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Effect of gene stacking and soil inoculum level on the resilience of resistant canola against *Plasmodiophora brassicae* (clubroot) pathotype 3H

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Background:

Clubroot, caused by the protist Plasmodiophora brassicae Woronin, is a destructive disease for canola/rapeseed (*Brassica napus* L.) production; severe infection has shown 100% yield losses in western Canada. Genetic resistance is a key factor in a clubroot management plan to maintain crop yield and restrict inoculum build-up in the soil. However, a clubroot resistance (CR) gene can quickly become ineffective when used repeatedly, due to the diversity of pathogen population.

Objective:

To evaluate resistance resilience of selected canola inbred/hybrid lines that carry a single (*Rcr1* or *Crr1*^{rutb}) or stacked (*Rcr1* and *Crr1*^{rutb}) CR genes against a field population of *P. brassicae* pathotype 3H, the predominant type found in western Canada. The assessment was carried out in multiple cycles of exposure under controlled-environment conditions, including the trend of clubroot severity and soil inoculum build-up over time.

Methods:

Canola inbred/hybrid lines carrying single or stacked CR genes as described above were repeatedly exposed to the same inoculum source at low (1 ×105/g soil) and high (1 ×107/g) initial resting-spore concentrations. Clubroot severity was assessed after six weeks. Then all roots (with/without symptoms) were buried in the same pot of soil that they came from for three weeks to allow galls to decompose before thoroughly mixing them into the soil. The planting was carried out for five cycles, with all roots returned to the same soil after each cycle. Soil inoculum was quantified for each pot (replicated) using droplet-digital PCR before each planting.

Results:

Relative to a susceptible control, lines carrying either single or double CR genes showed strong resistance initially, especially under the low initial pathogen inoculum. The resistance declined slightly over time, especially for lines carrying the CR gene *Crr1*^{rutb} alone and under the high initial inoculum condition where disease severity index (DSI) increased from initial 9% to 39% before the planting of cycle five. The line with *Rcr1* fared slightly better, with DSI being consistently below 20%. In contrast, the line carrying both *Rcr1* and *Crr1*^{rutb} showed much greater resilience; DSI was at <3% regardless of the inoculum level or exposure cycle. All resistant lines reduced the P. brassicae inoculum in soil, with more substantial reductions observed for the treatments with the higher initial spore concentration. Continuous planting of susceptible canola increased the soil inoculum level over time.

Conclusions:

The canola line carrying the combined CR genes *Rcr1* and Crr1^{rutb} demonstrated stronger resistance resilience to a field population of *P. brassicae* pathotype 3H than those carrying only one of the resistance genes. The decline of single-gene resistance occurred more rapidly under high soil inoculum levels. Therefore, any practice that lowers *P. brassicae* resting spores in soil, such as a >2-year break between canola crops, would likely help resistance resilience, especially for canola carrying only a single CR gene. Growing a resistant canola cultivar, with either a single or multiple CR genes, helps decrease *P. brassicae* inoculum build-up in soil, especially in fields where the inoculum level is very high.