

**PATHOLOGICAL RELATIONSHIP OF *MELOIDOGYNE HAPLA* AND  
*PHYTOPHTHORA MEGASPERMA* F. SP. *MEDICAGINIS*  
IN TWELVE *MEDICAGO SATIVA* L. CULTIVARS<sup>†</sup>**

G. D. Griffin and F. A. Gray<sup>2</sup>

Nematologist, USDA ARS, Forage and Range Research Laboratory, Utah State University, Logan, UT 84322-6300, U.S.A.,<sup>1</sup> and Professor, Department of Plant, Soil and Insect Sciences, University of Wyoming, Laramie, WY 82071, U.S.A.<sup>2</sup>

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ABSTRACT

Griffin, G. D., and F. A. Gray. 1994. Pathological relationship of *Meloidogyne hapla* and *Phytophthora megasperma* f. sp. *medicaginis* in twelve *Medicago sativa* L. cultivars. *Nematropica* 24:35-43.

Effect of prior inoculation with *Meloidogyne hapla* on the performance of 12 cultivars of alfalfa (*Medicago sativa*) with varying resistance to *M. hapla* was studied under greenhouse conditions. Inoculation with the fungus alone caused plant mortality in the fungus-susceptible cultivar Deseret but not in susceptible Vernal. None of the *P. megasperma* f. sp. *medicaginis*-resistant cultivars died from Phytophthora root rot (PRR). Inoculation with *M. hapla* alone resulted in mortality in nine of 12 cultivars. Combined inoculation of the two pathogens increased mortality in 10 of the 12 cultivars; only Nevada Synthetic XX (Nev Syn XX) germplasm and Chief with dual resistance were unaffected by the addition of the nematode. Inoculation with the fungus alone suppressed the dry shoot weight in four cultivars, including Vernal and Deseret, both susceptible to *P. megasperma* f. sp. *medicaginis*. *Meloidogyne hapla* reduced dry shoot weights and reproduced on roots of all entries except Nev Syn XX. *Meloidogyne hapla* reduced root weights of four of six alfalfa cultivars resistant to the nematode and five of six cultivars susceptible to the nematode. The fungus suppressed root weights in four cultivars including Vernal and Deseret. Severity of PRR was lowest in the resistant cultivar Kingstar and highest in the susceptible cultivar Deseret. Overall, *M. hapla* increased the susceptibility of alfalfa to *P. megasperma* f. sp. *medicaginis*, but the fungus did not affect the resistance or susceptibility to *M. hapla*.

*Key words:* alfalfa, cultivars, *Medicago sativa*, nematode reproduction, Phytophthora root rot, shoot weight, resistance, root-knot nematode, susceptibility.

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RESUMEN

Griffin, G. D. y F. A. Gray. 1994. Relación patológica de *Meloidogyne hapla* y *Phytophthora megasperma* f. sp. *medicaginis* en 12 cultivares de *Medicago sativa*. *Nematropica* 24:35-43.

Se estudió el efecto de pre-inoculaciones con *Meloidogyne hapla* sobre el desarrollo 12 cultivares de alfalfa (*Medicago sativa*) con variada resistencia a *M. hapla*, en condiciones de invernadero. La inoculación solo con el hongo produjo mortalidad en el cultivar susceptible al hongo Deseret pero no en el Vernal también susceptible. Ninguno de los cultivares resistentes a *P. megasperma* f. sp. *medicaginis* murió de pudrición radical por *Phytophthora* (PRF). Nueve de los 12 cultivares inoculados solamente con *M. hapla* murieron. La inoculación con hongo y nematodo causó muerte en 10 de los 12 culti-

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vares; los que sobrevivieron fueron Nevada Synthetic XX (New Syn XX) y el Chief. La inoculación solamente con el hongo disminuyó el peso seco de los rebrotes de cuatro cultivares incluyendo el Vernal y el Deseret, ambos susceptibles a *P. megasperma* f. sp. *medicaginis*. *Meloidogyne hapla* redujo el peso seco de rebrotes y se reprodujo en 11 cultivares excepto en el New Syn XX. También *M. hapla* redujo el peso radical de cuatro de los seis cultivares de alfalfa resistentes al nematodo y cinco de los seis susceptibles al nematodo. El hongo redujo el peso radical de cuatro cultivares incluyendo el Vernal y el Deseret. En síntesis, *M. hapla* incrementó la susceptibilidad de la alfalfa a *P. megasperma* f. sp. *medicaginis*, pero el hongo no afectó la resistencia o susceptibilidad a *M. hapla*.

