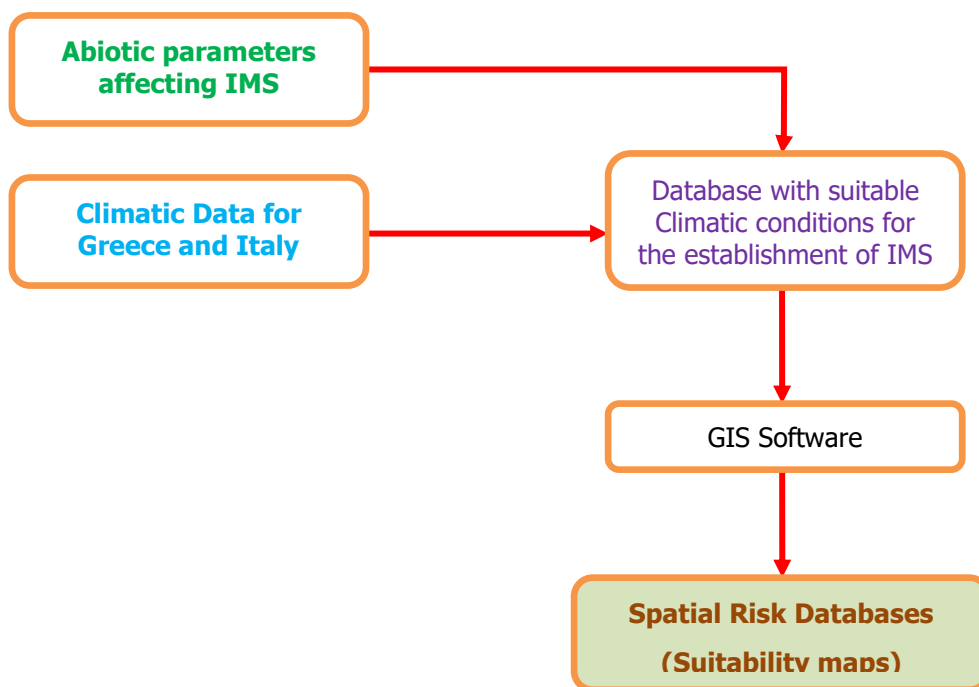
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##### **Action A.2: Analysis of climatic & environmental parameters influencing the invasive mosquitoes introduction & establishment**

Foreseen start date: 01/07/2013      Actual start date: 01/07/2013

Foreseen end date: 30/06/2014      Actual end date: 30/06/2014

Achievements and description of the progress: Aim of this Action is to provide the Spatial Risk Databases for the establishment of IMS in Greece and Italy. In order to achieve this goal, a methodological approach was designed and applied according to the following scheme:



The outcome of the abovementioned approach is the development of the IMS Spatial Risk Databases (Suitability Maps) for Greece and Italy which are given attached in the Inception Report.

According to the approved proposal, based on these databases and the respective maps, a smaller grid analysis would be performed. The results of this analysis would lead to the selection of the most representative areas/places for the pilot monitoring actions and especially for the installation of the prototype MDs (Action B.2).

From the analysis of the suitability maps developed in Action A.2, it was obvious that it was not possible to select specific areas for the smaller grid analysis since the largest part of both countries appear to have an important suitability factor. Therefore, another methodological approach was followed, based on multi-criteria analysis of specific areas. Following this “new” procedure, the 12 Pilot Monitoring Areas (PMAs) were grouped in 3 major categories:

- Ports
- Airports
- Other important areas

The report describing the methodological approach and the specific characteristics of the 12 PMAs is given in a deliverable submitted in Annex A.2.2.

This Action was implemented by TERRA NOVA with the contribution of BPI, , NCSR Democritos and CAA.

Problems/Delays: No problems or delays occurred during the 9-month period that caused any delays of other actions of the project. Deliverables: Deadline of deliverables was on 31/12/2013 and 30/06/2014.

**Action A.3: Socio-economic impacts of the problem targeted (01/07/2013 – 31/03/2014)**

Foreseen start date : 01/07/2013      Actual start date: 01/07/2013

Foreseen end date: 31/03/2014      Actual end date: 31/12/2014

Achievements and description of the progress: During the first months of implementation the scientific team focused on the identification of state-of-the-art methods relevant to the problem targeted. This was followed by a "session" of meetings and discussions with experts and stakeholders for the evaluation of the cost category regarding “public prevention costs”. The Italian beneficiaries collected data related with the costs of the Regional Plan of Emilia-Romagna in order to define the costs sustained by Municipalities, updated their estimation on the amount of expenses and provided us with relevant figures for the preparation of the first report on "The Public prevention costs". In the meantime, LIFE CONOPS team dealing with A.3 has been expanding its collaboration network, both through the first workshop in Greece as well as through personal contacts, with other important stakeholders, private companies and experts. This expansion was necessary for both the evaluation of other cost categories, such as medical costs, and the enhancement of cost valuation methods and tools applied in relevance with the problem targeted. For the estimation of health impacts, our team prepared "Tables of Databases" concerning the cost categories of health impacts, medical costs and productivity losses, in order to evaluate the cost of certain disease outbreaks in Greece and Italy. Based on the results of the aforementioned analysis the first report of the public impacts and costs caused by the ‘IMS problem’ was prepared presenting the socio-economic cost imposed by the IMS problem in selected areas of Greece and Italy and identifying the crucial parameters of the economic burden associated with the overall mosquito problem. What is more, a primary cost benefit analysis of the prevention strategies was conducted based on the main findings of the first report. In the meantime, LIFE CONOPS team has been working in collaboration with experts for designing the appropriate methodological framework and specialized questionnaires concerning the estimation of the IMS impact on the private level. A “Report of the private (households) impacts and costs caused by the ‘IMS problem’” was prepared by LIFE CONOPS team based on the results of a small-scale survey which was implemented in selected parts of

Greece and Italy. The survey was implemented in November 2014 and was conducted in two geographical levels, Greece and Italy. Questionnaires were partly distributed through the web as well as in hard copy. The aim of LIFE CONOPS scientific team is the continuation of more precise indexes of the economic magnitude of the Asian tiger mosquito through a well-designed survey using specialized methodological tools based on contingent valuation work which will be implemented in the frames of Action C.3.

Problems/Delays: The initial duration of Action A.3 (9 months), ending at the end of March 2014, proved too limited for the thorough identification and evaluation of the socio-economic impacts. One of the main reasons is re-organization of the municipalities according to the “Kallikratis” plan which took place at the 2011 in Greece. This re-organization made the identification and mining of costs’ data incurred by local authorities difficult. Moreover, we have identified several stakeholders, both private and public, that are involved in the management of the costs. Lastly, prevention costs and, to some extent, the ex-post costs cannot be easily allocated between invasive and native mosquito species. Therefore, an extension was claimed regarding the duration of Action A.3 until 31/12/2014. This implied an extension of 9 months. This extension did not require any modification on the budget and other details of WP A.3.

### **Measures to overcome the problems**

As already indicated in the monthly progress reports and at the inception report, we established systematic contacts with all the involved stakeholders. These stakeholders were able to provide us with useful information for LIFE CONOPS project, and therefore, we have established a permanent networking with them. These stakeholders are:

- KEELPNO (Hellenic Center of Diseases Control & Prevention),
- Aristotle University of Thessalonica (Department of Urban-Regional Planning and Development Engineering),
- Ecodevelopment S.A., Private Company specialized in Mosquito Control and
- University of Bologna (Department of Agro-food Science and Technology)

Through this network, we achieved a thorough identification and evaluation of the costs incurred. The careful evaluation of the costs, imposed by Invasive Species (IS), is necessary since they are interrelated with costs imposed by native species and therefore, we plan to use their knowledge in order to allocate them.

For the aforementioned reasons an extension for the duration of Action A.3 has been essential, so as to have the necessary time for an in-depth investigation and analysis of the costs. This was also supported by the earlier beginning of Action C.3 on the 1<sup>st</sup> of April 2015 (previous starting date January 2016). This extension does not require any modification on the budget and other details of Action C.3. This is also beneficial for the Action B.6 (also starting at the 1<sup>st</sup> of April 2015). Consequently, the findings of Action A.3 and a longer duration for Action C.3 (without affecting its endpoint) will help the correlation between Actions B.6 and C.3.

Deliverables: Reports of the impacts and costs caused by the ‘IMS problem’ both in the households and public prevention are presented in Annexes A.3.1 and A.3.2 respectively.

## **5.1.2 Action B: Implementation actions**

### **Action B.1: Design and development of the prototype IMS monitoring (surveillance) devices**

Foreseen start date: 1/10/2013  
Foreseen end date: 30/9/2014

Actual start date: 1/10/2013  
Actual end date: 30/12/2016

Achievements and description of the progress: Although the 3<sup>rd</sup> MD (demonstrator) generation had been developed fully operational until June 2015, it took almost 18 months for the completion of the construction of the rest 11 MDs. This was part of a quality control process decided by the relevant beneficiaries of the specific Action (BPI, TERRA NOVA and ONEX) to gradually construct the rest MDs in order to give time to each MD to operate on field conditions and thus further optimize the next MDs incorporating to them the latest mechanical and software improvements. During this period, each constructed MD was installed in the selected PMA, where was tested in real conditions in terms of mosquitoes catches, operational integrity, behaviour in outdoor conditions in a wide climatological range, response in unforeseen situations (e.g. power loss, extreme weather conditions, etc). A series of improvements were identified, designed and applied to the already constructed MDs and incorporated in the construction of the remaining MDs: Motor sampling piston, which replaced the initial pneumatic one, CO<sub>2</sub> flow metering system, metallic cases for the lactic acid, improvement of the mechanical automations, finalization of the PLC working cycle, design of 8 alternative sampling funnels.

Eventually, the construction of all 12 MDs was completed at the end of 2016 and 10 of them have already been installed in the respective PMAs.

At this point it has also to be mentioned that the casing parts of the demonstrator MD were reconstructed between November and December 2015 for demonstration purposes at the Mid-term workshop of LIFE CONOPS that was held in AUA premises on the 10<sup>th</sup> of December 2015.

The NMS test/evaluation was held in parallel with the operation/ testing trials of the MDs that were gradually constructed and installed at the respective PMAs. This action was essential, since the NMS is a functional system of the MD itself and cannot operate without the MD. In this context, various optimization modifications were implemented to the NMS regarding the user interface, the reporting template, the scheduling options, the alarms reporting, the interaction with the MD's PLC, etc.

This Action was implemented by TERRA NOVA, BPI and ONEX.

#### Problems/Delays:

A major delay was faced in Action B.1, as the whole process of constructing and testing the MD proved to be much more complex, labor demanding and time consuming than expected. Thus, instead of foreseen completion date of September 2014, we were able to have completed and installed by the end of June 2015 a 3<sup>rd</sup> generation of the MD in the premises of BPI for field testing. This 9-month delay was unforeseen but impossible to avoid and consequently led to delays in the upcoming Actions (mainly B.2 and C.1). Thus, we asked for a prolongation of the project duration until November 2018, which was granted by the European Commission on December 2017.

#### Deliverables:

Deliverables B.1.5 and B.1.7 are resubmitted following the EC comments.

