Exogenous dsRNA applications to identify novel candidate susceptibility genes to downy mildew

Elisabetta Sergi[1*], Giuliana Maddalena[1], Valentina Ricciardi[1], Demetrio Marcianò[1], Beatrice Lecchi[1], Osvaldo Failla[1], Silvia Laura Toffolatti[1], Gabriella De Lorenzis[1]

[1] Affiliation Department of Agricultural and Environmental Sciences, via G. Celoria 2, 20133 Milan (Italy)

[*] Corresponding author: elisabetta.sergi@unimi.it*

One of the major threats to viticulture is represented by fungal pathogens. *Plasmopara viticola*, an oomycete causing grapevine downy mildew, is one of the principal causes of grape production losses. The most efficient management strategies are represented by a combination of agronomical practices, fungicides' applications, and use of resistant varieties. Plant resistance is conferred by the presence of resistance (R) genes. Opposed to them, susceptibility (S) genes are encoded by plants and exploited by pathogens to promote infection. Loss or mutation of S genes can limit the ability of pathogens to infect the host. By exploiting post-transcriptional gene silencing, known as RNA intereference (RNAi), it is possible to knock-down the expression of S genes, promoting plant resistance. Gene silencing is induced upon exogenous application of dsRNA, which can interfere with protein synthesis. With the aim of identifying new candidate genes to be employed in breeding programs, three novel candidate S genes to downy mildew, *VviLBDlf7*, *VviAS1* and *VviB3*, have been identified. Candidates' validation was carried out through RNAi on the susceptible cultivar Pinot noir. Disease severity was estimated by experimental inoculation of *P. viticola* on leaves sampled at different timepoints after the treatment with dsRNA, while gene expression was evaluated by real time RT-PCR. Successful downregulation of candidate genes resulted in a significant reduction of plants susceptibility to downy mildew, suggesting our candidates as downy mildew S genes, leading to the possibility of employing an RNAi-based strategy as a more sustainable alternative to conventional management strategies.

Keywords: S genes, RNAi, gene silencing, *Plasmopara viticola*, *Vitis vinifera*.