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Progress in Mapping and Cloning Qualitative and Quantitative Resistance Against *Phytophthora infestans* in Potato and Its Wild Relatives

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Abstract Cultivated potato is susceptible to many pests and pathogens, none of which is more of a threat to potato agriculture than the late blight disease, caused by the oomycete *Phytophthora infestans* (Mont.) de Bary. To date all efforts to thwart this most adaptive of pathogens have failed, and early attempts to deploy '*R* genes' introgressed from the wild Mexican hexaploid *Solanum demissum* ended in abject failure. With the advent of facile gene mapping and cloning, allied to knowledge of plant resistance gene structure, renewed efforts are leading to mapping and isolation of new sources of late blight resistance in potato wild species, many of which are being performed under the auspices of the BIOEXPLOIT project (Sub-project 2). We document recent advances in late blight resistance gene mapping and isolation, and postulate how these genes, allied to knowledge of pathogen effectors and their recognition specificity, may greatly enhance our chances of halting the progress of late blight disease in potato crops worldwide.

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Introduction

Since the dawn of potato genetics, several resistances against the most serious disease, late blight, caused by the oomycete *Phytophthora infestans* (Mont.) de Bary, have been identified in wild potato species, with an increasingly large number being identified in recent years. Moreover, significant progress has been made in mapping and isolating genes conferring resistance to *P. infestans* and, with the availability of pathogen effectors, recognition specificity of resistance (*R*) genes can now be determined at a molecular level. However, many resistances to this rapidly evolving and highly adaptable pathogen have been quickly overcome by pathogen variations and truly durable resistance against *P. infestans* remains elusive. This state of affairs has led to new screens of potato germplasm for potentially novel sources of *P. infestans* resistance, several of which are being studied in the context of the BIOEXPLOIT project.

High quality maps, both genetic and physical, are indispensable to the localization, identification, and analysis of disease resistance genes. The main objectives of BIOEXPLOIT Sub-project 2 are to generate potato populations segregating for novel *P. infestans* resistances and to map and isolate the genes present. Other BIOEXPLOIT sub-projects, reported elsewhere in this issue of Potato Research, aim to identify and study pathogen-derived effectors, respectively. Linking the different BIOEXPLOIT strands, *P. infestans* effectors derived from these studies are now being increasingly incorporated into the characterisation of *R* gene specificity. In this paper, we provide an overview of the ongoing work on mapping potato resistance loci to *P. infestans*, isolating the underlying genes, and put these efforts into the context of previous work on analyzing *P. infestans* resistance genes.

Previous *P. infestans* Resistance Breeding

Early potato breeding for *P. infestans* resistance resulted in introgression of 11 resistance (designated *R1-R11)* genes from the Mexican hexaploid species *S. demissum* (Black et al. 1953; Malcolmson and Black 1966). However, as early as in the 1950s breeders realized that deployment of these genes, even in combination, did not provide durable resistance to the rapidly changing populations of *P. infestans.* In the 1960s, the parallel breakdown of three *R* genes was witnessed following the release of the potato cultivar Pentland Dell, containing *R1, R2* and *R3*. Released in 1961, Pentland Dell succumbed to extensive infection by new late blight races only 6 years later. This was prior to the introduction of the *P. infestans* A2 mating type, and a number of highly virulent pathogen genotypes, following a drought in the late 1970s (Drenth et al. 1994). Up to this point *P. infestans* populations of the A1 mating type in Europe showed little variation and isolates overcoming *R1*, *R2* and *R3* had not been identified. Importantly, it appears that selection pressure imposed upon the pathogen population by the widespread use of Pentland Dell increased the frequency of more complex

isolates (Shattock et al. 1977). Alongside other studies, Pentland Dell has illustrated the risks of relying on host resistance for disease control without due consideration of how the pathogen population may respond to its deployment, and has also sparked intensive searches for potentially more durable forms of resistance that focus on *R* genes and quantitative sources of resistance from other wild, *P. infestans* resistant, *Solanum* species (Bradshaw et al. 2004a; Bradshaw and Birch 2006).

