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Transcriptome profiling of two olive cultivars infected by *Xylella fastidiosa*

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A devastating outbreak of a strain of *Xylella fastidiosa* is ravaging the olive trees of southern Puglia, Italy (Saponari *et al.*, 2015). The bacterium, discovered in 2013, causes the Olive Quick Decline Syndrome (OQDS), a destructive disease characterized by apical and marginal leaf scorching, extensive branch and twig dieback, followed by death of the trees (Martelli *et al.*, 2016).

Genome studies (Giampetruzzi *et al.*, 2015; Loconsole *et al.*, 2016;) have shown that the olive-infecting strain of *X. fastidiosa* belongs to the subspecies *pauca*, sequence type 53 (ST53), whereas epidemiological investigations and transmission tests proved that the xylem-feeding meadow froghopper *Philaenus spumarius* is the vector of the bacterium (Saponari *et al.,* 2014).

The impossibility of curing *X. fastidiosa*-infected plants calls for the implementation of management strategies aimed at preventing infection and containing bacterial spread in the field. The international experience acquired with some of the best known *Xylella*-induced disorders, such as Pierce's disease of the grapevine and Variegated chlorosis of citrus, indicates that disease containment relies on the mechanical and/or chemical control of the vector(s), and the elimination of inoculum sources, i.e. infected hosts. It is also common knowledge that these measures can hardly eradicate the pathogen, when it has entrenched itself in areas with favourable climatic and environmemtal conditions. In these cases, co-existence with the disease, e.g through the identification and use of resistant plants would represent a desirable option.

With this aim in mind, field surveys carried out for scouting resistant germplasm allowed the identification of a possible source of resistance in olives of cv. Leccino which, in areas with a very heavy inoculm pressure, showed limited signs of infection, contrary to the extensively grown local cvs Cellina di Nardò and Ogliarola Salentina.

Quantitative determination of the bacterial population in infected olive trees showed that cv. Leccino hosts a much lower concentration of bacterial cells than cv. Ogliarola salentina, suggesting that it may possess genetic constituents and/or regulatory elements counteracting *X. fastidiosa* infection.

A global transcriptome profiling comparing healthy and infected trees of the two above-mentioned cultivars, was performed for determining whether a tolerant/resistant condition of cv. Leccino exists. Statistical analysis of the differentially expressed genes (Figure 1) from plants of the cv. Leccino (LC) and Ogliarola salentina (OG) healthy (H) or infected by *Xylella fastidiosa* (X), showed a distinct clustering of healthy and infected plants of both cvs and locates the infected cv. Ogliarola salentina more distantly than the cv. Leccino from the corresponding healthy olives.

Quantitative analysis revealed that 659 and 447 genes were differentially regulated upon *Xylella fastidiosa* infection, in cvs Leccino and Ogliarola salentina, with respect to the healthy plants of the same cultivars (Figure 2). The differential gene expression of infected *vs* healthy plants demonstrates that olives of both cvs perceive the presence of *Xfp*. Moreover, in depth analysis of main altered genes of the susceptible cv Ogliarola salentina indicated that they are related to water stress and to an intense activation of the defense response imposed by *Xylella fastidiosa* infections.

These findings suggest that cv. Leccino is endowed with an intrinsic tolerance to *Xylella fastidiosa*, which makes it eligible for further studies aimed at investigating molecular pathways underlying its different defense response.