#### **Action B.2: Pilot implementation of the surveillance plan**

Foreseen start date: 01/07/2014	Actual start date: 01/07/2014
Foreseen end date: 30/11/2018	Actual end date: 30/11/2018

#### Achievements and description of the progress:

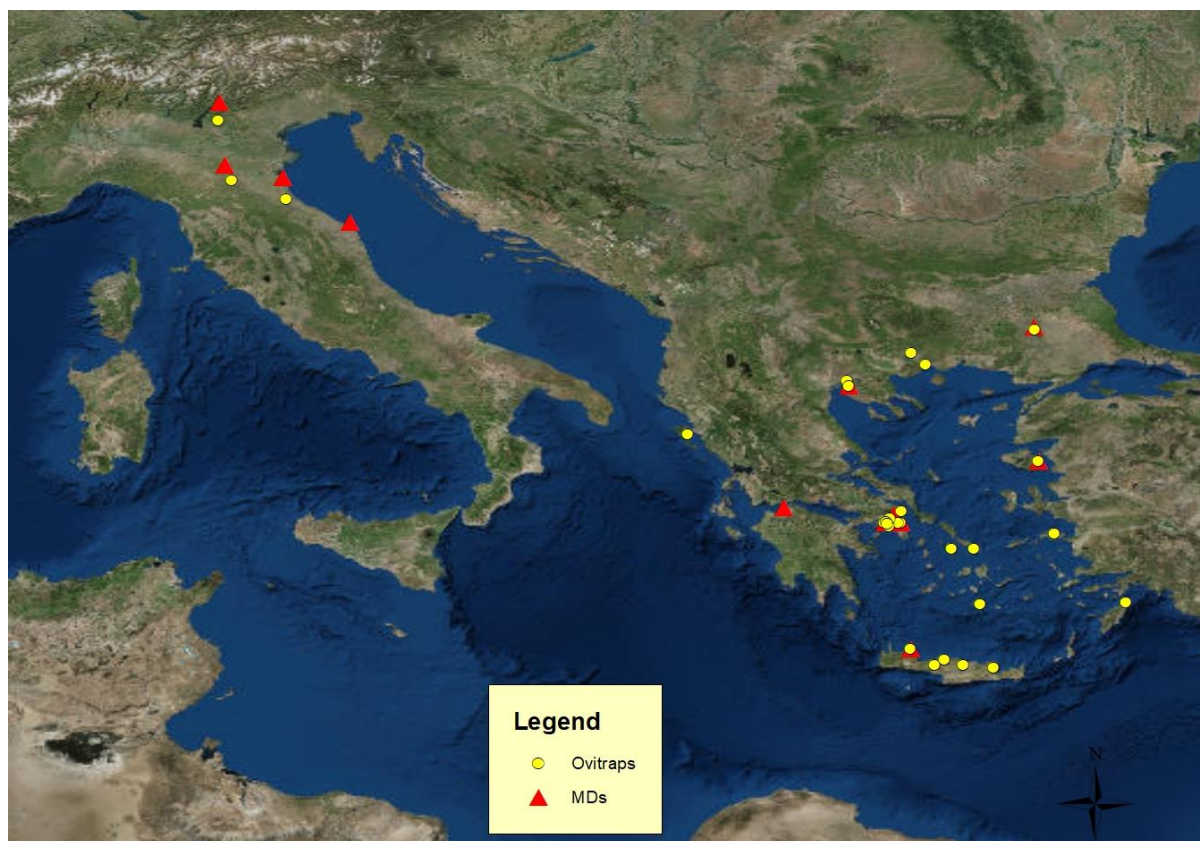
In Action B.2, the Prototype Monitoring Device (MD) has been installed in the selected locations, which are considered high risk points of entry for IMS in Greece and Italy. As described in previous reports, the selected areas were a result of Action A.2. According to the proposal, five pilot areas would be selected, three of them in Greece and the other two in Italy, to host the MDs. In each area, two MDs would be installed and there was the provision to keep two MDs as a back-up. Nonetheless, due to the large number of points of entry and the larger

than expected size of the MD, instead of the foreseen distribution of the MDs according to the proposal, it was decided to follow a slightly different pattern. Indeed, in fear of the invasion and establishment of *Ae. aegypti* in Europe it was decided by LIFE CONOPS team that it would be more useful to cover a larger number of points of entry with the MDs produced. This decision was further strengthened by the fact that there were many Stakeholders (mainly the managers of airports and ports) who expressed their wish to host the MD in their premises. As a result, it was decided to choose 12 Pilot Areas (instead of 5 foreseen), eight in Greece and four in Italy. For all Pilot Areas, authorization licenses were issued. The Pilot Areas are presented in the map below. At this point, we should note that the initial plan was to install two MDs in Thessaloniki (Greece), one in the airport and one in the port, however the big influx of refugees in the Aegean islands persuaded the LIFE CONOPS team to install an MD on the island of Lesbos in Aegean Sea, instead of the port of Thessaloniki. The following table presents the locations in Greece and Italy, where the MDs (all of which included a weather station that recorded temperature, wind speed & direction, humidity, rain levels) were installed. In parenthesis, the name of each MD is presented which refers to the names of the Ancient Greek and Roman Gods.

<b>Greece</b>	<b>Italy</b>
Athens' airport (Athena)	Ancona (Hera/Juno)
Athens' port (Poseidon)	Crevalcore (Aphrodite/Venus)
BPI (Hestia)	Ravenna (Apollo)
Chania (Zeus)	Rovereto (Artemis/Diana)
Thessaloniki's airport (Hermes)	
Mytilene (Hephaestus)	
Orestida (Ares)	
Patra (Demeter)	

The delays in the construction of MD (Action B.1) had an impact on the installation of the MDs (Action B.2). However, the LIFE CONOPS team was able to overcome the delay and cover the foreseen activities due to the prolongation of the project's duration, which was granted by the European Commission in December 2017. Thus, the MDs operated under field conditions and the LIFE CONOPS team was able to collect data for the three mosquito periods in almost all Pilot areas (namely in 2016, 2017 and 2018). Meanwhile, in order to cover the initial delay in Action B.2, LIFE CONOPS expanded the oviposition surveillance network, especially in Greece where there was no regular surveillance data so far (the ovitraps' distribution is presented in the map below). Before 2014, in Greece, only sporadic data were collected in relation to the detection of imported human cases of CHIKV and DENV. Thus, it should be highlighted that LIFE CONOPS for the first time initiated a systematic entomological surveillance for IMS *Aedes* species and the obtained data were available for all policy makers (e.g. Ministries of Health and Rural Development and Food, KEELPNO etc.). It is noteworthy,

that thanks to this extensive network of ovitraps, *Ae. cretinus* was recorded again in Greece. Before the invasion of *Ae. albopictus* this species was the only day-biting mosquito that has been recorded in several areas in Greece. Now, based on our IMS collections, this species is located only in two areas in Greece: in Rethymno (regional unit of Crete) and in Leros island. This indicates that our surveillance system is considered a reliable tool for the entomological surveillance of IMS and the fact that we identified *Ae. cretinus* shows the high sensitivity of these traps and their ability to capture insects at very low densities.



**Figure 1:** Surveillance network of MDs and ovitraps in Greece and Italy

Before their installment under field conditions, all MDs were tested for their functionality. However, after their installation some problems occurred mainly due to the particular environmental conditions in each area. For example, in many ports LIFE CONOPS team had to face the huge amounts of saline dust which covered the mechanical parts of the MD and led to malfunctioning. The problem was overcome with regular cleaning of the MD. Another important problem was the malfunctioning of the MD in Lesvos, which was installed in the port. There, the refugees unplugged the MD in order to use the electrical current to charge their cellphones. After contacting the Port authority and Municipality of Mytilene the problem was partially overcome.



Thus, the operational problems were gradually resolved and the surveillance plan in the points of entry was implemented as foreseen. The action foresaw three monitoring periods (mosquito periods), however, due to the delay in the construction of the MD, the results of the 1<sup>st</sup> monitoring period were only based on the oviposition network and not on the MDs. The project's prolongation enabled the LIFE CONOPS consortium to also acquire mosquito surveillance data from the MDs' operation during the 2018 mosquito period (Deliverable B.2.10). Thus, the third foreseen mosquito period was covered sufficiently. The MDs samplings were usually taken with a five-hour step, i.e. from 8.00 am to 1.00 pm, from 1.00 pm to 6.00 pm and from 6.00 pm to 11.00 pm). Samplings were not scheduled during the night hours because *Aedes sp.* are active during day (day-biting mosquitoes). In some cases, based on the unfavorable weather conditions, samplings were scheduled with a daily step (from 8.00 am to 11.00 pm, night hours excluded). Sampling from ovitraps was scheduled with different periodicity depending on the local availability of LIFE CONOPS collaborators. For example, in the main point of entries samples were collected every month (established the first week every month), while in Chania samples were established and collected every week. The dense sampling schedule adopted in Chania was induced by the initial plan to implement there the Sterile Insect Technique (Action B.6).

Mosquito distribution differs in time and space due to seasonal variations and environmental heterogeneity. Based on the available entomological data we can study the mosquito density in different ways trying to understand their distribution in time.

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The deliverables of Action B.2 include the database with the results of the MDs and ovitraps surveillance network for the three monitoring periods. The Final Report (Deliverable B.2.10 and Deliverable B.2.11) includes data for the monitoring periods of 2017 and 2018 (due to the granted prolongation).

All adult females collected by the MDs were tested afterwards for their CHIKV and DENV status. All tested samples were found negative to the viruses (Deliverable B.2.12). Virus circulation in mosquitoes is expected to be zero in areas where no past or present evidence of virus circulation exists.

At the end of 2018, LIFE CONOPS had obtained a large amount of data about the presence, distribution, seasonal abundance and virus status (actually, no virus was found in the selected samples) from the points of entry in Greece and Italy. The following figure presents the mean number of eggs based on the surveillance data collected during LIFE CONOPS implementation in Chania (Greece). Results shows when density starts to increase and when reaches the pick

for every year. This information is essential for mosquito control programs and therefore entomological surveillance could produce reliable data for mosquito density, distribution etc.

[CONFIDENTIAL INFORMATION ERASED]

Problems/Delays: A 9-month delay was faced due to the complex, labor demanding and time-consuming construction of the Prototype monitoring device. The prolongation of the project duration, which was granted by the European Commission on December 2017, enabled LIFE CONOPS team to cover the delay and implement the Action as foreseen. Deliverables: B.2.10- Database with the collected IMS along with their seasonal abundance in pilot areas (Final report), B.2.11- Database with the oviposition activity of female IMS in pilot areas (Final report), B.2.12- Database with the CHIKV and DENV status in adult females IMS in pilot areas (Final report). Also, the revised deliverables following the comments of EC are also submitted.

### **Action B.3: Development and production of biodegradable substances to control**

#### **invasive mosquitoes**

Foreseen start date: 01/07/2013

Foreseen end date: 31/03/2015

Actual start date: 01/07/2013

Actual end date: 30/09/2015 (extended due to the limited seasonal availability of plant material of the selected EOs)

Achievements and description of the progress: LIFE CONOPS team previous research results have determined that the Essential Oils (EOs) of *Juniperus* sp. plants and *Citrus* sp. fruits display potentials for upgrading and large-scale application for the control of mosquito's population. In this context, according to project's proposal the investigation of two plant sources of EO were proposed: i) wastes from *Citrus* juicing processes and ii) parts of *Juniperus* plants (taxa *J. drupacea* and *J. phoenicea*). These materials were collected from distinct parts of Greece such as Sterea Hellas, Mt Parnassus (*Juniperus phoenicea*), Peloponnesus, Mt Parnon (*Juniperus drupacea*), Peloponnesus, Argos (*Citrus limon*, *Citrus X paradisi*, *Citrus reticulate*, *Citrus sinensis*) and the Ionian Islands, Corfu (*Citrus japonica*). All aforementioned samples accounted 18 EOs from *Citrus* sources and 36 from *Juniperus* taxa. Since during the implementation of Action B.3, some of these plant sources were not proved efficient for wide application, mainly due to their yield and availability, we replaced them by investigating 16 additional samples from several plant families' samples, originating from diverse parts of various locations across Greece. The main method used for EOs isolation was the hydro-distillation (conventional and/or microwave) while the cold pressing method was applied for processing the *Citrus* juice industry wastes. The identification of EOs chemical composition was performed using Gas Chromatography-Mass Spectrometry (GC/MS). Subsequently, the

bioactivities of the EOs were assessed in respect both their mosquito larvicidal and repellent properties. The selection of the most efficient EOs for the pilot scale preparation and experimentation (Action B.6) was somehow difficult, since results determined many EOs as actives. Thus, we developed a tool (calculation methodology), which is capable to enumerate the sustainability of natural compounds production in conjunction with their respective bioactivity performance. Application of this methodology offered, for first time, solid and documented arguments in respect to sustainability claims of natural products utilization as substitutes of synthetic chemical compounds. The following three EOs have been identified as most suitable for further evaluation in the terms of the forthcoming pilot field tests: I) the EO of *Origanum onites* (V12), which displayed the highest simultaneous repellent and larvicidal properties, II) the EO obtained from the summer collection of unripe *J. phoenicea* berries (J17) selected for the implementation of pilot scale repellency tests, and III) the EO C15 originated from the processed industrial by-products of *Citrus sinensis*, selected for the larvicidal pilot scale tests. More details regarding the methods utilized for the isolation, analyses and bioactivity-efficiency assessments of the EOs are presented in Annexes B.3.1 to B.3.6.

Problems/Delays: No problems or delays occurred, except the already presented delay in the large-scale preparation of the EOs, due to the seasonal unavailability of the selected plant material. Deliverables: Deadline of deliverables was on 30/06/2014, 30/09/2014, 31/10/2014 and 31/03/2015. All dated were met except the large-scale preparation of the EOs which were concluded on September 2015 with the preparation-delivery of the large scale preparation of the three EOs selected.

#### **Action B.4: Future climatic and environmental data projection**

Foreseen start date: 01/07/2013	Actual start date: 01/07/2013
Foreseen end date: 30/06/2014	Actual end date: 31/12/2014

Achievements and description of the progress: The aim of this Action is the prediction of future climatic and environmental data in Greece and Italy, as well as the development of the spatial databases and relevant maps presenting the future IMS suitability of both countries.

