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FINAL REPORT OF AN AUDIT
CARRIED OUT IN
ITALY
FROM 18 TO 25 NOVEMBER 2014
IN ORDER TO EVALUATE THE SITUATION AND OFFICIAL CONTROLS FOR XYLELLA
FASTIDIOSA

Executive Summary

This report describes the outcome of an audit carried out by the Food and Veterinary Office (FVO) in Italy, from 18 to 25 November 2014, following an earlier audit in February 2014.

The objective of the audit was to evaluate the plant health situation and control measures applied for Xylella fastidiosa (Well and Raju), in particular, the implementation of Commission Implementing Decision 2014/497/EU of 23 July 2014.

The audit team found that:

Extensive human and financial resources have been made available for research and containment of Xylella fastidiosa, and there is a good level of awareness about the problem.

With one exception, none of the eradication measures required by Decision 2014/497/EU, have been carried out. The situation has deteriorated since the last audit and Xylella fastidiosa continues to spread rapidly. The current Italian policy for the Infected Zone is now containment of Xylella fastidiosa and measures aiming at full eradication of the pest are not carried out.

The current controls do not ensure that host plants not fulfilling the requirements of the EU Decision remain in the Demarcated Area. All the existing garden centres located in the Demarcated Area have not been identified and, therefore are not officially controlled.

There is a possibility that not all host plant species have been identified and pathogenicity tests for a range of genera (including Vitis and Citrus) have not been concluded. Until the precise host range of Xylella fastidiosa is known, the movement restrictions in place (although applied to a wider range of species than required in the Decision) do not provide adequate security that no infected plants leave the area.

The proposed intensive surveys in the Eradication Zone, Buffer Zone and Security Zone, will help in the early detection of Xylella fastidiosa and enable the implementation of rapid eradication. This strategy could also limit the natural spread of the insect vector to new areas. However, taking into account the high populations and the passive mobility of the insect vector (vehicles, wind), the protective function of the two zones is questionable.

There is a significant risk of further spreading of Xylella fastidiosa outside the Demarcated Area by the movement of host plants and by the high populations and mobility of the insect vectors.

The report makes a number of recommendations to the competent authorities, aimed at rectifying the shortcomings identified and enhancing the control measures.

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ABBREVIATIONS AND DEFINITIONS USED IN THIS REPORT

Abbreviation	Explanation
ARECBC	Agriculture Research and Experimental Centre Basile Caramia
BZ	Buffer Zone
cm	centimeters
DA	Demarcated Area
ELISA	Enzyme-Linked Immunosorbent Assay
EPPO	European Plant and Mediterranean Plant Protection Organisation
EU	European Union
EZ	Eradication Zone
FVO	Food and Veterinary Office
GPS	Global Positioning System
ha	Hectare
ISO	International Organisation for Standardisation
IT	Information technologies
IZ	Infected Zone
MAIB	Mediterranean Agronomic Institute of Bari
km	kilometre
m	meter
MIPAAF	Ministry of Agriculture, Foodstuffs and Forest Policies
PAO	Provincial Agricultural Offices
PCR	Polymerase Chain Reaction
RPS	Regional Plant Health Services
RAI	Regional Agency for Innovation
RAIF	Regional Agency for Irrigation and Forestry
SA	Single Authority
SFB	State Forestry Body
SZ	Safety Zone
<i>Xf</i>	<i>Xylella fastidiosa</i>

1 INTRODUCTION

This audit took place in Italy from 18 to 25 November 2014. It was added to the 2014 Food and Veterinary Office's (FVO) planned audit programme as response to the emergency situation in Italy.

The FVO team consisted of two auditors from the FVO and one expert from a European Union (EU) Member State. Representatives from the Single Authority (SA), Central Plant Health Service of the Ministry of Agriculture, Foodstuffs and Forest Policies (MIPAAF), and the Regional Competent Authority from Puglia accompanied the FVO team during the audit.

An opening meeting was held on 18 November at the headquarters of the Regional Plant Health Services (RPS) in Bari during which, the objective, scope and itinerary for the audit were confirmed by the FVO team.

2 OBJECTIVES

The objective of the audit was to evaluate the plant health situation and control measures applied for *Xylella fastidiosa* (Well and Raju), in particular, the implementation of Commission Implementing Decision 2014/497/EU of 23 July 2014.

The following table lists the sites visited and meetings held in order to achieve these objectives:

Meetings/visits		No.	Comments
	Regional	2	Regional Plant Health Services (RPS) of Puglia. During the meeting representatives of the Single Authority were present.
	Laboratories	1	Laboratory of the Agriculture Research and Experimental Centre 'Basile Caramia' - Locorotondo
	Research	1	Meeting with researchers of Puglia region, who are involved in the research activity related to <i>Xf</i> . The meeting was organised by the RPS of Puglia.
Plant health control sites			
Infected olive fruit production sites		3	Gallipoli, Casarano
Eradication sites		2	Sternatia, Lecce
Sites of production of olive trees, fruit trees and ornamental plants for planting		2	Taviano, Maglie

3 LEGAL BASIS

The audit was carried out under the general provisions of EU legislation, in particular Articles 21 and 27a of Council Directive 2000/29/EC.

3.1 RELEVANT EU LEGISLATION

Council Directive 2000/29/EC provides for protective measures against the introduction into and spread within the EU organisms harmful to plants or plants products. The legal reference for this Directive is listed in the Annex to this report.

Commission Implementing Decision 2014/497/EU of 23 July 2014, as regards measures to prevent the introduction into and the spread within the Union of *Xylella fastidiosa* (Well and Raju).

References to EU legislation in this report are to the latest amended version, where applicable.

4 BACKGROUND

Xylella fastidiosa strain CoDiRO was detected in Lecce province, Puglia region of Italy in October 2013. It is associated to the 'Olive Quick Decline Syndrome' causing severe damage and death to olive trees.

The FVO had previously carried out one audit in February 2014 in order to evaluate the situation and official controls for *Xylella fastidiosa* (*Xf*), ref DG(SANCO)/2014/7260. The report is available on http://ec.europa.eu/food/fvo/ir_search_en.cfm as are the Single Authority's comments on the report and its response to the recommendations. The FVO audit identified several shortcomings concerning mainly: laboratory tests, identification of host plants, surveys and control measures.

Immediately after the audit, the Decision 2014/87/EU was published. The Decision established a set of rules for the movement of all plants for planting outside Lecce province.

In April 2014, *Xf* was known to be present mainly in the Gallipoli area and in five outbreak sites, based on the outcome of an extensive survey carried out since November 2013.

