EVALUATION OF PIGEONPEA (*CAJANUS CAJAN*) ACCESSIONS FOR RESISTANCE TO *MELOIDOGYNE INCOGNITA* RACE 3 AND *M. JAVANICA*[1]

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ABSTRACT

Moore, K. M., J. R. Rich, K. L. Buhr, and D. A. Knauft. 1988. Evaluation of pigeonpea (*Cajanus cajan*) accessions for resistance to *Meloidogyne incognita* race 3 and *M. javanica*. Nematrópica 18: 129–136.

Twenty-eight pigeonpea (*Cajanus cajan*) accessions were evaluated for resistance to *Meloidogyne incognita* race 3 and *M. javanica*. In a greenhouse seedling test, accessions were inoculated with either *M. incognita* or *M. javanica*. Four of these accesions, (ICP 8866, ICP 11289, ICP 11291, and ICPH-2) were selected, based on the level of resistance they exhibited, for further evaluation in a field microplot test. In the greenhouse, 24 of the accessions inoculated with *M. incognita* were free of galls and egg masses. When inoculated with *M. javanica*, 10 of 28 pigeonpea accessions were not galled and egg masses were not observed. In the microplot test, the resistant accessions ICP 8866, ICP 11289, ICP 11291, and ICP H-2 caused population densities of both *M. incognita* and *M. javanica* to decrease. Yield of the four accessions was not affected by these two *Meloidogyne* species.

Key words: Cajanus cajan, Meloidogyne incognita, M. javanica, pigeonpea, resistance, root-knot nematode.

RESUMEN

Moore, K. M., J. R. Rich, K. L. Buhr y D. A. Knauft. 1988. Evaluación de selecciones de gandul (*Cajanus cajan*) para resistencia a *Meloidogyne incognita* raza 3 y *M. javanica*. Nematrópica 18: 129–136.

Veintiocho selecciones de gandul (*Cajanus cajan*) fueron evaluadas para su resistencia a *Meloidogyne incognita* raza 3 y *M. javanica*. En una prueba de invernadero, las selecciones fueron inoculadas con *M. incognita* o con *M. javanica*. Cuatro de estas (ICP 8866, ICP 11289, ICP 11291 e ICPH-2) fueron seleccionadas, según el nivel de resistencia que mostraron, para ser evaluadas adicionalmente en microparcelas en el campo. En el invernadero, 24 de las selecciones inoculadas con *M. incognita* no presentaron agallas o masas de huevos. En las inoculaciones con *M. javanica*, 10 de las 28 selecciones no presentaron agallas; tampoco se observaron masas de huevos. En la prueba de campo realizada en microparcelas, las selecciones resistentes ICP 8866, ICP 11289, ICP 11291 e ICPH-2 redujeron las densidades poblacionales de ambas especies de *Meloidogyne*. El rendimiento de las selecciones no fue afectado por ninguna de las dos especies del nematodo.

Palabras claves: Cajanus cajan, gandul, *Meloidogyne incognita, M. javanica,* nematodo agallador, resistencia.

INTRODUCTION

Pigeonpea (*Cajanus cajan* L. Millsp.) is a short-lived perennial pulse crop grown throughout the tropics (15.) Valued as a food crop in most regions of the world, pigeonpea has been grown in the United States mainly as either a green manure or forage crop (10). In recent years there has been an increase in the use of pigeonpea for human consumption in the United States. Cultivars suitable for commercial production have not been available in the southeastern United States and plant breeders at the University of Florida are developing the crop to meet the specific requirements of agriculture in that region.

