Interaction of *Pratylenchus zeae* with Four Soil Fungi on Sorghum^{1, 2}

Domingo Bee-Rodríguez and Alejandro Ayala³

ABSTRACT

In a greenhouse experiment, a population consisting of 1,500 Pratylenchus zeae in 20-cm pots were pathogenic on sorghum, and suppressed top and root growth. Pronounced necrosis of the roots resulted. Top growth was retarded by combinations of *P. zeae-Curvularia* spp., *P. zeae-Fusarium* moniliforme, *P. zeae-Rhizoctonia solani*, and *P. zeae-Macrophomina* sp., and by *F. moniliforme* and *R. solani*, alone. All nematode-fungi combinations and all fungi alone suppressed root growth. The combination *P. zeae-Curvularia* spp. produced most damage. *P. zeae-R. solani*, Curvularia spp., and *F. moniliforme* produced severe necrosis of sorghum roots. An initial inoculum of 750 *P. zeae* in 20-cm pots was not pathogenic on sorghum in a second experiment. Only the combination of *P. zeae-F. moniliforme* affected the fresh root weights; dry root weights were retarded significantly by *P. zeae-F. moniliforme* and by *F. moniliforme*, alone. The intensity of necrosis also varied with the different inocula.

INTRODUCTION

Sorghum [Sorghum bicolor (L.) Moench] is affected by a number of diseases (5, 7, 11), pests, aluminum toxicity, and low soil pH. Several nematode species, especially *Pratylenchus zeae*, are associated with a purplish color of the foliage 2 or 3 weeks after germination, wilting, and rapid death of the plants that resembles a die-back, few roots, which turn reddish, and a root cortex that peels easily. This condition is especially evident in clay soils of the Coto (Oxisol) series. Several soil fungi (*Macrophomina* sp., *Fusarium moniliforme*, *Curvularia* sp. and *Rhizoctonia solani*) were also isolated from root cultures.

Pathogenicity of *Pratylenchus* sp. has been demonstrated on several agricultural crops (2,3,4,6). Endo (4) demonstrated that *P. zeae* reproduced readily on corn and sorghum roots.

Pammel et al. (11) observed that *Fusarium* caused a root rot on corn which is expressed as a reddish color. These plants could be easily pulled from the soil. This pathogen also attacked sorghum; the main symptom was stem breaking at the first node, but the roots were not severely injured. Leukel and Martin (7) demonstrated the pathogenicity of *F. moniliforme* on sorghum; the incidence of the disease was greater at higher temperatures, which was corroborated by Johnson et al. (5).

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³ Graduate Student, Department of Agronomy, Faculty of Agriculture, and Nematologist, Agricultural Experiment Station, Mayagüez Campus, University of Puerto Rico, Río Piedras, P.R. The effects of interactions between nematodes and fungi are well known on several crops. Norton (9) reported an association of *Pratylenchus hexincisus* with charcoal rot of sorghum. Fungi, especially *Fusarium* sp. (1,5,7,12) and *Curvularia* sp. (1), have been isolated from sorghum seeds.

The objectives of these experiments were to determine the effect of P. *zeae*, alone or in combination with four soil fungi on sorghum.

MATERIALS AND METHODS

Fusarium moniliforme, Macrophomina spp., Curvularia spp., and R. solani were isolated from the roots of sorghum obtained from the experimental farm of the Mayagüez Institute of Tropical Agriculture (MITA) at Isabela. They were cultured in PDA (potato dextrose agar) and, after 10 days of growth, used to inoculate the test plants.

To apply the fungi, the content of a petri dish was blended with 150 cm³ of water for 10 s and the suspension added to the soil surface around the plant. The nematode inoculum was extracted from roots of sorghum plants in a mist chamber. The nematodes present in a suspension were calculated, and a predetermined number pipetted into holes in the soil around each plant.

Eight sorghum (T.E. Haygrazer) seeds were planted per 20-cm clay pot, containing equal parts of top soil, filter-press cake, and river sand, and later thinned to two plants. The treatments used were: 1,500 *P*. *zeae; Macrophomina* sp.; *Curvularia* spp.; *R. solani; F. moniliforme;* 1,500 *P. zeae* and *Macrophomina* sp.; 1,500 *P. zeae* and *Curvularia* spp.; 1,500 *P. zeae* + *R. solani;* 1,500 *P. zeae* and *F. moniliforme;* and the controls (free of microorganisms).