*Palabras clave:* alfalfa, cultivares, *Medicago sativa*, reproducción del nematodo, pudrición radical de *Phytophthora*, peso de rebrotes, resistencia, nematodo agallador, susceptibilidad.

## INTRODUCTION

Phytophthora root rot (PRR) caused by *Phytophthora megasperma* sp. *medicaginis* Kuan & Erwin (4) is an important disease of alfalfa (*Medicago sativa* L.) in the United States (17). It is particularly severe in flood-irrigated fields in the western United States (3,5,13). Stand reductions of up to 75% in the midwest (16) and the Intermountain Region (5) were found where the fungus attacks seeds, seedlings, older plants, and *Rhizobium* nodules (7).

The incidence of fungal diseases, including PRR, increases in the presence of root-knot nematodes, *Meloidogyne* spp., including the northern root-knot nematode, *M. hapla* Chitwood (6,9,12,15,18). There are differences in the incidence of PRR on 'Apollo II' alfalfa, which is resistant to *P. megasperma* f. sp. *medicaginis* and is susceptible to *M. hapla*, and on 'Deseret' alfalfa, which is susceptible to both pathogens (6). New alfalfa cultivars with PRR resistance are being developed and released at a rapid rate and have been classified for resistance or susceptibility to nematodes and the fungus by the Certified Alfalfa Seed Council (1).

Differences in the resistance and susceptibility of alfalfa cultivars and germplasm to *M. hapla* have been reported (8,11). Hence, this study was conducted to determine the host-parasite relationship

between *M. hapla* and *P. megasperma* f. sp. *medicaginis* on commercial alfalfa cultivars for possible differences in resistance and susceptibility of alfalfa to single and combined inoculations.

## MATERIALS AND METHODS

*Plant Materials:* Alfalfa entries used in the study were chosen for resistance and susceptibility to *M. hapla* and *P. megasperma* f. sp. *medicaginis* and classified as highly resistant, resistant, marginally resistant, or susceptible (1). The germplasm Nevada Syn XX (Nev Syn XX) and cultivars Chief, Kingstar, Lobo, and WL 316 are classified as resistant to both *M. hapla* and *P. megasperma* f. sp. *medicaginis* (Table 1). Trident, Apollo II, Commander, Chief, Cimarron, and Vernal are resistant to *P. megasperma* f. sp. *medicaginis* but not to *M. hapla*, and Deseret is susceptible to both pathogens.

*Nematode inoculum:* Eggs of *M. hapla* used as inoculum were originally obtained from lettuce plants (*Lactuca sativa* L.) collected in northern Utah and cultured in the greenhouse on tomato (*Lycopersicon esculentum* Mill. cv. Rutgers). Inoculum was collected with the sodium hypochlorite method (14) and refrigerated at 5 C for less than 24 hr. in deionized water until applied to plant roots.

*Fungal inoculum:* Stock cultures of a Wyoming isolate of *P. megasperma* f. sp. *medicaginis* obtained from diseased alfalfa were maintained on cornmeal agar. The inoculum was prepared by placing two 5-mm-diam plugs of mycelium from stock cultures into 1,000-ml roux culture bottles containing 160 ml of sterilized, liquid V-8 juice medium (6). After 5 days, cultures were shaken vigorously to fragment mycelia and placed in a horizontal position for an additional 9 days. Inoculum was prepared by blending the mycelial mat from one culture bottle in 238 ml of sterilized deionized water the day of plant inoculations.

