

Host status and susceptibility of sorghum to *Pratylenchus* species (1)

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SUMMARY

The host status of sorghum for the lesion nematodes *Pratylenchus brachyurus*, *P. crenatus*, *P. coffeae*, and *P. zaeae* was tested in greenhouse and field microplot experiments. The susceptibility of sorghum to *P. brachyurus* and *P. zaeae* was also examined. Sorghum cultivar Pioneer 8222 (P 8222) was a good host for *P. zaeae*, which had reproduction indices (Pf/Pi) of 62.8 and 44.3 in greenhouse and microplot tests, respectively. *Pratylenchus brachyurus* and *P. coffeae* had limited reproduction on P 8222 with reproduction indices of less than 3.0 in greenhouse and microplot tests. In greenhouse tests, P 8222 was a nonhost for *P. crenatus*, which had a reproduction index of 0.1. In greenhouse tests with ten different sorghum cultivars, all but one cultivar supported higher levels of reproduction of *P. zaeae* than of *P. brachyurus*. Sorghum growth in greenhouse tests was unaffected by initial populations as high as 8 000 *P. brachyurus*/seedling. *In vitro* inoculation of sorghum seedling roots with *P. brachyurus* resulted in extensive necrotic lesion development that was suggestive of a hypersensitive reaction. In contrast, initial populations as low as 600 *P. zaeae*/seedling suppressed sorghum growth in greenhouse tests. The relationship between initial nematode populations and sorghum growth was described adequately by quadratic models ($R^2 = 0.63$ and 0.67 for root and shoot growth, respectively). In microplot tests, however, initial populations as high as 600 *P. zaeae*/500 cm³ had no detectable effect on sorghum growth. Necrotic lesion development was sparse and limited when sorghum seedling roots were inoculated with *P. zaeae in vitro*. Thus, while sorghum is a good host for *P. zaeae*, it is relatively tolerant or insensitive to the nematode's feeding activities.

RÉSUMÉ

Qualité d'hôte et sensibilité du sorgho aux espèces de *Pratylenchus*

La qualité d'hôte du sorgho envers les nématodes *Pratylenchus brachyurus*, *P. crenatus*, *P. coffeae* et *P. zaeae* a été testée en serre et en microparcelles expérimentales. La sensibilité du sorgho à *P. brachyurus* et à *P. zaeae* a été également étudiée. Le cultivar Pioneer 8222 (P 8222) est un bon hôte pour *P. zaeae* qui montrait des taux de reproduction (Pf/Pi) de 62,8 et 44,3 en serre et en microparcelles respectivement. *P. brachyurus* et *P. coffeae* avaient un taux de reproduction de moins de 3,0 en serre et en microparcelles. En serre, le cv. P 8222 ne s'est pas montré hôte pour *P. crenatus* (taux de reproduction : 0,1). Sur dix cultivars de sorgho testés en serre, neuf ont montré un taux de reproduction de *P. zaeae* plus élevé que celui de *P. brachyurus*. Lors des tests en serre, la croissance du sorgho n'était pas affectée par des populations initiales aussi élevées que 8 000 *P. brachyurus* par plantule. *In vitro*, l'inoculation de racines de plantules de sorgho avec *P. brachyurus* produit des lésions nécrotiques indicatrices d'une réaction d'hypersensibilité. À l'opposé, en serre, des taux d'inoculum aussi peu élevés que 600 *P. zaeae* par plantule empêchent leur croissance. La relation entre population initiale du nématode et croissance du sorgho est traduite de façon adéquate par des modèles quadratiques ($R^2 = 0,63$ et $0,67$ pour les racines et les pousses, respectivement). Cependant en microparcelles des taux de population aussi élevés que 600 *P. zaeae* pour 500 cm³ de sol n'ont eu aucun effet sur la croissance du sorgho. L'inoculation *in vitro* de *P. zaeae* sur des racines de plantules de sorgho cause des lésions nécrotiques éparses et limitées. Ainsi, alors que le sorgho est un bon hôte pour *P. zaeae*, il demeure relativement tolérant ou insensible à la prise de nourriture par les nématodes.