Mapping of Resistance Genes

The majority of *S. demissum* blight resistance genes (*R1* to *R11*), have been mapped (Table 1), and are primarily located on linkage groups (LG) IV (*R2*), V (*R1*) and XI (*R3a*, *R3b* and *R5-R11*) (Leonards-Schippers et al. 1992; Li et al. 1998; Huang et al. 2004; Huang 2005; Park et al. 2005a; Bradshaw et al. 2006a). Following the observation that the race-specific *R* genes from *S. demissum* did not confer durable resistance to *P. infestans*, extensive screening programmes of potato gene banks for new sources of resistance have been conducted. Subsequent population development and mapping have led to the localization of several *Rpi* (for Resistance to *P. infestans*) genes from wild *Solanum* species (Table 1). Good examples of these are: *S. pinnatisectum* (Kuhl et al. 2001), *S. bulbocastanum* (Naess et al. 2000; Song et al. 2003; van der Vossen et al. 2003; Park et al. 2005a, b; van der Vossen et al. 2005) from Mexico and *S. berthaultii* (Rauscher et al. 2006), *S. microdontum* (Sandbrink et al. 2000), *S. mochiquense* (Smilde et al. 2005) and *S. paucissectum* (Villamon et al. 2005) from other South American countries. Moreover, the primitive cultivated diploid *S. phureja* has been described to contain valuable resistance (Śliwka et al. 2006). These efforts have shown that wild and primitive cultivated potato germplasm are a rich source of novel resistances to *P. infestans* that can be exploited in potato breeding programmes.

Some of these resistances are being studied in the context of the BIOEXPLOIT project. For example, *Rpi-mcq1* from *S. mochiquense* maps to the distal region of the long arm of chromosome IX, close to the location of a previously mapped *P. infestans* resistance in tomato (Smilde et al. 2005). Similarly, blight resistance in the non-tuberising species *S. caripense* is currently being studied. Extremely low levels of genetic variations have required the development of SNP-based allele-specific markers (Nakitandwe et al. 2007a) and aided the construction of a map that has positioned the *Rpi* gene to LG IX (Nakitandwe et al. 2007b). These findings suggest that research groups mapping genes to locations of previously mapped *Rpi* genes can make extensive use of information from these previous findings, a key feature of the BIOEXPLOIT project, which should help to accelerate gene isolation.

Further population development is ongoing within the BIOEXPLOIT project, whereby several species are being used to establish crosses for future mapping work (E. Ritter, A. Carrasco, V. Lefebvre, personal communication; Danan et al. 2009). These species include *S. boliviense*, *S. gourlayii*, *S. avilesii*, *S. spegazzinii*, *S. sparsipilum*, *S. venturii* and *S. berthaultii* and other ones. In addition, an integrated linkage map of the potato transcriptome has been constructed utilizing cDNA-AFLP technology, and subsequently anchored to the ‘bins’ of the Ultra High Density

Table 1 Late blight resistance genes, their origin, linkage group (LG) and cloning strategy

