

The impact of entry exclusion on conjugation of *Agrobacterium tumefaciens* virulence plasmid

Agrobacterium tumefaciens is the causative agent of crown gall disease. Infection by this plant pathogen leads to the formation of tumors which adversely affect plant functions and stunt growth, ultimately leading to economic losses for nursery owners and crop farmers. Pathogenic *A. tumefaciens* carry a tumor inducing (Ti) plasmid, which encodes virulence genes and an entry exclusion system. The entry exclusion system antagonizes conjugation of the Ti plasmid into an *A. tumefaciens* strain already containing a similar plasmid, and is thought to be encoded by the *trbJK* region on the Ti plasmid. This study investigates the question: What is the impact of the entry exclusion system on the conjugation of the Ti plasmid into *Agrobacterium tumefaciens*? To examine this question, I will perform conjugation assays to compare the efficiency of Ti plasmid conjugation into two different stains of *A. tumefaciens*. To prepare for these conjugation assays, I have created a Δ *trbJK* mutant strain, ABD1, which lacks the genes putatively encoding the entry exclusion system. I will measure the rate of conjugation of the Ti plasmid into this ABD1 strain, and compare it to the rate of conjugation into the wildtype 15955 strain, which has a functional entry exclusion system. In order to track the movement of the plasmid and quantify donor, recipient, and transconjugant densities, I will label the chromosome and Ti plasmid of 15955 and ABD1 with antibiotic markers. Thus far, my project progress has included creating the ABD1 mutant and making significant headway in marking the ABD1 and 15955 strains. After completing construction of these strains, I will begin the conjugation assays to determine how entry exclusion influences Ti plasmid conjugation.