*Experimental procedure:* Alfalfa transplants were grown from scarified seeds treated with Captan®, and pregerminated on filter paper. Bacterial inoculum (*Rhizobium meliloti* Dang.) was placed with the seed at planting to insure nodulation and infection (6,7,12). Fifteen-day-old alfalfa seedlings were transplanted singly into individual plastic containers (6-cm-diam × 21-cm-deep) filled with 540 cm<sup>3</sup> of a 1:1 mixture of steam-sterilized Kidman fine sandy loam (coarse-loamy mixed mesic Calcic Haploxeroll; 91% sand, 5% silt, 4% clay; pH 7.2; 1.0% organic matter) and Logan clay loam (mixed mesic Typic Calciaquollid; 27% sand, 31% silt, 42% clay; pH 7.8; 2.5% organic matter) (6).

Twenty days after transplanting, plants were treated as follows: plants in treatments 1 and 3 were inoculated with 1 000 *M. hapla* eggs in 10 ml deionized water; those in treatment 2 were inoculated with a 12.5-ml suspension of fragmented fungal mycelia of *P. megasperma* f. sp. *medicaginis* per plant (6). Plants in treatment 4 served as the uninoculated controls. Twenty-eight days after inoculation, plants in treatment 3 were inoculated with a 12.5 ml suspension of fragmented mycelia per plant. Plants were inoculated by pouring nematode eggs and (or) fragmented mycelia

into four holes 10 cm deep in the soil around the hypocotyl base. Plants were grown at a controlled greenhouse bench temperature of  $26 \pm 3^\circ$  C. Supplemental light for a 19-hr daylength was provided by high-output fluorescent lamps. The experiment design was a two-way factorial (12 cultivars × four inoculations) arranged in a randomized complete-block design. The four treatments per alfalfa entry contained 12 replicates each.

Shoots of each plant were clipped and weighed after 45 days, when 10% of the plants had flowered. The experiment was terminated 96 days after the final inoculation and the percentage of plants surviving was determined. Plants were then removed from pots, shoots were reclipped, and roots were washed free of soil. Dry weights of both shoots and roots were determined. Roots were rated for PRR severity and *M. hapla* galling (6). Phytophthora root rot severity was rated on a scale of 1 to 6: 1 = none; 2 = slight (fine roots destroyed or minor lesions); 3 = moderate (1–2 distinct lesions per root); 4 = severe (many elongated lesions or 25–50% taproot rotted); 5 = very severe (more than 50% tap root rotted, plant nearly dead); 6 = plant dead. Each plant was evaluated for the percentage of the root system galled on a scale of 1 to 6: 1 = no galls, 2 = 1 to 10%, 3 = 11 to 30%, 4 = 31 to 50%, 5 = 51 to 80%, and 6 = 81 to 100% galled roots. Nematode eggs were extracted from root tissue (12), and the nematode reproductive indices (Pf/Pi = final nematode population divided by initial nematode inoculum) were determined. Data were analyzed using an analysis of variance test. When the F value was significant, means were separated at  $P \leq 0.05$  using Duncan's new multiple range test. Percentage plant survival data were transformed using arcsine transformation. The study was repeated with similar results, and data presented are from the first experiment.

## RESULTS

All plants of the alfalfa entries except Deseret (susceptible check) survived a single inoculation of the fungus (Table 1). Plant survival in cultivars inoculated with *M. hapla* alone was greater in nematode-resistant cultivars (except in Lobo, WL 316, and Vernal) than in susceptible cultivars. In 10 of 12 cultivars, fewer plants inoculated with a combination of both pathogens survived than did plants inoculated with either fungus or nematode alone.

As with plant survival, inoculation with *M. hapla* alone suppressed dry shoot weight of surviving plants of all cultivars

except Nev Syn XX (Table 2). Inoculation with *P. megasperma* f. sp. *medicaginis* alone suppressed the dry shoot weight of Trident, Cimarron, Vernal, and Deseret. Dry shoot weight of plants inoculated with a combination of the two pathogens was affected more by resistance to *M. hapla* than by resistance to *P. megasperma* f. sp. *medicaginis*. Vernal (marginally resistant to *M. hapla*) and Deseret (susceptible to *M. hapla*) had the greatest suppression in shoot weight following the combined inoculation with both pathogens.

