MINIREVIEW

Oomycete genomics: new insights and future directions

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Received 16 March 2007; revised 1 May 2007; accepted 2 May 2007.
First published online 7 June 2007.

DOI:10.1111/j.1574-6968.2007.00786.x

Editor: Richard Staples

Keywords
oomycetes; genomics; plant pathogens; evolution.

Abstract

The oomycetes form a distinct phylogenetic lineage of fungus-like eukaryotic microorganisms that are relatively closely related to photosynthetic algae such as brown algae and diatoms. Plant pathogenic species, notably those of the genus *Phytophthora*, are the best-studied oomycetes. The genomes of four *Phytophthora* and one downy mildew species were recently sequenced resulting in novel insights on the evolution and pathogenesis of oomycetes. This review highlights key findings that emerged from these studies and discusses the future challenges for oomycete research.

Introduction

The oomycetes form a diverse group of fungus-like eukaryotic microorganisms, also known as water molds, that include saprophytes as well as pathogens of plants, insects, crustaceans, fish, vertebrate animals, and various microorganisms (Margulis & Schwartz, 2000). Oomycetes form a distinct phylogenetic lineage of stramenopile eukaryotes and are relatively closely related to photosynthetic algae such as brown algae and diatoms (Sogin & Silberman, 1998; Baldauf *et al*., 2000; Adl *et al*., 2005). A multitude of saprophytic oomycetes primarily inhabit aquatic and moist soil habitats and play key roles in decomposition and recycling of organic matter (Margulis & Schwartz, 2000). However, plant pathogenic species, notably those of the genus *Phytophthora*, are the best-studied oomycetes (Kamoun, 2003, 2006; Judelson & Blanco, 2005). Species of the genus *Phytophthora* (the 'plant destroyer' in Greek) are arguably the most devastating pathogens of dicotyledonous plants (Erwin & Ribeiro, 1996). They cause enormous economic damage on important crop species such as potato, tomato, pepper, soybean, and alfalfa, as well as environmental damage in natural ecosystems. Virtually every dicot plant is affected by one or more species of *Phytophthora*, and several monocot species are infected as well.

The study of oomycetes has reached new heights with the completion of the genome sequence drafts of five species that are among the most notorious plant pathogens (Tyler *et al*., 2006) (Table 1). *Phytophthora capsici*, *Phytophthora infestans*, and *Phytophthora sojae* infect diverse crops, such as pepper, potato, and soybean, respectively (Birch & Whisson, 2001; Hausbeck & Lamour, 2004; Kamoun & Smart, 2005; Tyler, 2007). *Phytophthora ramorum* affects native and ornamental woody plants resulting in environmental damage (Rizzo *et al*., 2005). The downy mildew, *Hyaloperonospora parasitica*, is a pathogen of *Arabidopsis thaliana* and figures prominently in research on this model plant (Slusarenko & Schlaich, 2003). The genomes of *P. sojae* and *P. ramorum*, the first to be sequenced, were recently described in a momentous *Science* paper (Tyler *et al*., 2006) and in a series of companion studies published in a 'special focus' issue of *Molecular Plant–Microbe Interactions* (December 2006). This review highlights key findings that emerged from these studies including new insights on the evolution and pathogenesis of oomycetes and discusses the future challenges for oomycete research.

Table 1. Oomycete genome projects and their associated web resources

Species	Genome size (Mbp)	Estimated number of genes	Resources
Hyaloperonospora parasitica	75	NA	Washington University, http://genome.wustl.edu/pub/organism/Fungi/Hyaloperonospora_parasitica/
Phytophthora capsici	65	12 011	JGI, in progress with expected release during 2007
Phytophthora infestans	240	22 658	Broad, http://www.broad.mit.edu/annotation/genome/phytophthora_infestans/Home.html
Phytophthora ramorum	65	15 743	JGI, http://genome.jgi-psf.org/Phyra1_1/Phyra1_1.home.html
Phytophthora sojae	95	19 027	JGI, http://genome.jgi-psf.org/Physo1_1/Physo1_1.home.html

