Società Chimica Italiana
Divisione di Spettrometria di Massa

Università di Catania

Book of Abstracts

Untargeted metabolomics approach to understand grapevine communication mediated by volatile organic compounds against downy mildew

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Summary: *Volatile organic compounds (VOCs) play crucial roles in the communication of plants with other organisms and are mediators of plant defense against pathogens. The objective of this work is to understand the mechanism of grapevine communications mediated by VOCs against grapevine downy mildew (caused by* Plasmopara viticola*), using an untargeted metabolomics approach.*

Keywords *volatile organic compounds, downy mildew, metabolomics, grapevine*

1. Introduction

Plants can produce a wide variety of volatile organic compounds (VOCs), which can play crucial roles in the regulation of plant responses against pathogens[1]. Different modes of action against phytopathogens have been attributed to VOCs, such as induction of plant resistance and direct inhibition of pathogen growth [1]. Recent studies showed that the amount of some volatile terpenes was higher in resistant than susceptible grapevine genotypes upon *Plasmopara viticola* inoculation, indicating their possible involvement in resistance mechanisms against this pathogen [2,3]. This work aims at identifying the metabolic response of VOC-treated grapevine leaves and the potential activation of VOC-mediated resistance mechanisms using an untargeted metabolomics approach.

2. Experimental

Susceptible grapevine leaf disks were treated with a pure terpene or with water (control) at one day before and one hour after *Plasmopara viticola* inoculation (*P. viticola*-inoculated) or water application (mock-inoculated). Downy mildew severity was assessed six days post inoculation (dpi) as a percentage of the leaf disk surface covered by sporulation [3]. Leaf samples were collected at two time points (one and six dpi) and an untargeted metabolomics approach was applied using ultra-high pressure liquid chromatography- electrospray ionization - high resolution- quadrupole-time of flight-mass spectrometry(UHPLC-ESI-Q-TOF-MS) analysis [4,5]. The Q TOF was operated in both positive and negative ion modes [5]. Raw data were processed using an in-house R script and statistical analysis was performed with the MetaboAnalyst online platform (https://www.metaboanalyst.ca/MetaboAnalyst/home.xhtml) [4]. Features with significant changes in abundance were identified according to the Volcano Plot analysis, using a *t*-test (adjusted p-value ≤ 0.05) and a fold-change higher than two in at least one comparison [6]. Putative annotations andmetabolic pathways of annotation features were obtained by KEGG (https://www.kegg.jp/kegg/)with *Vitis vinifera* as a reference organism using the webserver MassTRIX(http://masstrix3.helmholtzmuenchen.de/masstrix3/start.html)[4].

3. Results

The assessment of disease severity confirmed that the pure volatile terpene reduced downy mildew severity on susceptible grapevine leaf disks. Principal component analysis applied on the features (specified by retention time and *mass to charge* ratio) discriminated samples according to VOC treatment and time point, indicating global metabolite changes in VOC-treated leaf disks. The annotation of selected features and the respective metabolic pathways will highlight grapevine defense responses activated by VOC treatments.

4. Conclusions

The untargeted metabolomics approach will identify the metabolic response of grapevine leaves to pure VOCs and will help to improve the knowledge of grapevine defense mechanisms activated by VOCs against pathogens.

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