ERWINIA OVERCOMES POTATO RESISTANCE BY ATTACKING ITS DEFENCES

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INTRODUCTION

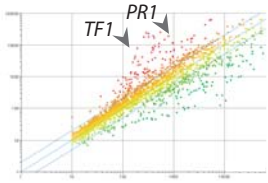
Erwinia carotovora subsp. *atroseptica* (*Eca*) is an economically important pathogen of potato, causing tuber soft rot and blackleg. There are no chemical treatments to control *Eca* in the field. Morever, there are no commercial cultivars with total resistance to *Eca*. Understanding mechanisms used by *Eca* during infection is crucial in the development of resistant cultivars.

The main weapons used by Eca to infect plants are plant cell wall degrading enzymes. However, the recently identified type III secretion system (T3SS) in *Eca* is also instrumental in injecting effector proteins into the host plant to suppress, manipulate or modulate defences during early stages of infection (Toth and Birch, 2005). The aims of this project were:

- To identify a mutant in *geneX,* a putative effector, and to assess its role in pathogenicity.
- To identify potato defence pathways modified by this putative effector.
- To use this knowledge for enhanced resistance to *Erwinia*.

Microarrays

To determine which other genes are up-regulated together with TF1 transcription factor, cDNA from leaves infiltrated with *Eca wt* vs. *geneX* mutant 0.5 hours post inoculation (hpi) were hybridised to an Agilent microarray. *PR1* gene was also shown to be up-regulated together with the TF1. *PR1* is a marker of salicylic acid-dependant pathways.



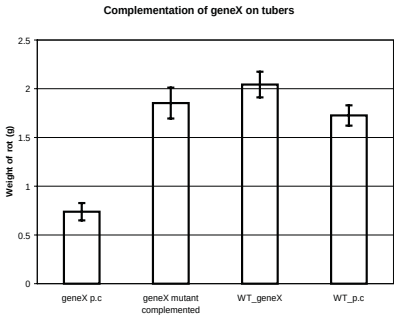
Results suggest that *geneX* product suppresses SA-dependent potato defence mechanisms

Improved resistance

- The above results suggest that constitutive expression of TF1 transcription factor can increase resistance to *Erwinia*.
- Transgenic lines with increased expression of the TF1 transcription factor were generated.
- Pathogenicity assays on Desiree vs. transgenic TF1 lines.
- Lesion measured from 2 to 17 days post inoculation (dpi).

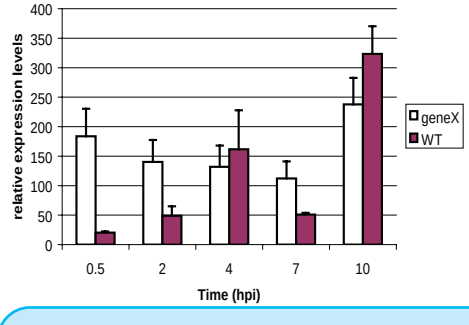
Characterisation of *geneX* mutant and Real-Time PCR

The role of *geneX* in virulence was determined by identifying a Tn5 insertion mutant in this gene. Thereafter, pathogenicity tests were carried out on both potato tubers and stems to compare virulence of *geneX* mutant vs *Eca* wild type (wt) strain. Results showed reduced virulence of mutant. *GeneX* mutant was also complemented with plasmid carrying *geneX* and complementation restored virulence of mutant to *Eca* wt levels.



geneX is required for virulence in *Eca*

Relative expression levels of TF1 in leaves infiltrated with *Eca* or *geneX* mutant



To study the effect of *geneX* products on potato plants, the expression of transcription factor1 (TF1) was monitored using Real-Time PCR. Initially, low levels of TF1 were observed in leaves infiltrated with *Eca* wt while a massive increase of the transcription factor occured at 10hpi. On the contrary, TF1 was highly upregulated in leaves infiltrated with *geneX* mutant within 0.5hpi of inoculation.

This result suggests possible involvement of *geneX* product in suppression of TF1 during early hours of infection by *Eca* wt

Improved resistance

Transgenic lines are significantly resistant to *Erwinia* compared to control plants

Desiree control (17dpi)

Transgenic TF1 plant (17dpi)

Overexpression of TF1 enhances resistance of of potato cultivar Desiree to Eca

References

Toth IK and Birch PBJ 2005 Rotting softly and stealthily. Current Opinions in Plant Biology 8(4):424-429

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