ABSTRACT

Plant resiliency and survival hinges on rapid, efficient, and synchronized responses to stress and expedient, coordinated execution of developmental processes. The environmental cues are often simultaneous and compounding. The plant must perceive and differentiate these stimuli and subsequently manifest an appropriate and integrated response. Therefore, communication across the whole plant is essential. The vascular system not only transports nutrients, water, and energy, but also serves as an information highway to traffic macromolecular signals. The phloem mobilizes nucleic acids, proteins, hormones, and lipophilic compounds from source to sink for targeted systemic signaling. A handful of phloem-mobile nucleic acids and proteins have been unambiguously linked to stress response and development: for example, BEL transcripts for tuber development and Flowering Locus T (FT) as a central regulator of the transition to flowering. However, given the expansive number of macromolecules identified in phloem sap, their functions in long-distance signaling remains an active area of research. Moreover, the presence of lipids and lipid-binding proteins (LBPs) in the hydrophilic environment of the phloem is an anomalous phenomenon that prompts further investigation. In this dissertation, I have elucidated the role of phloem-mobile lipid binding proteins in lipid-mediated long-distance signaling for abiotic stress response. First, I surveyed several small lipid-binding proteins previously identified in the phloem. Annexin 1 (ANN1), Major Latex Protein-like Proteins 43 and 423 (MLP43 and MLP423) and Bet v1 Allergen bind neutral and negatively charged phospholipids and predominantly localize to the periphery of the cell. Their genetic expression profiles differ from one another in response to various abiotic stress factors, indicating that they act in distinct mechanisms.

Further, the expression patterns correlate with those of phospholipases that generate phosphatidic acid, a known regulator of stress response. Second, I expand upon the function of Phloem Lipid-Associated Family Protein (PLAFP) in lipid-mediated stress response and signaling. I characterized the phosphatidic acid (PA) binding activity of PLAFP and identified a receptor candidate for the PLAFP-PA signal. PLAFP comprises a single PLAT/LH2 domain, which can act to bind lipids and facilitate protein-protein interaction. Using homology modelling, mutagenesis, and lipid overlay studies, I show that the conserved basic residues Lysine-42 and Ariginine-82 and an adjacent tryptophan-enriched hydrophobic groove within PLAFP contribute to PA binding and possible solubilization. Consistent with a predicted function in protein-protein interaction, PLAFP co-localizes with receptor-like kinase Vascular Highway 1 Kinase (VH1K) in vivo. RNA-Sequencing discovered several dozen differentially expressed genes when PLAFP expression levels are altered, which suggests PLAFP elicits downstream transcriptional changes. Finally, I developed a novel optogenetics-based method for the investigation of long-distance translocation in plants, which revealed PLAFP is systematically transported. Taken together, PLAFP participates as a phloem-mobile signal in a tightly regulated and specific mechanism for PA-mediated systemic stress response. This represents an emerging field of research to understand the role of lipids in long-distance signaling both as components of the membrane and as signals themselves.