Temperature and precipitation are the main climatic parameters that are related to the suitability of a region for the establishment and seasonal abundance of the IMS. Climate models suggest changes in future temperature and precipitation rates. In this work changes in temperature and precipitation rates due to climate change are assessed over Greece and Italy since Mediterranean region is considered to be the most prominent climate response Hot-Spot. The NASA GISS GCM ModelE is used to simulate current and future climate under the IPCC-A1B emissions

scenario. However, the outputs from the GCM are relatively coarse (i.e.,  $2^{\circ} \times 2.5^{\circ}$ ) for applications in regional and local scales. The need for regional climate projections in a finer grid size is assessed, here, using the WRF model to dynamically downscale GCM simulations. Temperature and precipitation rates for three current years (i.e., 2009-2011) are compared with values for three future years (i.e., 2049-2051) under IPCC-A1B emissions scenario. Results from the global model suggest that the selected years are representative for the relevant current (i.e., 2006-2015) and future (i.e., 2046-2055) years.

In order to develop the spatial databases and the respective IMS suitability maps and to be able to compare them to the relevant databases and suitability maps developed in Action A.2, the methodological approach of Action A.2 was also followed in this Action.

Using the abiotic factors that affect the IMS entry, spread and establishment as well as the future climatic data produced by the NASA GISS GCM ModelE, the future spatial databases and their relevant suitability maps were developed for 3 species (*Ae. aegypti*, *Ae. albopictus* and *Ae. triseriatus*). Especially for *Ae. albopictus* and *Ae. triseriatus* the developed future suitability maps are very similar to the current suitability maps developed during the implementation of Action A.2. In order to overcome this difficulty and also to be able to have comparable results, a map for each species was developed. Each map is presenting the comparison of the suitability factor between current and future years.

As it is concluded from the study of the produced deliverables, the suitability of Greece and Italy for all 3 species (*Ae. aegypti*, *Ae. albopictus* and *Ae. triseriatus*), will increase in the following years. More specifically:

- regarding the *Ae. aegypti* suitability, the areas that will be most likely affected in the future years by the climate change, will be the southern coastal areas of both countries,
- for *Ae. albopictus*, the suitability factor in the forthcoming years, in both Greece and Italy will increase. In Italy this increase appears to be equable with only some increase peaks at small areas of medium and high altitude. On the contrary, Greece presents more intense suitability factors' difference for the future years. The areas affected more in terms of *Ae. albopictus* future suitability, are the ones in medium and high altitudes and especially the mountainous areas of Pindos and Rodopi and
- for *Ae. triseriatus* mainly the northern parts of Greece and Italy present an increased suitability taking into account the fact that both countries currently present a high suitability factor.

Problems/Delays: Due to the fact that the expected computational time for part of Action work was underestimated and proved to be inadequate, the Action's duration was extended until 31<sup>st</sup> of December 2014. Deliverables: All B.4 deliverables are submitted with previous reports.

### **Action B.5: Design of management plans to control IMS**

Foreseen start date: 01/07/2014	Actual start date: 01/07/2014
Foreseen end date: 31/03/2015	Actual end date: 31/08/2015

Achievements and description of the progress: *Aedes albopictus* (Asian tiger mosquito) is the IMS already well established both in Greece and in Italy (and other EU countries), causing high concern in public health for its vectorial capacity of pathogens causing human diseases such as Dengue, Chikungunya and Zika viruses. The developed management plan has been structured as a comprehensive practical technical handbook to assist local authorities in organizing the vector control activities in the best possible way. It is focused on *Ae. albopictus* as the IMS has already been well established both in Greece and in Italy, to which the terms “control” therefore applies as the only option we currently have to reduce the density of the species

The management plan to control *Ae. albopictus* is a complex system that includes coordinated actions to adequately face the most important aspects involved into the problem such as:

- standardized quantitative monitoring by specific ovitraps to obtain regular information on the mosquito population density dynamic,
- the mosquito population density data will also serve the risk assessment for arboviruses like Dengue, Chikungunya and Zika causing serious threat in public health,
- the local community should be involved in the control campaign in private areas where most of the *Ae. albopictus* breeding sites are available and simple actions adopted by citizen may have a significant impact on the mosquito density,
- standard control measures in public areas should be organized regularly using larvicides in the road drains to cover the whole breeding season. It is also necessary to conduct independent quality control operations on the larval treatment in order to assure high efficacy and promptly adopt possible corrective actions (e.g. emergence of resistance),
- an emergency vector control plan should be prepared, and responsibilities clearly assigned to the stakeholders to face the epidemic risk in case of importation of infected persons and
- attention is also devoted in a pilot door-to-door control strategy to be adopted locally in case the regular control campaign does not achieve sufficient results.

Specific annexes to practical organize the activities such as: standard operational procedures for ovitraps field managements; standard operational procedures for eggs counting; quality control procedure for the *Ae. albopictus* monitoring; mayor ordinance scheme; standard operational procedures for emergence vector control operations in case of Dengue, Chikungunya and Zika cases detection; quality control procedure for larval treatments efficacy in road drains; standard operational procedures for bioassays on insecticide sensibility; template for public tender for PCO, are also provided.

The final version of LIFE CONOPS management plans were submitted in August 2015. However, draft versions had been prepared few months earlier. This enabled LIFE CONOPS team to begin implementing the basic actions of the management plans before the submission of the final version (Action B.7). Additionally, during the mid-term workshop (December 2015) one of the Working Groups (WG) was responsible to evaluate and suggest more actions and/or activities. This WG consisted from experts/policy makers, such as entomologists, epidemiologists and medical doctors.

LIFE CONOPS management plans have been legally operating since August 2016: a ministerial circular note produced by Hellenic Ministry of Health and distributed to all public health units in Greece (stakeholders for public health, available only in Greek language: [http://www.conops.gr/wp-content/uploads/timeline/16082016/Management%20plans\\_Zika\\_Dengue\\_Chik\\_58894-2016.pdf](http://www.conops.gr/wp-content/uploads/timeline/16082016/Management%20plans_Zika_Dengue_Chik_58894-2016.pdf)). To assist local authorities in organizing the vector control activities, LIFE CONOPS management plans have been structured as a comprehensive practical technical guideline to ensure quality and efficiency. For all imported cases, in 2015 and 2016, LIFE CONOPS management plans (via ministerial circular note) helped to implement an immediate and capillary mosquito control activity, beginning within 24 hours from the case reporting.

Problems/Delays: Due to the delays in Action B.1 the management plans were finalised on 31<sup>st</sup> of August 2015. Deliverables: Management plans for *Aedes albopictus* and *Aedes aegypti* were submitted with previous reports.

#### **Action B.6: Pilot implementation of management plans to control IMS**

Foreseen start date: 01/04/2015      Actual start date: 01/04/2015

Foreseen end date: 30/09/2018      Actual end date: 30/09/2018

Achievements and description of the progress: The first version of LIFE CONOPS management plans was submitted in August 2015 and during the mid-term workshop (in December 2015) the first Working Group (WG1, in total 3 WGs) was responsible to evaluate and suggest more

actions and/or activities. WG1 included LIFE CONOPS representatives and experts/policy makers, such as entomologists, epidemiologists and medical doctors (direct involvement of stakeholders). Dissemination of messages through professional network, such as LIFE CONOPS website and social networks, also helps increase public awareness and education in mosquito control. Thus, to raise public and stakeholder awareness in order to better focus the control activities on the target IMS breeding sites, LIFE CONOPS “built” a website in which useful information about bio-ecology of the IMS, guidelines, updated news and videos were provided in three languages (Greek, English and Italian). Our website counts more than 99,100 website visitors and more than 248,000-page views and a video with basic information about mosquitoes, personal protective measures and how to take prevention steps to avoid mosquitoes breeding sites counts more than 44,000 views.

Among the activities, in the pilot implementation of the management plan designed during Action B5, LIFE CONOPS team developed and evaluated for the first time in Greece and Italy the “door-to-door” strategy. Door-to-door strategy can trigger community participation aiming at the elimination of mosquito breeding sites. In Greece during the period 2017-2018 an education campaign in Palaio Faliro, was evaluated to find possible correlation between public awareness and abundance of breeding sites. Additionally, the “SIT” strategy (sterile insect technique) was evaluated during the period 2017-2018, in Greece. The SIT is an environmentally friendly insect pest control method involving the mass-rearing and sterilization, using radiation, of a target pest, followed by the systematic area-wide release of the sterile males by air over defined areas, where they mate with wild females resulting in no offspring and a declining pest population. The main scope of these trials was to analyse the effectiveness of both “door-to-door” and SIT control measures in reducing the *Ae. albopictus* population density. Hatching rate is an index that is commonly used in SIT projects to show the decreasing of fertilised eggs in target area. So, an index closes to 1 means fertilised eggs and closes to zero non-fertilised eggs. The design, implementation and evaluation of SIT in Greece for the management of *Aedes* invasive species resulted in very good outcomes; hatching rate decreased by more than 70% compared to control areas (**Figure 6**, hatching rate index).

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Finally, for the mosquito resistance status in Emilia-Romagna (Italy), LIFE CONOPS studied the geographical distribution at fine scale of the resistance alleles recently discovered in *Cx. pipiens* (susceptible to diflubenzuron-DFB, **Figure 7**). The insurgence of DFB resistance in *Cx. pipiens* populations in the Eastern Emilia-Romagna provinces, the high mutation frequencies recorded, and the limited number of available larvicides highlight the necessity for the

development of appropriate insecticide resistance management programs in these settings. Since the invasion of *Ae. albopictus* in urban areas in Europe (in the last 10-15 years), DFB based products have been used intensively. Resistance status/evaluation is one of the components of LIFE CONOPS management plans, to ensure the sustainability of current control interventions for all mosquito species and safeguard public health.

[CONFIDENTIAL INFORMATION ERASED]

Ongoing *Cx. pipiens* resistance surveillance in Northern Italy and other countries where DFB applications take place, including the monitoring of other vector species such as *Aedes albopictus* for DFB resistance mutations are prerequisites for the development and execution of insecticide resistance strategies resulting in efficient and effective arbovirus vector control. DFB is currently one of the most efficient larvicides and in the past it has been also used extensively for agricultural pest control. A major problem associated with the limited selection of available insecticides and their extensive use in mosquito and agricultural pest control is the development of insecticide resistance.

Consequently, contemporary legislation tends to increase limitation on the use of all synthetic pesticides, promoting bio-pesticides as a safer alternative. Bio-prospecting efforts for bio-pesticides provide results, which rarely reach the industry. In Action B.6 we evaluated potential biocides to chart the path from the laboratory bench (Action B.3) to field assessment. As subjects of bioprospecting were defined Mediterranean culinary plants, while as valorization target was selected the *Ae. albopictus* control. Of increased significance was the transition between experimentation phases that presented three major challenges; first, the identification of a broadly available carvacrol rich essential oil (CREO) as indicated by the bio-prospecting results (Action B3); second, the delineation of the suggested concentration in order to assure efficacy and environmental safety (Action B3); third, the selection of a CREO formulation that would facilitate field application (Action B6). Our results present for the first time the repellent properties of Conehead Thyme, Cortuk, Savory, Greek Sage, Monk's Pepper, and Fennel EOs against *Ae. albopictus*, as well as the larvicidal properties of Cortuk and Conehead Thyme EOs against the same target. The results obtained through field tests indicated that the emulsified CREO might be considered as a potent *Ae. albopictus* larvicidal and/or repellent agent. In summary, we believe that our approach successfully addressed the identified challenges and represents a methodological example for the exploitation of EOs as mosquito control agents. At last but not least, one of the major components of the LIFE CONOPS management plans was raising awareness among citizens. Towards this end, the action "A 'network' of students against the Tiger Mosquito and others IMS" has been implemented in Italy, with involvement



of 12 classes from 3 high schools, 2 from Cesena and 1 from Rimini. In total 275 students have attended the activities of the project. The project is an educational path with the aim to sensitize teachers, students and families about the effects of their habits on the biology, behavior and the related health risks due to the mosquitoes' spread (For more details, see Annex B.6.3).

The project activities are organized in 3 steps: 1<sup>st</sup> STEP in the class: theoretical lesson with specialist (entomologists, biologists and specialists involved in the mosquito's field) to explore the biology and ecology of mosquitoes and to highlight the relevance of the surveillance and the risks of IMS introduction related with the human activities and climatic changes.

2<sup>nd</sup> STEP in the field: in the school garden or in the surroundings, to identify and recognize the suitable sites for the development of the mosquito's larvae and to collect the samples to analyze together in the science laboratory. 3<sup>rd</sup> STEP (the summer work): investigation in the own garden or in the surroundings. The students become the "detectives" of the environment near their houses and spread acquired knowledge with families, relatives and neighbors.

