Spatio-temporal dynamics in the composition and function of the CCaMK/CYCLOPS complex

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Summary

Plant root symbioses with arbuscular mycorrhiza (AM) fungi and nitrogen-fixing bacteria bear huge potential for sustainable agriculture by reducing the chemical fertilizer input required to maintain high crop yields. The regulation and signal transduction mechanism leading to AM and the nitrogen-fixing root nodule symbiosis (RNS) share common components including the calcium and calmodulin dependent protein kinase (CCaMK) and its phosphorylation target CYCLOPS, a DNA binding transcriptional activator (Tirichine et al., 2006; Yano et al., 2008; Singh et al., 2014). The CCaMK/CYCLOPS complex is a central regulatory hub in symbiosis signaling. It controls the expression of three transcriptional regulators of three distinct developmental programs. NIN controls nodule organogenesis and, together with ERN1, infection thread formation while RAM1 is indispensable for arbuscule development (Singh et al., 2014; Pimprikar et al., 2016; Cerri et al., 2017). The corresponding promoters control distinct timing, expression domains and response to different stimuli. The promoter choice and activity of CCaMK/CYCLOPS must therefore be coordinated at a spatio-temporal and a stimulus-specific level to trigger appropriate cell developmental programs. In the past, we identified additional putative complex components that may contribute to binding of diverse cis regulatory elements within the known target promoters of CCaMK/CYCLOPS. The doctoral student will study the relevance of the identified additional complex components using a range of techniques, including reverse genetics utilizing transposon insertion populations or CRISPR/CAS genome editing technology. The spatio-temporal composition of the complex and its structural rearrangement will be studied via in vivo FRET-FLIM in root hair nuclei in response to signals emanating from arbuscular mycorrhiza fungi or nitrogen-fixing bacteria. Biochemical in vitro measurements will be used to quantify protein-protein and protein-DNA binding affinities. We expect to unravel key steps in the molecular dynamics of the CCaMK/CYCLOPS complex underlying the specific activation of the appropriate and distinct developmental programs in response to fungi and bacteria and thus the establishment of AM and root nodule symbioses.