*Meloidogyne hapla* alone suppressed dry root weights of all alfalfa cultivars except Nev Syn XX, Kingstar, and Trident (Table 3). *Phytophthora megasperma* f. sp. *medicagi-*

Table 1. Effect of the interaction between *Meloidogyne hapla* (Mh) and *Phytophthora megasperma* f. sp. *medicaginis* root rot (PRR) on the survival of plants in resistant and susceptible alfalfa entries.<sup>u,x</sup>

Alfalfa entry	Fall <sup>y</sup> dormancy	Reaction to <sup>z</sup>		Plant survival (%) after 4 months			
		Mh	PRR	Mh	PRR	Mh+PRR	Control
Nev Syn XX	4	HR	R	100 aA	100 aA	100 aA	100 aA
Chief	4	MR	HR	100 aA	100 aA	100 aA	100aA
Kingstar	3	MR	R	100 aA	100 aA	83 bB	100 aA
Lobo	6	R	R	92 bB	100 aA	83 bC	100 aA
WL 316	4	MR	MR	92 bB	100 aA	83 bC	100 aA
Trident	4	S	HR	83 cB	100 aA	75 cC	100 aA
Apollo II	4	S	HR	83 cB	100 aA	75 cC	100 aA
Commandor	4	S	R	92 bB	100 aA	75 cC	100 aA
Shield	3	S	R	83 cB	100 aA	75 cC	100 aA
Cimarron	4	S	MR	92 bB	100 aA	83 bC	100 aA
Vernal	2	MR	S	83 cB	100 aA	75 cC	100 aA
Deseret	5	S	S	83 cC	92 bB	67 dD	100 aA

<sup>u</sup>Values are the means of 12 replications (one plant per replicate), and means not followed by the same letter differ ( $P \leq 0.05$ ) according to Duncan's new multiple range test (lower case letters for columns, capital letters for rows). Data were transformed using arcsine transformation.

<sup>x</sup>Initial inoculation of plants with single inoculations of *M. hapla* and *P. megasperma* f. sp. *medicaginis* at 35 days. *P. megasperma* f. sp. *medicaginis* followed *M. hapla* by 28 days for combined inoculations.

<sup>y</sup>Fall dormancy is rated on a 1–9 scale, 1 = most dormant (winterhardy) and 9 = least dormant.

<sup>z</sup>S = susceptible; MR = moderately resistant; R = resistant; HR = highly resistant according to Certified Alfalfa Seed Council, 1992.

Table 2. Effect of the interaction between *Meloidogyne hapla* (Mh) and *Phytophthora megasperma* f. sp. *medicaginis* root rot (PRR) on shoot weight of resistant and susceptible alfalfa entries.<sup>a,b</sup>

Alfalfa entry	Reaction to <sup>c</sup>		Dry shoot weight (g)			
	Mh	PRR	Mh	PRR	Mh+PRR	Control
Nev Syn XX	HR	R	2.7 aA	2.5 abA	2.4 aA	2.8 aA
Chief	MR	HR	2.2 abB	2.8 abA	2.3 aB	3.0 aA
Kingstar	MR	R	2.1 abcB	2.7 abA	2.2 aB	3.0 aA
Lobo	R	R	2.0 bcC	2.5 abB	2.1 aC	2.9 aA
WL 316	MR	MR	1.8 bcB	2.8 abA	1.6 bB	3.0 aA
Trident	S	HR	1.9 bcB	2.3 abcB	1.3 bC	2.9 aA
Apollo II	S	HR	2.0 bcB	2.9 aA	2.1 aB	3.0 aA
Commandor	S	R	1.8 bcB	2.5 abA	2.1 aB	2.8 aA
Shield	S	R	1.6 bcB	2.5 abA	1.6 bB	2.9 aA
Cimarron	S	MR	1.7 bcC	2.2 bcB	1.3 bC	2.8 aA
Vernal	MR	S	1.6 bcB	1.8 cB	0.8 cC	2.9 aA
Deseret	S	S	1.5 cB	1.8 cB	0.8 cC	2.8 aA

<sup>a</sup>Values are the means of 12 replications (one plant per replicate), and means not followed by the same letter differ  $P \leq 0.05$ ) according to Duncan's new multiple range test (lower case letters for columns, capital letters for rows).