Problems/Delays: A delay occurred due to problems faced during the development of the Prototype monitoring device. The prolongation of the project duration, which was granted by the European Commission on December 2017, enabled LIFE CONOPS team to cover the delay and implement the Action as foreseen. Deliverables: A detailed report for the pilot implementation of the listed actions of the management plans for Greece and Italy was submitted in Deliverables B.6.4. Especially for Greece, the number of pilot actions in 2017 and 2018 were increased compared to previous years. In deliverable B.6.5 the final report on plan activities for the period 2015-2018 according to the designed management plans are presented.

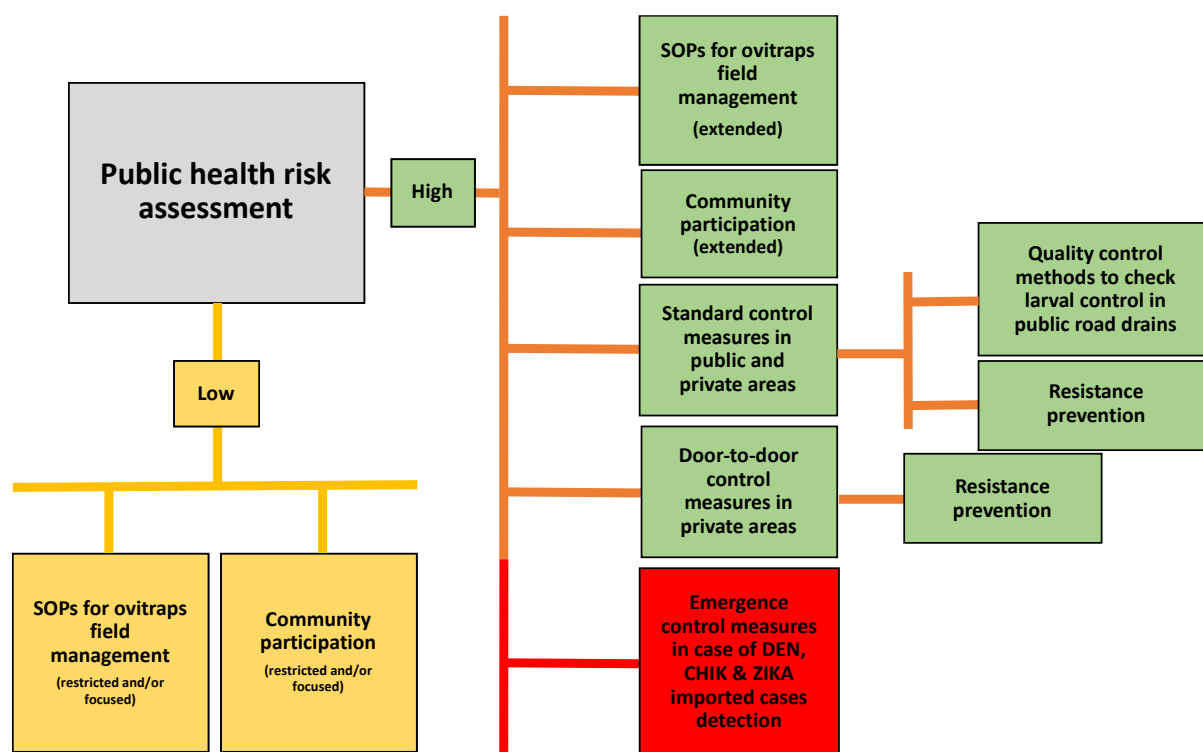
#### **Action B.7: Development of integrated management plans to control IMS**

Foreseen start date: 01/04/2017      Actual start date: 01/04/2017

Foreseen end date: 30/11/2018      Actual end date: 30/11/2018

Achievements and description of the progress: *Aedes (Stegomyia) albopictus* (Skuse 1894) (Diptera: Culicidae), is the only IMS already well established in many EU and neighbour countries. Its management is a complex system that includes coordinated actions to adequately face the most important aspects involved into the problem, such as nuisance and vector-borne diseases (VBDs). Action B.5 presented a detailed design of management plans, which was focused on *Ae. albopictus* and *Ae. aegypti*. The latter species is not present yet in European continent, and other IMS (*Aedes atropalpus*, *Aedes japonicus*, *Aedes koreicus*, *Aedes triseriatus*) are present in limited areas, so specific and different approaches are implemented. The management plan for *Ae. albopictus* has been structured as a comprehensive practical

technical guideline to assist local authorities in organizing the field activities in the best possible way. The LIFE CONOPS scientific team, during Actions B.6 and C.1, applied the produced management plan and together with other experts and stakeholders review and finalized it. A significant evaluation of this management plan was conducted during the standard operational procedures for emergence vector control operations in case of CHIKV, DENV and Zika cases detection (Circular from Ministry of Health, Greece, ΑΔΑ: ΩΜΙ7465ΦΥΟ-Χ4Θ). In the framework of LIFE CONOPS project a detailed management plan to control the invasive mosquito species *Ae. albopictus* was developed. The management plan (**Figure 8**) includes coordinated actions such as standardized control measures and quality control activities, monitoring protocols, the involvement of local community and stakeholders and an emergency vector control plan to reduce the risk of an epidemic in case of detection of infected persons. Moreover, the management plan provides useful material for activities in “prevention stage” that are compulsory from stakeholders to manage IMS, such as quality control procedure for the *Ae. albopictus* monitoring; and template for public tender for Pest Control organizations (PCO). Furthermore, attention is also devoted to communication aspects and actions to be adopted in case of emergency. Consequently, the design of management plan was constructed on a “two-choice” risk for each area: areas that are considered as “low risk” or as “high risk”.



**Figure 8.** LIFE CONOPS Management Plans

Problems/Delays: There were no delays. Deliverables: An integrated management plan to control the major IMS in Europe (*Ae. albopictus*) and neighbour countries has been developed. This management plan to control *Ae. albopictus* in areas where the species is well established, includes several activities which may be modulated following local resource availability and cost-benefit evaluation.

The management plans produced in the framework of LIFE CONOPS are complex systems including several components (and displayed in Figure 1) which are clustering in 3 major groups:

1. Entomological and VBDs surveillance
  - a. Standardized quantitative monitoring
  - b. Public health risk assessment
2. Standard control measures in public and private areas
  - a. Emergence control measures,
  - b. Efficacy & Quality control methods
  - c. Resistance prevention
3. Public/stakeholders' awareness
  - a. Community participation
  - b. Pilot door-to-door control measures in private areas

Each component is explained in detail to provide a comprehensive, practical and technical guidance to assist local authorities in organizing the vector control activities in the best possible way. Together with the routine actions aiming at reducing population density of *Ae. albopictus*, the management plan includes also an emergency vector control plan to reduce the risk of an epidemic in case of detection of infected persons.

Compared with the deliverable in Action B.5, the final deliverable was modified after reviewed by European experts in the area of mosquito control. One major area of concern centered on reform the entomological surveillance with ovitraps. In more details, two methods suggested during the sampling with the use of ovitraps: a) If the collected eggs need to be hatched the ovitraps should be filled with tap water and remain in the field for only 7 days and b) If there is no necessity for egg hatching, the operators may fill the ovitrap with the solution of *Bacillus thuringiensis* var. *israelensis* and remain in the field for 14 days. Finally, we used the phrase “oviposition substrate” since every country uses different tool e.g. tongue depressor (Greece), masonite wood (Italy), polystyrene blocks (France), filter paper (Albania).

Another change (minor) is to organize the management plan as a main text, which briefly explain the activities, while specific annexes were produced to practical organize the activities.

Our intention (LIFE CONOPS team and experts) is to make a document as practical as possible so, each stakeholder can easily read the main text (basic information) and then choose the Annex/activity that has to implement. In this way the reader/stakeholder does not have to read the whole document (main text and all Annexes) but only selected parts of it.

### 5.1.3 Action C: Monitoring of the impact of the project actions

#### Action C.1: Monitoring of the performance of the pilot implementations

Foreseen start date: 01/07/2014	Actual start date: 20/05/2015
Foreseen end date: 30/09/2018	Actual end date: 30/09/2018

Achievements and description of the progress: In Action C.1 the monitoring of the performance of the MD, the network management system (NMS) and the effectiveness of the implementation of management plans were foreseen.

The performance of the MD and the NMS was assessed in one unified Deliverable (Deliverable C.1.1 – C.1.2) because MD and NMS are designed to be an integrated system, to ensure distant management of the trap and thus the MD cannot operate without the NMS and vice versa. Data from the ovitraps located as close as possible to the MD and data from other adult traps (e.g. BGs) installed also near the MD, were used for comparison with the data collected from the MD to assess the performance of the monitoring system. The MD and BG-Sentinel traps were baited with the same attractants. In most cases, data from the MDs converge with or surpass the number of catches from other adult traps. This is obvious in the case of Chania, where the mosquito population was greater and thus the number of mosquito catches was greater as well.

[CONFIDENTIAL INFORMATION ERASED]

Based on the results obtained from ovitraps (see also Annexes C.1.1 and C.2.2) show that the only IMS detected in Chania (and in Greece generally) was *Ae. albopictus* and the highest number of mosquito eggs was recorded in urban areas, which probably favored the creation of a more suitable microhabitat for the Asian tiger mosquito. However, in these areas **both BGs and MD**, which were placed as close as possible to these sites, presented a low number of collected mosquitoes. It is important to note that in the regions where the mosquito population density was very low both BG and MD presented very low catches, as expected. According to previous research (Yiji Li et al., 2016) BG-Sentinel trap is more sensitive for monitoring the

population density of *Ae. albopictus* during the peak months of the year, which is probably true for MD as well, since it is an adult trap with similar operation principle. However, more research should be done on this issue. In 2018, MDs were operated for longer periods and therefore we were able to collect more entomological data compared to previous years. Therefore, we were able to see the seasonal abundance of the IMS, concerning the comparison between MDs and the other commonly available adult traps such as BG-sentinel. The threshold for adults' traps (including MDs) is difficult to estimate because of several biases. LIFE CONOPS findings will help us to examine any possible relationship between mosquito density and environmental conditions. Furthermore, we will try to identify any relationship between the mosquito density and human nuisance perception (e.g. a quantification of a threshold for nuisance perception by humans).

During LIFE CONOPS implementation period the effectiveness of management plans was assessed every time an imported case of CHIKV, DENV and Zika was recorded (Deliverable C.1.4). LIFE CONOPS management plans were legally in force since September 2016 and this gave the opportunity to evaluate the management plans in terms of their implementation. Thus, in case of an imported case, LIFE CONOPS team was involved to ensure along with the HCDCP/KEELPNO and the National School of Public Health the foreseen procedures according to management plans, to avoid transmission of the virus.

[CONFIDENTIAL INFORMATION ERASED]

In Italy the “National plan for surveillance and prevention of mosquito borne arboviruses with specific focus on Chikungunya, Dengue and Zika viruses – 2016” include actions similar to the ones developed by the LIFE CONOPS. The Emilia-Romagna region as well as in other Northern Italy regions such as Piedmont, Lombardy, Veneto, Tuscany, the active surveillance and preventative control operations following the detection of CHIKV, DENV or ZIKAV imported cases are regularly and routinely applied, while in other regions the Public Health system is still under organization. The publication of the Practical Management Plan for Invasive Mosquito Species in Europe: I. Asian Tiger Mosquito (*Aedes albopictus*) will be instrumental to further stimulate the adoption of preventative measures in all the Regions.

Furthermore, to evaluate LIFE CONOPS management plans implementation, the door to door (D-t-D) was implemented in the Municipality of Vravrona. In terms of the mosquito population, very soon after the first visit of door-to-door, we recorded a relevant decrease in mosquito

density. In Vravrona, LIFE CONOPS team managed to decrease the total number of fertilized eggs and consequently the annoyance from *Ae. albopictus* without using biocides (Figure 10). This was very “strong message” for residents to realize their responsibility as a part of an integrated mosquito management plan in their area. This outcome is very important since limited resources at national and local level are available. In 2018 almost none of the local authorities (Regions, Regional Units, Municipalities) were able or interested to perform door-to-door programs. However, for 2019, many Municipalities are planning to include this strategy in their mosquito control programs. Therefore, we uploaded a brief description of this strategy (<http://www.conops.gr/door-to-door/>) in LIFE CONOPS website, to help them design and implement the new strategy in a proper and effective way.