Oomycete evolution: plant killers with a photosynthetic past

As stramenopiles, the oomycetes share an evolutionary history with photosynthetic organisms and therefore may have evolved from phototrophic ancestors (Sogin & Silberman, 1998; Baldauf *et al.*, 2000; Adl *et al.*, 2005). In fact, modern phylogenetic analyses provide robust support for the chromalveolates, a super-group of algae and protists that brings together the stramenopiles (oomycetes, diatoms, and brown algae) and the alveolates (apicomplexans, ciliates, and dinoflagellates) (Schlegel, 2003; Adl *et al.*, 2005; Harper *et al.*, 2005). The chromalveolate lineage is postulated to have derived from a common ancestor that acquired a chloroplast endosymbiont from a red algae (Cavalier-Smith, 2002). One prediction of the chromalveolate hypothesis is that nonphotosynthetic lineages like oomycetes have lost chloroplasts throughout their evolution. In such a case, the endosymbiont may have left a genomic footprint through the transfer of chloroplast genes to the host nucleus. The genome sequences of *P. sojae* and *P. ramorum* provided strong support for this hypothesis. A total of 855 *Phytophthora* genes showed similarity to photosynthetic organisms such as red algae or cyanobacteria (Tyler *et al.*, 2006). Phylogenetic analyses indicated that several *Phytophthora* genes for biosynthetic enzymes group in two distinct phylogenetic branches: one with affinity to proteobacteria indicative of a mitochondrial origin, and another with affinity to cyanobacteria suggestive of a plastid origin (Tyler *et al.*, 2006). Thus these analyses support the view that the stramenopiles were founded by a photosynthetic ancestor and are consistent with the chromalveolate hypothesis. It appears that devastating pathogens, such as *Phytophthora*, have evolved from benign phototrophic ancestors.

The oomycetes are more closely related to the apicomplexans, the alveolate taxon that includes *Plasmodium* and *Toxoplasma* parasites, than to any other eukaryotic parasite, including major groups like fungi and trypanosomids (Schlegel, 2003; Adl *et al.*, 2005; Harper *et al.*, 2005). Striking similarities in pathogenicity mechanisms between oomycetes and apicomplexans have been noted with regard to host-translocation signals (see below) (Rehmany *et al.*, 2005; Bhattacharjee *et al.*, 2006), secretion of protease inhibitors (Tian *et al.*, 2004; Torto-Alalibo *et al.*, 2005), and attachment to host tissues (Robold & Hardham, 2005; Torto-Alalibo *et al.*, 2005). These shared mechanisms of pathogenesis between oomycetes and apicomplexans may be a reflection of their common evolutionary history (Haldar *et al.*, 2006).

The peculiar phylogenetic affinities of oomycetes are also reflected in their distant relation to the true fungi despite apparent similarities, such as filamentous growth habit, heterotrophic lifestyle, and specialized infection structures (Latijnhouwers *et al.*, 2003). However, comparative analyses of *Phytophthora* and fungal genome sequences identified a number of related genes that cluster together in phylogenetic analyses (Gotesson *et al.*, 2002; McLeod *et al.*, 2002; Torto *et al.*, 2002; Randall *et al.*, 2005; Richards *et al.*, 2006; Win *et al.*, 2006). Richards *et al.* (2006) explain these findings by multiple horizontal gene transfer (HGT) events that enabled the oomycetes to acquire fungal genes. Alternatively, differential gene loss might be the most parsimonious explanation for the observed biased phylogenetic distribution, particularly if the candidate HGT genes turn out to occur in a wider range of eukaryotes (Win *et al.*, 2006). Nonetheless, the observed overlap among plant pathogenic filamentous microorganisms in genes encoding enzymes involved in cell wall hydrolysis (Gotesson *et al.*, 2002; McLeod *et al.*, 2002; Torto *et al.*, 2002; Randall *et al.*, 2005; Win *et al.*, 2006), virulence (Gotesson *et al.*, 2002; McLeod *et al.*, 2002; Torto *et al.*, 2002; Randall *et al.*, 2005; Win *et al.*, 2006), and osmotrophy (Richards *et al.*, 2006) points to a complex evolutionary history for the oomycetes that resulted in a distinctive gene content.