Gene	Origin	LG	Cloning strategy	Additional information	References
R* genes from *S. demissum					
R2	*S. demissum*	4	Allele mining	Cloned	Li et al. 1998; Park et al. 2005a, b; Lokossou et al. 2009
Rpi-dmsf1	*S. demissum*	4	Map based and allele mining	Potential ortholog of *Rpi-blb3*	Hein et al. 2007
R1	*S. demissum*	5	Map based	Cloned	Leonards-Schippers et al. 1992; Ballvora et al. 2002
R3a	*S. demissum*	11	Map based	Cloned	Huang et al. 2004; Huang et al. 2005
R3b, R5-R11	*S. demissum*	11?	Map based and allele mining	Alleles of *R3a*	El-Kharbotly et al. 1996; Huang et al. 2004; Huang 2005; Bradshaw et al. 2006b
***R* genes from other wild *Solanum* species**					
R2-like	*unknown*	4	Allele mining	Ortholog of *Rpi-blb3*	Lokossou et al. 2009
Rpi-abpt	*S. bulbocastanum*	4	Allele mining	Ortholog of *Rpi-blb3*	Lokossou et al. 2009
Rpi-blb3	*S. bulbocastanum*	4	Map based	Ortholog of *Rpi-blb3*	Lokossou et al. 2009
Rpi-bst1	*S. brachistotricum*	4	Map based and allele mining	Potential ortholog of *Rpi-blb3*	J. Jones and Z. Chu, personal communication
Rpi-mcd	*S. microdontum*	4	Map based	Accession MCD167	Sandbrink et al. 2000
Rpi-mcd1	*S. microdontum*	4	Map based	Potential ortholog of *Rpi-blb3*	Tan et al. 2008
Rpi-blb2	*S. bulbocastanum*	6	Map based	Cloned	van der Vossen et al. 2005
Rpi1	*S. pinnatisectum*	7	Map based		Kuhl et al. 2001
RB/Rpi-blb1	*S. bulbocastanum*	8	Map based	Cloned	Naess et al. 2000; Song et al. 2003; van der Vossen et al. 2003
Rpi-pta1	*S. papita*	8	Allele mining	Cloned ortholog of *Rpi-blb1*	Vleeshouwers et al. 2008; Wang et al. 2008
Rpi-plt1	*S. polytrichon*	8	Allele mining	Ortholog of *Rpi-blb1*	Wang et al. 2008
Rpi-sto1	*S. stoloniferum*	8	Allele mining	Cloned ortholog of *Rpi-blb1*	Vleeshouwers et al. 2008; Wang et al. 2008
Rpi-mcq1	*S. mochiquense*	9	Map based		Smilde et al. 2005
Rpi-vnt1.1 Rpi-vnt1.3	*S. venturii*	9	Allele mining	Tm-2 homologs	Foster et al. 2009; Pel et al. 2009
Rpi-phu1	*S. phureja*	9	Map based		Śliwka et al. 2006
R_{ber}	*S. berthaultii*	10	Map based		Rauscher et al. 2006

Table 1 (continued)

Gene	Origin	LG	Cloning strategy	Additional information	References
Rpi-mcd	*S. microdontum*	10	Map based	Accession MCD178	Sandbrink et al. 2000
Rpi-pcs	*S. paucissectum*	11	Map based	Other found on LG 10 and 12	Villamon et al. 2005
	S. microdontum	?	Map based	Incomplete map	Bisognin et al. 2005
	S. caripense	9	Map based	Incomplete map	Nakitandwe et al. 2007a; Nakitandwe et al. 2007b

(UHD) potato reference map (Ritter et al. 2008). The map and the additional markers generated will aid the mapping of *R* genes.

Cloning of Resistance Genes by Map-based Cloning and Allele Mining Strategies

Map-based cloning of four *Rpi* genes has been published prior to BIOEXPLOIT: *R1* (Ballvora et al. 2002) and *R3a* (Huang et al. 2005) from *S. demissum* as well as *RB* or *Rpi-blb1* (Song et al. 2003; van der Vossen et al. 2003) and *Rpi-blb2* (van der Vossen et al. 2005) from the Mexican diploid species *S. bulbocastanum*. The latter two genes confer resistance to a wide range of *P. infestans* isolates, giving hope that these genes will provide a more durable resistance than the *S. demissum R* genes. All four of these genes possess a 'coiled coil' domain, nucleotide binding site and leucine rich repeats (CC-NB-LRR), one of the more frequent classes of plant *R* genes.

