DOTTORATO DI RICERCA IN BIOLOGIA CELLULARE E DELLO SVILUPPO

40th CYCLE
Project proposal for a Sapienza PhD scholarship

Main research line

Title: Formation and homeostasis of oligogalacturonides (OGs) in immune signalling

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Summary

Plant immunity is activated through plasma membrane receptors that recognize microbe-associated molecular patterns (MAMPs) and/or pathogen-derived effectors (Ngou et al., 2022). However, a primary danger for the plant cells is represented by any alteration to their encapsulating protective structure formed by the extracellular matrix (i.e. the cell wall, CW). CW is the cell compartment most exposed to damage and alterations of its integrity pose a high risk by offering an easy passageway to the pathogens into the plant tissue (Savatin et al., 2014; Molina et al., 2024; Pontiggia et al., 2024). Protection and repair of injured sites must have been a primary need of plants, and it is conceivable that a key step in the evolution of plant immunity was the acquisition of the ability, also in the absence of pathogens, to respond to cell and tissue damage, by perceiving damage-associated molecular patterns (DAMPs) generated during injuries (De Lorenzo et al., 2018). The oligogalacturonides (OGs), i.e. the first discovered and best characterized DAMPs in the living kingdom, are generated through a specific interaction between a plant protein named PGIP (polygalacturonase inhibiting protein), and microbial polygalacturonases (PGs) (Kalunke et al., 2015; De Lorenzo and Cervone, 2022). The crystal structure of the complex has recently shown that PGIP is not a classical inhibitor that hinders the active site of PG but rather as an enzyme modifier that creates with PG a dimeric enzyme and a novel active site especially designed for the formation of immune-reactive OGs, thus demonstrating that the formation of OGs is not a casual event (Xiao et al., 2024). Moreover, a dedicated system represented by redundant, developmentally regulated and oxidases (OGOXs) specifically maintain the homeostasis of OGs and play a role in the response to pathogens and wounding (Benedetti et al., 2018; De Lorenzo and Cervone, 2022)(manuscript submitted). Notably, OGs have been recently shown to be crucial elements in the regulation of the dynamics of plasma membrane (PM) receptor kinases (RKs) in response to the peptides known as RAPID ALKALINIZATION FACTORs (RALFs) mediated by the RK FERONIA (Liu et al., 2024), reinforcing the view that OGs are key regulatory molecules in the plant.

This PhD project wants to investigate whether and how the formation of OGs by a PG-PGIP mechanism and subsequent OG homeostasis by OGOX are at the basis of plant responses also in the absence of pathogenic microbes, i.e. upon wounding and during developmental processes.

OBJECTIVE 1. The structural information gained from the recently solved crystal structure of the *Pv*PGIP2-*Fm*PG complex will be exploited to search for yet unexplored specific OG-forming combinations of PGs and PGIPs that regulate wound healing as well as other physiological processes in Arabidopsis. In parallel, biochemical approaches will be used also to isolate Arabidopsis PGs that interact with the Arabidopsis PGIP1 and PGIP2. PGs of interest will be further characterized, also through reverse genetic approaches.

OBJECTIVE 2. The role of OGOX1 and its membrane-associated form will be elucidated in the context of immune signalling and the dynamics of PM signalling receptor complexes.

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