PLANT-PARASITIC NEMATODES ASSOCIATED WITH VEGETABLE CROPS IN BENIN: RELATIONSHIP WITH SOIL PHYSICO-CHEMICAL PROPERTIES

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Summary. Surveys were conducted in vegetable-producing communes of Benin between May 2004 and November 2005 to study the diversity and incidence of plant-parasitic nematodes affecting these crops and to assess the effects of soil physico-chemical properties on the nematodes. A total of 171 plant root and 171 soil samples collected from 26 vegetable crops were examined. Eleven species of plant-parasitic nematodes were recorded, of which six are reported for the first time in the country. When analyzed by crops sampled, communes surveyed and agro-ecological zones, the most frequently observed and widely distributed nematode species were root-knot nematodes, *Meloidogyne* spp., followed by *Helicotylenchus dihystera* and *Scutellonema clathricaudatum*. These nematodes infected 96.1, 76.9 and 53.9% of the crops sampled, respectively, and were observed on vegetable crops in 100, 94.4 and 44.4% of surveyed communes, respectively. Mean population density of *Meloidogyne* spp. (juveniles and males) was greater than that of the other nematode species. Prevalence, relative abundance and mean intensity were also higher for *Meloidogyne* spp. (79, 62.1 and 99.3%, respectively) than for other nematode species. The largest population densities of *Meloidogyne* spp. were observed in celosia, cucumber, green pepper, carrot, the African garden eggplant, okra, basil, and crin-crin (West African sorrel) (174, 103, 101, 98, 96, 96, 91, 79 nematodes/g sample, respectively). Correlation analysis indicated a weak relationship between soil physico-chemical properties and nematode population density (maximum value of the coefficient of correlation r = 0.41). The results indicate that, apart from the direct influence of the host plant, soil properties play an important role in the abundance, distribution and structure of plant parasitic nematode communities. This validates the potential of nematodes as bio-indicator organisms of soil health.

Keywords: Diversity, incidence, prevalence, relative abundance, survey.

A wide range of local and introduced leaf, fruit and root vegetables and pulses are grown throughout the world. Vegetable production and consumption have expanded rapidly in the past two decades in most areas of the world, with production significantly outpacing population growth since 1990. Surprisingly, the total production has actually decreased slightly since 1990 (Sikora and Fernandez, 2005), which is speculated to be due to a number of constraints that hamper vegetable production, with nematodes, especially root-knot nematodes, *Meloidogyne* spp., amongst the most important (Netscher and Sikora, 1990). Plant-parasitic nematodes (PPN) are an extremely important limiting factor in vegetable production, and in many areas a major factor requiring extensive use of pesticides. In the urban and peri-urban (UPU) areas of southern Benin, approximately nineteen species of indigenous tropical and exotic leafy vegetables are grown (James *et al.*, 2006), but the only existing information regarding the diversity and distribution of PPN on vegetable crops is limited (Dodego, 1979; Sikora, 1991). However, the studies of these authors covered only a few vegetable-producing sites and crops, indicating the lack of countrywide information on nematodes affecting vegetable crops in Benin.

The role nematodes play in limiting vegetable production depends to a large extent on the farming system employed (Taylor, 1976), on the physical and/or chemical environment in the soil (Quénéhervé, 1988; Kandji *et al.*, 2001), and on climate (Curran *et al.*, 1986). Popovici and Ciobanu (2000) reported that the strength of the relationship between nematode community and soil type varies with the nematode species and that nematode reproduction is positively correlated with relative humidity and negatively correlated with air temperature. Also, they observed that soil texture is the most important factor explaining the presence of some nematode species and that environmental factors affect the ability of nematodes to parasitize and reproduce on their host.