Diseases are important in limiting production of *Sorghum bicolor* (L.). Moench and those caused by fungi, bacteria, and viruses are well documented (Frederiksen, 1982). Several genera of plant parasitic nematodes are reported to reproduce on sorghum (Chevres-Roman, Gross & Sasser, 1971; Carter & Nieto, 1975; Bee-

Rodriguez & Ayala, 1977; Birchfield, 1983) and some have been implicated in yield suppression (Lamberti, 1969; Chevres-Roman, Gross & Sasser, 1971; Marks & Townsend, 1972; Smolik, 1977; Pinto & Lordello, 1980). Although quantitative surveys of sorghum fields have indicated a high frequency of occurrence of *Pra-*

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tylenchus species (Chevres-Roman, Gross & Sasser, 1971; Bee-Rodriguez & Ayala, 1977; Pinto & Lordello, 1980; Sharma & Medeiros, 1982; Cuarezma-Teran, Trevathan & Bost, 1984) specific information regarding host status and pathogenicity of this genus on sorghum is limited. Sorghum was reported to be a host for *Pratylenchus brachyurus* (Endo, 1959; Good, Murphy & Brodie, 1972), *P. hexincisus* (Norton, 1958), *P. penetrans* (Marks & Townsend, 1972), *P. sudanensis* (Yassin, 1973), *P. thornei* (O'Brien, 1982), and *P. zaeae* (Endo, 1959; Cuarezma-Teran, Trevathan & Bost, 1984, 1985). Sorghum has also been reported to be a non-host for populations of *P. hexincisus*, *P. neglectus* and *P. scribneri* (Dickerson, Frantz & Lash, 1978). Cuarezma-Teran, Trevathan and Bost (1984) reported that sorghum was both a host for, and susceptible to, *P. zaeae*.

The objectives of this study were (i) to determine the host suitability of sorghum to the lesion nematodes, *P. brachyurus*, *P. coffeae*, *P. crenatus*, and *P. zaeae*; (ii) to determine the pathogenicity of those species for which sorghum is a host; and (iii) to examine sorghum cultivars and breeding lines for variation in reaction to these *Pratylenchus* species.

Materials and methods

Populations of *P. brachyurus*, *P. coffeae*, *P. crenatus*, and *P. zaeae* used in these studies were cultured monoxenically on alfalfa callus tissue (Riedel & Foster, 1970). The sorghum cultivar Pioneer 8222 was used for all tests unless noted otherwise.

Inoculum for initial greenhouse and *in vitro* tests was obtained by incubating callus cultures on Baerman funnels for 48 hours. In all other tests inocula was prepared by a modification of the technique of Martin, Riedel and Rowe (1983). Callus tissue was separated from the agar medium and homogenized for 5 seconds in a blender. The resulting slurry was mixed with a moist, pasteurized loamy sand (85 % sand, 8 % silt, 7 % clay, pH 7.5) in a rotary soil mixer. Nematode populations in this highly infested soil was determined by incubating triplicate 50-cm³ samples on Baerman funnels for seven days. Sufficient amounts of this infested soil was then mixed with uninfested soil in pots or field microplots to obtain desired nematode populations. The soil used for inoculum preparation was identical to that used in greenhouse tests and that contained in field microplots.

Estimates of nematode populations for all tests were determined by combining counts from the soil and root fractions of the population. In greenhouse tests roots were separated from the soil, washed with water, blotted dry, and weighed. Nematodes were extracted from root tissues by incubating a 3-gram sample in a mist chamber for four days. Nematodes were extracted from soil by elutriation-centrifugation (Byrd *et al.*, 1976) of a

500-cm³ sample. Small root fragments recovered during elutriation were also placed in a mist chamber for four days. Counts of nematodes from all three fractions (soil, harvested roots, and roots recovered during elutriation) were combined to determine total nematodes per pot. Nematode populations in soil and roots in microplot tests were determined similarly, following elutriation of 500-cm³ soil samples. Individual samples were collected from each microplot and consisted of eight soil cores, each 2.5-cm diameter by 25-cm deep. Reproduction indices, the final population (*P_f*) divided by the initial population (*P_i*), were calculated for all tests.