[CONFIDENTIAL INFORMATION ERASED]

In Action C.1, according to the proposal, sampling of immature mosquitoes in breeding sites (larvae or pupae) should be collected to evaluate the implementation of management plans. However, this proved to be unfeasible since *Aedes* species are container-inhabiting mosquitoes largely in private areas, which are difficult to access. Instead, two different methodologies were chosen to evaluate the efficiency of LIFE CONOPS management plans. In Italy (Deliverable C.1.3) a molecular protocol was developed, for the early detection (e.g. as egg or larval stage) of which mosquito species is present in a target area. Because the most popular surveillance method is currently based on ovitraps, there is the need to develop a rapid and sensitive method allowing the discrimination of *Aedes* species laying eggs in the ovitraps. The abovementioned protocol, enabled the evaluation of the entomological surveillance implemented with ovitraps, as suggested in Appendix 1 of LIFE CONOPS management plans (Deliverable B.5). In Greece, a protocol was developed (1) to determine the pattern of genetic variability within the Greek *Ae. albopictus* populations based on analyses of the sequence diversity of the mitochondrial gene cytochrome oxidase I (COI) and (2) to establish the evolutionary relationships of these Greek populations with other *Ae. albopictus* populations and (3) to determine the geographic origin of the populations that colonized Greece. The above targets were set in order to be ready in case of a new IMS introduction (e.g. *Ae. aegypti*) and investigate a possible resistance strain. Such a case took place in Madeira, in 2013, when after an imported case was detected, *Ae. aegypti* mosquitoes were found to carry mutations associated with insecticide resistance. Thus, the abovementioned protocol was developed to evaluate the resistance prevention described in LIFE CONOPS management plans (Deliverable B.5). However, fortunately, we did not have the opportunity to use the developed protocols during the implementation of LIFE CONOPS

since no resistance was found in the mosquito populations collected. Nevertheless, the developed protocols will be a valuable tool in case of resistance development. To summarize, these two methods, the early detection of mosquitoes in egg stage and their pattern of genetic variability, are molecular tools that will enhance the entomological surveillance activities. The obtained results, the identification, in early stage, new *Aedes* invasive mosquito species and their resistance status, will help the management plans through the establishment of a trap network in areas that are considered points of entry with high risk.

Problems/Delays: A delay was faced due to the delay in the construction of the Prototype monitoring device. The prolongation of the project duration which was granted by the European Commission on December 2017 enabled LIFE CONOPS team to cover the delay and implement the Action as foreseen. Deliverables: Deliverable C.1.1 & Deliverable C.1.2 constitute a unified deliverable with the evaluation of the performance of the MD. Deliverable C.1.3 includes the evaluation of the implementation of LIFE CONOPS management plans in terms of entomological surveillance, resistance prevention and annoyance level of residents due to adult mosquitoes. Deliverable C.1.4 includes the evaluation of the effectiveness of LIFE CONOPS management plans as implemented in case of imported cases.

### **Action C.2: Assessment of the environmental impacts of the management plans**

Foreseen start date: 01/04/2015      Actual start date: 04/09/2013

Foreseen end date: 30/09/2018      Actual end date: 30/09/2018

Achievements and description of the progress: An investigation regarding the main characteristics of the PMAs in Greece and Italy including the parameters enhancing the establishment of IMS as well as the expected impacts of the implementation of the management plans was conducted.

The collected data include land uses, meteorological data (temperature, relative humidity, wind speed and direction, rainfall), a set of risk areas (based on the current land uses) and the main water bodies identified in each area, since water is the main natural mean that can be potentially affected by the use of mosquito population control substances. The different land use categories that are also characterized as high-risk points are mainly IMS entry points or places with a high concentration of people:

- Public transportation places (bus stations, train stations, marinas/ ports)
- High population places (shopping malls, exhibition centres, sports areas, parks)
- IMS potential entry points (airports, industrial areas, end-of-life vehicles' and waste tyres' temporary storage and recycling units)

- Recognized sites with high IMS population density (cemeteries, greenhouses).

These data were mapped for each PMA and the main IMS pathways within each area were identified. This analysis was focused on *Ae. albopictus*, the IMS with the highest presence in both countries. In order to develop the *Ae. albopictus* pathways, its flying range of active dispersal (200 m) was used.

The results of the above investigation, data collection and analysis were presented in appropriate Cartographic Data [Deliverable C.2.1] supported by the relevant databases [Deliverable C.2.2].

Furthermore, for each PMA, microclimatic data were collected from the meteorological stations of the MDs. These microclimatic data include temperature, relative humidity, wind speed and direction, and rainfall.

Therefore, for the broader area of each PMA there is availability on the local meteorological characteristics, which can be combined with the IMS related geographical data. All these data can be used every time the LIFE CONOPS management plans are activated in each specific area.

Moreover, and since the main conventional biocide that is commonly used is Diflubenzuron (DFB), a thorough research regarding its properties, its toxicological profile and its application impacts was conducted [Deliverable C.2.3]. Water samples from canals collecting sewage waters from Bologna city area, were collected after the application of DFB and analysed to check for possible residual level of DFB and the expected impact on the water macrofauna for the years 2015 and 2016. The scope of this study was to measure, during summer months, the DFB concentration in rainwater collection canals as well as to evaluate the risk of DFB for aquatic fauna. The main outcome from this study was that if the pest control operators follow strictly the label dosage regarding the use of DFB, it is not considered probable that using DFB in this particular context raises significant environmental risks and involves impacts on non-target fauna present in aquatic ecosystems connected to the urban drainage network.

In addition, the possible environmental impacts deriving from the proposed application of the alternative larvicidal essential oil (EO) of *Origanum* that was developed in the context of Action B.3, was also performed. More specifically, the results of the first stage of the environmental impact assessment [Deliverable C.2.3] indicated that although the *Origanum* EO presents lower aquatic toxicity levels compared to the respective ones of DFB, its 3 main substances (**Carvacrol**, **p-cymene**, **γ-terpinene**) are not completely environmentally safe in terms of aquatic ecotoxicity. For this reason, it was proposed at the next steps of Action B.3, (a) various compositions of *Origanum* EO and (b) various concentrations of the EO in the application



solution, to be tested in order to define the ones that achieve effective control of IMS while simultaneously present minimized aquatic ecotoxicity. Indeed, that guideline was followed, and the completion of Action B.3 led to the production of the final larvicide product (CREO), which was field tested during Action B.6. CREO was assessed in terms of its environmental performance during the second stage of the environmental impact assessment, which concluded [Deliverable C.2.4] that CREO presents excellent properties in terms of mosquito's larvae confrontation (comparable to those of synthetic pesticides currently in use), while the environmental impacts of its proposed application practice are minimized in terms of aquatic ecotoxicity.

Finally, calculation and analysis of the environmental footprint of the designed and implemented IMS Management Plans, was performed for the entire LIFE CONOPS pilot implementation period [Deliverable C.2.4]. The comparison of the LIFE CONOPS Management plans performed against the so called "zero solution" which, in the context of IMS population control, is the widely used application of chemical substances (standard control measures in public and private areas). Based on this fact, it is obvious that the application of several preventive actions prior to the "Standard control measures in public and private areas" has the benefit of lower chemical substances released into the environment and subsequently lowers environmental impacts.

This Action was implemented by TERRA NOVA with the contribution of CAA, AUA and BPI.

Problems/Delays: There were no delays regarding the implementation of the specific Action.

Deliverables: Deliverable C.2.1 Spatial Mapping

Deliverable C.2.2: Database of Environmental and Meteorological Data

Deliverable C.2.3: 1st Report of the Assessment of the environmental impacts from the pilot implementation of the management plans

Deliverable C.2.4: Final report of the Assessment of the environmental impacts of the management plans

### **Action C.3: Assessment of the socio-economic impacts of the management plans**

Foreseen start date: 01/01/2016      Actual start date: 01/04/2015

Foreseen end date: 30/09/2018      Actual end date: 30/09/2018

Achievements and description of the progress: The scope of Action C.3 is the evaluation of the economic effectiveness of the proposed management plans to control the IMS problem.

The key variable for such an evaluation is the potential benefits induced by the management plans to trace and elucidate these benefits.

The scientific team of LIFE CONOPS selected a variety of methods for a holistic estimation of the socioeconomic aspects of the problem both from a citizens' and an experts' point of view.

The synthesis of results produced by the current report are expected to act as a guide for the estimation of the effectiveness of present control and management strategies and the examination of possible societal welfare in the design of future control strategies.

One of the main methods used by for the Greek case study was that of the “Choice Experiment Method” for the elicitation of household preferences to control IMS, along a range of other available methods. The advantage of using the Choice Experiment Method is actually the elicitation of household preferences along various attributes. Specifically, the attributes examined are:

- IMS vs Native Species
- Nuisance vs Disease Levels
- Daily Nuisance vs Nuisance during the Night
- Willingness To Pay among different suggested programs

A web-based questionnaire through a popular meteorological data website ([www.meteo.gr](http://www.meteo.gr)) took place from September to October 2016 with a total of approximately 1,200 answers. The scope of this questionnaire was the validation of specific parameters regarding the private prevention costs for IMS and the preferences for the application of improved mosquito control programs in a national level. In addition, the economic evaluation of the proposed management plans was also evaluated through “a stakeholders’ opinion” survey. This qualitative survey has been designed for the evaluation of the socioeconomic impacts of the management plans by key stakeholders such public policy makers, medical practitioners, public health experts and regional delegates. The implementation of a pilot questionnaire for stakeholders for the Greek case took place in May 2016. In total a pool of 81 experts were approached for the (58) Greek and (23) Italian cases. This survey was completed in May 2017. Several smaller scale questionnaires have been implemented in both countries and are presented in the final integrated report. The economic efficiency of improved management plans was also evaluated in the frames of different Control Cost Options based on different disease outbreak scenarios. Briefly state the main results.

[CONFIDENTIAL INFORMATION ERASED]

Based on the findings on the stakeholders' survey it was found that Health Impacts are regarded as more important than nuisance impacts in Greek case, while the overall harmfulness of

mosquitoes appears to be the most important factor for Italian Stakeholders, while diseases from invasive species were considered a serious threat in both cases. The results of the online survey (meteo.gr) showed that nuisance from mosquito: (a) is significant all over Greece, although with some regional differences, thus indicating areas of higher priority for future policy actions (b) is similar for both invasive and native species and (c) is the main reason for taking individual prevention measures.

[CONFIDENTIAL INFORMATION ERASED]

Problems/Delays: A 6 months (prior to initial start date) extension has been demanded (from 01/04/2015 to 30/09/2015), in order to keep the design /data of the C.3 survey in line with the results of Action A.3. This extension did not require any modification on the budget and other details of Action C3. This was also beneficial for the Action B.6 (also starting on April 1<sup>st</sup>, 2015). Consequently, the findings of Action A.3 and a longer duration for Action C.3 (without affecting its endpoint) has been useful to better analyze the correlation between Actions B.6 and C.3.

Deliverables: In Annex C.3.1 the final questionnaires are presented.

## **5.1.4 Action E: Project management and monitoring of the project progress**

### **Action E.1: Project management by BPI**

Foreseen start date: 01/07/2013      Actual start date: 01/07/2013

Foreseen end date: 30/11/2018      Actual end date: 30/11/2018

Achievements and description of the progress: Detailed description of the project management procedures are presented in the Administrative and Financial part of this report (Annex E.1.1 to Annex E.1.8). The project management included discussion and cooperation between partners about documents' completion, milestones, financial issues and project deliverables as well as several working meetings between the beneficiaries to prepare and organise the progress of the project. In particular for Action B.1 and B.2, where we were faced with the major delays, communication between beneficiaries involved in their implementation was constant aiming at evaluating the operational performance of the MDs and at investigating alternative interventions to optimize their performance.

### **Action E.2: Monitoring of project progress**

Foreseen start date: 01/07/2013      Actual start date: 01/07/2013

Foreseen end date: 30/11/2018      Actual end date: 30/11/2018

Achievements and description of the progress: A Quality Assurance/Quality Control (QA/QC) System was developed in order to monitor the progress of the implementation of the LIFE CONOPS project. Towards this direction a QA/QC manual was developed, and a QA/QC Committee was formed consisting of one representative by each project beneficiary. Its role is to monitor and control the progress of each project Action by compiling and evaluating every nine months the corresponding report. The report evaluation outcomes –especially early warnings- are being transferred to the scientific, technical and financial committees for further actions (if necessary). In total 7 reports were produced by the QA/QC Committee. The members of this committee, headed by the project manager-coordinator (Dr Antonios Michaelakis), are listed in Table 2. The most recent reports, 6<sup>th</sup> and 7<sup>th</sup>, are included in the Deliverables E.2.7 and E.2.8, respectively.

Deliverables: E.2.7 is the 6<sup>th</sup> QA/QC Report and E.2.8 is the 7<sup>th</sup> QA/QC Report. The reports were developed by BPI with the contribution of all beneficiaries that provided the required data of their organisations.