Oomycete pathogenesis: an inordinate fondness for effectors

Diverse prokaryotic and eukaryotic plant pathogens secrete effector proteins that modulate plant defense circuitry and enable parasitic colonization (Birch *et al.*, 2006; Chisholm *et al.*, 2006; Kamoun, 2006). In the oomycetes, two classes of

effectors target distinct sites in the host plant: apoplastic effectors are secreted into the plant extracellular space, and cytoplasmic effectors are translocated inside the plant cell, where they target different subcellular compartments (Birch *et al*., 2006; Kamoun, 2006). One class of cytoplasmic effectors, the so-called RXLR effectors, has been the subject of much research in the last 2 years (Rehmany *et al*., 2005; Birch *et al*., 2006; Kamoun, 2006; Tyler *et al*., 2006). These effectors, first identified as avirulence proteins in *H. parasitica* (Allen *et al*., 2004; Rehmany *et al*., 2005), *P. infestans* (Armstrong *et al*., 2005), and *P. sojae* (Shan *et al*., 2004), carry a conserved motif, termed RXLR (arginine, any amino-acid, leucine, arginine), that is located downstream of the signal peptide and has been implicated in host translocation (Rehmany *et al*., 2005; Bhattacharjee *et al*., 2006; Birch *et al*., 2006; Kamoun, 2006). Remarkably, the RXLR motif is similar in sequence and position to the plasmodial host translocation (HT)/Pexel motif that functions in delivery of parasite proteins into the red blood cells of mammalian hosts (Hiller *et al*., 2004; Marti *et al*., 2004). A *c*. 30 amino-acid region encompassing the RXLR motif of *P. infestans* RXLR proteins AVR3a and PH001D5 mediates the export of the green fluorescent protein (GFP) from the *Plasmodium falciparum* parasite to the host red blood cell, suggesting that the RXLR and HT/Pexel domains are functionally interchangeable (Bhattacharjee, *et al*., 2006). Altogether this and related findings led to the view that oomycete RXLR effectors are modular proteins with two major functional domains (Bos *et al*., 2006; Kamoun, 2006). While the N-terminal domain encompassing the signal peptide and RXLR leader functions in secretion and targeting, the remaining C-terminal region carries the effector activity and operates inside plant cells.

The genome sequences of plant pathogenic oomycetes enabled genome-wide cataloging of RXLR effectors using computational approaches (Kamoun, 2006; Tyler *et al*., 2006). The RXLR effector secretomes turned out to be more complex than expected consisting of hundreds of candidate effectors. Tyler *et al*. (2006) reported 350 RXLR effectors each in the genomes of *P. ramorum* and *P. sojae* using iterated similarity searches. Analyses performed using combinations of motif and hidden Markov model searches uncovered at least 50 candidates in the downy mildew *H. parasitica* and more than 200 each in *P. capsici*, *P. infestans*, *P. ramorum*, and *P. sojae* depending on the stringency of the method used (J. Win & S. Kamoun, unpublished) (Table 2).

Comparative analyses of the RXLR effectors indicate that they undergo birth and death evolution resulting in divergent sets of effectors in the three species (Jiang *et al*., 2006; Tyler *et al*., 2006). Patterns of accelerated rates of gene loss and duplication at some RXLR effector loci were reported for *P. sojae* and *P. ramorum* (Jiang *et al*., 2006; Tyler *et al*., 2006). In another example, *Avr3a* of *P. infestans* and *ATR1* of *H. parasitica* occur in conserved syntenic chromosomal regions but are highly divergent in primary sequence (Armstrong *et al*., 2005). Rapid evolutionary rates in these effector genes may reflect evolutionary adaptations to host plants.

The remarkably large numbers of candidate RXLR effectors identified in the genome-wide analyses suggest that oomycetes extensively modulate host processes during infection. The next challenge is to unravel the virulence activities of these effectors to understand how they perturb plant processes to increase the reproductive success of the pathogen (Birch *et al*., 2006; Kamoun, 2006). For instance, a possible virulence function was ascribed to *P. infestans* RXLR effector Avr3a, which is able to suppress the hypersensitive cell death induced by another *P. infestans* protein, INF1 elicitin (Bos *et al*., 2006). The extent to which other RXLR effectors suppress plant cell death and defenses remains to be determined.