Various pests, including insects, fungi, bacteria, viruses, weeds, and nematodes, constitute a threat to successful pigeonpea production. Among these pests, plant-parasitic nematodes are widespread in agronomic crops and may pose a serious constraint to production of pigeonpea. *Meloidogyne* spp. are prevalent in Florida and are potentially damaging to pigeonpea. Two species, *M. incognita* (Kofoid & White) Chitwood and *M. javanica* (Treub) Chitwood, were reported as parasites of pigeonpea (2,5,11,14). Because of the potential problems that may be associated with these *Meloidogyne* spp., incorporation of resistance into commercial cultivars is desirable. Our objectives were to evaluate pigeonpea germplasm for resistance to *M. incognita* race 3 and *M. javanica* in greenhouse and microplot tests.

MATERIALS AND METHODS

A greenhouse evaluation of seedlings was conducted in the fall of 1984 and repeated in the summer of 1985. In both tests, the same 28 pigeonpea accessions were evaluated for their reaction to *M. incognita* race 3 and *M. javanica* (Table 1). Fifteen accessions were acquired from the world collection of pigeonpea germplasm at the International Crops Research Institute for the Semi-Arid Tropics (ICRISAT) in India and were selected for screening on the basis of preliminary work that suggested possible resistance to *Meloidogyne* spp. (13). Two of the accessions were cultivars commonly grown in Puerto Rico, whereas four accessions were acquired from Honduras and five from Haiti.

Seed of the accessions were germinated on wet filter paper in petri dishes. The 3-day-old seedlings were planted into 150-cm^3 Super-Cell Cone-tainers (Ray Leach Cone-tainers, Canby, OR. 97013). Each Cone-tainer had 20 cm^3 of vermiculite placed in the bottom and the remaining volume was filled with methyl bromide-fumigated Arredondo fine sand (loamy, siliceous, hyperthermic, grossarenic, paleudult). When 2-wk-old,

Table 1. Accessions evaluated for resistance to *Meloidogyne incognita* race 3 and *M. javanica*.

Accession	Region of Origin	Accession	Region of Origin
ICP 8858	India	ICP 11297	India
ICP 8859	"	Kaki	Puerto Rico
ICP 8860	"	2-B-Bushy	"
ICP 8861	"	ICPL-234	Honduras
ICP 8862	"	M-59-1	"
ICP 8865	"	ICPH-42[z]	"
ICP 8866	"	ICPH-2[z]	"
ICP 8867	"	ICPL-297[z]	"
ICP 8869	"	QPL-46	"
ICP 10958	"	ICPL-374[z]	Haiti
ICP 11286	"	Local	"
ICP 11289	"	SNO-44	"
ICP 11291	"	SNO-57	"
ICP 11295	"	PDM-1	"

[z]May have originated in India

each plant was inoculated with eggs of either *M. incognita* race 3 or *M. javanica*. Eggs were collected from tomato roots (*Lycopersicon esculentum* Mill., 'Rutgers') using the sodium hypochlorite extraction method (7). Inoculations were made with a syringe calibrated to deliver 3 ml of suspension containing approximately 750 eggs + second-stage juveniles (J2) of the appropriate *Meloidogyne* species. Two 3-ml injections were made per Cone-tainer. The experimental design was a randomized complete block (RCB) with six replications. Each experimental unit was a Cone-tainer with one plant. In addition, 'Rutgers' tomato seedlings served as susceptible controls. Sixty-five days after inoculation, plant heights were recorded. Plant root systems were rinsed in tap water and stained with Phloxine B to make the egg masses more discernible (6). The root systems were then rated separately for egg mass formation and galling. The roots were rated using a scale of 0–5 where 0 = 0; 1 = 1–2; 2 = 3–10; 3 = 11–30; 4 = 31–100; and 5 = more than 100 egg masses or galls per root system (16).

Based on the results of the greenhouse evaluation, four accessions (ICP 8866, ICP 11289, ICP 11291, and ICPH-2) were selected for a microplot test. These four accessions exhibited some resistance to either or both *M. incognita* race 3 and *M. javanica*. The field microplot test was initiated at the University of Florida Agricultural Research Center, Live Oak, Florida on 25 August 1985. The microplots were 76-cm-diam fiberglass cylinders embedded 50 cm into the soil (9). As in the greenhouse test, the experimental design was an RCB replicated six times with the experimental unit being one microplot. The soil was a Lakeland fine sand (thermic, coated typic quartzipsamments). Four wk

prior to beginning the test, soil in the microplots was treated with 1,3-dichloropropene at the rate of 121 L/ha to reduce plant-parasitic nematode contamination. After 2 wk, soil in the plots was turned to a depth of 23 cm and allowed to aerate for 2 wk to ensure that the fumigant had dissipated completely.