[CONFIDENTIAL INFORMATION ERASED]

### **Action E.3: Networking activities with other relevant EU projects**

Foreseen start date: 01/07/2013      Actual start date: 01/07/2013

Foreseen end date: 30/11/2018      Actual end date: 30/11/2018

Achievements and description of the progress: during this Action, both Greek and Italian beneficiaries have performed a thorough search in order to identify projects relevant to LIFE CONOPS. A total of sixteen (16) LIFE, FP7 and other EU projects have been identified as linked to our project and LIFE CONOPS has established collaboration with most of them and this effort was reinforced through the dissemination activities [e.g. LOVCEN (Montenegro), LIFE E.I.D (LIFE99 ENV/F/000489, France), AIM COST (many partners, Italy coordinator), Infravec2 (many partners, France coordinator), etc]. Moreover, Prof. R. Bellini (CAA) and Dr A. Michaelakis (BPI) participated in many expert panels in EU where they presented the LIFE CONOPS management plans (e.g. Zikalliance, IAEA etc). In addition, the LIFE CONOPS team participated in various conferences and other meetings for the presentation of the project's methodology and results. It was also reinforced by the participation of numerous stakeholders in the LIFE CONOPS workshops. A specific connection was established with the International Atomic Energy Agency (IAEA), which enabled the implementation of the Sterile Insect

Technique in Vravrona in 2018 and will enable the pilot application of the SIT in 2019 (after-LIFE activities). Moreover, because of the previous networking activities, LIFE CONOPS established collaboration with COST Action CA17108 in which the major researchers in mosquito control are included. Dr A. Michaelakis is an MC member and the coordinator in WG2 (three WGs, WG2 – Conventional and Innovative Control Tools). Furthermore, as a result of the collaboration and data exchange with the Pest Control Association in Greece, LIFE CONOPS team managed to ensure the implementation of the door-to-door activity in the Municipality of Moschato as part of a public tender for the first time in Greece. The communication and exchange of knowledge continued until the end of the project and will be continued after the end of the project, as an after-LIFE activity. This new activity led to the reward of the P. Faliro Municipality with the “Best City Award” in category “Response actions and studies to protect citizens from emergencies ”.

Problems/Delays: No problems or delays occurred. Deliverables: Deliverable E.3.3 describes the networking activities of LIFE CONOPS.

#### **Action E.4: Development of project’s After-LIFE Communication Plan**

Foreseen start date: 01/07/2013      Foreseen end date: 31/12/2017

Foreseen end date: 30/11/2018      Actual end date: 30/11/2018

Achievements and description of the progress: The main priority of the communication activities carried out during the project were: i) Information activities targeting at health stakeholders, decision makers and policy makers ii) Information activities targeting at the general public, iii) Networking and result dissemination activities targeting at competent authorities, other programmes, organizations, scientific community, etc.

The After-LIFE Communication Plan was designed based on the experience gained during the implementation of the project and the more than effective dissemination activities that took place throughout the project. The detailed After-LIFE Communication Plan is described in Deliverable E.4.1. The After-LIFE includes updating and maintaining of all communication channels of the project (website, Facebook, twitter, mosquito weather forecast, etc.), participation in national and international conferences and workshops and publication of newly acquired results from the implementation of the project to peer review journals. Moreover, the operation of 3 Monitoring Devices in Benaki Phytopathological Institute (Kifissia, Greece), in COSCO (Piraeus, Greece) and CAA (Crevalcore, Italy) will continue for at least 3 years after the end of the project. In this way, data monitoring will continue for long-term data analysis.

As far as the stakeholders' engagement is concerned, the Greek Pest Control Association, SEAME, has accepted to become a LIFE CONOPS communication channel by re-presenting all videos, articles, news, etc. from the LIFE CONOPS website to their website.

Apart from the abovementioned actions, important efforts were made towards the replicability of methodologies included in the LIFE CONOPS management plans, which are legally in force since September 2016 with a Ministerial Circular. Towards this end, the sterile insect technique (SIT), is planned to be repeated in 2019 in Vravrona for the whole mosquito period (from April to October). SIT is an environmentally friendly insect pest control method involving the mass-rearing and sterilization, using radiation, of a target pest, followed by the systematic area-wide release of the sterile males in defined areas, where they mate with wild females resulting in no offspring and a declining pest population. (IAEA website, <https://www.iaea.org/topics/sterile-insect-technique>).

Furthermore, replication of the door-to-door methodology is planned to new geographical areas.  
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Finally, in LIFE CONOPS website we created webpages (sections) which are dedicated to activities that could be useful to all stakeholders such as door-to-door and entomological surveillance activity (<http://www.conops.gr/door-to-door/>, <http://www.conops.gr/protaseis-gia-entomologiki-epitirisi/>). The goal of these sections is to provide them with a better understanding of these activities' concepts, while providing useful tips.

Problems/Delays: No problems or delays occurred. Deliverables: Deliverable E.4.1 describes the detailed After-LIFE Communication Plan of LIFE CONOPS.

#### **Action E.5: Monitoring of project Carbon footprint**

Foreseen start date: 01/07/2013      Actual start date: 01/07/2013

Foreseen end date: 30/11/2018      Actual end date: 30/11/2018

Achievements and description of the progress: Scope of Action E.5 is the monitoring of the carbon footprint of the LIFE CONOPS Project. On March 31, 2014 the 1<sup>st</sup> Carbon Footprint Report was delivered according to the project timetable. In order to estimate, calculate, monitor and reduce if possible, the Greenhouse Gas emissions resulting from the implementation of the project, the steps followed are:

- i) Ensuring the support of all participating Beneficiaries,
- ii) Collection of primary quantitative data related to recognized sources of direct and indirect emissions required for the calculation of carbon footprint, such as:

- a. Energy use for heating and cooling purposes of the beneficiaries' premises, for lighting purposes and for computer use
  - b. Transportation emissions related to the project
  - c. Emissions from organization of LIFE CONOPS events
- iii) Calculation of the carbon footprint resulting from the project's implementation and compilation of the Carbon Footprint Report. The Carbon Footprint Committee through the coordinator communicates these results to all participating Beneficiaries.

The 5th Carbon Footprint Report (Deliverable E.5.3), the 6th Carbon Footprint Report (Deliverable E.5.4) and the 7th Carbon Footprint Report (Deliverable E.5.5) are submitted with the current Final Report. These reports were developed by TERRA NOVA with the contribution of all beneficiaries that collected the required data regarding their organizations.

In order to calculate the overall carbon footprint during the LIFE CONOPS implementation, all calculations from the 7 Carbon footprint reports implemented during the project were taken into account. For the evaluation of the carbon footprint, the above-mentioned calculations were compared to the respective carbon footprint anticipated in the approved proposal of LIFE CONOPS project. This comparison revealed that from the implementation of the LIFE CONOPS project a total amount of **11.36 tCO<sub>2</sub>e were not emitted to the atmosphere** compared to what was anticipated in the approved proposal (Analytical data can be found in the Deliverable E.5.5).

Problems/Delays: No problems or delays occurred. Deliverables: i) 5<sup>th</sup> Carbon Footprint Report (Deliverable E.5.3), ii) 6<sup>th</sup> Carbon Footprint Report (Deliverable E.5.4) and iii) 7<sup>th</sup> Carbon Footprint Report (Deliverable E.5.5).

### **Action E.6: Audit of project financials**

Foreseen start date: 01/07/2013	Actual start date: 01/07/2013
Foreseen end date: 30/11/2018	Actual end date: 30/11/2018

Achievements and description of the progress: Two Independent Auditors, one for Greek beneficiaries and one for the Italian beneficiaries, were appointed by BPI and CAA respectively to verify the compliance of all beneficiaries' expenditures documentation with the national legislation and the respective accounting rules, the Grant Agreement and the LIFE+ Common Provisions. The conclusions of the Italian Auditor were incorporated to the final audit report submitted with Deliverable E.6.1.

Problems/Delays: No problems or delays occurred. Deliverables: Deliverable E.6.1.

## 5.2 Dissemination actions

### **5.2.1 Objectives**

According to the project proposal, the implementation of the dissemination actions was expected to produce the following quantified results per dissemination Action, throughout the project duration:

#### **Action D.1: Creation of project logo**

- Creation of CONOPS's logo

#### **Action D.2: Development, launching and maintenance of project's website**

- LIFE CONOPS's website launched and being continuously updated.
- At least 100 visitors per month
- For the projects' Facebook the number of "likes" are expected to be at least 1,000
- For the twitter account the number of followers is expected to be at least 500.

#### **Action D.3: Dissemination of project's progress and results**

- Organization of 2 Stakeholder workshops (Greece, Italy) at the beginning of the project
- Organization of 1 Mid-Term workshop (Greece).
- Organization of 2 Final conferences (Greece, Italy).
- At least 20 meetings (in both countries),
- At least 14 articles in newspapers-journals,
- 4 scientific journals,
- At least 10 interviews in TV and radio stations.

#### **Action D.4: Development of project's notice boards**

- At least 15 Informative Boards

#### **Action D.5: Development of Layman's Report**

- Layman's Report of the LIFE CONOPS project in Greek, Italian and English available and downloadable by the project's website.

### **5.2.2 Dissemination: overview per activity**

For each dissemination action (Action D.1, Action D.2, Action D.3, Action D.4 and Action D.5) a description in quantifiable terms and a comparison to the expected results, as defined in the proposal, is presented hereinafter.



In Action D.3, which is the core dissemination Action of LIFE CONOPS, an extended summary of the dissemination activities is presented.

#### **Action D.1: Creation of project logo**

Foreseen start date: 01/07/2013      Actual start date: 01/07/2013

Foreseen end date: 30/09/2013      Actual end date: 30/09/2013

Achievements and description of the progress: The Action was already successfully finished.

#### **Action D.2: Development, launching and maintenance of project website**

Foreseen start date: 01/07/2013      Actual start date: 01/07/2013

Foreseen end date: 30/11/2018      Actual end date: 30/11/2018

Achievements and description of the progress: The design of the website and the development of its content began in August 2013. The website was launched on the internet in three languages (Greek, English, and Italian) in September 2013 ([www.conops.gr](http://www.conops.gr)). Simultaneously, links to the LIFE CONOPS website were inserted in beneficiaries' websites ([www.bpi.gr](http://www.bpi.gr); [www.caa.it](http://www.caa.it); [www.uehr.gr](http://www.uehr.gr); [www2.ipta.demokritos.gr/](http://www2.ipta.demokritos.gr/), [www.terranova.gr/](http://www.terranova.gr/), [www.onexcompany.com/](http://www.onexcompany.com/)). The LIFE CONOPS website offers information about the project and its actions, the beneficiaries and the project activities, such as events, workshops and conferences that are held during the implementation of the project. In addition, it contains useful links, and links to the official Facebook page and Twitter account of the project. The website includes a link to the “timeline of the project” which is an add-in that gives short and quick information about the dissemination activities of LIFE CONOPS. Another add-in was added in LIFE CONOPS website, which offers a link to the LIFE CONOPS' mosquito weather forecast which was incorporated into METEO weather forecast ([www.meteo.gr](http://www.meteo.gr)) since June 2014.

Furthermore, a particular area was dedicated to the detailed presentation of **LIFE CONOPS management plans** (<http://www.conops.gr/management-plans/>) and exhaustive instructions for the **entomological surveillance with the use of ovitraps** (<http://www.conops.gr/management-plans/ovitraps/>) with videos and images.

During the last year of the project and after the successful implementation of the methodologies suggested by LIFE CONOPS management plans, namely **door-to-door** (<http://www.conops.gr/door-to-door/>) and **Sterile Insect Technique** (<http://www.conops.gr/sit-technique/>), details and information about them were uploaded to inform stakeholders who are interested in adopting the methodologies. Indeed, the result of the uploads was immediate and various municipalities contacted LIFE CONOPS to get further

information especially about door-to-door methodology. This was particularly important and useful for the successful design of LIFE CONOPS after-LIFE Communication Plan.

Until August 2015, almost 52,250 unique visitors had been recorded. The website features the budget, EC contribution and an explicit acknowledgement to the support of the LIFE+ financial instrument of the EU. More information about the website structure and analytics regarding the audience overview are presented in Annex D.2.

Until the end of November 2018, about 126.991 sessions of website of LIFE CONOPS project had been recorded. Based on the results of the visitors' analytics, visits occurred more frequently in July 2014 and September 2014. This is obviously related to the release of mosquitoes' activity application (on July 2014) and to the broadcast of LIFE CONOPS video as social message after the approval by the Greek National Council for Radio and Television (NCRTV) (September 2014), respectively.

