Root-Knot Nematodes Associated with Beans in Colombia and Peru and Related Yield Loss

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ABSTRACT

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A survey of root-knot nematodes associated with bean (*Phaseolus vulgaris*) roots and soils in the major bean production regions of Colombia and Peru was conducted during 1988–1989. Over 150 samples were processed for these parasites, and specimens from more than 100 of the collections were identified to species. Samples were collected from departments of Valle, Cauca, Nariño, Antioquia, and Tolima of Colombia and from Ica, Lima, Ancash, La Libertad, and Cajamarca of Peru. Only *Meloidogyne incognita* and *M. javanica* were detected in Peru, at frequencies of 66 and 7%, respectively, of the samples collected. In Colombia, *M. incognita, M. javanica, M. hapla*, and *M. arenaria* were detected at frequencies of 62, 21, 14, and 14%, respectively. Mixtures of root-knot species were common in samples from Colombia but rare in samples from Peru. Yield loss due to root-knot nematodes (*M. incognita* and *M. javanica*) was assessed by a field-infestation technique, and comparable plots of the commercial bean cultivar Calima and the CIAT advanced bean line PVA 916 were planted in infested plots and in uninfested plots 15 m from the infested ones. The nematodes induced yield losses of 45–63% to Calima and 26–32% to PVA 916, as determined by dry seed weight. Yield loss due to nematodes was greater with concurrent leafhopper (*Empoasca kraemeri*) damage.

Root-knot nematodes (*Meloidogyne* spp.) are distributed worldwide and have a collective host range that includes nearly all crop plants (11). Yield losses of beans due to these nematodes may reach 60–90% (17,20,21). The extent of damage depends on nematode species and population density, bean cultivar, and environmental conditions.

An investigation of the root-knot problem in any given location must consider the nematode species involved and the population density and must determine the effects of the nematodes on growth and yield of the target host plants. *Meloidogyne* spp. have been studied more than any other plant-parasitic nematode, but as yet only limited information is available concerning their host-parasite relationships. The International Meloidogyne Project (IMP) was established in 1975 to study *Meloidogyne* spp. on a world basis (12). However, it did not specifically address any particular crop species. Dry edible beans (*Phaseolus vulgaris* L.) were included in only a small proportion of the total host samples collected during the IMP. Beans are an extremely important food crop in the developing world, especially in Latin America and in the highlands of eastern and southern Africa.

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In the Andean nations of Colombia and Peru, beans are grown under a wide range of environmental conditions and crop production systems. They are a major protein component in the diets of consumers with low to middle incomes in these countries (9).

Plant diseases are a major factor limiting bean production in the Andean zone countries (16). Root-knot nematodes are among those pathogens causing considerable yield losses to beans in this region. Despite their importance, few studies have focused on the biology and management of *Meloidogyne* spp. infecting beans in these countries.

This investigation was conducted to identify the species of *Meloidogyne* infecting beans within the major production regions of Colombia and Peru and to quantify bean yield losses caused by these nematodes.

MATERIALS AND METHODS

Determination of nematode distribution and identification. Bean root and soil samples were collected from the major bean production regions of Colombia and Peru during 1988 and 1989. Regions sampled in Colombia included the departments of Valle, Cauca, Nariño, Antioquia, and Tolima, which represent a wide range of ecological zones in the country and the predominant bean production systems (Fig. 1A). In Peru, samples were collected from the coastal regions (irrigated desert valleys) in the departments of Ica, Lima, Ancash, and La Libertad (Fig. 1B). Two samples were also collected from beans grown with maize (*Zea mays* L.) in the mountainous department of Cajamarca.

Wherever galled bean roots were encountered, root samples were collected, loose soil was shaken off, and samples were taken to the laboratory in a plastic bag. Galled roots obtained from several locations within a field were pooled as one sample. In some cases, soil samples were collected by pooling four or more subsamples from a field into a single 1-kg (approximately) sample. After this sample was mixed, a 250-cm^3 subsample was mixed with an equal amount of pasteurized greenhouse soil mix or sand. The resulting mixture was then planted with a susceptible host (either bean cv. Canario Divex or tomato [*Lycopersicon esculentum* Miller] cv. Rutgers) in a 10-cm-diameter plastic pot to allow the development of adult females for identification. Perineal patterns from adult females were used as the primary character for species identification (3,18). When available, a minimum of 16 perineal patterns were prepared for the identification of nematode species from each collection.