To observe root symptom development on sorghum, 2-day-old seedlings were placed in Petri dishes with the roots between two pieces of absorbant paper (three seedlings/dish) and a suspension of 200 individuals of *P. brachyurus* or *P. zaeae* pipetted over the roots of each seedling. Inoculated roots, and non inoculated controls, were incubated on the laboratory bench at 24° and symptoms noted at 24 and 48 hours after inoculation. At 48 hours roots were stained with 0.1 % acid fuchsin in lactophenol and examined microscopically to confirm the presence of nematodes within the roots.

Results

When estimating nematodes populations from greenhouse tests, most nematodes were found in the finer roots (< 2-mm diameter) as opposed to the more coarse roots (> 2-mm diameter). The nematodes recovered from the three grams of root samples were always fewer than those in the finer root fragments recovered during elutriation. More nematodes were recovered from the total root fraction (ca 70 % of the total population) than from the surrounding soil.

REPRODUCTION OF PRATYLENCHUS SPP. ON SORGHUM

In greenhouse tests of reproduction, a single 3-day-old sorghum seedling was transplanted into a 20-cm diameter pot containing a loamy sand soil; 300 individuals of either *P. brachyurus*, *P. coffeae*, *P. crenatus*, or *P. zaeae* were then pipetted onto the soil around the base of each seedling. There were eight replications of each treatment and the test was repeated once. The tests were terminated after ten weeks. Reproduction indices for *P. brachyurus* and *P. crenatus* in these tests were less than 1.0 while that for *P. coffeae* was only slightly greater than 1.0. In contrast, *P. zaeae* had a significantly (*P* = 0.01) higher reproduction index of 62.8 (Tab. 1).

To determine reproduction under near field conditions, field microplots were established in 55-cm diameter by 45-cm deep plastic cylinders buried in the ground and filled with a loamy sand soil. The soil was infested with individual *Pratylenchus* species at the rate of 60 nematodes/500 cm³ by the modified technique of

Martin, Riedel and Rowe (1983) and ten sorghum seed immediately planted into each microplot. There were four replicated microplots for each *Pratylenchus* species. Fourteen weeks after planting the reproduction indices for *P. brachyurus* and *P. coffeae* were 1.7 and 2.2, respectively (Tab. 1). In this test the reproduction index for *P. zaeae* was 44.3 and was significantly greater ($P = 0.05$) than that of *P. brachyurus* or *P. coffeae*.

Table 1

Reproduction of *Pratylenchus* species on sorghum (Pioneer 8222) in greenhouse and microplot tests

Pratylenchus species	Greenhouse (a)		Microplots (b)	
	Pf	Pf/Pi	Pf	Pf/Pi
<i>P. brachyurus</i>	280	0.9	100	1.7
<i>P. coffeae</i>	400	1.3	130	2.2
<i>P. crenatus</i>	40	0.1	—	—
<i>P. zaeae</i>	18850	62.8	2660	44.3
LSD 0.01	1360	45.3	1230	20.5

(a) Initial populations were 300 nematodes/pot; Pf values are numbers/pot.

(b) Initial populations were 60 nematodes/500 cm³ soil; Pf values are numbers/500 cm³ soil.

PATHOGENICITY OF *P. BRACHYURUS* AND *P. ZEAEE* ON SORGHUM

To test pathogenicity in the greenhouse, individual 3-day-old sorghum seedlings were transplanted into 15-cm diameter pots containing a loamy sand soil infested with either *P. brachyurus* or *P. zaeae* by the modified technique of Martin, Riedel and Rowe (1983). There were six different initial populations for each nematode species and eight replications of each treatment.

Initial populations of *P. brachyurus* as high as 8 000 nematodes/pot had no detectible effect on sorghum growth after six weeks (Tab. 2). A slight root necrosis was observed on plants grown in soil infested 8 000 nematodes/pot. In contrasts, initial populations as low as 600 *P. zaeae* per pot significantly ($P = 0.05$) suppressed plant heights, and shoot dry and root fresh weights. Growth differences between control plants and plants in *P. zaeae*-infested soil were observed as early as two weeks after transplanting. Roots of plants that had received the highest Pi were severely stunted but no root necrosis was observed. The relationship between shoot dry weight and the natural logarithm of Pi [$\ln(Pi + 1)$] for *P. zaeae* fit the quadratic equation $y = 4.07 + 0.53x - 0.11x^2$