In July 2014 the EU Decision 2014/497/EU was published. It requires surveys for detection of the harmful organism. It also establishes a set of provisions and measures for the Demarcated Areas (DA) namely, conditions for the movement of host plants within and from the DA, the establishment of DA and measures to be taken in the DA.

The host plants for planting (except seeds) listed as 'specified plants' (hereafter host plants) in Decision 2014/497/EU are: *Catharanthus* (Madagascar periwinkle), *Nerium* (oleander), *Olea* (olive), *Prunus* (stone fruits), *Vinca* (periwinkle), *Malva* (mallow), *Portulaca* (purslane), *Quercus* (oak) and *Sorghum* (Johnson grass).

At the beginning of July the SA notified the EU Commission of three new outbreaks sites, in addition to the initial outbreaks identified until April 2014. In the same letter three new plant species were notified as *Xf* host plants: *Prunus avium* (sweet cherry), *Polygala myrtifolia* (sweet pea shrub) and *Westringia fruticosa* (coastal rosemary).

At the beginning of August two new outbreak sites were notified. This was immediately followed by another notification of the wide presence of *Xf* in nine new municipalities of Lecce province and of a new host plant, *Acacia saligna* (acacia).

In the middle of September 2014 a new host plant species was notified to the Commission, *Spartium junceum* (Spanish broom).

In 14 October 2014, a national decree was published allowing for the implementation of the EU Decision 2014/497/EU.

Until October 2014, Puglia region issued several regional resolutions defining provisions and requirements for the control of *Xf*.

5 FINDINGS AND CONCLUSIONS

5.1 ORGANISATIONAL ASPECTS OF PLANT HEALTH CONTROLS

Legal requirements

Article 1(4) of Council Directive 2000/29/EC provides that Members States shall ensure a close, rapid, immediate and effective cooperation between themselves and the Commission in relation to matters covered by this Directive and that, to this end, each Member State shall establish or designate a single authority, which shall be responsible, at least, for the coordination and contact in

relation to such matters.

Article 2(1)(g) of Council Directive 2000/29/EC requires that the responsible official bodies in a Member State shall either be the official plant protection organisation established under the IPPC, or any other State authority established at national level or at regional level, under the supervision of the national authorities. Article 2(1)(i) of the same Directive requires Member States to ensure that their public servants and qualified agents have the qualifications necessary for the proper application of the Directive.

Findings

5.1.1 Competent authorities

The SA stated that there have been no changes to the organisational aspects of plant health controls, as described in the FVO country profile for Italy (http://ec.europa.eu/food/fvo/last5_en.cfm?co_id=IT), and in the reports of previous FVO audits, in particular that of a general audit carried out in Italy from 1 to 12 March 2010 (Ref: DG(SANCO) 2010/8601).

The structure and roles of the national and competent authorities remain as described in the February audit report.

In summary, the SA is the Central Plant Health Service of the Ministry of Agriculture, Foodstuffs and Forest Policies (MIPAAF). The RPS of Puglia implements the plant health policy in the region. The region is divided in Provincial Agricultural Offices (PAO), where the plant health inspectors are working.

Taking into account the extension of *Xf* spread and the implementation of official control measures, the regional and central governments have recently involved other organisations in *Xf* control:

- The State Forestry Body (SFB) is a national organisation, with enforcement powers, belonging to MIPAAF. The SFB is in charge of the implementation of several national decrees in particular in the following areas: environmental, forest, forest nurseries, land management and waste management. A total of 50 SFB staff work in Lecce province and if needed, the team can be reinforced with staff from neighbouring provinces. So far, the SFB has participated in eradication activities, in particular in relation to enforcement and access to private property, but it is intended that in the future they will collaborate also in plant movement control (road controls), nursery control and possibly in the surveys. At the moment, a protocol between the region and the SFB is being prepared and will be signed in the near future.
- InovaPuglia, the Regional Agency for Innovation (RAI), is in charge of technological innovation (information and communication technologies). It manages all IT based automated systems of the regional administration (payments, budget) and actively participates in regional projects.
- The Regional Agency for Irrigation and Forestry (RAIF) is participating now in the control of *Xf*. RAIF works mainly in the public forest sector of the region, including afforestation, forest management and forest fire protection. The agency also deals with irrigation, wells and water management in Puglia. They have around 100 staff in Lecce province. They offer services e.g. machinery for the eradication.

The RPS stated that important financial resources have been made available. In 2014 the total regional budget for *Xf* was 6 000 000 € from which, part will be transferred to 2015. In addition, Puglia was granted 2 500 000 € from the central government for eradication, phytosanitary treatments, elimination of grass and eradication of ornamental host plants in roads.

5.1.2 Communication and information

Wide publicity and technical information about *Xf* is available to stakeholders and the general public. There is systematic coverage by regional and national media about the situation of *Xf* in Lecce, raising awareness of the problem.

The research institutions organised the 'International Symposium on the European outbreak of *Xf* in olive' in Gallipoli, which was attended by more than 200 participants from 18 countries. Some researchers have participated in several scientific meetings in other Member States, Italy and third countries where the situation and knowledge on *Xf* was discussed.

There are monthly *Xf* specific meetings between the RPS, PAO phytosanitary inspectors and phytosanitary agents. Also periodical meetings with stakeholders and producers to discuss topics related to *Xf* control, take place in the region.

During the audit, the FVO team met with plant producers and traders, and noted that they were fully familiar with the official requirements and control measures for *Xf*.

5.1.3 Laboratories and testing

The RPS stated that, in relation to February 2014, the same four laboratories are participating in the *Xf* testing.

The audit team visited the laboratory of the Agriculture Research and Experimental Centre 'Basile Caramia' (ARECBC) in Locorotondo. The ARECBC laboratory has been, since the beginning very active in testing samples from the surveys and from nurseries. The laboratory is accredited by a national certification body but it is not accredited under the ISO 17025 standard.

The laboratory uses exclusively the ELISA test for detection of *Xf* in plant tissues according to a protocol elaborated by the Institute of Plant Virology, Research National Centre /Department of Plant and Soil Sciences and Food, University of Bari. The *Xf* detection method is based in the standard PM 7/98, *Specific requirements for laboratories preparing accreditation for a plant pest diagnostic activity* of the European Plant and Mediterranean Plant Protection Organisation (EPPO).

All the positive results are confirmed by the reference laboratory in Bari. The laboratory has participated in two ring tests that were carried out amongst the four regional laboratories that participate in the *Xf* detection.

The total number of samples tested since November 2013 until the moment of the audit is listed in table 1.

