

Genetic mapping of resistance to *Aphanomyces* root rot in alfalfa

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Aphanomyces root rot, caused by *Aphanomyces euteiches*, is one of the most important diseases of alfalfa in the United States. Two races of the pathogen are currently recognized using differential varieties, WAPH-1, resistant to race 1, and WAPH-5, resistant to race 1 and race 2. Although most varieties are resistant to race 1, fewer have resistance to race 2. Bioassays of soil samples from Minnesota and New York, as well as those published from other states, indicate that race 2 is the predominant race in North America. However, distinguishing races is limited by the differential varieties available. The goal of this project is to characterize the race-specific resistance to *Aphanomyces* root rot and map resistance genes in order to facilitate breeding for resistance and to clarify race/resistance gene relationships. Microscopic analysis of race-specific resistance identified a hypersensitive response of individual epidermal or cortical cells upon pathogen attack followed by suberization of cells surrounding the stele and strong autofluorescence in cortical cells, indicating the presence of phenolic compounds. The severity of root rot symptoms corresponded with amount of pathogen DNA and oospore density in roots. Resistant and susceptible plants were identified from three varieties, WAPH-1, WAPH-5 and 53V52, and used as parents to produce F1 mapping populations. DNA from 373 F1 plants segregating for resistance was used for genotyping using genotyping-by-sequencing followed by genotype calling using FreeBayes pipelines. SNP markers significantly associated with resistance to strain MF-1 (race 1) was identified on chromosome 1 with candidate NBS-LRR genes within 38 kbp and markers associated with resistance to strain MER-4 were located on chromosome 2. Phenotyping the F1 population with six additional strains of *A. euteiches* identified plants with resistance to single strains. These results indicate that many races of the pathogen exist in North America. Ongoing QTL mapping will identify the loci for resistance to additional strains of *A. euteiches* and associated markers may be useful in marker-assisted selection for breeding alfalfa. The results of this study support the use of diverse strains when selecting disease resistant parental materials so that the maximum number of resistance genes are represented in the final variety.