<sup>b</sup>Initial inoculation of plants with single inoculations of *M. hapla* and *P. megasperma* f. sp. *medicaginis* at 35 days. *P. megasperma* f. sp. *medicaginis* followed *M. hapla* by 28 days for combined inoculations.

<sup>c</sup>S = susceptible; MR = moderately resistant; R = resistant; HR = highly resistant according to Certified Alfalfa Seed Council, 1992.

*nis* alone suppressed root weights of Cimarron, WL 316, Vernal, and Deseret, which have PRR ratings of MR, MR, S and S, respectively. Combined inoculation with both pathogens suppressed root weight in 11 cultivars (compared to control) and in Kingstar and Trident (compared to inoculation with *M. hapla* alone). Only when both pathogens were present was there a suppression of root weight in Kingstar. Root weights of the resistant germplasm Nev Syn XX were not suppressed by either single or combined inoculations of the nematode and fungus.

All alfalfa entries exhibited root galling to some extent by *M. hapla* (Table 4). Between 27 and 28% of the root tissue of those cultivars classified as moderately resistant (1) were galled (average gall rating of 3.8). Adding the fungus to the inoc-

ulation did not reduce the root gall indices of any entry despite resistance classification. The Pf/Pi ratio in cultivars inoculated with *M. hapla* alone (excluding Nev Syn XX) ranged from 79 in Lobo and Chief (resistant to *M. hapla*), and WL 316 (marginally resistant) to 111 in Shield and Deseret (susceptible to *M. hapla*). Combined inoculations suppressed nematode reproduction. The Pf/Pi ratio in the combined inoculation of the two pathogens ranged from 49 in WL 316 to 95 in Shield, whereas the greatest suppression in nematode reproduction was on Deseret. The Pf/Pi ratio for *M. hapla* in Nev Syn XX alone and combined inoculations was < 1.

Although the severity of PRR was low in this study (Table 5), many plants had typical root-rot lesions. Following inoculation with *P. megasperma* f. sp. *medicaginis* alone,

Table 3. Effect of the interaction between *Meloidogyne hapla* (Mh) and *Phytophthora megasperma* f. sp. *medicaginis* root rot (PRR) on root weight of plants in resistant and susceptible alfalfa entries.<sup>a,b)</sup>

Alfalfa entry	Reaction to <sup>z</sup>		Root weight			
	Mh	PRR	Mh	PRR	Mh+PRR	Control
Nev Syn XX	HR	R	3.3 aA	2.9 bcA	2.9 aA	3.4 abA
Chief	MR	HR	2.4 bcB	3.4 aA	2.2 cdeB	3.3 abcA
Kingstar	MR	R	3.5 aA	3.3 abA	2.6 abcB	3.5 aA
Lobo	R	R	2.3 bcdB	3.4 aA	2.7 abB	3.4 abA
WL 316	MR	MR	2.4 bcB	2.4 deB	2.1 deB	3.0 abcA
Trident	S	HR	2.8 bA	2.9 bcA	2.0 deB	3.0 abcA
Apollo II	S	HR	2.1 cdB	3.0 abA	2.3 bcdB	3.2 abcA
Commandor	S	R	2.1 cdB	2.9 bcA	2.2 cdeB	3.1 abcA
Shield	S	R	2.0 cdB	2.7 cdA	1.9 deB	2.9 abcA
Cimarron	S	MR	1.9 cdB	2.2 eB	1.8 efB	2.8 abcA
Vernal	MR	S	1.8 cdB	2.0 eB	1.4 fC	2.6 cA
Deseret	S	S	1.7 dB	2.0 eB	1.6 fC	2.7 bcA

<sup>a)</sup>Values are the means of 12 replications (one plant per replicate), and means not followed by the same letter differ ( $P \leq 0.05$ ) according to Duncan's new multiple range test (lower case letters for columns, capital letters for rows).