Table 1: Number of samples tested in the ARECBC laboratory

	Until April 2014		After April 2014		
	No Samples	Positive	No Samples	Positive	
Surveys	7 639	234 olive	1 011	-	8 650
		2 oleander			
		6 sweet pea			
Nurseries	4 470	-	130	-	4 600
Total	12 109	242	1141	-	13 250

The laboratory tested 13 250 samples in total, mainly originated from the surveys carried out until April 2014. It is foreseen that the number of samples from the current surveys will increase in the near future.

The laboratory can test an average of 150 samples per day and has a permanent staff of three

technicians. If needed, the team may be reinforced with extra staff. The RPS pays 10 € for each sample received and tested.

The audit team noted that the laboratory has a skilled, competent and very motivated staff and has a general set of equipment necessary for the analysis.

The audit team noted that the sample codification and administration of the test results is not simple and straightforward. The code is copied by hand several times in the laboratory. The photometric readings are assessed without the aid of a colour code system to highlight results. There is no verification system for the work of the technicians. Taking into account the large number of samples the applied procedures are prone to mistakes and errors.

5.1.4 Research

The RPS stated that, in relation to February 2014, the same research institutions are participating in *Xf* testing.

The audit team met a group of researchers, from several entities, that are involved in research activities related to *Xf*. These are: National Research Center, University of Bari, Mediterranean Agronomic Institute of Bari, Agriculture Research and Experimental Centre 'Basile Caramia' and University of Foggia. The meeting had the objective of updating the audit team with the last developments on *Xf* research carried out in the region.

The research activities are now concentrated in two main topics:

- the bacteria: epidemiology, pathogenicity, insect vector
- area management: surveys, sampling, mapping, specific software

The bacteria was successfully isolated in March 2014 from several plant species. The genome sequencing is finished, indicating that the local strain, *Xylella fastidiosa* strain CoDiRO, is very close to *Xylella fastidiosa* pv. *pauca*.

The same identity sequence was obtained from several host plants in the region. This very low genetic variation indicates a recent introduction of *Xf* in the area.

Field observations showed that young olive trees are infected by *Xf*, which confirms that the bacteria by itself can cause the decline and death of the olive trees. This is a new finding, since previously there was a belief that the trees were dying when *Xf* was present and enhanced by a synergy with fungi and insects.

Inter laboratory tests were carried out for validation of the ELISA and PCR test protocols, these showed a very good consistency of result and now the level of detection is clearly defined.

The visual symptoms in several host plants were described and documented.

The survey of 100 weed species did not increase the known list of weed host plant infected by *Xf* and there are now indications that they play a minor role in the epidemiology of *Xf*. There are on-going pathogenicity tests to confirm the host plant range in particular in *Vitis* and *Citrus*, the final results will be available in the second semester of 2015.

The researchers informed the FVO team that the former positive findings on *Quercus*, *Malva*, *Portulaca* and *Sorghum*, could not be confirmed by additional tests or transfer experiments. They could have been false positive results due to test methods in development at the time.

In order to identify potential insect vectors, monitoring and transmission tests of insects were carried out. At the moment three vector insects species have been found capable to acquire *Xf*: *Philaenus spumarius* L., *Neophilaenus campestris* Fallen and *Euscelis lineolatus* Brulle. However, from the three, only *Philaenus spumarius* has been proven capable of transmitting *Xf*, (transmission research work is on-going for other two species). The three insect species will be monitored and

used for assessing the presence of *Xf* in free areas, before symptoms appear in host plants.

The life cycle of the spittlebug *Philaenus spumarius* is now better known and it starts earlier in Puglia than in Central and North of Europe. There is one generation per year and the insect overwinters in the egg stage. The eggs laid on weeds close to the ground, hatch in February and originate the nymphs, which start immediately to suck sap from the plant and form the foam where they live. They remain on the weeds until they mature in June, as adults. The adults appear in early June and continue to fly until late October however, some can survive throughout the winter. The adults hop on and feed on olive trees where the infection stage begins. The peak of the insect population is from June to September, which corresponds to the active spreading of the disease.

Transmission studies in the field have shown that *Philaenus spumarius* adults acquire *Xf* in olive trees (and not on weeds) sometime early June and then they are able to spread the bacteria to other olive trees. The foregut of the insect is the area where the bacteria lays.

The active flight of the insect seems to be limited however, the passive transport by vehicles or wind seems to be a very important factor for *Xf* spread.

Two additional on-site techniques for *Xf* detection are being developed: Direct Tissue Blot Immuno Assay (DTBIA) for plant tissue and Loop-Mediated Isothermal Amplification (LAMP) for insects. Both are practical, quick, cheap and very effective detection methods.

There are new developments on the use of remote sensing techniques to support the survey field work, using photointerpretation of high resolution infrared images for detection of olive trees with symptoms. In order to have updated information aerial images will be taken with a certain periodicity. This will allow for a more targeted survey since the surveyors would go straight to the trees showing symptoms.

Two new IT applications were developed: Xylweb and Xylapp for assisting the field surveys. The first, still under progress, is a regional system for data storage and processing. The Xylapp is used on tablets and provides inspectors with a regional cartographic grid allowing a fast identification of the survey site and recording data related to the observation point. The data collected by Xylapp is uploaded daily to the central server (Xylweb) for storage and processing (see section 5.3.2).

Conclusions on organisational aspects of plant health controls

The competent authorities remain the same since last audit. Three new organisations are now participating in *Xf* control. Substantial regional and national financial resources have been made available for control and research.

Trained and skilled technicians are available in the laboratory however, the possibility of false negative test results cannot be excluded due to weaknesses in sample codification and reading of test results.

New and important developments in research (vector life cycle, host plant range) became available, which will help in controlling further spread of the disease. Some studies are still in progress: pathogenicity tests, latency and symptom expression, vector control and olive tree tolerance/resistance to *Xf*. Therefore, the control measures and the movement control of host plants, should be regarded with precaution.

5.2 LEGISLATION

Legal requirements

The Italian National Decree of 26 September *Emergency measures for the prevention, control and eradication of Xylella fastidiosa (Well and Raju) within the territory of the Italian Republic*, transposes the Commission Implementing Decision 2014/497/EU.

Several regional resolutions and determinations establish the provisions and additional requirements for the control of *Xf*.

Findings

The SA stated that it was decided to implement the EU Decision in a national decree with specific requirements for the Demarcated Area (DA) in the Lecce province. The decree had a favourable opinion of the National Phytosanitary Committee (NPC).

In addition, two national decrees were also approved. One establishing a national technical/scientific committee and a second one granting 2 500 000 € financial support for different actions in Puglia.

