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Genomic tools for the management of clubroot of canola (Brassica napus)

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Clubroot, caused by the soilborne pathogen Plasmodiophora brassicae, is an important disease of canola (Brassica napus) and other members of the Brassicaceae. This disease has been spreading in western Canada in recent years and is a significant threat to the $26.7 billion canola industry. Strategies including crop rotation, sanitization of field equipment and the application of soil amendments can help to control the pathogen, but the planting of clubroot resistant cultivars has proven to be the most effective management approach. Unfortunately, new pathotypes of P. brassicae have emerged since 2013 which can overcome host resistance, and growers require new tools to mitigate the impact of clubroot.

Genomic data provide novel ways to complement traditional breeding to increase host resistance and to explore pathogen diversity. We used RNA-seq to evaluate differential gene expression through a time-course in cultivars with contrasting responses when challenged with P. brassicae. At 7 days after inoculation (dai), abiotic stress, secondary metabolism, cell wall and hormone biosynthesis were regulated in both cultivars, while heat-shock proteins were mainly upregulated in the resistant cultivar. At 14 dai, auxin-related genes were significant in both cultivars, and specific pathogenesis-related proteins and transcription factors were upregulated in the resistant cultivar. At 21 dai, a strong general gene downregulation was evident in the susceptible cultivar, while biotic-stress-related genes remained active in the resistant cultivar. We are using bioinformatics to evaluate specific disease resistance proteins and receptors, cell wall genes, transcription factors and auxin-related genes as potential candidates that can be used for functional validation and to increase host resistance through gene editing.

To study relationships among P. brassicae pathotypes and design pathotype-specific detection assays, we obtained full genome and variant information for 45 P. brassicae single-spore and field isolates. Clustering using full genome variant analysis showed that most isolates classified as variants of pathotypes 5 and 6 on the Canadian Clubroot Differential grouped together, while they grouped separately from variants of pathotypes 2 and 3. Single nucleotide polymorphisms and insertion/deletions are currently being surveyed for use in standard PCR and RNase-H-dependent PCR (rhPCR) assays, to accurately distinguish P. brassicae clusters, pathotypes and isolates. Preliminary results show rhPCR is promising as a detection method, allowing quick diagnostics of pathotype composition.