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# PR Proteins and Induced Resistance against Pathogens and Insects

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# Laser microdissection of grapevine leaves infected by *Plasmopara viticola* reveals site-specific defence-related processes.

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Downy mildew, caused by *Plasmopara viticola*, is one of the most important diseases of grapevine. *P. viticola* infects grapevine leaves and young berries by stomata and develops intercellular mycelium in the mesophyll. Gene expression analyses are commonly carried out on whole grapevine leaves at the early stages of infection. However, only a small fraction of leaf cells are in contact with the pathogen, and the large portion of non-infected cells could mask the transcriptional changes related to defence reactions. More accurate information on the modulation of defence-related genes at the site of *P. viticola* infection could help in better understanding the regulation of the defence at the site of infection and to clarify the reaction of the surrounding tissues.

Laser microdissection was used to precisely isolate cells at the site of *P. viticola* infection and at the adjacent layers from inoculated leaves of *in vitro*-grown grapevines. Protocols for sample fixation, laser microdissection and RNA isolation from group of cells were optimized, and the expression of genes encoding pathogenesis-related (PR) proteins, transcription factors, and enzymes involved in defence processes was analysed by real-time RT-PCR. The expression of defence-related genes was induced by *P. viticola* in stomata and in the adjacent cells, and their expression level was greater at the site of infection compared to the whole infected leaf. Our results demonstrated specific activation of defence-related processes at the sites of *P. viticola* infection, which were masked in the whole-leaf analysis. This optimized protocol can be used for site-specific transcriptomic analysis and it may be also suitable to study plant cell interactions with other pathogens.