The audit team discussed with Italian authorities the national decree and noted that:

- due to the large number of new findings during the summer, the decree declares almost all the territory of Lecce province as DA. The DA comprises an Infected Zone (IZ) and a two km Buffer Zone (BZ);
- in addition to the EU Decision, the decree foresees the creation of a two km Security Zone (SZ) located five to six km beyond the BZ. It also declares a one km strip located in the IZ immediately before the BZ, as the Eradication Zone (EZ) (see section 5.3.1);
- the State Forestry Corps (SFC), the Central Inspectorate for Quality Controls and Fraud Prevention for Agri-food Products and other regional agencies may participate in the control of *Xf*;
- the decree provides details for risk based surveys and, sampling and testing of confirmed or potential insect vectors.

Although the decree incorporates most of the provisions of the EU Decision correctly, the audit team noted that:

- the new host plants identified after the publication of EU Decision have been included in the decree however, they have been listed as species and not as genera.
- in the IZ in Lecce (including the EZ), a derogation allows for not taking the eradication measures in line with the EU Decision, there is only reference to measures deemed appropriate in order to contain *Xf* and destroy vector populations.

Conclusions

Specific national legislation for the control of *Xf* implementing Decision 2014/497/EU is in place.

The national decree lists the new identified host plants as species. Until further scientific evidence about the susceptibility of other species become available, the precautionary principle should apply and genera should have been listed in the decree, following the same approach of the EU Decision.

In the IZ, including the EZ, there is no mention in the decree of the eradication measures that have to be taken in accordance with the Article 7 (3) and Section 2 of Annex III of Decision 2014/497/EU.

5.3 DEMARCATED AREA AND SURVEYS IN LECCE

Legal requirements

Article 4 (1) of Decision 2014/497/EU requires that, Member States shall conduct annual surveys for the presence of *Xf* in their territory on the host plants and on other possible host plants. Those surveys shall be based on sound scientific and technical principles, and shall be carried out at appropriate times with regard to the possibility to detect the *Xf*. The surveys shall take account of

the available scientific and technical evidence, the biology of *Xf* and its vectors, the presence and biology of host plants or plants likely to be host plants of *Xf*, and any other appropriate information, concerning the presence of *Xf*.

Article 7 (1) requires that, where the results of the surveys show the presence of *Xf*, or where such a presence is confirmed, the Member State concerned shall without delay demarcate a 'demarcated area'.

Article 7 (2) requires that the demarcated area shall consist of a zone in which *Xf* was found to be present, 'the infected zone'. That zone shall be defined in accordance with Section 1 of Annex III. The demarcated area shall further consist of a zone surrounding the infected zone, 'the buffer zone'. That zone shall be defined in accordance with Section 1 of Annex III.

Findings

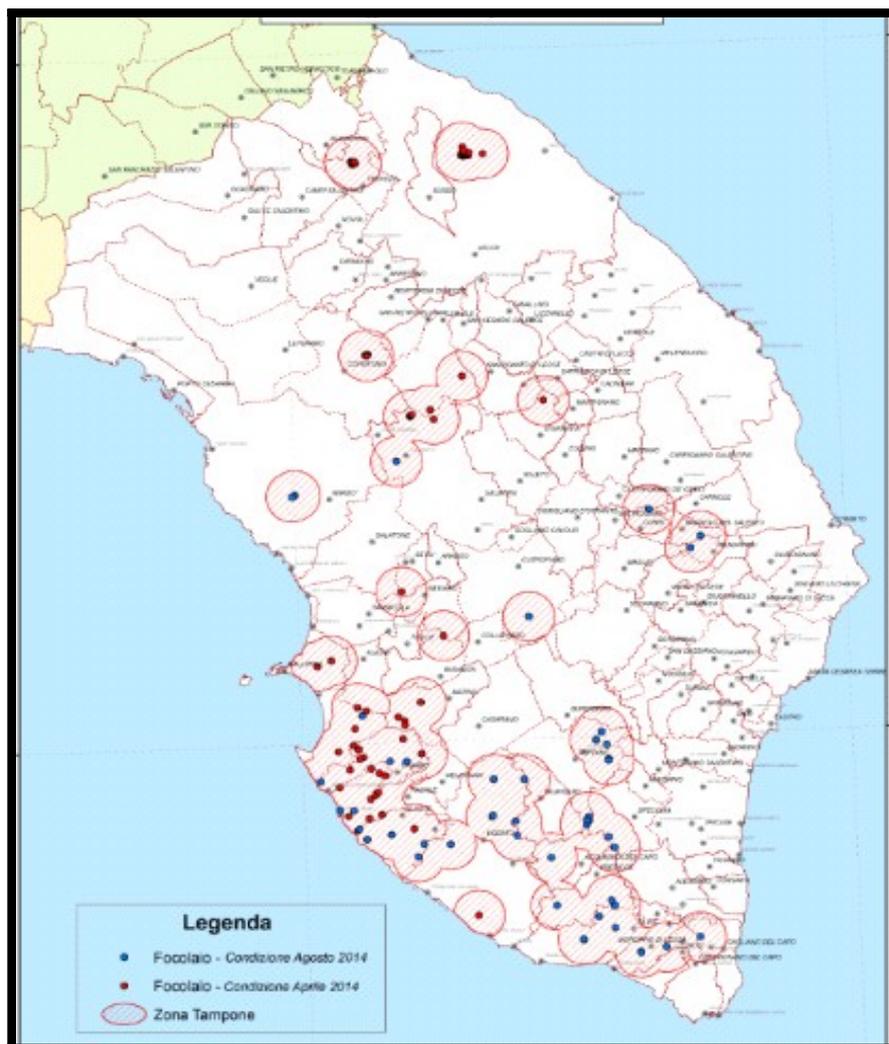
5.3.1 Establishment of DA

Xf was found initially in the area of Gallipoli. At the end of the surveys carried out until April 2014 and after testing around 16 000 samples, *Xf* was subsequently found in other five outbreak sites to the north: Trepuzzi, Lecce, Copertino, Galatina, Sternatia. The RPS established a DA for these sites, see figure 2.

At the time, there were no specific EU requirements for establishing DA and Italy decided to establish a DA consisted of the IZ and a one km BZ. Eradication measures were implemented in the five small outbreak sites (see section 5.4.2).

Mainly through information provided by farmers, at the end of the summer 2014 the number of trees showing symptoms had increased, in particular in the area south of Gallipoli. The outbreak situation worsened and the infected area increased dramatically since February 2014, see figure 1.