In Benin, vegetable crops are cultivated in the UPU areas of the country, which are characterised by a wide diversity of soil types (mineral, ferruginous, ferralitic, hydromorphic, "vertisols"), agro-ecological zones (humid forest, Guinea savannah, sub-humid savannah and Sudan savannah), climates (beninese, sub-sudanian, tropical sudanian, atacorian) and vegetation (fallow, mangrove swamp) (Adam and Boko, 1993). But no data

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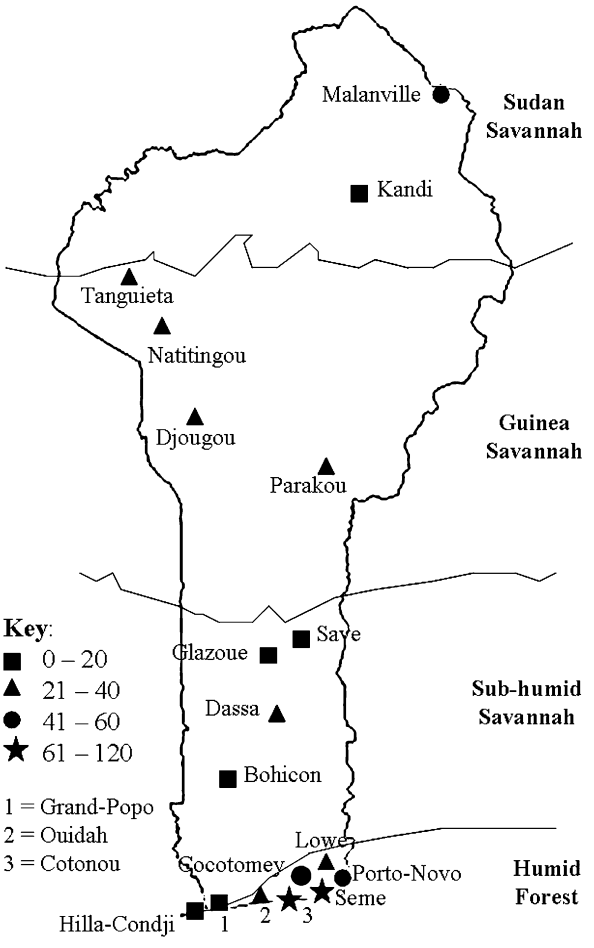

Fig. 1. Benin map showing vegetable production communes surveyed during May 2004 and November 2005 and *Meloidogyne* spp. population density/g of sample at each commune.

were available on the relationship between PPN on vegetable crops and soil physico-chemical properties. A countrywide survey is, therefore, necessary to establish correctly the diversity and distribution of nematodes affecting vegetable crops in order to develop adequate management options. Consequently, the objectives of the current study were to *i*) determine the diversity and incidence of potentially harmful PPN on vegetable crops by means of surveys of areas of current and potential vegetable production in Benin (James *et al.*, 2006), and *ii*) to monitor the effect of soil physico-chemical properties on the community structure of PPN assemblages. The first of the objectives is an addition to information provided by Dodego (1979) and Sikora (1991) on PPN associated with vegetable crops produced in Benin.

MATERIAL AND METHODS

Sampling sites and procedure. The surveys were undertaken in eighteen communes (Table III) representative of intensive vegetable production areas, between May 2004 and November 2005. The surveyed vegetable sites were located in the UPU areas of those communes, which were located in four agro-ecological zones (AEZ) (Table III and Fig. 1). These sites were selected on the basis of the importance of production, variability of vegetable crops produced and geographic distribution. The geographic coordinates of each of the sites were recorded using a Global Positioning System (GPS) and used to plot the map presented in Fig. 1. A total of 171 soil and 171 plant root samples were collected from 26 different vegetable crops. At each vegetable site, for a selected crop, three beds were randomly selected and five plants sampled randomly per bed by uprooting plants using a garden trowel (Coyne *et al.*, 2007). Samples were bulked per bed for roots and soil (at least 1 kg per bed) and stored in plastic bags until extraction of the nematodes and soil physico-chemical analyses.

Extraction and preservation of nematodes. Nematodes were extracted separately from roots and soil for each of the samples collected. The root systems of each sample were washed free of soil, chopped into pieces (*ca.* 0.5 cm) and nematodes extracted from a 5-g sub-sample using a modified Baermann method for 48 h (Coyne *et al.*, 2007). Nematodes were also extracted from 50 g soil per sample using the same extraction method. The nematode suspensions were collected in beakers, allowed to settle for two hours and the supernatant poured off. The final volume, which was *ca.* 1-1.5 ml for each suspension, was poured into a 7 ml vial and hot (70 °C) formalin (4%) added. The vials were maintained in a fridge at 4 °C until nematodes were identified and their population densities assessed.