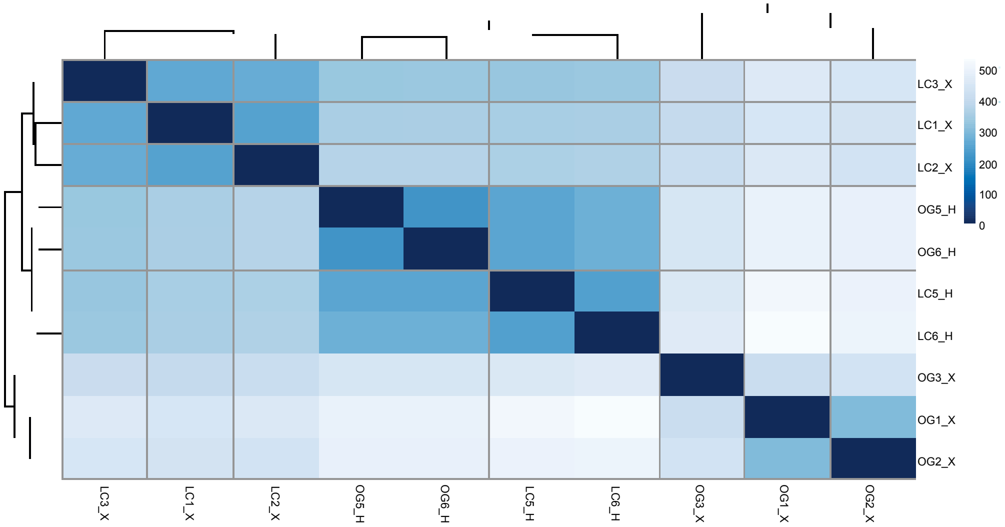

Figure 1. Statistical analysis of the transcripts libraries using rlog trasformation dato of gene expression. The heatmap shows the hierarchical clustering of biological replicates using sample-to-sample distances. Blue and white colors represent low and high values of distance, respectively. Healthy (LC5_H, LC6_H, OG5_H, OG6_H) and *Xylella fastidiosa*-infected plants (LC1_X, LC2_X, LC3_X, OG1_X, OG2_X, OG3_X) plants of cvs Leccino (LC) and Ogliarola salentina (OG) are showed.

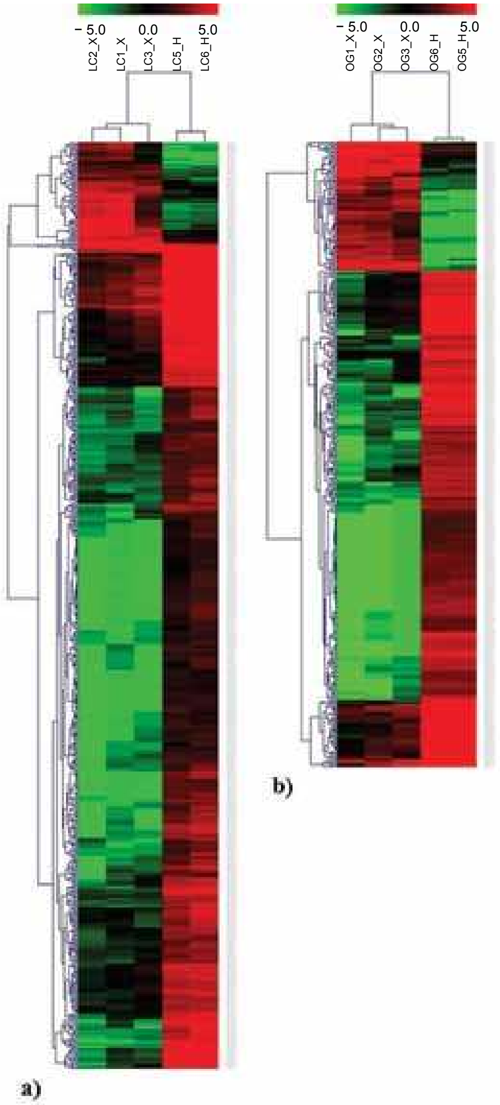

Figure 2. Heat maps of expression levels (log2) created with MeV v4.9 (http://www.tm4.org/mev) showing the 659 (a), 447 (b) differential expressed transcripts of the Xf-infected cv. Leccino (LC1_X; LC2_X; LC3_X) vs healthy (LC5_H; LC6_H), the Xf-infected cv. Ogliarola (OG1_X; OG2_X; OG3_X) vs healthy (OG5_H; OG6_H). Fold expression values are indicated by different colours.

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