$- 0.11x^2$ ($P = 0.001$, $R^2 = 0.67$) (Fig. 1), where $x = \ln(Pi + 1)$ and $y =$ shoot dry weight. The relationship between fresh root weight and $\ln(Pi + 1)$ fitted the quadratic equation $y = 9.57 + 1.56x - 0.31x^2$ ($P = 0.001$, $R^2 = 0.63$) (Fig. 2), where $x = \ln(Pi + 1)$, and $y =$ fresh root weight.

Table 2

Growth response of sorghum (P 8222) to different initial populations (Pi) of *Pratylenchus brachyurus* in greenhouse tests (a)

Pi	Plant height (cm)	Shoot dry wt (g)	Root fresh wt (g)
0	54.0	7.1	15.1
500	44.3	5.6	11.8
1 000	43.3	5.4	12.2
2 000	51.2	6.5	11.7
4 000	45.4	6.0	10.8
8 000	47.2	6.5	13.9
LSD 0.05	NS	NS	NS

(a) Plants grown for 6 weeks after transplanting seedlings into infested soil, values are means of 8 replicates.

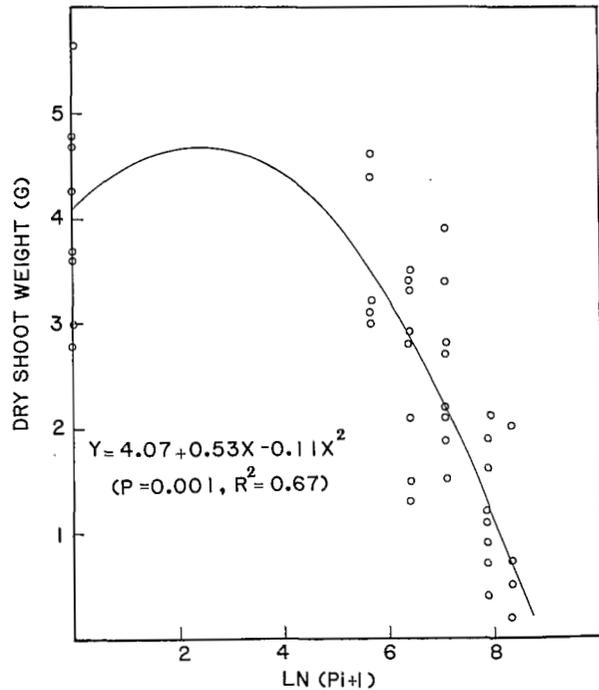


Fig. 1. The relationship between sorghum (P 8222) shoot dry weight after six weeks and initial populations of *Pratylenchus zaeae* in greenhouse tests.

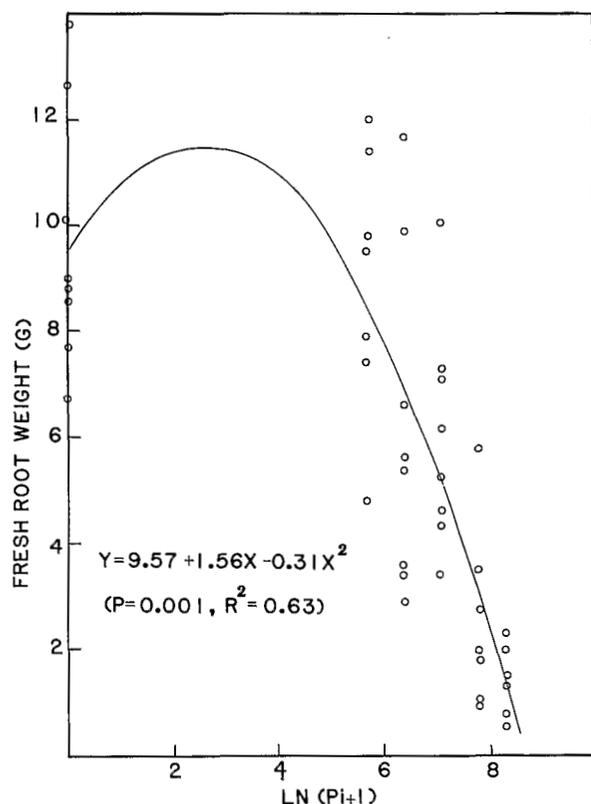


Fig. 2. The relationship between sorghum (P 8222) fresh root weight after six weeks and initial populations of *Pratylenchus zaei* in greenhouse tests.