Figure 1 – *Xf* Demarcated Areas in Lecce (August 2014)

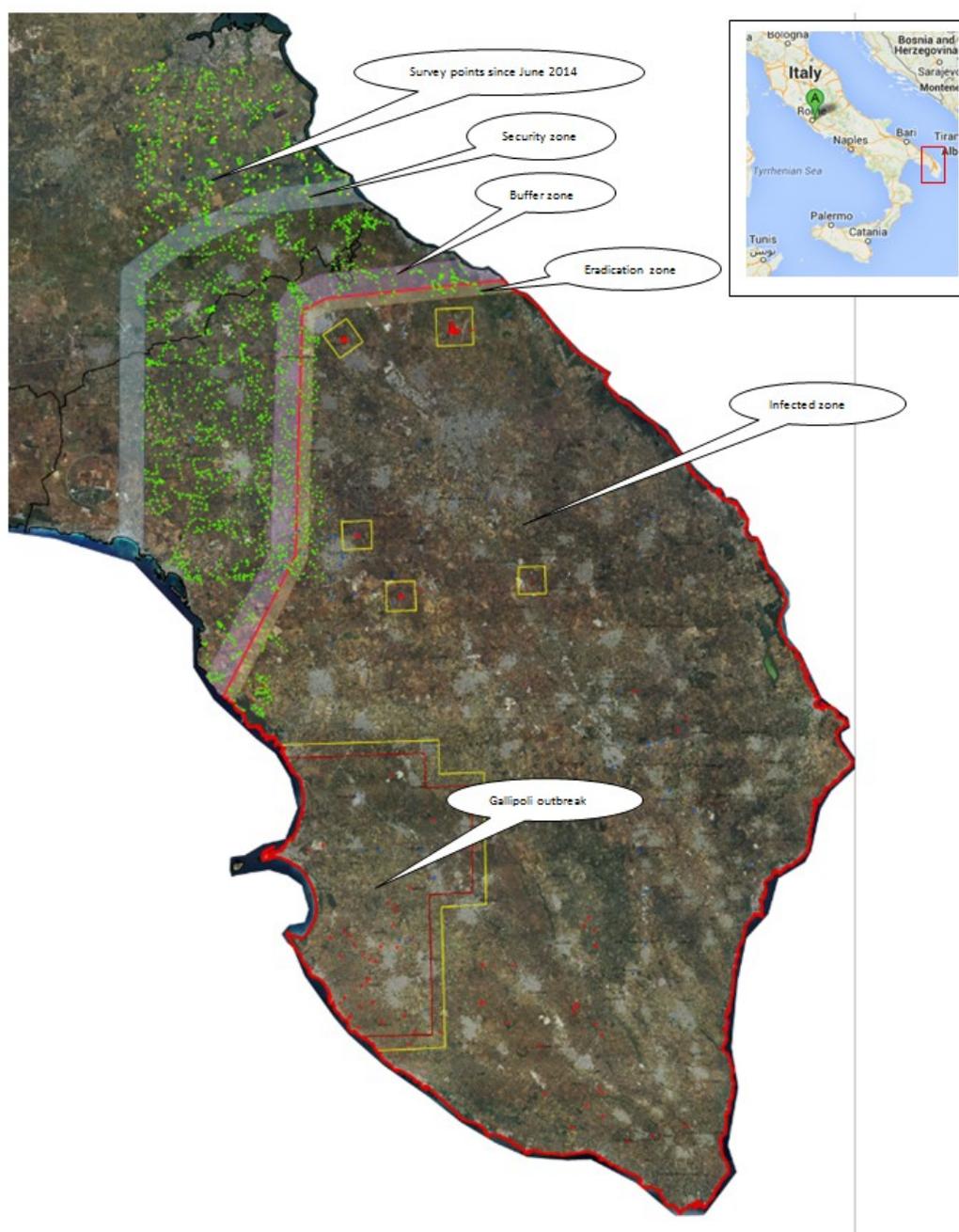


In October 2014, intensive surveys were carried out on the limits of Lecce province in order to establish a BZ outside the last known outbreak site. Such surveys are still being carried out and will be finished in March 2015. So far, and after 2 400 samples have been tested, the results have confirmed that the pest is not present in the survey area.

As requested by the national decree, in addition to the definition of the DA and establishment of the IZ and BZ, a SZ and EZ were also defined. The SZ is located at a distance of five to six km beyond the BZ. The EZ is the last one km of the IZ immediately before the BZ, see figure 2.

The RPS stated that, in addition to the BZ, the creation of a SZ at the border of Lecce province, seems like the only feasible option left for preventing the natural spread of the disease. The proposed intensive survey in the EZ, BZ and SZ, will help in the early detection of *Xf* and enable the implementation of rapid and efficient eradication measures. This strategy could limit the natural spread of the insect vectors to new areas, as well.

Fig. 2 – *Xf* Demarcated Area, Security Zone and Eradication Zone in Lecce (November 2014)



The total area of the DA is now 241 000 ha (IZ, 231 000 ha and BZ, 10 000ha), with around 90 000 ha of olive orchards containing around 9 000 000 olive trees. The RPS explained that the large dispersion of *Xf* is due to the enhanced vector activity and could not indicate the number and distribution of infected olive trees in the DA.

The RPS stated that in case of a detection *Xf* in the BZ, in the SZ or in the area between the two, the plant health situation will be assessed. If it is a small outbreak and the immediate eradication seems to be successful the border of the infected zone will not be changed. However, in case of large outbreak with many infected trees, the border of the zone will be changed accordingly.

Conclusions on the *Xf* DA in Lecce

The DA in Lecce is established in line with Article 7(2) of Decision 2014/497/EU.

It is intended not to change the border of the DA in case of the detection of small outbreaks outside the IZ. This is not in line with the Section 1 of Annex III of the EU Decision 2014/497/EU, which requires that, for any finding the limits of the IZ have to be reviewed and changed accordingly.

5.3.2 Surveys

The previous survey plan was based on a 2.7 km x 3.2 km grid, where at least, 13 samples from seven plant species are collected in each grid unit: five samples of olive, two of oleander, two of mallow, one of Bermuda grass and one of Johnson grass. It is also foreseen to collect three samples of insect vectors per each grid unit.

The RPS stated that the survey plan had had substantial changes since February 2014. The plan is being revised, taking into account the new tools that are now available from the research in particular, detailed mapping and new software.

The RPS of Puglia intends to carry out official surveys on the host plants every year, and on *Acacia saligna*, *Polygala myrtifolia*, *Spartium junceum*, *Westringia fruticosa*, and other potential host plants to check for the presence of *Xf*. The surveys should target the areas considered to be most at risk of being infected by *Xf*, such as:

- areas in which the host plants show visual symptoms;
- roads planted with host plants as roadside trees;
- areas in which the host plants are produced and traded;
- uncultivated land and wastelands, parks and similar areas, tourist areas.