Each plot was infested with an egg + J2 suspension of *M. incognita* race 3, *M. javanica,* or left as a noninoculated control. The source and preparation of inoculum were the same as described for the greenhouse evaluations. Each plot received 100 eggs + J2/100 cm^3 of soil (the soil volume was based on a depth of 23 cm). Inoculum was administered by pouring 1 L of the appropriate suspension of eggs + J2 over the soil surface of the microplot and then mixing the soil with a spade. After the soil was infested, five uniformly spaced holes, 4-cm-deep, were made in each microplot and two seeds were placed into each hole. Two wk after emergence, plants were thinned to five plants per plot. Each microplot received one pigeonpea genotype and one nematode treatment. After 150 days, the plants were cut 2 cm above the soil surface, dried, and weighed. One soil core (2.5 × 30 cm deep) was collected from the root zone of each of the five plants per microplot. Cores from each microplot were bulked; the soil was mixed; and nematodes were extracted from a 250-cm^3 aliquant using centrifugal-flotation (3,8). Three root systems from each microplot were selected at random and rated for root galling as described earlier. Data were analyzed as a factorial arrangement in an RCB design. Treatment variables were blocks, nematode inoculum, and accessions. Response variables analyzed were gall ratings, numbers of J2, and yields of dry matter.

RESULTS AND DISCUSSION

When inoculated with *M. incognita,* 24 of 28 accessions exhibited either minimal galling and egg mass production or no galling and egg mass production (Table 2). Ten of the pigeonpea accessions challenged with *M. javanica* in the greenhouse test were not galled and no egg masses were observed (Table 3). Other accessions inoculated with either *M. incognita* or *M. javanica* exhibited low to very high egg mass and root gall production. Susceptible tomato checks confirmed inoculum viability by their gall rating of 5. Greenhouse results corresponded with those of Sasser and Hartman (13) who previously had screened 15 of the accessions tested in this study.

Some variability occurred between the results of the summer and fall tests. The accessions inoculated with *M. javanica* showed a significant difference in root galling and egg mass production between seasons. In a previous study, 'Kaki' and '2-B-Bushy' were galled severely when infected with *M. javanica* (1). The summer gall scores obtained in this

Table 2. Mean gall scores and standard deviations of fall and summer evaluations of pigeonpea accessions inoculated with *M. incognita* race 3.

Accession	Root index[z]	
	Fall	Summer
ICPL-42	5.0 ± 0.0	5.0 ± 0.0
ICP 8858	4.5 ± 0.8	5.0 ± 0.4
ICP 8859	0.0	0.5 ± 0.8
ICPL-374	0.0	0.0
ICP 11295	0.0	0.5 ± 0.8
2-B-Bushy	0.0	0.0
ICP 8861	0.0	0.0
ICP 11289	0.0	0.0
ICP 8865	0.0	0.0
ICP 8866	0.0	0.0
ICPL-297	0.0	0.0
QPL-46	0.0	0.0
ICP 11291	0.0	0.0
Local	0.0	0.0
ICP 11297	0.0	0.0
Kaki	0.0	0.0
M-59-1	0.0	0.0
SNO-57	0.0	0.0
ICP 8867	0.0	0.0
ICP 8862	0.0	0.0
ICP 8869	0.0	0.0
ICPL-234	0.0	0.0
ICP 10958	0.0	0.0
ICP 8860	0.0	0.0
SNO-44	0.0	0.0
PDM-1	0.0	0.0
ICP 11286	0.0	0.0
ICPH-2	0.0	0.0