As is often the case with plant *R* genes, the isolated genes are found within clusters of evolutionarily-related paralogs and/or other *R* genes. For example *Rpi-blb1* resides on LG VIII in a resistance gene analogue (RGA) cluster of four members (Song et al. 2003; van der Vossen et al. 2003). *Rpi-blb2*, located on LG VI, was identified within a cluster of at least 15 RGAs with homology to *Mi-1* which, in tomato, confers resistance to nematodes, aphids and white flies (van der Vossen et al. 2005). Similarly, *R1* is located within an *R* gene rich region on LG V that not only comprises different, fast evolving *R1* paralogs but also two other *R* gene families with homology to *Prf* and *Bs4* from tomato (Ballvora et al. 2002; Kuang et al. 2005). In line with this finding, at least two genes (*H1*, *GroV1*) conferring resistance towards potato cyst nematode (PCN) as well as *Rx2* and *Nb* for resistance to potato virus X (PVX) were also found on LG VIII (reviewed by Gebhardt and Valkonen 2001).

Similarly, previous studies have shown that *R6, R7, R10* and *R11* co-localize with *R3a* and *R3b* on LG XI (El-Kharbotly et al. 1996; Huang et al. 2004; Huang 2005; Bradshaw et al. 2006a). Further phenotypic studies of allelic variations of the *R3* complex identified nine allelic versions of the *R3a* gene, implying that several of the as yet uncloned *S. demissum R* genes may be allelic variants of *R3a* (Huang 2005). In tomato, the complex *I2* locus yielding resistance towards *Fusarium oxysporum* f

sp *lycopersici* has been cloned from this region and contains one active gene and six RGAs (Simons et al. 1998).

The property of *R* gene clustering, allied to the observation that the potato genome appears to contain a relatively small number of resistance 'hot spots', suggests a way forward in the isolation of more *Rpi* genes in the near future. For example, 14 of 19 important *R* genes for resistance to pathogens such as viruses, nematodes and *P. infestans* were found in five *R* gene-rich regions (Gebhardt and Valkonen 2001). It is possible that other genes, unrelated to *R* genes, found within or proximal to such clusters may also play a role in quantitative resistance. Clustering makes it difficult to resolve the specific contribution of these "other genes" to observed resistance. Unbiased approaches such as the use of regional genomic arrays are currently being used within the BIOEXPLOIT project to identify novel candidate genes within known regions containing genes for quantitative resistance. In support of this concept natural variant alleles of a potato allene oxide synthase 2 (STAOS2) mapping to a quantitative resistance locus (QRL) on chromosome XI have been shown to complement a null Arabidopsis *aos* mutant, increasing levels of various defence-related proteins, such as jasmonates as well as resistance to Arabidopsis pathogens (Pajerowska-Mukhtar et al. 2008).

The often conserved gene sequence structure within *R* gene clusters has led to the concept of "allele mining" for *Rpi* genes, as map-based cloning is still a relatively labour intensive approach. The high levels of sequence conservation between RGAs at any given *R* gene locus, suggest the possibility that PCR can be used to amplify homologous/paralogous candidate *Rpi* genes from any given locus, once the resistance has been mapped and sequence information about the type of *R* genes within the loci is available. As outlined below, allele mining has been applied to identify paralogous (homologous genes within the same species) and orthologous genes (homologous genes between species).

It has been known for some time that certain domains such as the P-loop (Saraste et al. 1990), kinase-2 motif (Traut 1994) and the GLPL motif (Meyers et al. 1999) within the NBS part of *R* genes are highly conserved. In potato, this information has been used to PCR-amplify, among others, *R* gene fragments linked to *P. infestans R7* resistance on LG XI (Leister et al. 1996) and subsequently further refined to obtain molecular markers based on *R* gene variations within genomes in an approach referred to as "NBS-profiling" (van der Linden et al. 2004).