The surveys are carried out by phytosanitary agents from the three provinces: Lecce, Brindisi and Taranto and are coordinated by the RPS. The agents are in charge of the detection of visual symptoms mainly on olive trees and collection and delivery of samples on the laboratories. The total number of staff involved in surveys is 18, divided in nine teams of two agents each. At the moment, seven teams work in the BZ and two on the SZ.

The research work developed by the RIA and the Mediterranean Agronomic Institute of Bari (MAIB) are now the basis for surveys.

The RAI is linked to the Territorial Portal of Puglia where access to cadastral and land use databases is possible. In addition, several airplane flights over the regional territory using different levels of photo resolution, were and will be scheduled. All data originating in the portal and from the photointerpretation, allows RAI to produce very specific mapping to be used on the *Xf* survey.

The RPS stated that, there is an intention of overflying all the area with aerial photography: 320 000 ha will be covered at 18 cm resolution and 70 000 ha (mainly the BZ and SZ) will be photographed at 7 cm resolution.

It is planned that the MAIB identifies trees with symptoms through infrared photointerpretation. Updated maps will be fed in the tablets running Xylapp, this allows the phytosanitary agents to work in the field with very detailed and updated mapping.

The Xylapp is an Android application that runs in tablets. The application aims to facilitate, the precise location of the surveyed site and store all the information related to it, such as: GPS, species, variety, presence of symptoms, phenological stage, sampling codification and laboratory result.

There are five modules in the application: sampling, navigation, find, store and vademecum. Standard information (as drop down menus) is uploaded in the application.

The Xylweb is still being developed and will be fully operational in the near future. At the moment, the stored data is sent to the MAIB and RAI, by the phytosanitary agents.

The RPS stated that the survey intensity, will be:

- in the BZ and SZ, a very dense 100 m x 100 m grid will be used;
- in the EZ, the survey will be even more intense since it is intended to monitor all the area, host plant by host plant.
- in the area located in between the BZ and the SZ and in the remaining *Xf* free area in Puglia, the previous grid 2.7 km x 3.2 km will be used;

The phytosanitary agents explained that visual symptoms will be checked in each grid and at least one olive sample will be taken, preferably from symptomatic trees. If there are no symptoms a sample will then be taken from an asymptomatic tree. If there are no host plants in a grid they will take more samples in the next grid.

The sample is composite of four twigs from the symptomatic branches, when the sample is taken from asymptomatic trees the twigs are taken from opposite sides of the tree.

They also mentioned that the survey will be more difficult in urban areas located in the BZ and SZ, in this case symptomatic plants may be overlooked, but the RPS counts with the support of a large awareness campaign in the province that would tackle these situations.

The intense survey in the BZ and SZ should be finished at the beginning of the spring 2015 and it is planned to test around 50 000 samples by the end of 2015.

The monitoring of confirmed or potential insect vectors is carried out by MAIB staff. The monitoring intends to detect infect insect vectors in a free area, before symptoms become visible in the trees.

Experts visit plots (including non-cultivated areas) where they collect insects with a net in a 40 m circle. Immediately after, a quick scan of the captured insects is carried out in order to separate the insect vectors which are immediately kept in ethanol. Later, the insects are tested in the MAIB laboratory by PCR to check whether they are infected with *Xf*.

Similarly, all the data related to surveys of insects is also recorded and saved in Xylapp/Xylweb.

At moment of the audit, 110 sites in the free zone beyond the SZ had been monitored and around 1 000 insects had been tested, with no positive findings. The plan for the insect monitoring is to sample 2 000 sites and collect from 4 000 to 10 000 insects.

The experts informed the audit team that they clean the vehicles and the windows are always kept closed, to prevent the spread of infected insect to new areas.

It is also intended to carry out a similar monitoring of insect vectors, using sticky traps located inside and outside nurseries.

Conclusions on the surveys for *Xf* in Lecce

Intensive surveys are planned in the BZ and SZ and they seem appropriate for detection of infections in these zones.

Although the survey seems to be well designed and consistent with the plant health risks, some concern arose about the full implementation of the surveys in urban areas.

The monitoring of confirmed and potential insect vectors is a useful tool for early detection of the presence of *Xf* in new areas.

5.4 CONTROL MEASURES IN THE DA

Legal requirements

Article 1 (a) of Decision 2014/497/EU defines as 'specified plants' all plants for planting, other than seeds, of *Catharanthus* G. Don, *Nerium* L., *Olea* L., *Prunus* L., *Vinca* L., *Malva* L., *Portulaca* L., *Quercus* L. and *Sorghum* L..

Annex II of the above mentioned Decision establishes the conditions for movement of host plants within the Union. The host plants which have been grown for at least part of their life in a demarcated area shall only be moved to and within areas other than infected zones if they are accompanied by a plant passport stating that they fulfill the following requirements:

- the site of production fulfils the following conditions: is free from the *Xf*, is registered, is physically protected against the introduction of the *Xf* by its vectors, is subject to phytosanitary treatments for freedom from vectors of the *Xf*, two official annual inspections are carried out and neither symptoms of the *Xf* nor its vectors were found or, if suspect symptoms are found it is subject to testing and *Xf* is absent;
- representative samples of each species of host plants from each site of production have been subject to annual testing, and the asymptomatic presence of *Xf* has been excluded;
- phytosanitary treatments against the vectors of *Xf* are applied in the close proximity of the site of production.

The host plants are transported outside the flight season of the vectors, or in closed containers or packaging.

Article 7 (3) requires that, Member States shall take measures in the demarcated areas, as set out in Section 2 of Annex III.

Section 2 of Annex III of Decision 2014/497/EU lists the following eradication measures to be taken in demarcated areas: remove as soon as possible all infected plants, all symptomatic plants and all plants likely to be infected. No plant material should remain to prevent *Xf* spreading; sampling and testing, with 99 % reliability that the level of presence of *Xf* in those plants is below 0.1 %, of host plants and plants of the same genus as the infected plants, and all symptomatic plants within a radius of 200 m around infected plants; destroy, *in situ* or in a nearby location within the DA, the entire plants, parts of plants or wood which may contribute to the spread of the *Xf*; destroy *in situ* or in a nearby location, any plant material originated from pruning of host plants and plants of the same genus as the infected plants; carry out phytosanitary treatments of host plants and of plants that may host the vectors of *Xf*; trace back to the origin of the infection and tracing forward of host plants, which may have been moved and inform authorities of the area of destination; prohibit planting of host plants and plants of the same genus as the infected plants in sites which are not vector-proof; carry out intensive monitoring by at least annual inspections, especially BZ on host plants and plants of the same genus as the infected plants, including testing, of any symptomatic plants; raise public awareness of the threat of the *Xf* and the measures adopted to prevent its introduction into and spread, including the conditions for movement of host plants from the DA; take specific measures to address any particularity or complication in particular those related to the accessibility and adequate eradication of all plants that are infected or suspected of infection; take any other measure, which may contribute to the eradication of the *Xf*, taking account of ISPM No 9 and ISPM No 14.