<sup>b)</sup>Initial inoculation of plants with single inoculations of *M. hapla* and *P. megasperma* f. sp. *medicaginis* at 35 days. *P. megasperma* f. sp. *medicaginis* followed *M. hapla* by 28 days for combined inoculations.

<sup>z)</sup>S = susceptible; MR = moderately resistant; R = resistant; HR = highly resistant according to Certified Alfalfa Seed Council, 1992.

PRR ratings ranged from 1.17 in the dual resistant cultivar Kingstar to 2.08 in the dual susceptible cultivar Deseret, and were greater in Deseret, Nev Syn XX, and WL 316 than in the other entries. Kingstar had the lowest PRR rating following inoculation with the fungus or with both pathogens. The addition of *M. hapla* increased PRR only in Deseret and reduced PRR in Nev Syn XX.

## DISCUSSION

The results of this study were consistent with the results of other studies concerning the relationship between plant-parasitic nematodes and fungal pathogens.

*Meloidogyne hapla* affected the resistance of alfalfa to *P. megasperma* f. sp. *medicaginis*, but the fungus did not affect the resistance or susceptibility to *M. hapla* (6,12). Plant growth and survival from single and combined inoculations of *M. hapla* and *P. megasperma* f. sp. *medicaginis* were similar to the reported resistance and (or) susceptibility of the alfalfa entries to these two pathogens.

The decrease in the PRR of Nev Syn XX in the presence of *M. hapla* could be a result of an antagonism between the nematode and fungal zoospores (6). This is consistent with previous findings where seedling damping-off decreased when inoculated simultaneously with both

Table 4. The effect of *Phytophthora megasperma* f. sp. *medicaginis* root rot (PRR) on root galling and reproduction (Pf/Pi) of *Meloidogyne hapla* (Mh) on plants in resistant and susceptible alfalfa entries.<sup>u,w</sup>

Alfalfa entry	Reaction to <sup>x</sup>		Root galling indices <sup>y</sup>		Reproduction <sup>z</sup>	
	Mh	PRR	Mh	Mh + PRR	Mh	Mh + PRR
Nev Syn XX	HR	R	1.8 dA	1.4 cA	< 1 cA	< 1 eA
Chief	MR	HR	4.3 abcA	3.8 aA	79 bA	55 cdB
Kingstar	MR	R	3.9 acbA	3.1 bA	85 bA	62 bcdB
Lobo	R	R	3.7 bcA	3.0 bA	79 bA	60 bcdB
WL 316	MR	MR	3.3 cA	2.9 bA	79 bA	49 dB
Trident	S	HR	4.8 aA	4.2 aA	108 aA	77 abcB
Apollo II	S	HR	4.2 abcA	4.0 aA	99 aA	76 abcB
Commandor	S	R	4.8 aA	4.2 aA	96 abA	80 abcB
Shield	S	R	4.4 abA	4.0 aA	111 aA	95 aB
Cimarron	S	MR	4.3 abcA	3.8 aA	98 aA	82 abB
Vernal	MR	S	4.8 aA	4.1 aA	97 aA	68 bcdB
Deseret	S	S	4.9 aA	4.2 aA	111 aA	63 bcdB

<sup>u</sup>Values are the means of 12 replications (one plant per replicate), and means not followed by the same letter differ ( $P \leq 0.05$ ) according to Duncan's new multiple range test (lower case letters for columns, capital letters for rows).

<sup>w</sup>Initial inoculation of plants with single inoculations of *M. hapla* and *P. megasperma* f. sp. *medicaginis* at 35 days. *P. megasperma* f. sp. *medicaginis* followed *M. hapla* by 28 days for combined inoculations.

<sup>x</sup>S = susceptible; MR = moderately resistant; R = resistant; HR = highly resistant according to Certified Alfalfa Seed Council, 1992.

<sup>y</sup>1 = no galls, 2 = 1 to 10%, 3 = 11 to 30%, 4 = 31 to 50%, 5 = 51 to 80%, 6 = 81 to 100% root tissue galled.