It is worthwhile to mention that approximately 92% of total visits are unique (new visitors). Approximately 245.535-page views were recorded with average visit duration about 2 minutes each. All the analytics data are presented in Appendix (as provided by Google Analytics).

Until recently (November 2018), the LIFE CONOPS Facebook profile had already received about 695-page likes, while on Twitter the total number of tweets and followers were 610 and 211, respectively. Facebook and Twitter profiles are constantly updated and are the main communication channels, along with LIFE CONOPS website, for the news of the projects. It is worth noting that during the award ceremony of the Best City award there was a live video presenting the award given to the Municipality of Palaio Faliro for the implementation of door-to-door methodology under the guidance of LIFE CONOPS team.

Problems/Delays: No problems or delays occurred. Deliverables: Deliverable D.2: LIFE CONOPS website designed, developed and launched (Annex D.2). The website is maintenance by BPI with the monthly contribution of all beneficiaries.

### **Action D.3: Dissemination of project progress and results**

Foreseen start date: 01/07/2013      Actual start date: 01/07/2013

Foreseen end date: 30/11/2018      Actual end date: 30/11/2018

Achievements and description of the progress: This Action aims at the implementation of activities that will ensure the dissemination of the project objectives, actions and results to relevant stakeholders, scientific community and public in general. The extensive informative campaign which started with the beginning of the project, continued throughout the whole duration of project implementation. It is noteworthy, that the dissemination activities in most

cases surpassed the target numbers foreseen in the proposal until the end of the project. In particular, LIFE CONOPS, after the progress report, organized two final Stakeholder Workshops (one in Chania, Greece and one in Bologna, Italy). The Benaki Phytopathological Institute and the Regional Unit of Chania under the auspices of the Municipality of Chania organized the **final Stakeholder Workshop (in Greece) of the Project LIFE CONOPS (LIFE12 ENV/GR/000466)** on the 15<sup>th</sup> of December 2017 over “Managing Mosquitoes Causing Diseases in the Mediterranean Basin” (Deliverable D.3.4). Representatives of the LIFE CONOPS project presented the project’s results while representatives of other public health institutes in Greece, Italy and Cyprus attended the workshop as key-lecturers on specific issues regarding mosquito management and disease control. For more details and participants’ lists, agenda, invitation, project presentations please see Annex D.3.4 and the relevant Appendix.

The city of Chania was chosen to host the final Stakeholder Workshop in Greece for the following reasons: a) LIFE CONOPS conducted an entomological surveillance in the island of Crete, in 2015, resulted in the 1<sup>st</sup> record of established population of *Ae. albopictus*; b) in Chania we conducted an entomological surveillance of *Ae. albopictus* in the airport of Chania and nearby areas evaluating different surveillance methods, hatching protocols and the MD; c) after the final workshop, BPI hosted the annual consortium meeting of the IAEA project “Establishing Genetic Control Programmes for Aedes Invasive Mosquitoes- RER5022”.

In total 108 persons attended the workshop (at least 200 persons were expected to attend the workshop however the number of attendees was considerably high if we take into account that the workshop was not held in Athens). It is important that the majority of stakeholders from the Municipalities in the areas and the public health services were highly represented. The implementation of an intensive *Aedes* surveillance in Chania and the organization of the final workshop (organized in collaboration with the Region of Crete, Regional Unit of Chania and under the auspices of the Municipality of Chania) resulted to change the mentality of policy makers in Prefecture of Crete and in 2018 they implemented an improved mosquito control program that includes also several management activities (not only the use of biocides).

**The final Stakeholder Workshop (in Italy)** of the Project LIFE CONOPS (LIFE12 ENV/GR/000466) was held in Bologna, Italy, on the 16<sup>th</sup> -17<sup>th</sup> of April 2018. The workshop was organized by the “Servizio Prevenzione collettiva e Sanità Pubblica Regione Emilia-Romagna”, AUSL della Romagna and Centro Agricoltura Ambiente “G.Nicoli” on “Sustainable invasive mosquitoes management following the LIFE CONOPS initiative” (Deliverable D.3.1). For more details and participants’ lists, agenda, invitation, project presentations please see Annex D.3.1 and the relevant Appendix. Representatives of the LIFE

CONOPS project presented the project's results to a large audience where the most important EU sponsored projects dealing with vectors and vector borne diseases were also represented. Around 120 persons attended the Workshop (at least 200 persons were expected to attend the workshop). Although we did not manage to reach the target of the 200 attendees, the number achieved, and the quality of attendees was still high enough.

**Sixty-two (62) general public articles** were published in national and local press (at least 14 articles were foreseen in the proposal throughout the project duration), **twenty-four (24) press releases** were circulated, **and thirteen (13) scientific articles** were published by LIFE CONOPS team (4 were foreseen in the proposal throughout the project duration). It is noteworthy that apart from the thirteen already accepted and published scientific papers, there are at least 7 scientific articles which are currently being prepared to be submitted for publication in peer-review-journals, not to mention the integrated LIFE CONOPS management plans which was the result of consultation with the European experts in the area of mosquito control and were submitted for publication in a peer-review journals (Travel Medicine and Infectious Disease journal, in June 2019 still under review) By the end of the project's duration, **twenty (20) interviews** were given in TV and radio stations (at least 10 were foreseen in the proposal throughout the project duration). In particular, thirteen of them were interviews in the TV and the rest of them in radio stations. All published articles and interviews are presented in the Deliverable D.3.2, while the published articles and videos after the progress report are given only in electronic form in the Appendix of the Deliverable. In addition, a total number of **78 presentations in conferences**, workshops, meetings and other press events were released during LIFE CONOPS progress (Deliverable D.3.3). It is reminded that 20 were foreseen in the proposal. All dissemination material is submitted in electronic form.

Furthermore, the 39'' video, which was already produced before the Mid-term report, with information about mosquitoes and protection measures against them currently counts **more than 44.400 views** in YouTube. LIFE CONOPS video has also been approved by the Greek National Council for Radio and Television (NCRTV) as social message and was broadcasted on October 2014, September and October 2015 – 2016-2017 and 2018. A second video was produced by **LIFE CONOPS team in collaboration with Eugenides Foundation** and it is a short educational video about mosquitoes. The particular video currently counts in VIMEO 400 views (LIFE CONOPS in VIMEO, <https://vimeo.com/user23595598>). LIFE CONOPS team produced a third video for the DVD production foreseen in the proposal. The video has a duration of 3'17'' and presents in animation the scope, the objectives and the results of LIFE CONOPS. All videos are submitted with Deliverable D.3.5.

Moreover, another dissemination activity took place in Northern Italy. A large number of placemats (42.500) was distributed in restaurants of the area (Forlì – Cesena – Rimini – Ravenna). The placemats contained information about mosquitoes' prevention. The distribution was implemented on July – August and September 2016.

An original copy of the placemat is given with the Final Report, while a number of photos from the distribution of the placemats are submitted in Deliverable D.3.2.



**Figure 11:** Placemats distributed in Romagna area



**Figure 12:** Screenshots from the three videos produced and distributed during LIFE CONOPS (Deliverable D.3.5)

The extensive dissemination activities throughout the whole duration of the project boosted publicity and recognition of the LIFE CONOPS project. This, in combination with the innovative methodologies implemented in Action B.6 as part of the implemented LIFE CONOPS management plans, resulted in the public appreciation of the LIFE CONOPS family. As a result, the Municipality of Palaio Faliro and LIFE CONOPS won the Best City award for the implementation of door-to-door methodology.

For LIFE CONOPS team, the award was received from Dr Panos Mylonas, Deputy Director of Benaki Phytopathological Institute in the ceremony which took place on the 30<sup>th</sup> of November 2018 (<https://bit.ly/2UqgClF>). Apart from the Best City award, LIFE CONOPS gained another distinction and it was chosen to be presented as the project of the month February 2019, in the Greek LIFE Task Force website (<https://bit.ly/2TCuEDs>).



**Figure 13:** Best City award received by LIFE CONOPS and the Municipality of Palaio Faliro for the door-to-door initiative.

Problems/Delays: No problems or delays

occurred. Deliverables: Deliverable D.3.1: Final Conference in Italy. Deliverable D.3.2: Published articles, press releases, interviews. Deliverable D.3.3: Presentations in workshops and conferences. Deliverable D.3.4: Final conference in Greece. Deliverable D.3.5: Project's DVD production.

#### **Action D.4: Development of project notice boards**

Foreseen start date: 01/07/2014, 01/04/2015	Actual start date: 01/07/2014, 01/04/2015
Foreseen end date: 30/09/2014, 30/06/2015	Actual end date: 30/09/2014, 30/06/2015

Achievements and description of the progress: The noticeboards were designed and produced as foreseen for the Pilot Implementation areas. The noticeboards were placed at the premises of all partners. The noticeboards were placed only in four out of the 12 Pilot Implementation areas, due to the sensitive character of the Pilot Implementation Areas. Thus, in the case of ports and airport, the Directors were negative in placing the noticeboards in their premises in the fear of raising concern about diseases in their areas. They thought that it



*Figure 14: The noticeboard in the Public Health Department in Chania*



would be a negative advertisement for the ports and airports in the middle of an increasing touristic period. Thus, LIFE CONOPS team made every possible attempt to find alternative places for the noticeboards. For this reason, the Metropolitan Park Antonis Tritsis (Attica Greece) was chosen, the Chemistry Laboratory of the Agricultural University of Athens and the Public Health Department in Chania. The alternative places were chosen because they are easily accessible by public, and they have usually a large number of visitors/students.

Problems/Delays: No problems or delays occurred. Deliverables: The Deliverable D.4.1 is submitted.

### **Action D.5: Development of Layman's Report**

Foreseen start date: 01/10/2013      Actual start date: 01/10/2013

Foreseen end date: 30/11/2018      Actual end date: 30/11/2018

Achievements and description of the progress: Following the extensive dissemination

activities throughout the whole project's duration, the Layman's report was designed in such a way as to communicate the objective, actions and results of the project to a wide audience in a comprehensive way. The results of LIFE CONOPS are rather technical and therefore require

adequate attention to

describe with simple

language some

methodologies like the

Sterile Insect Technique.

For this reason, images and

explanatory definitions

have been added to make

the language fluent and

more comprehensive. In

practice, a first draft was

designed and distributed to

the beneficiaries of the project. The first draft of the Layman's report consisted of the

following sections: The problem, The challenge, Methodology, Innovation, Results, Benefits,

Publicity, Networking, Contact. All sections were written in simple language, aiming at

informing the readers, who are not familiar with the scientific terminology of the subject. The

1<sup>st</sup> draft of the Layman's report was distributed also to key stakeholders in order to have a

broad review about the context. Following the assessment of the comments received from



**Figure 14:** Layman's report

both the beneficiaries and the stakeholders, LIFE CONOPS team produced the final version of the Layman's report. After the finalization, the report was translated in Greek and Italian and uploaded in LIFE CONOPS website. The first version of the Layman's report is submitted with Deliverable D.5.1 and the final version of the Layman's report is submitted with the Deliverable D.5.2. An original copy of the Layman's Report is given with the Final Report.

Problems/Delays: No delays occurred.

Deliverables: Deliverable D.5.1: The 1<sup>st</sup> version of the Layman's Report. Deliverable D.5.2: Final version of the Layman's Report.

### 5.3 Evaluation of Project Implementation

The project's main objectives were on one hand to develop a network of 12 prototype devices and also to design and implement the integrated management plans specifically dedicated to the IMS. In order to achieve these results, a number of Actions and Tasks have been implemented.

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### 5.4 Analysis of long-term benefits

#### Environmental benefits

LIFE CONOPS initiated in Greece and strengthened in Italy, the systematic entomological surveillance for *Ae. albopictus* and other IMS. Before 2014, only sporadic entomological data were collected in Greece depending on the imported CHIKV and DENV human cases detection. The availability of more standardized field data serves the adoption of more cost-effective mosquito control methods thus reducing the waste of insecticides and better targeting their use. Moreover, thanks to the extensive network of ovitraps, *Ae. cretinus* was found again in Greece. *Ae. cretinus* is a species that used to be present in Greece but was thought to be eliminated due to the competition by the new invasive *Ae. albopictus*.

For three consecutive monitoring periods, the MDs installed in selected points of entries in Greece and Italy, collected samples which were tested afterwards for their virus status. Thus, decision makers in Greece and Italy consulted LIFE CONOPS team regularly (e.g. imported cases of DENV or CHIKV) on the vector presence and population density along with their virus status. As a whole, the installation of the MDs leads to increased capacity for early detection of new IMS in points of entry allowing the adoption of more cost-effective measures. Early



detection of IMS and entomological surveillance, results in the overall reduction in the use of biocides (insecticides and/or repellents) through a better understanding of the IMS phenomenon and improved capacity to prevent the problem from becoming an area-wide issue.

