

Abstract: Spittlebugs (Hemiptera; Aphrophoridae) natural enemies appear to be a not so effective guild. Olive orchard now invaded by *X. fastidiosa pauca* OQDS strain or threatened of, do not constitute the exception. *Zelus renardii* (Zr) (Hemiptera; Reduviidae) originate in Nearctic but spontaneously acclimated in Europe. We attempted the assassin bug breeding from 2015 starting with *M. gladiata* adults and nymphs, and have had the opportunity to offer many preys belonging to several insect orders to the assassin bug. Adult *Z. renardii* demonstrated to successfully attack and feed on almost all the offered insects, including large Coccinellidae as *Harmonia axyridis* (Pallas, 1773) (the Asian ladybeetle) and comparatively enormous *Gromphadorhina portentosa* (Schaum, 1853) (Blattodea Blaberidae; the Madagascar hissing cockroach) juveniles. Recently we offered to Zr for food adult, and juveniles of several phytophagous Hemiptera Taxa, including *Philaenus spumarius*, all of those have successfully preyed. Evidence suggest that Zr may serve as a biological control agent in olive orchards, in an inoculative or inundative program if bred in sufficient number. To have the opportunity we systematically tried several different dietary regimes all functional to a Zr mass-breeding program: from living insect preys to a purposely formulated meridic artificial diet. Revealing our about a one-year long experience, we discuss predator bionomics about a full artificial breeding possibility.

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Session 6 – Detection and identification

From transnational research collaboration to regional Standards: the EPPO diagnostic protocol for *Xylella fastidiosa*

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Abstract: In 2013 the EPPO Secretariat received the official report of *X. fastidiosa* by the National Plant Protection Organisation of Italy. A regional Diagnostic Protocol on the bacterium had been published in 2004 but it focussed mainly on *Citrus* spp. and *Vitis*. Because of the significant changes in the bacterium's geographical distribution and its list of plant host species, the revision of the EPPO Diagnostic Protocol was considered a priority; an expert working group was established and the EPPO Standards on *X. fastidiosa* was published in September 2016.

As the scientific knowledge on the bacterium, its vectors and host species has advanced and diagnostic tests have been and are being developed and validated in the framework of national and transnational collaborative research projects, work has started on a further revision of the protocol. The presentation will focus on a comparison of the different versions of the EPPO Diagnostic Protocol on *X. fastidiosa* and will show how research evidences support diagnostic activities and more broadly, regional policy.

First international proficiency testing for laboratory performance on *Xylella fastidiosa* detection

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Abstract: A proficiency test (PT) to evaluate the performance of laboratories involved in molecular and serological detection of *X. fastidiosa* was carried out in early 2017; 35 laboratories from EU/non-EU Countries tested 4 different methods to purify DNA, conventional and qPCR assays, and 2 ELISA tests. The number of resultant positive agreement/negative agreement/positive deviation/negative deviation was used to determine the laboratory performance (i.e. accuracy 100%). The overall results showed that all laboratories were able to correctly diagnose *X. fastidiosa* in the blind samples

containing the highest *X. fastidiosa* concentrations, whereas the performance of several laboratories was negatively affected by the lack of detection in the samples with the lowest concentrations, both through molecular and serological tests. Accuracy level of 100% (laboratory conformed to the PT) was successfully recovered in the majority of the laboratories performing qPCR and PCR assays on DNA purified using at least 2 of the 4 tested protocols. The use of automated platform ensured higher laboratory performance. As expected, results of the ELISA tests generated lower performance values in the majority of the laboratories, due to the lack of detection of positive samples containing the lowest the bacterial concentration. This study provides a good overview on the laboratory performance for the diagnostics currently used in the EPPO countries and indicate useful improvements that laboratories can adopt to achieve a better performance.

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Towards an efficient sampling procedure for early detection of *Xylella fastidiosa* in asymptomatic plants

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Abstract: In the past, *X. fastidiosa* was restricted to a number of countries through the America's (Almeida and Nunney 2015). This situation has been dramatically altered in Europe as *X. fastidiosa* has been found now in various host plants in several European countries. Movement of plants for planting is considered to be the most important pathway for entry of the pathogen into Europe, especially when present without symptoms (EFSA PLH Panel 2015). The significance of this pathway is enhanced by the enormous host range of *X. fastidiosa* and the high volumes of plants for planting used to be imported from countries where *X. fastidiosa* was known to occur. Currently there is an urgency for guidelines for sampling, specifically when asymptomatic *X. fastidiosa* infections can easily escape laboratory testing, due to its heterogeneous distribution in plant, that may lead to false-negative results. Experiments have been initiated towards an efficient sampling for early detection of *X. fastidiosa* in asymptomatic plants under confined conditions, by simulating different levels of infection in plants and sampling at different % and different sample size. Strains PD 7202 (ST 53) and PD 7211 (ST 73) belonging to *X. fastidiosa* subsp. *pauca* and originating from *C. arabica* are used (Bergsma-Vlami et al 2017). Particular attention is given to *P. myrtifolia*, *P. avium*, *P. domestica*, *N. oleander* and *C. arabica*. In our system, pre-defined *X. fastidiosa* inoculum per unit (f.w of petioles/midribs in g) is being mixed with healthy units at different infection levels and analysed based to EPPO diagnostic standard (EPPO 2016). Preliminary results will be presented.

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