Assessment of population density and identification of nematodes. Nematodes were concurrently counted and identified at the Biosystematic Division of the Agricultural Research Center, Plant Protection Research Institute (ARC/PPRI), Pretoria, South Africa. Nematode suspensions were allowed to settle for two hours in the vials and the volume of the supernatant reduced to 1-2 ml as described earlier; tap water was then added to obtain a final volume of 5 ml. Nematode population density was assessed in three 1 ml aliquots. Nematodes were counted and recorded separately by genus/species in a counting dish using a Leica Wild M3C stereomicroscope. Nematode population densities from the roots and the soil were transformed to their equivalents in 1 g roots and 1 g soil. Whenever a nematode could not be identified in the counting dish, it was mounted on a slide and identified using a Wild Leitz AG microscope. Nematodes were identified to species level based on their morphological characters by Dr. Esther van der Berg at ARC/PPRI.

Occurrence and incidence of plant-parasitic nematodes. Nematode prevalence, relative abundance and mean intensity were calculated according to Boag (1993):

Table I. Population density of plant-parasitic nematode genera detected in vegetable crops in urban and peri-urban areas of Benin, between May 2004 and November 2005.

Crop	Number of samples[a]	Number of nematode genera[b]	*Aphelenchoides*	*Ditylenchus*	*Helicotylenchus*	*Hemicriconemoides*	*Meloidogyne*	*Pratylenchus*	*Rotylenculus*	*Scutellonema*	*Quinisulcius*	*Tylenchulus*	*Xiphinema*
Akaya (*Cleome gynandra* L.)	1	2	0	0	0	0	22	0	0	1	0	0	0
Amaranth (*Amaranthus* spp.)	11	6	*0*[f]	0	15	1	9	2	0	0	0	7	0
Aubergine (*Solanum melongena* L.)	6	5	*0*	0	9	*0*	39	0	*0*	0	0	0	0
Basil (*Ocimum basilicum* L.)	1	3	0	0	1	0	91	0	0	0	0	6	0
Bell pepper (*Capsicum annuum* L)	3	2	0	0	0	0	9	0	0	1	0	0	0
Cabbage (*Brassica* sp.)	8	6	1	0	7	0	18	1	0	1	0	*0*	0
Carrot (*Daucus carota* L.)	15	6	0	*0*	7	0	99	0	*0*	0	5	*0*	0
Celosia (*Celosia argenta* L.)	10	5	6	0	33	0	174	4	0	0	0	0	1
Courgette (*Cucurbita pepo* L.)	2	3	0	0	1	0	13	0	0	1	0	0	0
Crin-crin (*Corchorus olitorius* L.)	7	5	0	0	6	4	79	*0*	0	2	0	0	0
Cucumber (*Cucumis sativus* L.)	9	4	*0*	0	13	0	103	0	0	0	1	0	0
Egusi (*Citrullus lanatus* (Thumb.) Matsum. et Nakai	1	1	0	0	0	0	4	0	0	0	0	0	0
Green bean (*Phaseolus vulgaris* L.)	2	4	0	0	5	*0*	3	0	0	3	0	0	0
Green pepper (*Capsicum annum* L.)	5	6	1	0	5	0	101	0	4	1	*0*	0	0
Hot pepper (*Capsicum fructescens* L.)	10	7	*0*	0	17	1	17	2	1	1	0	0	0
Lettuce (*Lactuca sativa* L.)	15	6	0	0	5	0	18	2	1	*0*	*0*	0	0
Melon (*Cucumis melo* L.)	1	3	0	0	3	0	28	0	0	10	0	0	0
Mint (*Salvia dorrii* (Kellogg) Abrams)	2	3	0	0	6	0	36	77	0	0	0	0	0
Eggplant[c] (*Solanum macrocarpon* L.)	31	6	0	0	6	0	96	*0*	1	*0*	*0*	0	0
Okra (*Abelmoschus esculentus* (L.) Moench)	6	5	*0*	0	12	0	96	1	0	4	0	0	0
Onion (*Alliup* spp.)	2	0	0	0	0	0	0	0	0	0	0	0	0
Parsley (*Petroselinum sativum* M.)	2	2	0	0	1	0	33	0	0	0	0	0	0
Pumpkin (*Cucurbita maxima* D.)	1	3	0	0	2	0	3	0	0	1	0	0	0
Tchiayo (*Occimum gratissimum* L.)	2	1	0	0	0	0	18	0	0	0	0	0	0
Tomato (*Solanum lycopersicum* L.)	14	9	2	*0*	13	1	45	3	1	3	1	0	0
Vernonia (*Vernonia amygdalina* D.)	4	2	0	0	2	0	3	0	0	0	0	0	0
Mean[d]			0.37 c	0.00 c	6.26 b	0.26 c	44.5 a	3.41 b	0.30 c	1.07 b	0.26 c	0.48 c	0.04 c
Percentage infected crops (%)[e]			19.23	7.69	76.92	23.08	96.15	34.62	23.08	53.85	23.08	7.69	7.69