Findings

5.4.1 Control measures in the nurseries

The RPS stated that at the moment, none of the nurseries existing in the DA of Lecce fulfil the requirements of Annex II of Decision 2014/497/EU therefore, no nurseries have been registered or allowed to issue plant passports.

Currently, the national legislation establishes a prohibition of trading outside Lecce any of the

genera listed in the EU Decision, as well as the four species listed in the national decree.

In February 2014, the RPS requested the nurseries to notify the existing stocks of host plants. At the time, a list including the name of nurseries and the amounts of olive trees per nursery, was provided to the FVO.

The RPS informed and provided legislation to the nurseries concerning the host plants that could and could not be traded.

The RPS stated that an official inventory was recently carried out in order to officially identify, quantify and give cash value to the host plants for planting that are seized in each nursery in the region. The total number of host plants is 213 937 and the intention is to destroy such plants and compensate the nurseries.

The amount of host plants stocked was only confirmed by the PAO inspectors in October/November 2014, and similarly the RPS provided the FVO with an updated list of seized olive plants for each nursery.

For the nurseries producing olive trees it was noted that the two lists did not match. The RPS stated that most of the difference is due to new producers or garden centres that have been identified recently.

Regarding the differences of quantities of olive plants stocked in the same nursery, the RPS informed that, in seven cases, where the nurseries had a much higher number of plants in November, the plants had not been declared by the nurseries in February. For the situations where the number of olive plants was lower in November, in two cases the counting was erroneous and on other two cases the nursery had destroyed plants. However, for the majority of cases the plants had been sold in the initial IZ (if the nursery was located there) or in the province (if the nursery was located in Lecce but outside the initial IZ).

For four nurseries with lower or higher number of plants identified in November the RPS did not give any explanation. Similarly, for the second nursery visited by the FVO team, no justification was given by the RPS for the additional higher number of bonsai olive plants (600 in February, 6 000 in November).

The RPS stated that the Italian legislation does not require the registration of garden centres. The RPS made efforts to identify them however, such task is still in progress. A letter was addressed to large retailers with garden centres informing about the requirements and the prohibition of movement of host plants not fulfilling the current requirements. It is intended that the SFB will start carrying out plant movement controls (road controls) in the near future,

The audit team visited two nurseries located in the IZ.

The first nursery sells ornamental plants and provides services for installing and maintaining private gardens. They have their own production of some plants for planting and the remainder are bought in neighbouring nurseries or in other nurseries in Italy.

The nursery has stocks of almost all the listed host plants: olive (including bonsai olive trees) oak and stone fruit and ornamentals (oleander, sweet pea shrub, acacia, Spanish broom and coastal rosemary), all these are seized.

The inspectors initially informed by e-mail the nursery of the host plants for planting that cannot be sold outside Lecce. At a later stage, they also provided the regional and national legislation banning the trade of such plants.

The inspectors stated that they visit the nursery quite frequently, two to three times per month. They carry out visual inspections of the plants, looking for *Xf* symptoms. After the inspection a report is sent to the RPS however, not always a signed copy is given to the producer. The inspection for

assessing the final number of host plants seized had taken place in 30 October 2014.

The owner informed the audit team, that he had participated in several meetings where the *Xf* topic was discussed, in addition extensive information had been disseminated by the local nursery association.

The audit team visited a second nursery also producing ornamental plants. The nursery has also stocks of almost all the listed host plants: olive (including bonsai olive trees) oak and stone fruit and ornamentals (oleander, sweet pea shrub, Spanish broom). The owner was aware that several plant genera and species cannot be traded. The nursery had been visited by PAO inspectors twice for *Xf* and on other occasions. The inspection for assessing the final number of host plants seized had taken place 8 November 2014.

Conclusion on control measures in the nurseries

As the February nursery stock declarations were not confirmed by official inspections until November 2014, the RPS had no comprehensive information about the host plant material stocked in nurseries therefore, the full enforcement of the movement prohibition of host plants was not ensured (e.g. bonsai olives) It cannot be excluded that host plants were traded outside the DA. There is the intention of creating a compensatory scheme for the destruction of the host plants seized in nurseries, this could be a solution for plants that will never be able to fulfill the new EU requirements and consequently be traded.

The garden centres in the province are still not fully identified and included in the official control system and no road controls have been carried out so far. Therefore, it cannot be excluded that host plants have been traded outside the DA.

5.4.2 Eradication measures

The RPS stated that, in the middle of April, eradication measures were carried out in the five outbreak sites known then (not in the large infested area of Gallipoli). At the time, the Decision 2014/497/EU was not approved, and the RPS decided to establish a DA comprising an IZ and a one km BZ.

The eradication actions were carried by RAIF and in total 104 trees were uprooted and destroyed. However, in four of the five sites further symptomatic trees were detected at a later stage.

The audit team visited two of the five outbreak sites and could confirm that trees were eradicated. The RPS provided copies of the legal dossier of the destruction (sampling, laboratory results, notifications etc). This is based on national legislation that implements Directive 2000/29/EC, which requires that eradication measures are taken when a listed harmful organism is found.

In the first outbreak site (one ha) visited, the PAO inspector was initially informed by the farmer of the presence of symptoms in olive trees. The inspector collected composite samples at the end of March, that were positive for five trees however, the results for the remaining trees in the orchard were negative. From 13 to 15 April and after the legal procedures had been completed, the RAIF uprooted and destroyed the five trees and the logs were handed to the farmer. No further symptomatic trees were detected.

The eradication seems to have worked in this site and the explanation given by the RPS was that the *Xf* detection took place at a very early stage of the infection.

In the second outbreak site visited, the infected area was 30 ha and included three farmers. The detection was made during the survey carried out by phytosanitary agents in March and the procedures and timing for the eradication was the same as in the previous outbreak.

During the summer, visual inspections carried out in the site indicated that symptoms were still present.

Taking into account the situation prevailing in the province at the time, it was decided not to carry out any further eradication actions.