[z]Mean of six plants and standard deviation. Root index based on a 0–5 rating where 0 = 0; 1 = 1–2; 2 = 3–10; 3 = 11–30; 4 = 31–100; and 5 = > 100 galls and/or egg masses per root system (14.)

study were very similar to that study, but galling in the fall test was considerably less pronounced. Generally, for all accessions there was more galling and egg mass formation in the summer than in the fall evaluation. Warmer temperatures in the summer probably enhanced nematode reproduction and was the likely cause of seasonal variation (17). The lack of a significant difference between seasons in the *M. incognita* treatment was probably a result of the large percentage of accessions tested that exhibited no galling in either season.

In both fall and summer tests a significant positive correlation was found between the egg mass rating and gall rating ($r = 0.91, P = 0.01$). This correlation indicates that gall rating alone may suffice as a parame-

Table 3. Mean gall scores and standard deviations for fall and summer seedling evaluations of pigeonpea accessions inoculated with *M. javanica*.

Accession	Root index[y,z]	
	Fall	Summer
ICP 8858	3.2 ± 0.4	5.0 ± 0.4
ICP 11295	1.7 ± 0.6	5.0 ± 0.0
ICPL-42	1.0 ± 0.6	4.6 ± 0.8
ICP 8862	0.8 ± 1.5	1.7 ± 0.6
ICPL-374	0.7 ± 0.5	0.0
ICP 11291	0.7 ± 0.5	0.0
2-B-Bushy	0.5 ± 0.8	4.1 ± 1.8
ICP 8866	0.5 ± 0.8	0.0
Kaki	0.5 ± 0.8	3.6 ± 0.7
ICPL-234	0.3 ± 0.5	1.7 ± 1.6
ICP 8867	0.3 ± 0.5	2.3 ± 0.8
ICP 11297	0.2 ± 0.4	0.0
M-59-1	0.2 ± 0.4	0.0
ICP 8860	0.2 ± 0.4	1.0 ± 0.9
ICP 8869	0.2 ± 0.4	0.0
ICP 11289	0.0	3.0 ± 1.3
ICP 8859	0.0	1.0 ± 0.9
ICP 8861	0.0	1.0 ± 1.3
ICP 11286	0.0	0.0
PDM-1	0.0	0.0
ICP 8865	0.0	0.0
ICPH-2	0.0	0.0
ICPL-297	0.0	0.0
QPL-46	0.0	0.0
ICP 1095	0.0	0.0
Local	0.0	0.0
SNO-44	0.0	0.0
SNO-57	0.0	0.0

[y]Mean of six plants and standard deviation.
[z]See Table 2 for explanation of index.

ter for measuring root-knot nematode reproduction. Resistance evaluations based on only gall ratings could be performed more rapidly and more conveniently than evaluations using egg mass ratings or egg counts.

No significant correlation was found either between gall rating and plant height or between egg mass rating and plant height, even though some accessions were severely galled and received the maximum rating of 5.0. This lack of a significant correlation suggests that some level of tolerance may be presnt in these accessions. Similar results were reported by Acosta et al. (1) who found that plant height was not affected by *M. javanica* even though galling was severe. In that study, shoot and

root weights also were not reduced by *M. javanica*. In a separate field study by Brachrein et al. (4), they concluded that only minor yield reductions were caused by root-knot nematodes on 10 different pigeonpea lines, even when average gall ratings were as high as 3.1 and there were 11–30 galls per root system (4).

In the microplot experiment, there was minimal galling and no population density counts were greater than the original inoculum level of 100 eggs + J2/100 cm^3. *F* values from the analysis of variance were not significant either for level of galling due to accessions or for nematode treatments. When population densities were compared there were no significant differences either among accessions or between nematode treatments. These microplot results indicated that the four accessions ICP 8866, ICP 11289, ICP 11291, and ICPH-2 have resistance to the nematode populations tested.