The pots were placed on a greenhouse bench in a complete randomized block design, replicated 5 times. Pots were separated and protected to minimize contamination during watering. Ambient temperature during the 5-week experiment was 24-36° C. The following parameters were evaluated at the end of the experiment: stem fresh and oven-dry weights, plant height, and disease index (0-5 scale). Fungi were reiso-lated and the nematode populations assessed in 100 cm³ of soil and 10 g of roots.

In a second experiment, the effects of the two fungi causing more damage in the first trial, (*Curvularia* spp. and *F. moniliforme*), and *P. zeae* were studied in different combinations. The treatments used were: 750 *P. zeae*; *Curvularia* spp.; *F. moniliforme*; 750 *P. zeae* and *Curvularia* spp.; 750 *P. zeae* and *F. moniliforme*; *Curvularia* spp. and *F. moniliforme*; 750 *P. zeae*, *Curvularia* spp. and *F. moniliforme*; and the control.

The soil was a Coto clay (Oxisol) brought from the MITA farm at

Isabela and partially sterilized with methyl bromide at the rate of 454 g/334.64 m^2 . Four seeds were planted per pot and later thinned to one.

The procedures described for the first experiment were employed for growing fungal and nematode inocula. The statistical design, number of replicates, and all other cultural procedures were similar to those employed in the first experiment.

RESULTS

The interaction of *P. zeae* with four soil fungi are shown in table 1. Top weight was suppressed (P = .05) by the combination *P. zeae*-*Curvularia* spp., by *F. moniliforme*, and by *P. zeae*. The top dry weight of plants inoculated with *P. zeae*-*Curvularia* spp. was signifi-

Treatments	<u>т</u> 1	Root	NT .	
	weights	Fresh weights	Fresh Dry weights weights	
	G	G	G	
P. zeae-Curvularia spp.	$25.1 c^2$	30.50 d	2.6 a	0.8 d
P. zeae-F. moniliforme	27.4 abc	32.25 cd	3.1 a	1.2 bcd
P. zeae-R. solani	27.5 abc	33.85 cd	3.2 a	2.2 abc
P. zeae-Macrophomina sp.	26.9 abc	35.60 bcd	3.3 a	1.0 bcd
F. moniliforme	26.4 bc	35.65 bcd	3.1 a	1.8 abc
R. solani	27.8 abc	37.80 bcd	3.0 a	1.6 bc
P. zeae	26.4 bc	39.20 abcd	3.9 a	3.2 a
Macrophomina sp.	28.1 abc	42.35 abc	3.8 a	1.6 bc
Curvularia spp.	28.9 ab	45.65 ab	3.7 a	2.4 ab
Control	29.7 a	49.25 a	6.0 b	0 d

 $^{\scriptscriptstyle 1}$ Index of necrosis based on classification of 0 (no necrosis) to 5 (extensive necrosis).

 2 Means with one or more letters in common do not differ significantly at P = .05 according to Duncan's Multiple Range Test.

cantly lower (P = .05) than that of plants inoculated with *Curvularia* spp. alone. The mean fresh root weight of the control plants was greater (P = .05) than that of plants infected with the combinations *P. zeae-Curvularia* spp., *P. zeae-F. moniliforme*, *P. zeae-R. solani*, *P. zeae-Macrophomina* sp., *F. moniliforme*, and *R. solani*. The adverse effect of the combination *P. zeae-Curvularia* spp. on plant roots was greater than the effects of *Curvularia* spp., and of *Macrophomina* sp. All treatments (P = .05) retarded root growth. Although not significantly different from the others, the plants infected with the combination *P. zeae-Curvularia* spp. appeared to be more severely damaged. *Pratylenchus zeae* produced more necrosis, although not significantly different from *Curvularia* spp., *F. moniliforme*, and *P. zeae-R. solani*.

The first two produced more necrosis alone than when inoculated together.

The population levels of P. zeae were much higher in the roots than in the soil. There was no apparent effect of competition from the different fungi on the reproduction of the lesion nematode as demonstrated by final populations.