Quantification of bean yield loss. Two experiments were conducted in a field at the CIAT-Palmira Station, in the Cauca Valley near Cali, Colombia, from April 1989 through April 1990. The field had been left fallow for two growing seasons without a weed control program. Prior to the two fallow seasons, beans had been planted in this field continuously for several years (two crops per calendar year). Soil in this field was a silty clay, with a pH of 7.3 and an organic matter content of 4.3%.

The field was infested artificially with nematodes. Three-week-old tomato seedlings (cv. Rutgers) grown in 5-cm-diameter peat pots were inoculated with a concentrated egg suspension of a mixed population of *Meloidogyne incognita* (Kofoid & White) Chitwood and *M. javanica* (Treub) Chitwood that had been collected from naturally infected beans in a nearby field in Candelaria, Valle Department, Colombia. After 2–3 wk, inoculated tomatoes were transplanted into raised beds in the experimental field. Peat pots were partially broken away and a complete fertilizer (10-30-10, 100 kg/ha) was applied at time of planting. Plant

spacing was 60 cm within and across rows. A second planting of infected tomato seedlings several weeks later doubled the length of the area being infested. A third planting 1 mo later replaced the first plantings and filled in the spaces where tomato plants had deteriorated. A border of crotalaria (*Crotalaria spectabilis* Roth) resistant to root-knot nematodes (10,14) was planted on three sides of the infested site as a living nematicidal barrier to reduce the likelihood of nematode spread; the fourth side was left open to allow entry to the field and in-furrow irrigation. To prevent nematode spread, irrigation was limited to the infested portion only and was prevented from progressing to the crotalaria border or beyond by mounding soil across irrigation furrows. Two months later, the shoot portions of the tomato plants were removed with a machete and the root systems left intact. The field was then carefully cultivated so that the infested, raised rows were maintained in situ. To increase nematode density and uniformity, CIAT advanced bean lines PVA 916 and PVA 476, susceptible to *Meloidogyne* spp., were planted into the rows (25–30 seeds per 1-m row) and allowed to grow for 6 wk before the shoot portions were removed. At this time the field was considered ready for use in conducting experiments to quantify yield loss of beans due to nematodes.

Initial nematode population density was assessed in this field from samples collected before and after the infestation. Average nematode numbers per plot were determined by the sugar flotation-centrifugation technique (2). No root-knot nematodes were detected in this field prior to infestation. At the time of planting for the first trial, soil samples were collected and nematode numbers were estimated by direct (flotation-centrifugation) and indirect (tomato bioassay) procedures. In the bioassay, 250 cm^3 of field soil was mixed with an equal amount of pasteurized sand and placed in a 10-cm-diameter pot with a tomato seedling. These seedlings were evaluated 6 wk later for severity of root galling and nematode egg mass production. Root galling severity was assessed on a 1–9 scale by estimating proportion of roots galled: 1 = no galling, 2 = ≤5% roots galled, 3 = 6–10%, 4 = 11–18%, 5 = 19–25%, 6 = 26–50%, 7 = 51–65%, 8 = 66–75%, and 9 = 76–100% roots galled (15). Egg mass production was also assessed on a 1–9 scale: 1 = no egg masses detected, 2 = 1 or 2 egg masses, 3 = 3–6, 4 = 7–10, 5 = 11–20, 6 = 21–30, 7 = 31–60, 8 = 61–100, and 9 = over 100 egg masses produced per plant (expanded from Sasser et al [13]).

In the first trial, the local bean cultivar Calima was established in plots of four rows, 4 m long, 15 seeds per meter, with four replications. Two buffer rows, also of Calima, were planted around the plots. Treatments consisted of plots planted in the infested site and in the uninfested site beyond the crotalaria border (about 15 m from infested treatment plots). In the second trial, Calima and the CIAT advanced bean line PVA 916 were planted in plots of four or 10 rows, 4 m long, with four replications, in infested and uninfested sites. Seeds were pretreated with a slurry of carbaryl and carboxin, using methylcellulose as a sticking agent, and a complete fertilizer (10-30-10, 100 kg/ha) was applied at the time of planting. Seeds were hand-planted, and a mixture of linuron (1 kg/ha) and pendimethalin (2 L/ha) was applied after seeding to all rows and alleys as well as to the plot perimeter. Beans were protected against foliar diseases and insects according to commercial recommendations, beginning 2 wk after germination. However, an early infestation of leafhopper (*Empoasca kraemeri* Ross and Moore) severely damaged young bean plants in the first trial, equally in nematode-infested and uninfested treatments. Root galling was rated at harvest by digging all plants and estimating average galling per replicate in the first trial and by excavating one plant per row 2 wk before harvest and evaluating mean plot galling in the second trial. Total weight of bean seeds harvested per plot was taken as yield, adjusted for moisture content, and expressed in kilograms per hectare. Treatment means were separated by Student's *t* test.