The RPS stated that after the approval of Decision 2014/497/EU, no eradication measures in accordance with the EU Decision, have been or will be taken. However, the RPS stated that, despite the fact that it is not clearly spelled in the national decree, in the EZ (which belongs to the IZ) eradication measures will be taken in line with the Article 7 (3) and section 2 of Annex III of Decision 2014/497/EU.

During the meeting with researchers, the FVO team noted there is a unanimous opinion that, no efficient and effective eradication is possible in the DA anymore, due to:

- the dimension of the outbreak and spread of the bacteria in the province;
- high populations and very intense vector activity;
- the possibility of having more host plants than those already identified and their location in private gardens.

The national decree of 26 of September requires that the RPS of Puglia shall implement the following measures:

Within the BZ and in addition to the requirements in Section 2 of Annex III of the EU Decision:

- weed elimination to control the nymphs of insect vectors;
- removal of all host plants along roadsides, road tabs, ditches, canals, green areas, etc.

Within the SZ, where there are no specific EU requirements:

- insecticide treatments to control confirmed or potential vector insect populations;
- weed elimination to control the nymphs of insect vectors;
- removal of all host plants along roadsides, road tabs, ditches, canals, green areas, etc.;
- intensive monitoring of host plants to check for *Xf* at the most appropriate time.

Within the EZ (part of the IZ), the national decree does not refer to the EU measures of Section 2 of Annex III of Decision 2014/497/EU:

- insecticide treatment to control confirmed or potential vector insect populations;
- weed elimination to control the nymphs of insect vectors;
- removal of all host plants along roadsides, road tabs, ditches, canals, green areas, etc.;
- intensive monitoring to check for *Xf* through the sampling of host plants;
- removal, without laboratory testing, of all infected host plants or plants considered to be infected on the basis of visual inspections or which show symptoms of *Xf*.

The RPS stated that although there is no specific mention in the decree of the measures to be taken in the IZ, they plan to advise growers in the remaining part of the IZ, to take the following containment measures:

- insecticide treatments and the control of insect vectors in three periods, January-April, May-August and September-December;
- regular pruning in the olive orchards;
- timely pruning of the asymptomatic branches when the initial symptoms appear;
- in the infected orchards, removal of plants strongly attacked and not anymore in production.

The RPS stated that, further mandatory plant health measures to be implemented in the IZ of the province of Lecce shall be adopted by subsequent decision of MIPAAF, in response to proposals of the National Plant Health Committee.

Conclusion on eradication measures

In April 2014, in the five small size outbreaks identified outside the main infected area of Gallipoli, eradication tasks were carried out. However, some months later further trees were found infected in four of these five outbreak sites.

After the large expansion of the outbreak area in the summer throughout Lecce province, no eradication measures have been taken in the IZ. This is not in line with Article 7 (3) and Section 2 of Annex III of the EU Decision 2014/497/EU. However, the decree of 26 September outlines eradication measures in the newly established EZ in Lecce.

6 OVERALL CONCLUSIONS

Extensive human and financial resources have been made available for research and containment of *Xylella fastidiosa* outbreak and there is a good level of awareness about the problem.

With one exception, none of the eradication measures required by Decision 2014/497/EU, have been carried out. The situation has deteriorated since the last audit and *Xylella fastidiosa* continues to spread rapidly. The current Italian policy for the Infected Zone is now containment of *Xylella fastidiosa* and measures aiming at full eradication of the pest are not carried out.

The current controls do not ensure that host plants not fulfilling the requirements of the EU Decision remain in the Demarcated Area. All the existing garden centres located in the Demarcated Area have not been identified and, therefore are not officially controlled.

There is a possibility that not all host plant species have been identified and pathogenicity tests for a range of genera (including *Vitis* and *Citrus*) have not been concluded. Until the precise host range of *Xylella fastidiosa* is known, the movement restrictions in place (although applied to a wider range of species than required in the Decision) do not provide adequate security that no infected plants leave the area.

The proposed intensive surveys in the Eradication Zone, Buffer Zone and Security Zone, will help in the early detection of *Xylella fastidiosa* and enable the implementation of rapid eradication. This strategy could also limit the natural spread of the insect vector to new areas. However, taking into account the high populations and the passive mobility of the insect vector (vehicles, wind), the protective function of the two zones is questionable.

There is a significant risk of further spreading of *Xylella fastidiosa* outside the Demarcated Area by the movement of host plants and by the high populations and mobility of the insect vectors.

7 CLOSING MEETING

A closing meeting was held on 25 November 2014 at the headquarters of the Regional Plant Health Service of Puglia in Bari, with representatives of the SA and the RPS visited during the audit.

During the meeting, the preliminary findings and conclusions of the FVO team were presented, which were, provisionally accepted by the authorities.

8 RECOMMENDATIONS

The Single Authority in Italy is recommended to:

N°.	Recommendation
1.	Ensure that, Italian national and regional legislation implements provisions that are fully in line with the Decision 2014/497/EU.
2.	Ensure that, if the presence of <i>Xylella fastidiosa</i> is confirmed outside the Infected Zone, the delimitation of the Infected Zone and Buffer Zone shall be reviewed and changed accordingly, in line with point 4 of Section 1 of Annex III of Decision 2014/497/EU.
3.	Ensure that, all garden centres located in the Demarcated Area are immediately identified and included in the official control regime, in order to prevent any movement of plants for planting outside the Demarcated Area, that do not fulfil the requirements of Annex II of Decision 2014/497/EU.
4.	Ensure that, in the Infected Zone, measures are taken in line with Article 7 (3) and section 2 of Annex III of Decision 2014/497/EU.
5.	Ensure that procedures in the ARECBC laboratory, particularly the coding of samples and the readings of test results are reliable, thus ensuring that any presence of <i>Xylella fastidiosa</i> can be confirmed in line with Article 6 (1) of Decision 2014/497/EU.

The competent authority's response to the recommendations can be found at:

http://ec.europa.eu/food/fvo/rep_details_en.cfm?rep_inspection_ref=2014-7327

ANNEX 1 - LEGAL REFERENCES

Legal Reference	Official Journal	Title
Dir. 2000/29/EC	OJ L 169, 10.7.2000, p. 1-112	Council Directive 2000/29/EC of 8 May 2000 on protective measures against the introduction into the Community of organisms harmful to plants or plant products and against their spread within the Community
Dec. 2014/497/EU	OJ L 219, 25.7.2014, p. 56–64	2014/497/EU: Commission Implementing Decision of 23 July 2014 as regards measures to prevent the introduction into and the spread within the Union of <i>Xylella fastidiosa</i> (Well and Raju)