To amplify full-length *R* genes, allele mining was first utilized to clone *RB* from *S. bulbocastanum* (Song et al. 2003). Following a map based cloning approach, it was found that all of the BAC clones containing the target locus corresponded to the susceptible *'rb'* allele. Gene sequences from these BAC clones allowed a 'long-range PCR' approach whereby the *RB* gene was identified amongst four RGAs from the resistant parent (Song et al. 2003). The allelic, but functionally equivalent *Rpi-blb1* gene (van der Vossen et al. 2003) and the unrelated *Rpi-blb2* gene (van der Vossen et al. 2005) were further used to PCR amplify orthologues in additional *Solanum* species showing late blight resistance (Wang et al. 2008). This study, which is part of BIOEXPLOIT, identified the *Rpi-blb1* homologs *Rpi-sto1*, *Rpi-plt1*, *Rpi-pta1* and *Rpi-pta2* from *S. stoloniferum*, *S. polytrichon* and *S. papita* respectively but did not yield homologs from *Rpi-blb2*. The authors confirmed genetically that *Rpi-sto1* and *Rpi-plt1* reside in the same position on LG VIII as *Rpi-blb1*. Further

effector based studies by Vleeshouwers et al. (2008) (detailed below) have shown that *Rpi-sto1* and *Rpi-pta1* are indeed functionally equivalent to *Rpi-blb1*. The implications of these studies are two-fold. Firstly, the genomic organisation of the *Rpi-blb1* gene cluster has been revealed and it is apparent that the *Rpi-blb1* locus was present prior to the divergence of tuber-bearing and non-tuber-bearing *Solanum* species. Furthermore, presence of *Rpi-blb1* orthologues is linked to the flanking RGAs *RGA1-blb* and *RGA3-blb* albeit both RGAs can be found without the presence of *Rpi-blb1*. Secondly, in terms of breeding prospects, introgression of *Rpi* genes from *Solanum* species such as *S. bulbocastanum* that cannot be crossed readily with cultivated *S. tuberosum*, can be circumvented by choosing more amenable species (e.g., *S. stoloniferum*) that carry functional *R* gene orthologues. One less optimistic conclusion from these studies is the possibility that, despite the diverse array of potato wild species available to the breeder, the number of distinct functional *Rpi* genes maybe somewhat smaller than was previously supposed. It also highlights the need to employ all available taxonomic information when designing introgression programmes for *Rpi* genes.

A very effective use of an allele mining approach has recently been demonstrated (Foster et al. 2009; Pel et al. 2009) in the BIOEXPLOIT project. These groups used sequence information from the Tm-2 locus of tomato, which confers resistance to *Tomato Mosaic Virus*, to isolate candidate genes from the potato species *S. venturii*. Mapping of the chromosome IX locus had been achieved with the help of NBS profiling. *Rpi-vnt1.1* and *Rpi-vnt1.3* belong to the coiled-coil NBS leucine-rich repeat (LRR) class of *R* genes, and their peptides are 75% identical to the tomato Tm-2 protein. These authors also isolated *Rpi-phu1*, from the primitive cultivated species *S. phureja*, and interestingly, this gene was identical to *Rpi-vnt1.1*, suggesting the possibility of introgression between the two species at some previous date.

Transcribed orthologues of *RB* have also been 'mined' from a set of *S. verrucosum* accessions differing in their responses to *P. infestans* infection (Liu and Halterman 2006). Stable transformation of a susceptible *S. tuberosum* by one of the *RB* orthologues from the late blight resistant *S. verrucosum* accession PI275260 conferred resistance to *P. infestans*. The functional *S. verrucosum RB* gene contains an insertion of a complete leucine rich repeat when compared to the *RB* gene from *S. bulbocastanum*, and differs from a susceptible *S. verrucosum* orthologue at only four amino acid residues (Liu and Halterman 2006).

In addition to the above described *R* genes, an additional race-specific *R* gene, *Rpi-blb3* from *S. bulbocastanum*, has been cloned using a map-based cloning approach and subsequently used for allele mining (Lokossou et al. 2009). *Rpi-blb3* has previously been mapped to LG IV and appears to be part of a large cluster of *R* genes comprising *R2* from *S. demissum*, *R2-like* from an *S. demissum*-free background and *Rpi-abpt* from *S. bulbocastanum* (Park et al. 2005a; Park et al. 2005b). Functional orthologues of *Rpi-blb3* have been identified in these species by allele mining. Similar to *Rpi-blb1, Rpi-sto1 and Rpi-pta1*, effector-based studies with *Avr2* (J.G. Morales et al., unpublished results) have shown that *R2, R2-like and Rpi-abpt* are indeed functionally equivalent to *Rpi-blb3* (Lokossou et al. 2009).