In microplot tests, however, there was no observed suppression of sorghum growth or yield due to initial population of *P. zaei* as high as 600 nematodes/500 cm³ soil. At midseason (seven weeks after planting), the reproduction indices for different treatments ranged from 3.5 to 4.7. Reproduction indices at harvest (fourteen weeks after planting) ranged from 11.8 to 29.4 and there was a significant ($P = 0.01$, $R^2 = 0.998$) negative correlation between the log P_i and the final reproduction indices.

In laboratory studies, *P. brachyurus* caused more root necrosis on sorghum than did *P. zaei* (Fig. 3). These differences were evident within 24 hours, when necrosis was observed only on roots infected with *P. brachyurus*. After 48 hours, extensive necrosis was observed on roots inoculated with *P. brachyurus* while only a slight necrosis was observed on roots infected with *P. zaei*. Microscopic examination of stained roots revealed that large numbers of nematodes had penetrated the roots in each treatment.

VARIATION IN THE REPRODUCTION INDICES OF *P. BRACHYURUS* AND *P. ZAEI* ON DIFFERENT SORGHUM CULTIVARS

Reproduction of *P. brachyurus* and *P. zaei* on ten different sorghum cultivars was tested in the greenhouse. Single 3-day-old sorghum seedling were transplanted into 20-cm diameter pots containing a loamy sand soil infested by the modified Martin, Riedel and Rowe (1983) technique. There were four separate tests; two for each nematode species. The first test included five sorghum cultivars while the second test included six cultivars. Sorghum cultivar Pioneer 8222 was included in each test as a reference standard and there were eight replications of each cultivar for each test. The tests were terminated after eight weeks and nematode populations determined as described previously.

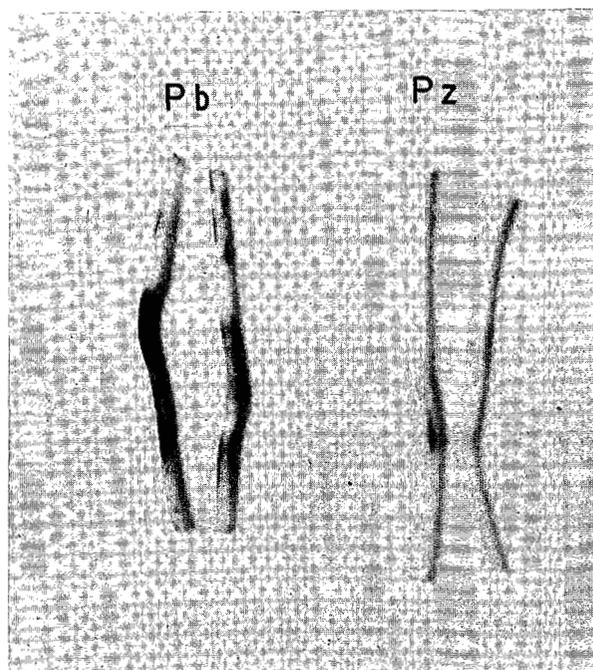


Fig. 3. Symptoms observed on seedling roots of sorghum (P 8222) 48 hours after inoculation with *Pratylenchus brachyurus* (Pb) or *P. zaei* (Pz) in laboratory tests.

No significant variation was observed among the sorghum cultivars with respect to their ability to support reproduction of *P. brachyurus* (Tab. 3). There was, however, a significant difference in total reproduction of *P. brachyurus* between Test I and Test II. In general, total reproduction was low for all sorghum cultivars, with the highest reproduction index being 2.5 for P 8222 in Test I. The reproduction indices for *P. zaei* on all

sorghum cultivars were greater than 1.0 (Tab. 3). The highest reproduction indices were on Topaz for Test I and P 8222 for Test II. These values were significantly higher ($P = 0.05$) than reproduction indices on RT × 430 and Greenleaf, respectively. Generally, the reproduction indices in Test II were much higher than those in Test I. The reproduction indices for *P. zaeae* were higher than those for *P. brachyurus* on all sorghum lines except RT × 430.

