

Plant lipids play a key role in plant growth and development, serving both as an energy and structural material and as essential signaling molecules involved in plant responses to biotic and abiotic stresses. Based on their structure, plant lipids can be divided into four major classes: 1) galactolipids, 2) phospholipids, 3) triacylglycerols (TAGs), and 4) sphingolipids. TAGs, stored in the form of lipid droplets (LDs), represent a major energy source, especially during seed germination and the early stages of seedling development. Although the processes of TAG biosynthesis and LD formation have been well characterized, the enzymatic mechanisms underlying their degradation remain largely unknown.

To date, two major metabolic pathways have been shown to participate in TAG degradation during seed germination: lipolysis, the primary route for TAG breakdown, and lipophagy, an alternative mechanism linked to autophagy. In lipolysis, TAGs stored in LDs are hydrolyzed by specific lipases into free fatty acids and glycerol, providing substrates essential for  $\beta$ -oxidation and energy production. In *Arabidopsis thaliana*, the main lipase responsible for TAG breakdown during seed germination is SUGAR-DEPENDENT1 (SDP1). However, the persistence of residual TAG degradation in *sdp1* mutants indicates the presence of additional, as-yet-unidentified lipases involved in this process.

Lipophagy is a specialized form of autophagy, the cellular recycling system responsible for removing damaged or obsolete structures. In lipophagy, LDs are directed to the vacuole, where they are degraded by vacuolar lipases. Two types of lipophagy are distinguished: macrolipophagy, in which LDs are sequestered into autophagosomes and delivered to the vacuole, and microlipophagy, in which LDs directly interact with the tonoplast (vacuolar membrane) and are subsequently engulfed into the vacuole. Despite intensive research efforts, the precise mechanisms of LDs degradation in plant cells remain unclear. Understanding these processes is essential for advancing our knowledge in plant lipid metabolism and may have practical applications in plant biotechnology—for instance, increasing oil content in seeds, modifying fatty acid composition, or improving plant stress resistance.

The main goal of this project is to comprehensively characterize the enzymatic activity, subcellular localization, and physiological function of a putative lipase potentially responsible for vacuolar TAG degradation in *A. thaliana*. Our preliminary studies have identified a promising candidate: a previously uncharacterized  $\alpha/\beta$ -hydrolase containing the conserved GX SXG motif and a catalytic triad (Ser–Asp–His) typical of TAG lipases. Bioinformatic analyses predict the presence of a signal peptide and target localization of this protein to the vacuole, strongly suggesting its potential involvement in lipophagy. Importantly, *A. thaliana* mutants lacking a functional version of this protein display delayed TAG degradation during seed germination and early seedling development, further supporting its role in lipid mobilization.

The results obtained in this project will significantly advance our understanding of the complex enzymatic network involved in lipid degradation in plants. Consequently, the project represents an important contribution to the field of plant lipid metabolism and opens new research perspectives related to lipid regulation in higher plants.