The challenge of oomycete genetics: past, present, and future

Despite their unique phylogenetic affinities and high economic importance, oomycetes have been chronically understudied at the molecular level. This is in large part due to the difficulty of completing genetic experiments. In particular, manipulation of the sexual stage is notoriously difficult with many oomycetes, especially species of *Phytophthora* (Erwin & Ribeiro, 1996). In the last decade or so, the development

Table 2. Number of candidate RXLR effectors identified in oomycete genomes

Species	Reference	Genome size (Mbp)	Total RXLR*	RXLR+EER†	HMM‡
Hyaloperonospora parasitica	Washington University	75	149	42	19
Phytophthora capsici	JGI	65	420	146	96
Phytophthora infestans	Broad Institute	240	716	229	191
Phytophthora ramorum	Tyler *et al*. (2006)	65	531	214	181
Phytophthora sojae	Tyler *et al*. (2006)	95	672	189	158

*Total number of candidate RXLR effectors predicted by the presence of a signal peptide and the RXLR sequence at amino acids 30–60.

†Number of candidates with a RXLR and EER sequence.

‡Number of candidates identified using a hidden Markov model for the RXLR-EER domain.

of cDNA and partial genomic sequences opened the door for the study of oomycete molecular genetics prompting the emergence of a group of researchers specializing in this field. The concerted efforts of this group (currently dubbed the Oomycete Molecular Genetics Network http://pmgn.vbi.vt.edu/) helped make possible the five whole genome sequences, and an increase in detailed descriptive genetic studies based on microarrays, genome resequencing, proteomics (Savidor *et al.*, 2006), and other global technologies is fully expected. Furthermore, it is expected that candidate genes important for spore production, stress, response to antibiotics, pathogenicity, virulence, and other important characters will be identified. As the challenges associated with identifying interesting candidates are reduced, the need for tools and resources for validation and further testing will be of the highest priority. In short, the need for genetic resources/abilities has been greatly heightened by this recent windfall of genomic sequence.

Technical developments, such as DNA transformation, use of reporter genes, and genetic manipulation using gene silencing, have facilitated the discovery and functional analyses of several genes, although application of these approaches is by no means 'routine' (Kamoun, 2003). More recently, the reverse genetic approach targeting induced local lesions in genomes (TILLING; McCallum *et al.*, 2000) has been applied to *P. sojae* providing an important new tool for obtaining mutant versions of interesting genes including knock-outs (Lamour *et al.*, 2006). TILLING can be applied to *P. infestans* and *P. capsici* but would be difficult or impossible with *H. parasitica* and *P. ramorum*. The key requirements are the ability to maintain a large population of mutants (which is very difficult with *H. parasitica*) and a tractable sexual cycle (lacking in *P. ramorum*). A significant cost of any technique such as TILLING that relies on random mutagenesis is the overall load of mutations in the individual genomes. For *P. sojae*, TILLING mutants are generated using the chemical mutagen ENU and individuals receive between 800 and 1000 random induced point mutations (Lamour *et al.*, 2006). *Phytophthora sojae* is self-fertile, which allows direct access to the segregating mutation and, if viable, to the homozygous mutant progeny. The only drawback is that along with the homozygous mutant allele, the mutant strain carries an additional 200–250 homozygous mutations that may or may not have a phenotypic effect. One way to limit this background effect is to recover a large set of mutant progeny and track the impact of the desired mutation in the midst of the recombining additional mutations. The situation with *P. capsici* and *P. infestans* is different because they are obligate out-crossers requiring a minimum of two crosses before a homozygous mutant can be recovered. This adds an additional cross but reduces the impact of collateral mutations by more than 60%.

A model for oomycete genetics?

It is apparent that the different oomycete species have varying levels of tractability and ease of use depending on the criteria examined (Table 3), and some species are

