"Uncovering gene expression patterns of a Leucine-rich Repeat Receptor Kinase during Root Nodule Symbiosis

Medicago truncatula is a clover-like plant of Mediterranean origin that is an autogamous annual species. This genetically amenable model plant species possesses a small genome and can be rapidly cultivated in laboratory and greenhouse settings. Importantly, M. truncatula can establish beneficial plant-microbe symbiosis with bacteria called rhizobia leading to the development of nitrogen fixing root nodules. As a model organism for economically important crops such as alfalfa (M. sativa), M. truncatula provides a platform for studying the molecular genetics of nitrogen fixation. In our study, we focus on two specific Leucine-rich Repeat Receptor Kinases (LRR-RKs), Medtr7g013680 and Medtr7g407130 located on chromosome 7 to understand their roles in root nodules symbiosis. With the help of bioinformatic approaches, we retrieved the 3kb upstream promoter sequences of these LRR-RKs, and designed specific primers for amplification using Polymerase Chain Reactions. Subsequently, the promoters were cloned using the Golden Gate assembly system and resulting plasmids transformed into Escherichia coli. In the end, the construct will be transformed into Agrobacterium rhizogenes and a hairy root experiment will be performed for 14 days to confirm the expression of these genes during various stages of nodule development. In this study due to time limitations we were unable to clone both vectors successfully to begin Agrobacterium hairy root transformation. We will continue to clone these vectors until golden gate assembly step two is successfully completed. This study will provide insights into the regulatory role of two LRR-RLKs in the early signaling pathways of root nodule symbiosis. We hope to better understand the symbiosis signaling pathway and provide potential applications in agriculture by improving nitrogen fixation in the environment.