Three BIOEXPLOIT projects are focusing on cloning *Rpi* genes from LG 4 and comprise *Rpi-bst1* from *S. brachistotrichum* (Z. Chu and J. Jones, personal communication), *Rpi-mcd1* from *S. microdontum* (Tan et al. 2008) and *Rpi-dmsf1*,

a gene comprising a large component of a quantitative source of 'field' resistance probably derived from *S. demissum* (Bradshaw et al. 2004a; Hein et al. 2007). Genetic studies indicate that these resistances map within close proximity of *Rpi-blb3* and the associated genes *R2*, *Rpi-R2-like* and *Rpi-abpt*. Interestingly, *Rpi-mcd1* and *Rpi-dmsf1* have both been associated with field resistance, a quantitative resistance thought to be race non-specific and polygenically inherited (Bradshaw and Birch 2006; Bradshaw et al. 2004a, b; Bradshaw et al. 2006b; Tan et al. 2008). To isolate *Rpi-dmsf1*, a BAC library from a diploid potato clone expressing quantitative field resistance has been generated and screened with RGA probes derived from a tomato BAC clone (accession AF411807) linked to the *R2* cluster (Park et al. 2005b). Full length *Rpi-blb3*-like genes have been amplified from positively identified BAC clones, genomic DNA and cDNA (Hein et al. 2007; I. Hein et al., unpublished data) and function for these candidate genes is currently being sought.

Perspectives

Recent potato growing seasons have brought new challenges to the quest for resistance as a radical change in the *P. infestans* population structure is currently taking place across many European potato growing regions (Cooke et al. 2007a, b). A marked increase in the frequency of the A2 mating type has been reported in the Netherlands, France and the UK (Cooke et al. 2007a; Detourne et al. 2007; van Raaij et al. 2007) with wide-ranging short and long-term implications to the sustainability of blight control. This increase in A2 is dominated by a single clonal lineage, referred to as 'blue 13' or 'BPC06_3928A' that appears to be more aggressive than resident pathogen genotypes. In Great Britain, for example, SSR genotyping of almost 2,500 isolates has confirmed an increase in frequency from 12 to 70% of the population over three growing seasons from 2005 to 2007 (Cooke et al. 2007b). This lineage is also widespread in Germany, France and the Netherlands and has recently been found in Northern Ireland, Spain and Portugal (Cooke et al. 2007a).

In response to this threat, material previously identified as resistant to *P. infestans* is currently being re-screened with contemporary isolates including 'blue 13' to establish if the underlying *R* genes are still able to confer resistance. Furthermore, continuous screening of gene bank material such as the Commonwealth Potato Collection (CPC), hosted at the Scottish Crop Research Institute (SCRI), routinely includes these new isolates in addition to complex older resident isolates.

It is a very exciting time to be working in the field of plant disease resistance as genome sequence initiatives for both the plant and the pathogen already have, and will continue to provide information for studying the complex processes leading to resistance (if the plant recognises the invading pathogen) or to susceptibility (if the plant's defences are evaded by pathogen variations). The discovery of a conserved amino acid sequence, RXLR, found originally in the oomycete effectors *Avr3a* (Armstrong et al. 2005) and *Atr1* (Armstrong et al. 2005; Rehmany et al. 2005; Whisson et al. 2007) provide molecular insights into the evolution of oomycete pathogenesis and the host response. Analysis of fully sequenced oomycetes *P. sojae*, *P. ramorum*, *Hyaloperonospora arabidopsidis* (formerly *H. parasitica*) and *P. infestans* has revealed hundreds of potential secreted effectors containing the