Table 3. Which model system for the oomycetes

Features	*H. parasitica*	*P. capsici*	*P. infestans*	*P. sojae*	*P. ramorum*
Culturing	Obligate parasite, can only be cultured on plants	Culturable, fast grower	Culturable, slow grower	Culturable, slow grower	Culturable, slow grower
Spore production	Easy but requires infected plants	Easy	Easy	Requires serial washes in buffer solutions	Not reported
Plant interaction assays, degree of difficulty	Easy, infects cotyledons and leaves	Easy, infects all plant tissues	Easy, infects leaves and potato tubers	Moderately difficult, requires root or hypocotyls	Moderately difficult?
Tractability of host plant	High, Arabidopsis	High, tomato and pepper	High, tomato and potato	Moderate, soybean	Low, woody plants
Genome complexity	Moderate	Moderate	High	Moderate to high	Moderate
DNA transformation	Currently not possible	Established through zoospore electroporation	Established through several methods	Established through several methods	Not reported but potentially feasible
Gene silencing, RNAi gene inactivation	Currently not possible	Not reported but potentially feasible	Established	Not reported but potentially feasible	Not reported
Potential for genetic manipulation	Genetic crosses and gene mapping possible	High, highly fertile in a variety of crosses	Genetic crosses and gene mapping possible but difficult	Genetic crosses and gene mapping possible but difficult	Not possible
Genetic analysis by TILLING	Not possible	Established	Possible	Established	Not possible
Community size	Moderate	Small	Moderate	Small	Small

particularly appropriate for certain types of analyses. For instance, *H. parasitica* is a pathogen of the model plant *A. thaliana* suggesting that functional studies of host–pathogen interactions should be easier in this pathosystem. On the other hand, *H. parasitica* is an obligate parasite that cannot be maintained in the laboratory as pure *in vitro* cultures and, therefore, cannot be genetically transformed or used for large-scale reverse genetic approaches. *Phytophthora ramorum* is a particularly intriguing new species (Werres *et al.*, 2001) that has a large host range. Although *P. ramorum* is heterothallic and both compatibility types have been isolated, successful crosses have not been reported (Werres & Kaminski, 2005). Even if suitable strains for crossing were discovered, *P. ramorum* makes large numbers of thick-walled asexual chlamydospores (a spore not made by the other sequenced oomycetes) that greatly interfere with the recovery of sexual spores, making it difficult to complete routine crosses.

So far, most genetic research has focused on *P. infestans* and *P. sojae*. As a consequence, tools for transformation are the most advanced for these species and gene silencing and RNAi knock-down are possible in *P. infestans*. Both species have limited host ranges making them ideal candidates for studying fine-scale pathogen/host interactions – particularly the gene-for-gene interactions at the cultivar level (Armstrong *et al.*, 2005). The recent report of a bacterial artificial chromosome (BAC)-based, integrated physical map of the *P. sojae* genome provides a new resource for genomic investigations and was used to show the high level of clustering for a superfamily of secreted effector genes (Zhang *et al.*, 2006). Genetic crosses and associated mapping populations have been developed for both species (Drenth *et al.*, 1995; van der Lee *et al.*, 1997; MacGregor *et al.*, 2002), but the overall use of mapping has been relatively limited. For *P. infestans* there appears to be an overall low fecundity for many parental and progeny strains and in some cases it appears that crossing has resulted in progeny with variable ploidy levels (Erwin & Ribeiro, 1996). For *P. sojae* there are two limiting factors; the overall pool of genetic variation available for map development is limited as a consequence of a self-fertilizing sexual stage (isolates are highly inbred) and outcrossing is difficult as many of the progeny from a cross are the products of self-fertilization (May *et al.*, 2002).

Phytophthora capsici is a newcomer to oomycete molecular genetics. It is attractive because it is easy to cross and there is a large reservoir of natural variation present in natural populations (Lamour & Hausbeck, 2000, 2002). In addition, *P. capsici* grows rapidly in axenic culture (Lamour & Finley, 2006), easily produces abundant sporangia and zoospores (crucial for many transformation protocols and the development of mutant libraries), and has a simpler genome structure compared with other *Phytophthora* spp. Recent crossing experiments with *P. capsici* and the closely related sister species *Phytophthora tropicalis* indicate that intra and interspecific recombinant inbred lines may be possible (K. Lamour, unpublished) – a genetic resource yet to be exploited for an outcrossing oomycete. *Phytophthora tropicalis* attacks primarily tropical woody perennials (e.g. cacao, macadamia) whereas *P. capsici* infects primarily annual vegetables such as cucurbits, peppers, and tomatoes. Progeny sets from back and sib-crosses display a wide range of phenotypic variations including the ability to infect widely different hosts (Fig. 1). This is similar to reports of *in vitro* interspecific hybridization between the closely related sister taxa *P. infestans* and *Phytophthora mirabilis* from Mexico (Goodwin & Fry, 1994). For both *P. infestans* and *P. capsici*, these crosses between closely related but evolutionarily distinct lineages present the unique opportunity to investigate the genetic factors driving the evolution of host preference. It is expected that *P. capsici* will receive more attention in the coming years as genetic analyses increasingly transition from descriptive to more critical functional analyses of oomycete genes.

