Function of Type III TAL effectors in Bacterial Canker of Mango

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Introduction

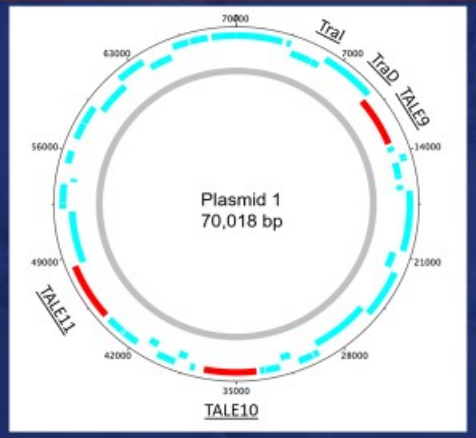
- *Xanthomonas citri* pv. *mangiferaeindicae* (Xcm) is a Gram-negative bacteria and the causal agent of the disease bacterial canker of mango. Common symptoms of Xcm on mango plants include black leaf lesions, chlorotic halos, and, in the most severe cases, premature fruit drop.

Background

- The complete genome of Xcm has been sequenced in our lab and is known to contain 11 TAL effector genes, which in other disease complexes, are known to contribute to virulence by targeting host genes for expression during infection. Three of the genes occur on a plasmid of Xcm (see below).
- The question remains as to which TAL effector genes of Xcm, if any, are involved in bacterial canker of mango.

Hypothesis

If TAL effectors have a function or are required for bacterial canker in mangos, mutations introduced into TAL effector genes of Xcm will result in phenotypic changes upon inoculation of mango leaves with the mutant strains.



Methods

To test the hypothesis, I will:

- Amplify sub-fragment of a TAL effector gene using Polymerase Chain Reaction (PCR) and clone into the vector pTOPOkn.
- The sub-fragment with pTOPO kn will be electroporated into Xcm and, since pTOPOkn cannot replicate in Xcm, rescue Kn^r strains that have sub-fragment integrated into TAL effector genes by homologous recombination and consequential disruption of TAL effector function.
- PCR will be used to validate integration into a TAL effector gene.
- TAL effector gene mutant strains will be assayed for pathogenicity on mango.

Expected Results

- Mutant strains for most, if not all, eleven TAL effector genes (the genes are highly similar in DNA sequence).
- Altered phenotypes of mutants in comparison to the wild type Xcm strain on mango.

Moving Forward

- Population of bacteria at the site of inoculation will be measured to determine if changes result in pathogen population changes.
- The next step will be to further characterize the TAL effector gene or genes whose loss results in changes in pathogenicity.

Sources

- Bai, F., Li, Z., Umezawa, A., Terada, N., & Jin, S. (2018). Bacterial type III secretion system as a protein delivery tool for a broad range of biomedical applications. *Biotechnol Adv, 36*(2), 482-493. doi:10.1016/j.biotechadv.2018.01.016
- Mak, A. N., Bradley, P., Bogdanove, A. J., & Stoddard, B. L. (2013). TAL effectors: function, structure, engineering and applications. *Curr Opin Struct Biol, 23*(1), 93-99. doi:10.1016/j.sbi.2012.11.001