Results obtained from a second experiment to evaluate the effect of P. zeae alone and in combination with *Curvularia* spp. and F. moniliforme are illustrated in table 2. Plant height was not adversely affected by nematodes or fungi alone or by combinations of them. Top fresh weights with F. moniliforme-infected plants was greater than those of the control and P. zeae, but top dry weight was similar in all treatments. Only the combination P. zeae-F. moniliforme suppressed root growth.

Treatments	Height	Root fresh weight	Top		Moonorio
			Fresh weight	Dry weight	index ¹
	Cm	G	G	G	
P. zeae	24.26 b ²	22.40 b	4.32 abc	0.84 ab	2.2 abc
P. zeae-F. moniliforme	26.98 b	27.96 ab	3.90 c	.53 b	2.6 abc
P. zeae-Curvularia sp.	24.60 b	24.46 ab	4.64 bc	.86 ab	2.4 abc
P. zeae-Curvularia spF. mo- niliforme	27.24 b	25.68 ab	6.92 ab	1.04 ab	3.4 a
F. moniliforme	32.38 a	33.92 a	5.72 bc	.55 b	1.6 bc
Curvularia sp.	26.02 b	23.54 ab	5.10 bc	.78 ab	1.8 abc
Curvularia spF. moniliforme	28.44 ab	30.74 ab	8.76 a	1.09 ab	3.0 ab
Control	28.20 ab	21.52 b	7.02 ab	1.46 a	1.0 c

 TABLE 2. - Growth and necrosis of sorghum plants inoculated with Pratylenchus zeae,

 Fusarium moniliforme, and Curvularia spp. alone and in different combinations

¹ Index of necrosis based on classification of 0 (no necrosis) to 5 (intensive necrosis).

 2 Means followed by one or more letters in common do not differ significantly at P = .05 according to Duncan's Multiple Range Test.

Both F. moniliforme alone and combined with P. zeae significantly reduced dry root (table 2). The combinations P. zeae-F. moniliforme-Curvularia spp.; and F. moniliforme-Curvularia spp. caused significantly more necrosis on sorghum roots than on the controls. There were no significant differences among the other treatments. Some necrosis developed also on the roots of control plants, which indicated the presence of a contaminant.

Final population levels of *P*. *zeae* were higher from the roots than in the soil samples.

DISCUSSION

Under the conditions of the first experiment, P. zeae and the fungi Curvularia spp., F. moniliforme, Macrophomina sp., or R. solani alone and in combinations suppressed sorghum root growth. Top dry weights were affected by *P. zeae*, *P. zeae-Curvularia* spp., and by *F. moniliforme*; *P. zeae*, *F. moniliforme*, *Curvularia* spp. and *P. zeae-R. solani* caused more necrosis on the plant roots. These results suggest that the lesion nematode and the four fungi are pathogenic on sorghum. However, the symptoms observed in the field were not produced in the greenhouse. Furthermore, results are different from those obtained by Norton (9) with *P. hexincisus* and *Macrophomina phaseoli* in the sorghum cultivar Combine 7078. The nematodes and the plant variety used and factors such as high soil temperature (24-36° C) and humidity were different. Norton (9) reported that the damage produced by both pathogens increased under conditions of low humidity.

Stanley B. King⁴ observed a sorghum disease in Nigeria similar to that described here. He isolated the four fungi genera used in our experiments but was unable to reproduce the disease under greenhouse conditions. He attributes this failure to sorghum plants never being exposed to conditions of stress. Johnson et al. (5) demonstrated that F. *moniliforme* infection and pathogenicity of sorghum roots are affected by temperature.

Some of the combinations in our experiments indicated antagonism: $Pratylenchus \ zeae$ alone caused necrosis, but not when combined with $F.\ moniliforme$ or Macrophomina sp. However, this apparent antagonism did not affect the reproductive potential of $P.\ zeae$. Litrell and Johnson (8) obtained similar results with $Pythium\ aphanidermatum$ and $Belonolaimus\ longicaudatus$.