[a]Number of root or soil samples collected during the surveys. [b]Number of nematode genera affecting each crop. [c]The African garden eggplant. [d]Each nematode population density (untransformed data) represents rounded value of [(nematode population density per g roots + nematode population density per g soil)/2]. [e]Number of crops infected by a particular nematode species divided by total number of crops sampled (26), expressed as a percentage. [f]Values in italics represent mean nematode population densities inferior to 0.5 and indicate the presence of nematodes in the samples. Statistical analysis and mean separations were undertaken on $\log_{10}$ (x+1) transformed data. Total number of roots or soil samples equals 171, collected from 18 intensive vegetable-producing communes in Benin.

Table II. Host susceptibility of seven vegetable crops sampled in previous studies by Dodego (1979) and Sikora (1991) in Benin to the six new nematode records observed during surveys in urban and peri-urban areas of eighteen vegetable producing communes of Benin between May 2004 and November 2005.

Crop	*Aphelenchoides*	*Ditylenchus*	*Hemicriconemoides*	*Scutellonema*	*Quinisulcius*	*Xiphinema*
Aubergine (*Solanum melongena* L.)	-	-	+	-	-	-
Carrot (*Daucus carota* L.)	-	+	-	-	+	+
Celosia (*Celosia argentea* L.)	+	-	-	-	-	+
Crin-crin (*Corchorus olitorius* L.)	-	-	+	+	-	-
Lettuce (*Lactuca sativa* L.)	-	-	-	+	+	-
Okra (*Abelmoschus esculentus* (L.) Moench)	+	-	-	+	-	-
Tomato (*Solanum lycopersicum* L.)	+	+	+	+	+	-
Percentage infected crops (%)	*42.9*	*28.6*	*42.9*	*57.1*	*42.9*	*28.6*

"-" indicates that the crop is not susceptible to the particular nematode genus,
"+" indicates that the crop is susceptible to the particular nematode genus

Table III. Identity and population density of plant-parasitic nematode genera detected in vegetable crops in eighteen vegetable-producing communes of Benin between May 2004 and November 2005.