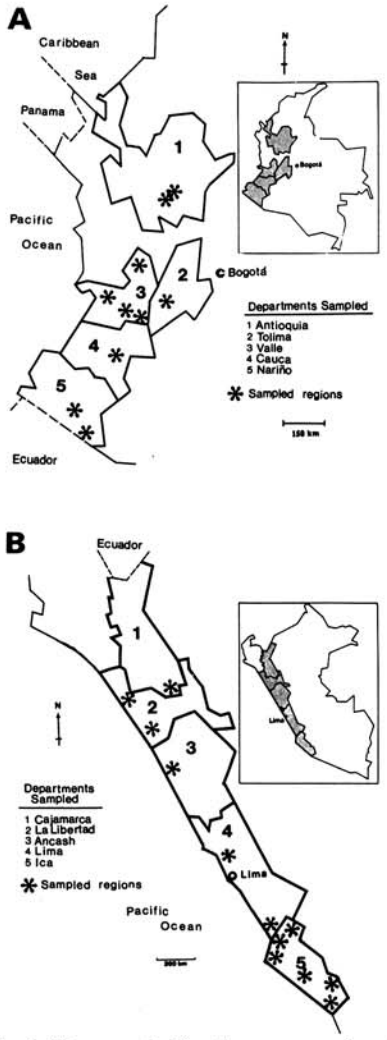

Fig. 1. Sites sampled for the presence of root-knot nematodes in **(A)** Colombia and **(B)** Peru.

RESULTS

Determination of nematode distribution and identification. Over 150 samples were collected from the bean production areas in Colombia and Peru (24 collec-

Table 1. Distribution and frequency of root-knot nematodes (*Meloidogyne* spp.) associated with *Phaseolus vulgaris* in Colombia and Peru

Location and elevation (m)	Number of collections	*Meloidogyne* species[a]					
		Mi	Mj	Mh	Ma	Mixture	Sp.
Colombia							
Valle, 900–1,000	2	0	0	0	0	2A	0
1,000–1,800	19	5	1	2	0	1B,2C,2D	6
Cauca, 1,800–1,850	4	1	0	1	0	1B	1
Nariño, 2,450–2,850	3	2	0	0	0	1A	0
Antioquia, 2,000–2,300	11	3	1	0	0	2A,1B,2C	3
Tolima, 1,000	2	1	0	0	0	0	1
Total	41	12	2	3	0	5A,3B,4C,2D	11
Peru							
Ica, 50–200	42	26	0	0	0	2A	13
Lima, 50–500	14	9	0	0	0	0	5
Ancash, 50	1	1	0	0	0	0	0
La Libertad 50–500	3	1	1	0	0	1A	0
Cajamarca, 2,200	2	0	0	0	0	0	2
Total	62	37	1	0	0	3A	20
Combined totals	103	49	3	3	0	8A,3B,4C,2D	31

[a] Mi = *M. incognita*, Mj = *M. javanica*, Mh = *M. hapla*, Ma = *M. arenaria*, Sp. = identified to genus only. Mixtures: A = *M. incognita* + *M. javanica*, B = *M. incognita* + *M. hapla*, C = *M. incognita* + *M. arenaria*, D = *M. incognita* + *M. javanica* + *M. arenaria*.

tions were from beans intercropped with maize) and assayed for *Meloidogyne* spp. Of these, 103 contained root-knot nematodes, which were identified to species in 72 samples. Two species of root-knot nematodes were identified from Peru: *M. incognita* (66% incidence) and *M. javanica* (7% incidence) (Table 1). Most of the samples of *M. javanica* were mixed with *M. incognita* (5% of Peru samples).

Because of lack of appropriate life stages or of nematode mortality in the collected samples, some collections could not be identified beyond the level of genus. Details of sample location, nematode identification, and additional host species as well as descriptions of the departments sampled are summarized elsewhere (6).