The general growth and development of the sorghum plants in a second experiment, using the same variety, was poor compared to those of the first experiment. The difference may have been due to the change in growth media used. Fusarium moniliforme alone or in combination with P. zeae retarded root growth by 63 and 64%, respectively. This response suggests that, under the prevailing conditions, only F. moniliforme was pathogenic to sorghum roots. Inhibition of root development by P. zeae was not significant, suggesting that the inoculum used (750 specimens/plant) was below the critical level; 1,500 had been used in the first trial.

Temperature (26-38° C) may also have exerted its effect on nematode reproduction and pathogenic capacity. Palmer et al. (10) found that at 24° C, dry weight of corn roots was 12% lower in *F. moniliforme*-infected plants, 30% in plants infected with the fungus and *P. scribneri*, and 39% when the nematode was alone.

RESUMEN

En un experimento de invernadero, 1,500 nematodos de la especie *Pratylenchus zeae* por tiesto de 20 cm. fueron patogénicos en sorgo, reduciendo el crecimiento del follaje y

⁴ Personal communication.

de las raíces. Se observó, además, necrosis marcada de las raíces. El crecimiento foliar fue adversamente afectado por las siguientes combinaciones de nematodos y hongos: P. zeae-Curvularia spp., P. zeae-Fusarium moniliforme, P. zeae-Rhizoctonia solani, y P. zeae-Macrophomina sp., y por hongos solos: F. moniliforme y R. solani. Todas las combinaciones nematodos-hongos y todos los hongos solos inhibieron el desarrollo radical. La combinación P. zeae-Curvularia spp. causó más daño. P. zeae, P. zeae-R. solani, Curvularia spp. y F. moniliforme indujeron la formación de una necrosis severa. El inóculo inicial de 750 P. zeae por tiesto de 20 cm. no fue patogénico al sorgo. Solamente la combinación P. zeae-F. moniliforme disminuyó el peso fresco de las raíces; el peso seco fue afectado por P. zeae-F. moniliforme y por F. moniliforme solo. La intensidad necrótica también varió con el inóculo.

LITERATURE CITED

- Bain, D. C., Fungi recovered from seed of Sorghum vulgare Pers. Phytopathology 40: 521-2, 1950.
- Bergeson, G. B., A report on the testing of onion varieties for resistance to lesion nematodes, Plant Dis. Rep. 46: 535-6, 1962.
- Dickerson, O. J., Darling, H. M., and Griffin, G. O., Pathogenicity and population trends of *Pratylenchus penetrans* on potato and corn. Phytopathology 54: 317-22, 1964
- Endo, B. Y., Responses of root-lesion nematodes, *Pratylenchus brachyurus* and *P. zeae*, to various plant and soil types. Phytopathology 49: 417–21, 1959.
- Johnson, D. L., Davison, A. D., and Heathman, E. S., A Fusarium root rot of Sorghum vulgare. Phytopathology 56: 148-9 (Abstr.), 1966.
- Koike, H., and Román, J., Pathogenicity of *Pratylenchus brachyurus* and *Pythium graminicola* to sugarcane. Phytopathology 60: 1562-5, 1970.
- Leukel, R. W., and Martin, J. A., Seed rot and seedling blight of sorghum. USDA Tech. Bull 839: 1–26, 1943.
- Litrell, R. W., and Johnson, A. W., Pathogenicity of Pythium aphanidermatum to Chrysanthemum in combined inoculations with Belonolaimus longicaudatus or Meloidogyne incognita. Phytopathology 59: 115-6, (Abstr.) 1969.
- Norton, D. C., The association of *Pratylenchus hexincisus* with charcoal rot of sorghum. Phytopathology 48: 355-8, 1958.
- Palmer, L. T., McDonald, D., and Kommedahl, T., The ecological relationship of *Fusarium moniliforme* to *Pratylenchus scribneri* in seedling blight of corn. Phytopathology 57: 825 (Abstr.), 1967.
- Pammel, L. H., King, C. M., and Seal, J. L., Studies on a *Fusarium* disease of corn and sorghum (Preliminary). Iowa State Col. Res. Bull. 33: 113-36, 1916.
- Swarup, G., Hansing, E. D., and Rogerson, C. T., Fungi associated with sorghum seed in Kansas. Phytopathology 46: 28 (Abstr.), 1956.