Commune	Agro-ecological zone[a]	Number of samples[b]	Number of nematode genera[c]	*Aphelen-choides*	*Ditylen-chus*	*Helico-tylenchus*	*Hemicrico-nemoides*	*Meloido-gyne*	*Praty-lenchus*	*Rotylen-culus*	*Scutel-lonema*	*Quinisu lcius*	*Tylen-chulus*	*Xiphi-nema*
Bohicon	SHS	5	4	0[d]	0	7	0	7	0	0	1	1	0	0
Cocotomey	HF	8	6	*0*[f]	0	8	3	121	*0*	1	0	0	0	0
Cotonou	HF	40	11	1	*0*	11	*0*	103	5	1	1	1	2	*0*
Dassa	SHS	6	*6*	*0*	0	9	0	34	*0*	*0*	6	0	0	0
Djougou	GS	13	*8*	*0*	0	10	*0*	34	1	*0*	1	0	*0*	0
Grand-Popo	HF	7	3	0	0	2	0	17	*0*	0	0	0	0	0
Glazoue	SHS	2	4	0	0	11	1	3	3	0	0	0	0	0
Hilla-Condji	HF	3	2	0	0	2	0	1	0	0	0	0	0	0
Kandi	SS	9	6	1	0	3	*0*	14	4	*0*	0	0	0	0
Lowe	HF	2	2	0	0	8	0	39	0	0	0	0	0	0
Malanville	SS	8	8	1	*0*	2	0	55	*0*	*0*	1	1	0	0
Natitingou	GS	15	4	0	0	4	0	39	0	0	2	*0*	0	0
Ouidah	SHS	2	2	0	0	2	0	26	0	0	0	0	0	0
Parakou	GS	13	7	0	0	20	0	44	1	3	*0*	2	0	*0*
Porto-Novo	HF	20	8	1	0	12	0	49	2	*0*	1	*0*	0	*0*
Save	SHS	7	4	0	0	17	*0*	19	0	0	0	0	*0*	0
Seme	HF	10	4	0	0	9	0	82	1	0	0	1	0	0
Tanguieta	GS	1	1	0	0	0	0	42	0	0	0	0	0	0
Mean				0.27 c	0.01 c	7.63 b	0.29 c	43.87 a	1.00 c	0.31 c	0.67 c	0.35 c	*0.15 c*	*0.02 c*
Percentage infested communes (%)[e]				38.89	11.11	94.44	33.33	100.00	50.00	44.44	44.44	38.87	*16.67*	*16.67*

[a]SHS = Sub-Humid Savannah, HF = Humid Forest, GS = Guinea Savannah, SS = Sudan Savannah; [b]Number of soil or root samples collected; [c]Number of nematode genera affecting each crop; [d]Each nematode population density (untransformed data) represents rounded value of [(nematode population density per 1g roots + nematode population density per 1g soil)/2]. [e]Number of crops infected by a particular nematode species divided by total number of crops sampled (26), expressed as a percentage. [f]Values in italics represent mean nematode population densities inferior to 0.5 and indicate the presence of nematodes in the samples. Statistical analysis and mean separations were undertaken on $\log_{10}$ (x+1) transformed data. Total number of roots or soil samples equals 171, collected on 26 vegetable crops.

– Prevalence = (number of samples having a particular nematode species)/(number of samples examined) x 100.
– Relative abundance = total number of individuals of a particular species per g soil and root sample in all the samples/number of samples including those with zero counts for that species.
– Mean intensity = number of individuals of a particular nematode species per g soil and roots in the positive samples/number of positive samples.

Physico-chemical properties of soils under vegetable production in surveyed sites. Soil samples were collected from eighteen communes during the surveys but, due to the insufficient weight of samples from four communes, only 92 soil samples collected from fourteen communes (Table VI) were used for assessment of their texture (clay, loam, sand) and chemical properties (organic matter, N, C/N, pH_{H2O}, exchangeable Ca^{++}, exchangeable Mg^{++}, exchangeable K^{+}, exchangeable Na^{+}, S, exchangeable cation capacity (ECC) and P_{total}). Additionally, a granulometric analysis was undertaken on the soil samples and the soil particle sizes: 0-2 µm (clay), 2-50 µm (loam) and 50-2000 µm (sand). Physico-chemical analyses were undertaken by the "Laboratoire des Sciences du Sol, Eaux et Environnement" of the Benin National Agricultural Research Institute (INRAB), Agonkanmey Station, and the relationships with nematode population density assessed.

Statistical analyses. Nematode population densities in roots and soil were normalized by transforming them to $\log_{10}(x+1)$ before they were subjected to analysis of the variance (Gomez and Gomez, 1984). For each sample, nematode population densities were assessed separately from roots and soil. However, mean nematode population densities calculated as (nematode population density per 1 g root + nematode population density per 1 g soil)/2 were considered. This was done for easier presentation of the results as all the nematodes identified (except *Xiphinema* spp.) are endoparasitic and are found in both soil and roots. For *Xiphinema* spp., which is an ectoparasite, only soil population densities were considered. Means were compared by Fisher's Protected Least Significant Difference Test (LSD), $P \leq 0.05$ using the SAS programme (SAS Institute, Inc., Cary, NC, USA. Version 9.1 for Windows 2003). The relationships between nematode diversity and population density and soil physico-chemical properties were assessed by correlation analysis using SAS.