Table 3

Reproduction indices (Pf/Pi) of *Pratylenchus brachyurus* and *P. zaeae* on different sorghum cultivars in greenhouse tests

	<i>P. brachyurus</i>		<i>P. zaeae</i>	
	Cultivar	Pf/Pi	Cultivar	Pf/Pi
Test I	P 8222	2.5	P 8222	3.9
	RT × 430	2.1	Topaz	8.4
	CS 3541	1.4	G 522 DR	4.9
	Top Hand	1.2	CS 3541	3.7
	Green Leaf	1.0	RT × 430	1.9
LSD 0.05		NS		5.0
Test II	P 8222	0.6	P 8222	49.2
	W 839 DR	0.3	W 839 DR	36.5
	P 8333	0.4	P 8333	29.0
	T × 2784	0.2	Top Hand	28.1
	Topaz	0.5	TX 2784	24.8
	G 522 DR	0.8	Green leaf	8.3
LSD 0.05		NS		30.5

Discussion

Sorghum is a good host for *P. zaeae* based on the high reproduction indices observed in both greenhouse and microplot experiments. The low reproduction index of *P. crenatus* indicates that sorghum is a non-host for this nematode species. Sorghum is a poor to moderate host for both *P. brachyurus* and *P. coffeae*. That the reproduction indices for these two species were slightly greater than 1.0, however, indicates the possibility of sorghum acting as a maintenance host. This may pose problems in rotation schemes when crops susceptible to these nematodes are grown after sorghum.

Since populations of *P. brachyurus* as high as 8 000 nematodes/pot had no effect on sorghum growth, it can be assumed that *P. brachyurus* is non-pathogenic to sorghum. It is highly unlikely that initial population densities in the field will exceed 8 000 nema-

todes/500 cm³ soil. The extensive root necrosis observed in response to invasion of sorghum roots by *P. brachyurus* is suggestive of a hypersensitive response and may be responsible, in part, for the limited reproduction of the nematode on sorghum. *P. zaeae* was pathogenic on sorghum in greenhouse tests. The relationships between *Pi* and dry shoot weight or fresh root weight were adequately described by quadratic models. Damage to sorghum at initial population densities of 600 nematodes/pot was similar to that observed previously (Cuarezma-Teran, Trevathan & Bost, 1985). That no effect of *P. zaeae* on sorghum growth or grain yield was observed in microplot tests, however, suggests that sorghum is relatively tolerant of the nematode under these conditions. The lack of significant necrotic symptom development on P 8222 in response to infection by *P. zaeae* in laboratory tests is further evidence that sorghum is tolerant of this nematode. Thus, *P. zaeae* may not be expected to cause significant yield losses in the field. Some variation in reproduction of *P. zaeae* on different sorghum cultivars was observed however. Thus, other sorghum cultivars may be less tolerant of *P. zaeae*. In previous studies (Starr, Newton & Miller, 1984), infection of seedling roots of the sorghum cultivar AT × 399 × RT × 430 by *P. zaeae* did result in extensive necrotic lesion development and a decrease in the hydrogen cyanide potential of infected tissue.