RXLR motif (Tyler et al. 2006; Whisson et al. 2007; Win et al. 2007). These predicted effectors provide invaluable tools to: a) characterise *R* genes and b) to identify resistance mechanisms that have the potential to be durable. Recently, Vleeshouwers et al. (2008) tested recognition of ten phylogenetically diverse *Solanum* genotypes from eight wild species, all resistant to various late blight isolates, towards 54 RXLR effectors that were transiently expressed in the respective genotypes using Potato Virus *X* (PVX). Interestingly, the small effector family *IpiO*, consisting of at least two members, *IpiO1* and *IpiO2,* is recognised by *Solanum bulbocastanum* carrying *Rpi-blb1*. Co-infiltration of *Rpi-blb1* and *IpiO* in *N. benthamiana* resulted in a strong hypersensitive response (HR) and confirmed that *IpiO* is *Avr-blb1* (Vleeshouwers et al. 2008). Transient expression of *IpiO* in the distantly related species *S. stoloniferum* and *S. papita* also induced a HR and allele mining for *Rpi-blb1*-like genes in these species identified *Rpi-sto1* and *Rpi-pta1,* respectively (Vleeshouwers et al. 2008; Wang et al. 2008). Sequence analysis and comparison with *Rpi-blb1* revealed only three and five non-synonymous nucleotide polymorphisms for *Rpi-sto1* and *Rpi-pta1* respectively, which have been shown to be functionally equivalent to *Rpi-blb1* as all three genes recognised *IpiO* in transient *Nicotiana benthamiana* co-infiltration assays and yielded an HR response. This study has given a surprising insight into *R* gene redundancy within *Solanum* species and has revealed that many of the resistances identified in wild species could potentially be mediated by orthologous genes and that effectors can be used to determine the recognition spectra of (orthologous) *R* genes. *IpiO1* and *IpiO2* appear to be present in the majority of European and North-American *P. infestans* isolates which could explain the relatively broad-spectrum resistance conferred by *Rpi-blb1* and the orthologues *Rpi-sto1*, *Rpi-pta1* as well as *Rpi-plt1* from *S. polytrichon* (Vleeshouwers et al. 2008; Wang et al. 2008).

In a paradigm shift and in line with the above findings, Birch et al. (2008) have proposed that understanding effector diversity in oomycete populations, as well as effector expression and function, can be used to drive a search for more durable sources of resistance. Invariant and functionally non-redundant effectors that are required for pathogenicity could provide the pathogen's 'Achilles heel'. Deployment of cognate *R* genes that recognise these effectors could yield a more durable form of resistance than *R* genes that target non-conserved and functionally redundant effectors, assuming that additional effectors from the pathogen do not interfere with the recognition event, or with processes downstream of the recognition event that lead, for example, to the HR (Birch et al. 2008).

Future deployment in potato improvement programmes of newly discovered resistance genes is an important issue. The aforementioned ongoing rapid shifts in *P. infestans* population structure render the conventional 'introgression' route, whereby it may take several years to introduce resistance genes into an adapted background, ineffective in combating this dynamic threat. The recent rapid progress in mapping and isolating newly discovered *P. infestans* resistance genes (notably in the BIOEXPLOIT project) leads to the possibility of more rapid deployment through a transgenic or 'cisgenic' route (Jacobsen and Schouten 2007; Schouten and Jacobsen 2008). The development of this option is dependent on changes in regulations regarding field deployment, current legislation being based on risk assessments with transgenes originating from non-crossable species. 'Cisgenes' are

genes derived from crop plants themselves or from crossable species. There are now several 'marker-free' methods for potato transformation, and it can be argued that cisgenesis is a 'cleaner' method for gene introgression than conventional methods, such as induced translocation and introgression breeding, which have problems associated with linkage drag. Thus, there is a compelling argument for exempting cisgenesis from current regulations on genetically modified organisms. This would greatly facilitate the rapid deployment of new resistances against *P. infestans*, either singly, in combinations or as 'mixtures'. Such a development would allow the modern potato breeder a realistic chance of combating the threat posed by *P. infestans*, and perhaps more importantly, would lead to a more sustainable potato crop (i.e. less applied chemicals) in regions where *P. infestans* is endemic.

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