Species composition of nematode collections from Colombia was more variable than that of collections from Peru. *M. incognita, M. javanica, M. hapla* Chitwood, and *M. arenaria* (Neal) Chitwood were detected in 62, 21, 14, and 14%, respectively, of the samples processed. In contrast to collections from Peru, a large proportion (33%) of the Colombian collections consisted of mixtures of two or three species. In all such cases, one of the species was *M. incognita.*

M. incognita not only was the most common species associated with beans in Colombia and Peru but also was the most widely distributed. All departments from which samples were collected had *M. incognita*, regardless of elevation, crop production system, or environment. *M. javanica* also was widely distributed in Colombia, although its frequency was lower than that of *M. incognita*. In Peru, *M. javanica* was detected only in the coastal departments of Ica and La Libertad, and the incidence of recovery was low (7%). *M. hapla* was observed on beans only in Colombia and exclusively at the higher elevations where cooler temperatures (average of 18 C) prevailed, such as in the mountainous portions of Valle, Cauca, and Antioquia. One-half of the *M. hapla* collections were mixed with *M. incognita. M. arenaria* was encountered only in mixtures with *M. incognita* or with both *M. incognita* and *M. javanica*, and only in a few locations in Colombia.

Quantification of yield loss. *Meloidogyne* spp. had a significant effect on bean yield in both trials, and yield loss was greater in the first trial when beans were subjected to the additional stress of leafhopper damage (Table 2). Bean yield losses ranged from 26 to 63%, depending on the trial and the bean cultivar. Plants grown in the infested plots showed heavy galling (Fig. 2). The yield of beans grown in the uninfested plots was approximately 400 kg/ha, whereas yield of beans grown in the nematode-infested plots averaged 145 kg/ha. The very low overall yield of beans in this trial was attributed to early infestation by *Empoasca*, but the nematode-induced yield loss was substantial and highly significant ($P < 0.01$). In the second trial, per-hectare yield values for the four-row and 10-row plots did not differ ($P = 0.05$). The bean cultivar Calima showed a 42–45% yield loss, whereas the improved bean line PVA 916 was apparently more tolerant, showing only a 26–32% yield loss. No root galling was detected on bean plants harvested from the uninfested plots (Table 3). Mean root galling of beans in the nematode-infested site was moderate in the first trial but relatively low in the second trial (average galling ratings of 5.2 and 3.2, respectively). The low root galling rating in the second trial may have been due to breakage of galled portions of the roots

Table 2. Influence of root-knot nematodes on yield of beans (local cultivar Calima and CIAT line PVA 916) at CIAT-Palmira, Colombia[a]

	Dry seed weight (kg/ha)				
	Trial 1	Trial 2			
	Calima	Calima		PVA 916	
Plots	4-row[b]	4-row	10-row	4-row	10-row
Infested[c]	147	1,054	1,018	1,462	1,400
Uninfested	398	1,820	1,840	1,979	2,051
	**[d]	**	**	**	**
Loss (%)	63	42	45	26	32

[a]Trial 1 was conducted from 24 November 1989 to 25 January 1990, and trial 2 from 1 February to 24 April 1990.
[b]Number of rows per replicate per treatment.
[c]Plots were infested by transplantation of tomato seedlings infected with *Meloidogyne incognita* and *M. javanica.*
[d]Highly significant ($P < 0.01$) effect of infestation on yield.

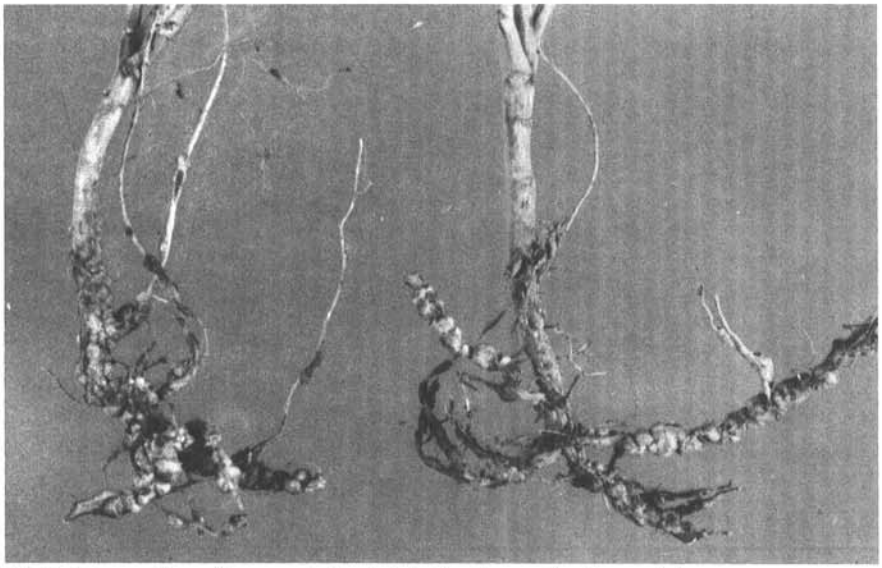

Fig. 2. Heavy galling of bean roots due to infection by root-knot nematodes.