REFERENCES

- BEE-RODRIGUEZ, D. & AYALA, A. (1977). Nematodes associated with sorghum in Puerto Rico. *Nematropica*, 7 : 16-20.
- BIRCHFIELD, W. (1983). Wheat and grain sorghum varietal reaction to *Meloidogyne incognita* and *Rotylenchulus reniformis*. *Pl. Dis.*, 67 : 41-42.
- BYRD, D. W., BARKER, K. R., FERRIS, H., NUSBAUM, C. J., GRIFFIN, W. E., SMALL, R. H. & STONE, C. A. (1976). Two semi-automatic elutriators for extracting nematodes and certain fungi from soil. *J. Nematol.*, 8 : 206-212.
- CARTER, W. W. & NIETO, S. (1975). Population development of *Meloidogyne incognita* as influenced by crop rotation and fallow. *Pl. Dis. Repr.*, 59 : 402-403.
- CHEVRES-ROMAN, R., GROSS, H. D. & SASSER, J. N. (1971). The influence of selected nematode species and number of consecutive plantings of corn and sorghum on forage production, chemical composition of plant and soil water use efficiency. *Nematropica*, 1 : 40-41.
- CUAREZMA-TERAN, J. A., TREVATHAN, L. F. & BOST, S. C. (1984). Nematodes associated with sorghum in Mississippi. *Pl. Dis.*, 68 : 1083-1085.
- CUAREZMA-TERAN, J. A., TREVATHAN, L. F. & BOST, S. C. (1985). Effects of *Pratylenchus zaeae* and *Quinisulcius acutus* alone and in combination on sorghum. *J. Nematol.*, 17 : 169-174.

- DICKERSON, O. J., FRANTZ, J. J. & LASH, L. D. (1978). Influence of crop rotation on nematode populations in Kansas. *J. Nematol.*, 10 : 284.
- ENDO, B. Y. (1959). Response of root lesion nematodes, *Pratylenchus brachyurus* and *P. zeae* to various plants and soil types. *Phytopathology*, 49 : 417-421.
- FREDERIKSEN, R. A. (1982). Problems in Sorghum Diseases. *Sorghum in the eighties, Proc. intern. Symp. Sorghum, 2-7 Nov. 1981, ICRISAT, India* : 263-271.
- GOOD, J. M., MURPHY, W. S. & BRODIE, B. B. (1972). Population dynamics of plant nematodes in cultivated soil : Length of rotation in newly cleared and old agricultural land. *J. Nematol.*, 5 : 117-122.
- LAMBERTI, F. (1969). Pathogenicity of *Longidorus africanus* on selected field crops. *Pl. Dis. Repr.*, 53 : 421-424.
- MARKS, C. F. & TOWNSEND, J. L. (1972). Multiplication of the root lesion nematode, *Pratylenchus penetrans* under orchard cover crops. *Can. J. Pl. Sci.*, 52 : 187-188.
- MARTIN, M. J., RIEDEL, R. M. & ROWE, R. C. (1983). A technique for quantitative use of nematodes from monoxenic cultures as soil inoculum. *Pl. Dis.*, 67 : 275-277.
- MITCHELL, R. E., MALEK, R. B., TAYLOR, D. P. & EDWARDS, D. I. (1973). Races of the barley root-knot nematode, *Meloidogyne naasi*. Characterization by host preference. *J. Nematol.*, 5 : 41-44.
- O'BRIEN, P. C. (1982). A study of the host range of *Pratylenchus thornei*. *Austral. Pl. Pathol.*, 11 : 3-5.
- NORTON, D. C. (1958). The association of *Pratylenchus hexincisus* with charcoal rot of sorghum. *Phytopathology*, 48 : 355-358.
- PINTO, N. F. J. A. & LORDELLO, R. R. A. (1980). Qualitative and quantitative survey of nematode in different experimental areas of the millet and sorghum. *Fitopatol. brasil.*, 5 : 439.
- RIEDEL, R. M. & FOSTER, J. C. (1970). Monoxenic culture of *Ditylenchus dipsaci* and *Pratylenchus penetrans* with modified Krusberg's and White's media. *Pl. Dis. Repr.*, 54 : 251-254.
- SHARMA, R. V. & DE S. MEDEIROS, A. C. (1982). Nematodes associated with sorghum in the Federal District of Brazil. *Pesq. Agropec. brasil.*, 18 : 49-53.
- SMOLIK, J. D. (1977). Effects of *Trichodorus allius* and *Tylenchorhynchus nudus* on growth of sorghum. *Pl. Dis. Repr.*, 61 : 855-858.
- STARR, J. L., NEWTON, R. J. & MILLER, F. R. (1984). Presence of dhurrin in sorghum root tissue and the effect of pathogenesis on hydrogen cyanide potential. *Crop Sci.*, 24 : 739-742.
- YASSIN, A. M. (1973). A root lesion nematode parasitic to cotton in Gezira. *Cotton Grow. Rev.*, 50 : 161-168.

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