Controlling Root Rot of Alfalfa Caused by Aphanomyces and Phypophthora

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Alfalfa is the fourth most important crop in the United States in terms of both cultivated acreage (greater than 20 million) and annual production value (greater than $6 billion). Alfalfa is typically grown as a perennial crop, with stands cultivated for three or more consecutive years. Effective control of root rot and crown rot diseases is critical to stand longevity, productivity and profitability. It is estimated that approximately 25% of the alfalfa hay crop of the U.S. is lost annually to disease, principally the result of soilborne pathogens. Soilborne diseases of alfalfa result in the death of both seedlings and mature plants, and also cause chronic sublethal disease. Soilborne diseases also chronically weaken root systems, which leads to additional losses due to winterkill of plants. A lack of acceptable levels of disease resistance also limits the use of alfalfa as a rotation crop in more sustainable crop production systems or as a profitable bridge crop during a transition period of land to organic status.

Isolation cages to produce seed of elite alfalfa populations in the field.

Most soilborne diseases of alfalfa are caused by fungi, water molds, and nematodes. These diseases cannot usually be controlled effectively by the application of fungicides or nematicides because chemical control is typically too expensive and the pathogens survive winter in dormant roots, stems, and leaves. The most economically effective method for controlling diseases of alfalfa is to grow varieties that are resistant to multiple diseases. Commercial alfalfa cultivars must be evaluated for resistance to nearly all significant soilborne diseases using standardized test protocols established by the North American Alfalfa Improvement Conference Committee (NAAIC) on Standardized Tests, for the resistance rating to be accepted by the National Alfalfa and Miscellaneous Legume Variety Review Board. The results of these tests are available to the public (www.alfalfa.org/falldormancy.html) and allow growers to compare levels of disease resistance among varieties.

Aphanomyces Root Rot (ARR) of alfalfa is a disease caused by the water mold *Aphanomyces euteiches*. Disease symptoms include stunted seedlings that have yellow cotyledons, damping off (seedling death) and poor stand establishment. This pathogen also causes a chronic decline in mature plants. This disease has been observed throughout the U.S. in all areas of alfalfa production except for those in the Southern Plains and Southwestern U.S. Isolates of *A. euteiches* that cause disease on alfalfa have been classified as being either one of two races based on the symptoms they produce on established resistant and susceptible check varieties. Both races (Race 1 and Race 2) have been detected in several Midwestern states, including Wisconsin and Minnesota. No effective chemical control for this disease of alfalfa is available. The pathogen produces spores that can survive for years in soil or in infected root debris. To minimize losses, the cultivation of resistant alfalfa cultivars is recommended, along with the avoidance of poorly drained and heavily infested fields. Inheritance of resistance in alfalfa to Races 1 and 2 of *A. euteiches* is controlled by different genes, and it is important for varieties to have resistance to both races when grown in areas where both races may be present. Several studies indicate that resistant varieties exhibit significantly better seedling health, yield, and persistence than more susceptible varieties when grown in naturally infested soils. A rapid test based on the detection of *A. euteiches* DNA can be used to determine if the pathogen is present in plant or soil samples, however, at present it is necessary to test isolates of *A. euteiches* against established resistant and susceptible check varieties to determine if an isolate is Race 1 or Race 2.

Table 1. *Comparison of means between bulked plant samples of alfalfa standard check populations[a] for disease severity index (DSI) ratings[b] and quantity (ng) of Aphanomyces euteiches DNA[c]. The correlations (ρ) between DSI and pathogen DNA quantity are presented.*

	***A. euteiches* MF-1**		***A. euteiches* NC-1**	
Population	ngDNA	DSI	ngDNA	DSI
WAPH-1	2.12 a	2.79 a	13.58 a	3.99 a
Saranac	8.75 b	3.92 b	12.66 a	3.96 a
WAPH-5	-	-	2.63 b	2.66 b
LSD (α = 0.05)	1.60	0.13	1.23	0.13
ρ (Prob>|ρ|)	0.78 (0.0004)		0.79 (<0.0001)	

[a] *A. euteiches MF-1:WAPH-1=resistant; Saranac=susceptible. A. euteiches NC-1: WAPH-5=resistant; WAPH-1 and Saranac=susceptible.*

[b] *Each experiment included four bulks of 15 plants each for each population. Prior to bulking, plants were individually scored using a DSI scale of 1 (healthy) – 4 (extensive necrosis of roots, hypocotyls and cotyledons, and severe stunting of stem).*

[c] *DNA was quantified using a polymerase chain reaction (PCR) assay specific for A. euteiches.*

Phytophthora Root Rot (PRR) of alfalfa is caused by the water mold *Phytophthora medicaginis*. This disease, which occurs throughout the alfalfa growing regions of the Northeastern and Midwestern U.S., can cause complete failure of stand establishment and is most prevalent when newly seeded fields are subjected to flooding early in the growing season. PRR of seedling alfalfa results in stunted seedlings and eventually, seedling death. The pathogen also causes wilt in mature plants. Similar to *A. euteiches*, the pathogen *P. medicaginis* produces spores that can survive for years in soil. PRR of alfalfa is best controlled by a combination of practices including water management, cultivation of resistant varieties, and use of the fungicide metalaxyl as a pre-plant seed treatment. Various studies have indicated that alfalfa cultivars with resistance to *P. medicaginis* have significantly higher yields than susceptible cultivars when grown in naturally infested soil.

P. medicaginis has been implicated in damping-off disease complexes of alfalfa with other soilborne pathogens including the northern root knot nematode (*Meloidogyne hapla*), southern root knot nematode (*Meloidogyne incognita*) and *A. euteiches*.

Field studies have indicated that alfalfa cultivars with resistance to both *A. euteiches* and *P. medicaginis* have significantly higher yields and greater plant vigor than cultivars that lack dual resistance.

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[c] *DNA was quantified using a polymerase chain reaction (PCR) assay specific for A. euteiches.*

Furthermore, both *A. euteiches* and *P. medicaginis* have been found together in alfalfa fields and in diseased plants. Studies suggest that at sites naturally infested with both *A. euteiches* and *P. medicaginis* there are greater losses in stand and yield than in sites infested with only one of the two pathogens. Field studies suggest that the use of varieties resistant to both pathogens likely provides the greatest plant persistence and hence, the best yield performance.

At the USDA-ARS Vegetable and Forage Research Unit, located in Prosser, WA, investigators are developing alfalfa populations that have high levels of resistance to multiple diseases, including *Aphanomyces root rot* and *Phytophthora root rot.*

This research has resulted in the development of DNA-based assays that can be used for detecting and quantifying both *A. euteiches* and *P. medicaginis* in infected plants (Tables 1 and 2). These new techniques will enable more precise identification of plants that are highly resistant to both pathogens. Improved methods for identifying plants with extreme levels of resistance to both diseases coupled with conventional methods (Table 2) for alfalfa breeding will result in timely development of these elite alfalfa populations.