South African Sclerotinia sclerotiorum research in the age of genomics

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clerotinia sclerotiorum is a fungal pathogen of many crop species that needs no introduction to anyone working in the agricultural industry. In South Africa, *S. sclerotiorum* causes disease on many economically important crop species including canola, soya bean and sunflower.

The persistent occurrence of this fungus in the field is attributed to its ability to form hardened, long-term survival structures called sclerotia, a trait it shares with other plant pathogens (e.g. *Claviceps purpurea* that causes ergot on cereal crops) and even some mushroom species (e.g. the psychedelic mushroom *Psilocybe Mexicana*). Sclerotia of *S. sclerotiorum* form the basis for its infection cycle, with severe outbreaks leading to significant yield losses.

Pathogenicity of S. sclerotiorum

Its notoriety as an economically important pathogen has led to *S. sclerotiorum* being a well-researched fungus. Although much is

known about its infection cycle, its ability to cause disease in a range of plants is still not well understood, which is a rather typical property among plant pathogens. This has led to many studies focusing on the way in which *S. sclerotiorum* causes disease and identifying a wide array of biological molecules that affect its pathogenicity, as well as ways to control it in the field.

For example, viruses and antagonistic fungal species that adversely affect *S. sclerotiorum* have been identified. These studies have boosted our understanding of the pathogen significantly, but as is commonly the case, the elusive 'silver bullet' has not yet been found.

Unravelling its genome sequence

Research is intricately linked to technology and following the development of so-called next generation sequencing platforms, we now have access to the full genome sequences of many living things. A genome sequence is a copy of all the genetic instructions present in a cell – it essentially represents the blueprint on what makes a species what it is.

The availability of full genome sequences has thus impacted all aspects of biology, including modern plant pathology where it has provided a plethora of new tools for enhancing plant health. For *S. sclerotiorum* (strain 1980), a high-quality draft of its genome sequence was produced in 2005 and, by using newer sequencing technologies, was completed in 2017.

The S. sclerotiorum genome has 38,7 million nucleotides, which is approximately 1,5% the size of the human genome. Despite its small size, the S. sclerotiorum genome contains roughly 50% as many genes as found in humans. However, the functions of most of these genes are not yet known, highlighting the complexity that underlies the functioning of something seen as a 'simple' fungus.

The tools provided by a genome sequence is useful for many studies related to fungal pathogens. Genomes offer the chance to develop populationlevel markers specific to a target species. This has significant benefits for epidemiological studies in that it allows estimation of genetic diversity and modelling of future infection risks. A genome sequence also provides a catalogue of the history of a species that can be picked apart for clues as to the origin and biological functioning of the fungus. Although it sounds very simple, these are complex tasks that require huge contributions from many scientific fields.

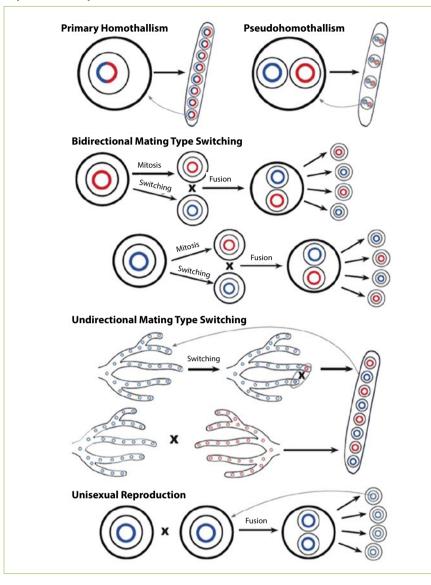
Reproduction ability of fungi

One study area that has benefitted from the availability of genome sequences is research into the reproductive strategies of fungal pathogens. The sexual process in fungi is controlled by genes present at a



Sclerotinia sclerotiorum on soya beans. (Photograph by Ken Wise, NY State IPM Program at Cornell University)

Figure 1: A representation of the various genetic mechanisms of homothallism in fungi. Red and blue circles represent haploid genomes of opposite mating type; large black circles represent single fungal cells; and small black circles within black ovals represent ascospores within asci. (Taken from Wilson et al., 2015)



specific location in the genome. Knowledge of the structure of this region provides insight into how these fungi reproduce, which in turn has important implications for the population dynamics of a species.

A species that employs an exclusively outcrossing or heterothallic reproductive mode (*Figure 1*) will need to find a compatible mating partner and subsequently produce genetically diverse progeny that can increase the adaptability of a species. In these fungi, if a compatible mating partner is not available, sexual reproduction will not occur, which could have a sizeable impact on the population genetics of the fungus. By contrast, in homothallic species (Figure 1), mating can occur in the absence of a partner. Here, an isolate can initiate solo sexual reproduction and produce thousands of sexual progeny by itself, resulting in a population of clones – individuals that are genetically identical. However, they retain the ability to outcross, and can thus benefit from outcrossing and the generation of variation similar to heterothallic species.

Reproduction of S. sclerotiorum

S. sclerotiorum is homothallic and can therefore reproduce in the absence of

a mating partner. During this process, the fungus produces the characteristic mushroom-shaped apothecia, which releases sexual spores that can establish an infection on the plant.

As expected, many field populations of the fungus show high levels of clonality due to homothallism. However, some population studies show moderate levels of genetic diversity and the genetic signatures of outcrossing, putting the reproductive strategy of this fungus under the microscope. From a molecular viewpoint, the sexual process is even more bizarre as it incorporates a change at the DNA level that structurally and functionally influences the sexual process. This leads to questions on the link between the DNA rearrangement process and the occurrence of unexpected levels of genetic diversity in the field.

Research on the local front

Our project aims to establish a South African-specific genomic resource for studies on *S. sclerotiorum*. The availability of a high-quality draft genome from a South African isolate of the species will prove invaluable for expanding our current understanding of the pathogen in a local context.

It would allow us to design genetic markers catered specifically to the South African population and target suitable regions for developing diagnostic tools, while at the same time providing insight into the evolutionary history of the pathogen in the country. With the genome information, we will also be able to study the reproductive strategy used by South African populations of the pathogen.

Insights gained from these studies will not only add to the growing debate around the population genetics structure of the fungus, but also contribute meaningfully to efforts aimed at combating this important pathogen in the field.

> For more information or a list of references, send an email to Dr Markus Wilken at markus.wilken@fabi.up.ac.za or Sikelela Buthelezi at sikelela.buthelezi@up.ac.za.