

Exploiting the *Phytophthora infestans* genome to determine targets for sustainable potato protection

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Introduction:

Phytophthora infestans, the causal agent of late blight disease remains, more than 160 years after instigating the Irish famine, the most devastating pathogen of potato. Breeding resistant potato cultivars is often undermined by rapidly occurring pathogen variations with the potential to overcome resistance (*R*) genes and quantitative field resistance. *R* gene-mediated resistance is manifested by the recognition of pathogen-derived effectors. Exploiting the recently sequenced *P. infestans* genome provides a unique opportunity to identify targets for potentially more durable resistance. Our strategy relies on the identification of secreted, invariant and functionally essential (and thus non-redundant) effectors that, as the pathogens 'Achilles heel', yield resistance that is more difficult to overcome by pathogen variants (Figure 1).

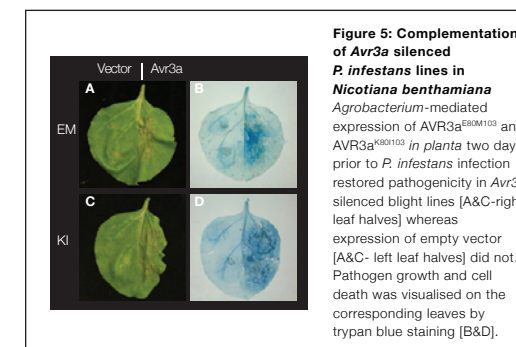
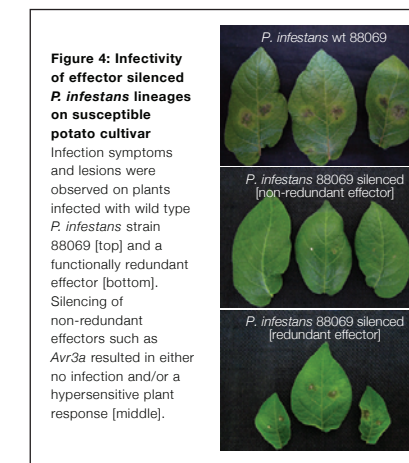
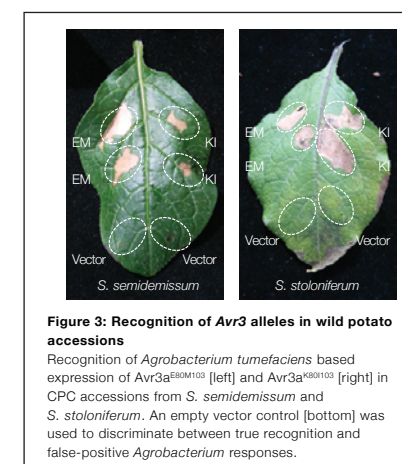
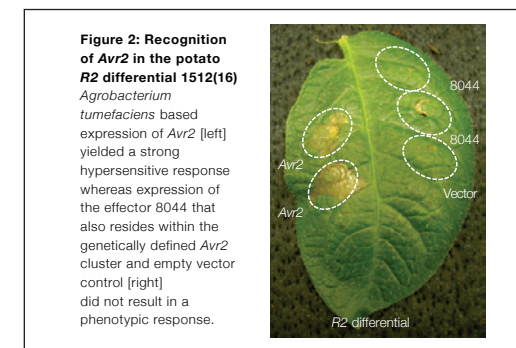


Figure 1: Pathogen effector driven pipeline to identify durable resistance
Developing genomic resources have allowed large-scale computational screening for a conserved RxLR/EER motif associated with oomycete effector transport across the host plasma membrane to reveal approximately 500 rapidly diverging *P. infestans* effectors. More than 100 RxLR/EER containing *P. infestans* effectors, induced during plant infection, have been amplified and cloned into binary vectors suitable for *Agrobacterium tumefaciens* mediated transient expression *in planta*. The conservation of effectors is studied within diverse *P. infestans* lineages and selected effectors are silenced. Silenced effectors that yield a loss of *P. infestans* virulence represent functionally non-redundant targets for durable disease resistance and are used in screening wild potato germplasm.

Results:

Expression of effectors in collections of cultivated and wild *Solanum* species maintained within the Commonwealth Potato Collection (CPC) have identified sources of *R* genes that recognise specific effectors, including AVR2, recognised by the *R2* resistance gene which maps to the more durable resistance locus present in cultivars Stirling & Lady Balfour (Figure 2) and *Avr3a*. *Avr3a*, the first effector characterized from *P. infestans*, exists in two forms: AVR3a^{K801103}, which is recognised by the cognate potato *R* gene *R3a* and AVR3a^{E80M103} that evades recognition by *R3a*. Transient and stable silencing of 25 effectors in *P. infestans* has identified 15 that are functionally essential and include *Avr3a* (Figure 4). Interestingly, the observed loss of pathogenicity in *Avr3a*-silenced *P. infestans* lines can be restored by transient, *in planta*, expression of both *Avr3a* alleles (Figure 5). Studying allelic variation of *Avr3a* in 82 diverse Mexican *P. infestans* isolates revealed that 77 contain *Avr3a*^{E80M103}.

Furthermore, CPC accessions from Mexico including *Solanum demissum*, *S. semidemissum*, *S. papita* and *S. stoloniferum* have been identified that recognise both the virulent and avirulent alleles of *Avr3a* (Figure 3). Blight tests have confirmed that these plants display high levels of resistance to diverse *P. infestans* lineages.



Conclusions:

Effector silencing and *in planta* complementation as shown for *Avr3a*, demonstrates that *P. infestans*, unlike some pathogenic bacteria, potentially contains functionally non-redundant effectors that are essential for pathogenicity. Introgression of genes from wild potato accessions that recognise such effectors could mark the way towards more durable resistance for potato cultivars.