



Grapevine- *Plasmopara viticola* Interactome: defining interacting partners through FRET-FLIM

Place of work:

Grapevine Pathogen Systems Lab, C2 building, 4th floor, lab 37, BiolSI at Faculdade de Ciências da Universidade de Lisboa

Supervisors: Rita B. Santos and Andreia Figueiredo (FCUL)

Contact: aafigueiredo@fc.ul.pt

Grapevine (*Vitis vinifera* L.) has high economic impact worldwide with plantation areas over 7.4 Mha (OIV data, 2021) and it also represents one of the most polluting agronomic industries with many tons of pesticides being used on each growing season. Current disease management practices for grapevine downy mildew rely on the intensive use of pesticides. However, directives from the European Union demand more sustainable agriculture practices, with low pesticide input. In the last years, our group has been studying the molecular mechanisms behind pathogen recognition and the activation of grapevine defenses. On that sense, we have identified a serine protease, VviSBT4.19, that we hypothesize that is linked to *Plasmopara viticola* recognition and activation of the signalling cascade. To assess our hypothesis, we have conducted a Yeast Two Hybrid (Y2H) assay that allowed us to define 4 possible interacting partners. We have also defined some *P. viticola* proteins that putatively interact with VviSBT4.19.

To confirm the interaction between VviSBT4.19 and the possible interacting partners identified by Y2H, we will use the Förster Resonance Energy Transfer by Fluorescence Lifetime Imaging (FRET-FLIM) technology. FRET involves the energy transfer through coupling of a donor and acceptor chromophore.

The main aim of this thesis proposal is to confirm the interaction between our candidates. For that the student will participate in the cloning of the candidate genes using the Gateway system, production of the recombinant candidate proteins in *N. benthamiana* by agroinfiltration and confirmation of its production. Finally, FRET-FLIM studies will allow the definition of the interacting partners of this grapevine serine protease.

Student may apply to a BiolSI Junior fellowship (6 months).