Oomycete genomics: what's next?

What lies ahead in oomycete genomics? First, it is important to address the limitations that are apparent in the current data sets. For instance, the quality of the gene calls is lacking in the published draft genome sequences. Most of the information about the *Phytophthora* gene structure was gathered from a handful of genes, and this knowledge has not always been optimally integrated into automated gene-calling programs. cDNA and expressed sequence tags have been generated for some species such as *P. infestans* (Randall *et al.*, 2005) and *P. sojae* (Torto-Alalibo *et al.*, 2007) and should be helpful in assisting in gene calling. Another essential approach is the development of full-length cDNA sequences as a resource to enable the validation of existing genome annotations as well as in the development of *ab initio* gene-calling programs (Win *et al.*, 2006). Similarly, genome assemblies can be improved by physical mapping data as reported for *P. sojae* (Zhang *et al.*, 2006). High-quality genome sequence assemblies and gene models are critical to ensure that comparative genomics analyses result in robust findings.

With the emergence of novel sequencing technologies, such as 454 sequencing and Illumina's sequencing by synthesis as well as the ever-decreasing cost of Sanger sequencing, it is expected that additional oomycete genomes will be sequenced in the near future. Which species should be sequenced next? Three approaches can be considered. First, species could be selected to cover the phylogenetic spectra of oomycete pathogens (Cooke *et al.*, 2000; Riethmuller *et al.*, 2002; Kroon *et al.*, 2004; Goker *et al.*, 2007). The species would represent major lineages within diverse clades like *Phytophthora* and the downy mildews (Goker *et al.*, 2007).