Greenhouse seedling evaluations indicated that pigeonpea accessions tested have varying levels of resistance to *M. incognita* and *M. javanica*. Resistance levels varied from highly resistant to highly susceptible. These pigeonpea accessions showed more resistance to *M. incognita* race 3 than to *M. javanica*. The four selected accessions which exhibited either immunity or a high level of resistance in the greenhouse also were resistant in microplots. Data from these tests indicate that resistance to *M. incognita* and *M. javanica* is related to genotype and resistant pigeonpea genotypes have been identified for use in breeding programs.

LITERATURE CITED

1. ACOSTA, N., N. VICENTE, and J. TORO. 1986. Susceptibility of pigeonpea (*Cajanus cajan*) cultivars and lines to *Meloidogyne javanica*. Nematrópica 16:1–10.
2. AYALA, A. 1962. Occurrence of the nematode *Meloidogyne javanica* on pigeonpea roots in Puerto Rico. Journal of Agriculture of the University of Puerto Rico 46:154–156.
3. AYOUB, S. M. 1980. Plant Nematology: An Agricultural Training Aid. Nema Aid Publications: Sacramento, California, U.S.A.
4. BACHREIN, S., G. M. PRINE, and R. A. DUNN. 1986. Soil and Crop Science Society of Florida Proceedings. 46:137–140.
5. DAS, S. N., C. D. MISHRA, and K. C. MOHANTY. 1979. Occurrence and host preference of some plant-parasitic nematodes of pulse crops. Indian Journal of Nematology 9:74.
6. DICKSON, D. W., and F. B. STRUBLE. 1965. A sieving-staining technique for extraction of egg masses of *Meloidogyne incognita* from soil. Phytopathology 55:497.
7. HUSSEY, R. S., and K. R. BARKER. 1973. A comparison of methods of collecting inocula of *Meloidogyne* spp. including a new technique. Plant Disease Reporter 57:1025–1028.
8. JENKINS, W. R. 1964. A rapid centrifugal-flotation technique for separating nematodes from soil. Plant Disease Reporter 48:692.
9. JOHNSON, J. T., J. R. RICH, and A. W. BOATRIGHT. 1981. A technique for establishing microplots in the field. Journal of Nematology 13:233–235.
10. KILLINGER, G. B. 1968. Pigeon peas (*Cajanus cajan* (L.) Druce), a useful crop for Florida. Soil and Crop Science Society of Florida Proceedings 28:162–167.

11. KRISHNAMURTHY, G. V. G., and N. A. ELIAS. 1968. Additions to host range of *Meloidogyne incognita* causing root-knot on tobacco in Hunsar. Indian Phytopathology 21:351–352.
12. RICH, J. R., C. HODGE, and J. T. JOHNSON. 1984. Population development and pathogenicity of *Meloidogyne javanica* on flue-cured tobacco as influenced by ethoprop and DD. Journal of Nematology 16:240–245.
13. SASSER, J. N., and K. M. HARTMAN. 1985. Evaluation of some pigeonpea lines for resistance to the root-knot nematode, *Meloidogyne* spp. International Pigeonpea Newsletter 4:44–45.
14. SINGH, N. D. 1973. Preliminary report of plant parasitic nematodes associated with important crops in Trinidad. Nematrópica 3:56–61.
15. SUMMERFIELD, G. J., and E. H. ROBERTS. 1985. Grain Legume Crops. William Collins Sons & Co., Ltd.: London.
16. TAYLOR, A. L., J. N. SASSER, and J. N. NELSON. 1982. Relationship of climate and soil characteristics to geographical distribution of *Meloidogyne* species in agricultural soils. Department of Plant Pathology, North Carolina State University and U. S. Agency for International Development: Raleigh, North Carolina, U.S.A.
17. THOMASON, I. J., and B. LEAR. 1961. Rate of reproduction of *Meloidogyne* spp. as influenced by soil temperature. Phytopathology 51:520–524.

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