<sup>z</sup>Final nematode population per plant/initial nematode population (inoculum) per plant.

pathogens (6). The low severity of PRR is probably a result of the short length of this study. PRR severity may have increased had the study continued for more than 4 months, which is characteristic of field conditions (7).

The major objective of this study was to determine if data generated with local nematode and fungal inoculum are consistent with the resistance classification given for commercial alfalfa cultivars (1). Commercial cultivars are selected for resistance based upon root gall indices and not on nematode reproduction. However, the Pf/Pi ratio for entries in this test with highly resistant, resistant, marginally resistant and susceptible levels of resistance were < 1, 79, 57, 77, respectively. Thus, only Nevada Syn

XX with a Pf/Pi ratio < 1 was resistant. Since root galling and reproduction are not always closely related, nematode reproduction is a more accurate means of determining resistance, and only plants with a Pf/Pi ratio < 1 should be classified as resistant (2,10). Since a high *M. hapla* Pf/Pi ratio was found on all entries except Nev Syn XX in this study, a reclassification of those cultivars selected for this study should be considered. Those plants showing normal plant growth with a high Pf/Pi ratio should be classified as tolerant and not resistant (2,10). Accurate determinations of plant resistance are important since the nematode can predispose resistant plants to fungal and bacterial pathogens (9,12,17).

Table 5. Effect of the interaction between *Meloidogyne hapla* (Mh) and *Phytophthora megasperma* f. sp. *medicaginis* root rot (PRR) on the development of *Phytophthora* root rot (PRR) of plants in resistant and susceptible alfalfa entries.<sup>u,x</sup>

Alfalfa entry	Reaction to <sup>y</sup>		PRR rating (1-5) <sup>z</sup>	
	Mh	PRR	PRR	Mh + PRR
Nev Syn XX	HR	R	1.91 aA	1.19 cB
Chief	MR	HR	1.32 bA	1.36 cA
Kingstar	MR	R	1.17 bA	1.00 cA
Lobo	R	R	1.68 bA	1.79 bA
WL 316	MR	MR	2.02 aA	2.41 bA
Trident	S	HR	1.45 bA	1.94 bA
Apollo II	S	HR	1.57 bA	1.50 cA
Commandor	S	R	1.36 bA	1.38 cA
Shield	S	R	1.45 bA	1.94 cA
Cimarron	S	MR	1.55 bA	1.38 cA
Vernal	MR	S	1.68 bA	1.94 bA
Deseret	S	S	2.08 aB	2.93 aA

<sup>u</sup>Values are the means of 12 replications (one plant per replicate), and means not followed by the same letter differ ( $P \leq 0.05$ ) according to Duncan's new multiple range test (lower case letters for columns, capital letters for rows).

<sup>x</sup>Initial inoculation of plants with single inoculations of *M. hapla* and *P. megasperma* f. sp. *medicaginis* at 35 days. *P. megasperma* f. sp. *medicaginis* followed *M. hapla* by 28 days for combined inoculations.

<sup>y</sup>S = susceptible; MR - moderately resistant; R = resistant; HR = highly resistant.

<sup>z</sup>1 = no disease; 2 = slight; 3 = moderate; 4 = severe; 5 = very severe.

Since Nev Syn XX germplasm is susceptible to a race of *M. hapla* from California (8), and all other entries were susceptible to the population of *M. hapla* used in this study, these populations may differ genetically from those used to develop *M. hapla* resistant alfalfa cultivars. It is important to determine whether cultivars of alfalfa may vary in resistance to *Meloidogyne* spp., including *M. hapla*, to avoid loss in field resistance. If possible, cultivars should be screened with nematodes from the region(s) where the alfalfa cultivars are to be grown.

Recurrent phenotypic selection based on resistance to a single pathogen should

increase levels of resistance in alfalfa to several pathogens including *Phytophthora megasperma* f. sp. *medicaginis* and *M. hapla*. Higher levels of resistance and more stable field resistance to *P. megasperma* f. sp. *medicaginis* may be possible if plants are selected after they have been sequentially inoculated with both the nematode and the fungus (6).

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