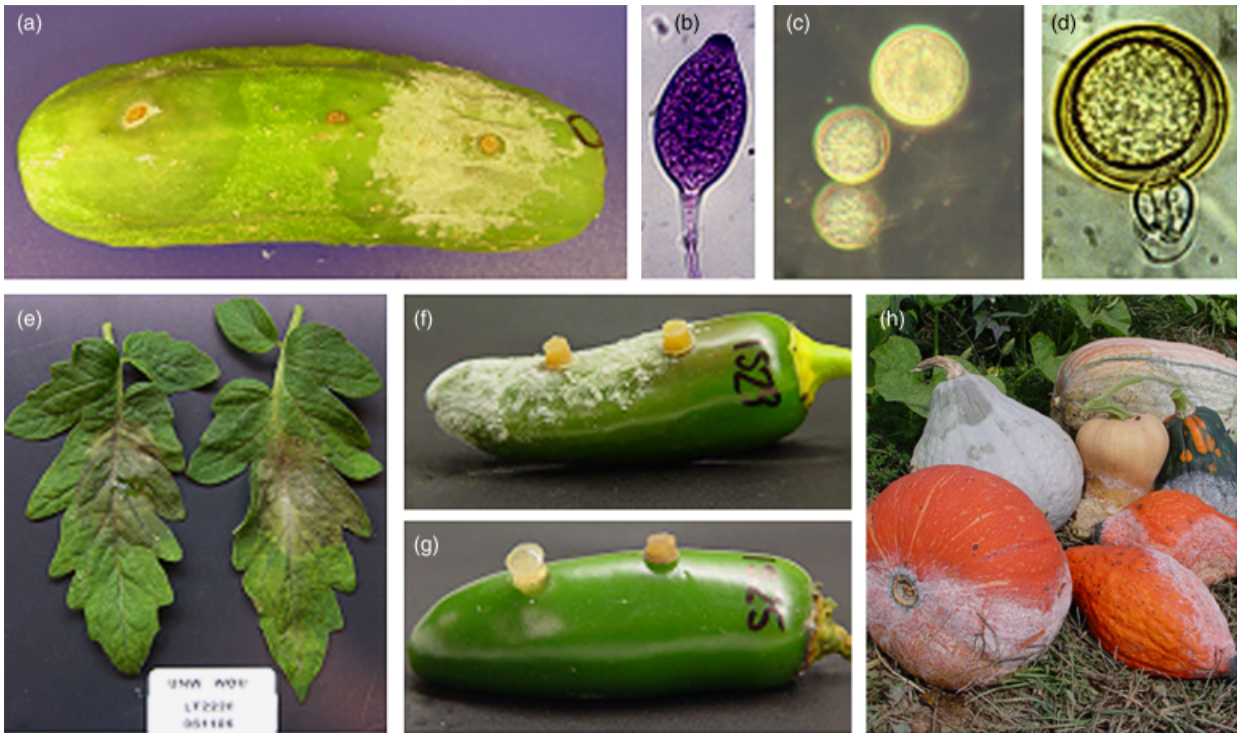

Fig. 1. Pictorial summary of traits segregating in a *Phytophthora capsici* sibcross and a *P. capsici* × *Phytophthora tropicalis* interspecific cross including (a) and (h) ability to infect and sporulate on cucurbits; (b) sporangia production; (c) chlamydospore production; (d) oospore production; (e) ability to infect tomato; (f) ability and (g) inability to infect jalapeno peppers among sib-progeny.

Second, species could be selected to represent the divergent lifestyles of oomycetes such as animal and fungal parasitism. Finally, clusters of sibling species or strains related to the sequenced species could be sequenced to enable comparative genomics. This should prove particularly productive for comparative analyses of effector genes. Comparative analyses of RXLR effectors in the currently sequenced *Phytophthora* genomes have not been particularly informative due to a high level of divergence and rapid evolutionary rates in these genes (Jiang *et al.*, 2006; Tyler *et al.*, 2006) (J. Win & S. Kamoun, unpublished). In contrast, comparative analyses of sibling species or strains, such as *P. capsici* and *P. tropicalis*, or *P. infestans* and *P. mirabilis*, would be more informative and will unravel candidate virulence and host-specificity genes (Goodwin & Fry, 1994).

Finally, it is hoped that oomycete genomics will move beyond the laboratory and into epidemic population studies. The oomycetes are a highly plastic group of organisms – notorious for responding quickly to human-mediated selection pressures, whether it is fungicide use or deployment of plant resistance genes (Erwin & Ribeiro, 1996). The patterns of gene conservation and expression dissected using the sequenced strains provide a compelling start point for testing hypotheses in the field. For example, *P. capsici* populations in the United States have been confined to cucurbit and solanaceous hosts since the 1920s and have only recently 'jumped' to snap and lima beans in the Midwest and Northeast (Leonian, 1922; Hausbeck & Lamour, 2004). Studies of *P. capsici* in the 1960s clearly demonstrate that snap and lima beans were nonhosts for *P. capsici* (Satour & Butler, 1968). What factors in the genome contributed to this dramatic host expansion? Did the overall complement of secreted effector proteins change? The oomycetes are diploid throughout their life history and the heterozygosis revealed by random shotgun genome sequencing provides an enormous set of candidate markers (especially for outcrossing oomycetes) (Tyler *et al.*, 2006) useful for tracking associations in populations and individual genomes – bridging the gap between experimental and population-level genomics.

Conclusions

In summary, these are exciting times for oomycete research! The five oomycete genomes comprise a treasure trove that has already impacted the understanding of these peculiar and devastating pathogens. Investigators have an unprecedented base from which to explore questions about oomycete pathology and evolution and the list of compelling questions is long. Are there common mechanisms of

infection among filamentous microorganisms? How did (and does) the virulence gene arsenal emerge and evolve? To what extent do these virulence genes vary between oomycetes and other pathogens? How is (and has) coevolution with host species shaping the structure of virulence genes? Clearly there is much work to be done to capitalize fully on these valuable resources, and one can only look at the future with a sense of excitement and opportunity.

Acknowledgements

Research in the author's laboratories was supported by NSF CAREER Grant No. 0347624 (KHL), The Gatsby Charitable Trust (JW), and NSF Plant Genome grant DBI-0211659, USDA-NRI project OHO00963-SS, and State and Federal Funds appropriated to OARDC, the Ohio State University (SK).

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