Table 3. Estimation of root-knot nematode population density and damage caused to beans in artificially infested plots at CIAT-Palmira, Colombia

	Trial 1				Trial 2
	Preplant[a]	Bioassay[b]		Field[c]	Field
Plots	J2	RGR	EMR	RGR	RGR
Infested	50	7	8	5	3
Uninfested	<1	1	1	1	1
	**[d]	**	**	**	**

[a]Number of second-stage juveniles (J2) of *Meloidogyne* spp. extracted from soil before establishment of trial 1.
[b]Root galling rating (RGR) and egg mass rating (EMR) on a scale of 1–9 (1 = no galls or no egg masses, 9 = 76–100% roots galled or >100 egg masses), evaluated 7 wk after planting tomato into soil samples collected from infested and uninfested plots before trial 1.
[c]RGR of beans planted in infested and uninfested field plots during trial 1 (evaluated at harvest) and trial 2 (evaluated 2 wk before harvest).
[d]Highly significant ($P < 0.01$) effect of infestation on rating.

from harvested plants as a result of the compacted condition of the soil.

DISCUSSION

This survey clearly illustrated the importance of *Meloidogyne* spp. on beans in Colombia and Peru with respect to their distribution and damage. In each of the sampled bean production zones of both countries, root-knot nematodes were present and infected bean roots. The nematode often caused substantial root galling, which reflected the high population densities of the pathogens. We strongly recommend that bean breeders working to incorporate root-knot nematode resistance in beans for Colombia and Peru include resistance to *M. incognita* and *M. javanica* in all developed bean germ plasm. One-fourth to one-fifth of Colombian root-knot nematode collections from beans contained *M. javanica*, usually in mixtures with *M. incognita*. Likewise, in collections from Peru, *M. javanica* was mainly found mixed with *M. incognita*.

Beans with resistance to *M. incognita* and grown in the Trujillo region of Peru have shown heavy root galling, probably caused by *M. javanica* (M. A. Pastor-Corrales, *unpublished*). Accordingly, resistance to *M. incognita* alone is not likely to be sufficient to protect beans against root-knot nematodes in many of the bean production regions in these two countries. Incorporation of resistance to *M. hapla* or *M. arenaria* in improved and adapted bean cultivars for Colombia or Peru is probably not justified at this time but may be necessary in the future. Approximately 200 pure lines and accessions have been evaluated for resistance to selected Colombian populations of root-knot nematodes containing *M. incognita* alone or mixed with *M. javanica*, and resistance in beans of a variety of seed types has been identified (6–8).

M. incognita is considered to be the most common species of root-knot nematode on a world basis (19). We concur in that *M. incognita* was identified in the majority of nematode collections obtained from the bean-growing regions of Colombia and Peru. *M. incognita* and *M. javanica* were detected in approximately 47 and 40% of root-knot nematode collections, respectively, obtained from a wide range of crops worldwide (19). In our investigation, *M. incognita* was recovered at a higher, and *M. javanica* at a lower, frequency from Colombia and Peru than in the IMP study worldwide. This suggests that *P. vulgaris* might be a more efficient and susceptible host for *M. incognita* and possibly a less suitable host for *M. javanica* than the average of crops sampled worldwide in the IMP study. Similar sample frequencies of *M. incognita, M. javanica*, and *M. arenaria* were detected on tomato planted after sugarcane cultivation in Hawaii (14).

Species diversity was also greater on beans sampled in Colombia than on those sampled in Peru. The sampled regions of Peru were relatively uniform, i.e., the coastal irrigated desert, which experiences no rain and relatively little cropping diversity (monocropped in rotation with cotton or sweet potato). In contrast, the sampled regions of Colombia were ecologically diverse, with wide variations in altitude, temperature, precipitation, soil types, cropping systems, and crop rotations (6). The uniformity of coastal Peru may render it an environment more suitable for *M. incognita* than for *M. javanica*. The variation in sample location characteristics and/or host crop species is probably related to the variation in nematode species. In surveys of other host crops in Peru, additional species of *Meloidogyne* have been detected (1,4,5).

Bean yield was substantially reduced by root-knot nematodes in the artificially infested plots. These data confirm the few reports in which root-knot nematodes have been experimentally shown to cause yield loss in beans (17,20). Depending on bean cultivar and additional factors, bean seed yield was reduced by 26–63%. Where poor farmers depend on their crop harvest for subsistence, as in much of the Andes, this level of yield loss may be devastating. As cropping practices in this region frequently favor the increase of soilborne pathogens, bean production in these regions is likely to continue to be affected unless active measures are taken to reduce the impact of these pathogens on beans.

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