On the other hand, the implementation of LIFE CONOPS management plans and in particular the implementation of the door-to-door strategy resulted in the increase of citizens' awareness on the issue of IMS and the adoption of sustainable practices for their management. Moreover, the implementation of the Sterile Insect Technique (SIT strategy) in Vravrona has resulted in a significant decrease of the *Ae. albopictus* population and for this reason the Municipality in agreement with the citizens requested from BPI to continue the SIT strategy (in 2019).

Furthermore, risk analysis on the current and future climatological conditions is delivered and presented in appropriate maps for the most important IMS in Greece and Italy and socioeconomic evaluation of the IMS problem and the management plans provides useful data to national authorities in order to acquire the required resources in case of emergency.

Moreover, several Mediterranean culinary plants were selected as subject of a bio-prospecting endeavor aiming the development of novel biodegradable mosquitos' repellents and larvicidal of natural origin. Their activity was evaluated in large scale field tests, performed for the first time in the terms of LIFE CONOPS project implementation, highlighting their potential use in future as "natural biocides" (biocides derived from plants).

LIFE CONOPS opened the way to the application of IMS active surveillance with the aim of early detecting harmful species such as *Ae. aegypti* in Southern Europe. The mosquito *Aedes aegypti* is the primary species responsible for transmitting viruses such as Zika, dengue, chikungunya and yellow fever between people. Based on ECDC, it is nowadays one of the most widespread mosquito species globally. Although its global establishment is currently restricted due to its intolerance to temperate winters, over the past 25 years there has been an increase in its distribution worldwide. After the LIFE CONOPS we are now better fitted and prepared in case of introduction of *Ae. aegypti*, to early detect it and to apply suppression actions, such as door-to-door and SIT, aimed to the prompt elimination before it spread onto a large area. In this case the advantage for the communities will be huge in terms of environmental impact, economy and life quality.

#### Long-term benefits and sustainability

The operation of the Monitoring Devices in Benaki Phytopathological Institute (Kifissia, Greece), in COSCO (Piraeus, Greece) and CAA (Crevalcore, Italy) will continue for at least 3 years after the end of the project. In this way, data monitoring continues for long-term data

analysis being thus an asset in the decision-making process. Moreover, surveillance and monitoring is a useful tool for the reduction of biocides as set out in the EU Regulation 528/2012.

The LIFE CONOPS management plans were legally in force before the end of the project (since September 2016) when a Circular from the Hellenic Ministry of Health for the adoption of LIFE CONOPS' Management Plans towards the confrontation of CHIKV, DENV and Zika virus in Greece, was issued. Among other, the implementation of LIFE CONOPS management plans contributes to the reduction of biocides (with the adoption of other actions for the elimination of breeding sites, such as door-to-door).

Moreover, the extensive dissemination activities throughout the whole duration of the project gave a great visibility to the project and our website is regularly visited by stakeholders who need information on IMS management. Furthermore, the Greek Pest Control Association (SEAME), agreed to use the LIFE CONOPS website as a communication channel after the end of the project by re-presenting all videos, articles, news, etc. via links from LIFE CONOPS website to their website.

Thanks to LIFE CONOPS, the surveillance of IMS has gained awareness in several Mediterranean countries such as Albania, Montenegro and Turkey, where actions are under development to increase the capacity to early detect and eliminate IMS.

#### Replicability, demonstration, transferability, cooperation

Important efforts were made by LIFE CONOPS team towards the replicability of methodologies included in the LIFE CONOPS management plans, which are legally in force. Towards this end, SIT, is planned to be repeated in 2019 in Vravrona area. Furthermore, replication of the door-to-door strategy is planned to new geographical areas in Greece and Italy.

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Furthermore, training seminars to professionals will continue in Greece and Italy with the cooperation of the Greek Pest Control Association (SEAME) and the Associazione Nazionale delle Imprese di Disinfestazione (ANID), respectively.

#### Best Practice lessons

The MDs were installed in the selected points of entry e.g. airports, ports, etc. For this reason, authorization licenses were issued due to the sensitive character of the areas. Each time the MD user had to visit the MD to collect samples or resolve a problem, a series of procedures, usually time consuming, had to take place for the issuing of the entry license. Although it was very

important to place the MDs at the particular points of entry, it would be more convenient to place them right beside the sensitive areas, possibly in vegetated shadow position, in order to avoid losing time in authorization processes.

#### Innovation and demonstration value

The following results of the project have gained a lot of attention by media and are considered as the main innovation aspect of the project:

1. **A prototype Monitoring Device (MD)** was designed and 12 MDs were manufactured and tested so as to play an important role in assisting efficiently and cost effectively the surveillance of IMS in high risk areas. Its main innovation aspects are:
  - a. continuous operation for a period of at least 1 month (one sample/day) or 95 sampling periods (on-demand)
  - b. assisted collection of mosquitoes through a continuous air-flow
  - c. equipped with the option of carrying appropriate attractants (lactic acid, CO<sub>2</sub>)
  - d. light weight sampling pot manufactured by a recyclable material
  - e. highly durable in extreme climatic conditions
  - f. remote monitoring, control and programming of its operation
  - g. preservation of the collected mosquitoes in temperature ranging from 0°C to 4°C
  - h. equipped with a meteorological station for the simultaneous monitoring of the existing climatic conditions.
2. **LIFE CONOPS integrated management plans** offer a comprehensive practical technical guideline to assist local authorities in organizing their vector control activities. LIFE CONOPS management plans are currently legally operating in Greece. Local authorities, that are responsible for organizing the vector control activities, implement LIFE CONOPS management plans (via the ministerial circular) for all imported cases (Vector-borne-Diseases from IMS). In Italy, improvements of the previously adopted mosquito control measures have been obtained throughout the LIFE CONOPS project such as the implementation of the door-to-door strategy, the quality control on the mosquito control treatment following imported cases of vector-borne-diseases and the evaluation of environmental impact of Diflubenzuron (most commonly used insecticide).
3. **Implementation of Sterile Insect Technique (SIT)** against the Asian tiger mosquito (*Ae. albopictus*). In autumn 2018, **for the first time in Greece**, the SIT pilot trial was launched in the area of Vravrona (Municipality of Markopoulo). The whole philosophy of SIT is based on the release of sterile male insects in order to halt the reproduction

process of the targeted mosquito species. The method of releasing sterile insects (included in LIFE CONOPS management plans) was combined with other mosquito management methods developed within LIFE CONOPS. In the case of Vravrona, the continued release of such sterile males in combination with other management methods eventually led to a reduction of *Ae. albopictus* population.

#### Long term indicators of the project success

The adoption of the door-to-door strategy by more Municipalities than the four which have already accepted and the development of SIT as an operational available tool will be a further success for the project. BPI will continue the collaboration with Municipalities and Prefectures in order to “transfer knowledge” for this new strategy and become part of mosquito control programs in Greece.

The After-LIFE Communication plan is presented in summary in paragraph “Action E.4: Development of project’s After-LIFE Communication Plan” and in detail in Annex E.4.1.

## 6. Comments on the financial report

### 6.1. Summary of Costs Incurred

PROJECT COSTS INCURRED				
	Cost category	Budget according to the grant agreement*	Costs incurred within the project duration	%**
1.	Personnel	1.870.695,00	2.207.280,43	117,99
2.	Travel	374.371,00	195.332,17	52,18
3.	External assistance	62.500,00	81.671,30	130,67
4.	Durables: total <u>non-depreciated</u> cost	330.000,00		
	- <i>Infrastructure sub-tot.</i>			
	- <i>Equipment sub-tot.</i>	56.000,00	2413,02	5,60
	- <i>Prototypes sub-tot.</i>	274.000,00	215.146,28	78,52
5.	Consumables	127.850,00	112.368,38	87,89
6.	Other costs	30.200,00	45.713,06	151,37
7.	Overheads	193.698,00	200.194,7251	103,36
	<b>TOTAL</b>	<b>2.989.314,00</b>	<b>3.060.138,50</b>	<b>102,39</b>

(\*) If the Commission has officially approved a budget modification indicate the breakdown of the revised budget. Otherwise, this should be the budget in the original grant agreement.

(\*\*) Calculate the percentages by budget lines: e.g. the % of the budgeted personnel costs that were actually incurred

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### 6.2. Accounting system

The accounting procedures permit a direct reconciliation of the costs and revenue declared in respect to the project with the corresponding supporting documents.

For that purpose, all beneficiaries established project codes in their analytical accounting system which operates as a unique Cost Center for the project and maintain separate accounting register for the economic monitoring, management and fulfillment of the project. The original documents for the expenditures of each partner make clear reference to the project. Where it is possible the reference to the project is made by the issuer, otherwise it is stamped with the acronym of the project and the project number. Clear reference to the project is also made in all

written agreements and contracts. Every three months the Coordinating Beneficiary (Department of Economic, BPI) gathers the project costs and financial forms submitted by each partner and assesses the legality and adequacy of supporting documents as well as their eligibility. Then, the coordinating beneficiary creates summary tables with the financial balance per beneficiary, per action and per cost category. Moreover, the coordinating beneficiary, also provides clarifications regarding the procedures of monitoring the financial project progress. The timesheets 'model used by all partners follows the template of LIFE projects. Timesheets are submitted by all beneficiaries every 3 months to the coordinating beneficiary.

The time record system of all beneficiaries is the same and follows the use template of LIFE projects. The working time is registered on a daily or weekly basis. The manual submission is monthly and the timesheets are signed by the employee and the supervisor.

### 6.3. Partnership arrangements (if relevant)

Partnership agreements between the Coordinator Beneficiary (BPI) and the associated beneficiaries of the Project were signed and submitted to the European Commission with Annex E.1.5 of the Inception Report. The first payment was made to the partners after signing the Partnership Agreement in accordance with the terms and conditions of financing as set out in the contract between the coordinator and the partners of beneficiaries. Associated beneficiaries complete the relevant financial tables according to the standards of LIFE + and send them to the Coordinating Beneficiary together with the supporting documents. The Coordinating beneficiary checks the costs relatively to the legality and adequacy of supporting documents as well as their eligibility compared to the budget of each partner. After the acceptance of the amendment of the project concerning the prolongation of the project's duration an amendment to the original grant agreement was signed, while the partnership agreements between BPI and AOO Ravenna and AOO Cesena (there was a change in their VAT number) were revised accordingly.

### 6.4. Auditor's report/declaration

An Italian auditor audited the Italian associated partners, and his Audit report was included in the Audit report of the Greek Auditor. For more details, please refer to Annex E.6.1.

### 6.5 Summary of costs per action

The following table presents the allocation of the costs incurred per action.

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## 7. Annexes

### 7.1 Administrative annexes

All partnership agreements have already submitted to the European Commission with the submission of the Inception report.

### 7.2 Technical annexes

#### 7.2.1 List of abbreviations

- BPI (Benaki Phytopathological Institute)
- AUA (Agricultural University of Athens)
- N.C.S.R Demokritos (National Centre for Scientific Research)
- UEHR (University Research Institute of. Environmental and Human Resources)
- CAA (Centro Agricoltura Ambiente “G. Nicoli”)
- PH-ER (Public Health Service of Emilia-Romagna)
- AUSL (Azienda Unità Sanitaria Locale)
- AOO (Area Organizzativa Omogenea)
- HCDCP-KEELPNO (Hellenic Centre for Disease Control and Prevention)
- IMS (Invasive Mosquito Species)
- DF (Dengue fever)
- WRF (Weather Research and Forecasting)
- MD (Monitoring Device)
- NMS (Network Management System)
- PMAs (Pilot Monitoring Areas)
- CHIKV (Chikungunya virus)
- DENV (Dengue virus)
- WNV (West Nile virus)

### 7.3 Dissemination annexes

#### 7.3.1 Layman's report

The development of the Layman’s report is presented in Annex D.5.1 (1<sup>st</sup> version) and Annex D.5.2 (Final version).



7.3.2 After-LIFE Communication plan – for LIFE+ Biodiversity and LIFE  
Environment Policy and Governance projects

The development of the After-LIFE Communication plan is presented in Annex E.4.1.

7.3.3 Other dissemination annexes

The extended dissemination activities are presented in Annexes D.3.1 to D.3.5.

## 7.4 Final table of indicators

The final table of indicators is presented in Annex E.1.7.

## 8. Financial report and annexes

For the following Annexes use the link to download the documents.

Annex 1: Payment Request

Annex 2: Consolidated Cost Statement for the Project

Annex 3: Financial Statement of each Beneficiary (submitted as excel file)

Annex 4: A justification letter from AUA for higher than foreseen daily rates