RESULTS

Assessment of population density and identification of nematodes. The number of samples collected per crop during the surveys varied depending on crop (Table I). Plant-parasitic nematodes of eleven genera were found associated with vegetable crops in Benin: *Aphelenchoides bicaudatus* Filipjev *et* Schuurmans Stekhoven, *Ditylenchus* sp., *Helicotylenchus dihystera* Sher, *Hemicriconemoides strictathecatus* Esser, *Meloidogyne* spp., *Pratylenchus coffeae* Sher *et* Allen, *Rotylenchulus reniformis* Linford *et* Oliveira, *Scutellonema clathricaudatum* Whitehead, *Quinisulcius capitatus* (Allen) Siddiqi, *Tylenchulus* sp., *Xiphinema* sp. (Tables I, III, IV, V).

Meloidogyne spp. were the most important in terms of population density (mean of 45 nematodes/g across crops), followed by *H. dihystera* (6 nematodes/g), *P. coffeae* (3 nematodes/g) and *S. clathricaudatum* (1 nematode/g). Across crops, population densities of these four nematode species were larger ($P \leq 0.05$) than those of other nematode species, for which less than one nematode was extracted per g of sample (Table I). The largest population densities of *Meloidogyne* spp. were observed on celosia, cucumber, green pepper, carrot, the African garden eggplant, okra, basil, and crin-crin (Table I). Among the surveyed crops, onion was the only one not infested by *Meloidogyne* spp. *Helicotylenchus dihystera*, *S. clathricaudatum* and *P. coffeae* were associated, respectively, with 76.9%, 53.8% and 34.6% of the crops. Species of *Tylenchulus*, *Xiphinema* and *Ditylenchus* were recovered from two crops each, i.e. 7.7% of total sampled crops. With the exception of egusi, onion and tchiayo, all crops were hosts of mixed populations of nematodes with nine species recovered from tomato and seven from hot pepper. Species of *Aphelenchoides*, *Ditylenchus*, *Hemicriconemoides*, *Scutellonema, Quinisulcius* and *Xiphinema* represent new records for the seven vegetable crops previously sampled in Benin by Dodego (1979) and Sikora (1991). The host susceptibility to these newly recorded nematodes depends on the nematode species, and percentage of host crops infested ranged from 28.6% to 57.1% (Table II).

Nematode species and population density varied between the communes. The largest nematode population density ($P < 0.05$) observed was of *Meloidogyne* spp. (44 nematodes/g), followed by *H. dihystera* (8 nematodes/g). The largest population density of *Meloidogyne* spp. was observed on samples collected at Cocotomey followed by Cotonou and Seme (121, 103 and 82 nematodes/g of sample, respectively) (Table III), all three communes being located in the humid forest (Table III and Fig. 1). *Meloidogyne* spp. were observed in all the communes followed by *H. dihystera* (94.4% of communes) and *P. coffeae* (50% of communes). The remaining nematode species were present in less than half of the surveyed communes. Except for Tanguieta, where only *Meloidogyne* spp. were observed, each of the surveyed communes was infested with mixed populations of nematodes. As many as eleven nematode species were observed in Cotonou and eight in Djougou, Malanville and Porto-Novo (Table III).

The number of samples (roots or soil) collected from the humid forest (HF), the Guinea savannah (GS), the sub-humid savannah (SHS) and the Sudan savannah

Table IV. Identity and population density of plant-parasitic nematodes detected in vegetable crops according to agro-ecological zones of Benin between May 2004 and November 2005.

Nematode species	HF[a] (90)	GS[a] (42)	SHS[a] (22)	SS[a] (17)
Aphelenchoides bicaudatus	1[b]c	0c	0c	1b
Ditylenchus sp.	0c	0c	0c	0b
Helicotylenchus dihystera	10b	11b	11b	2b
Hemicriconemoides strictathecatus	1c	0c	0c	0b
Meloidogyne spp.	86a	39a	20a	35a
Pratylenchus coffeae	3c	0c	0c	2b
Rotylenchulus reniformis	0c	1c	0c	0b
Scutellonema clathricaudatum	0c	1c	2c	0b
Quinisulcius capitatus	1c	1c	0c	0b
Tylenchulus sp.	1c	0c	0c	0b
Xiphinema sp.	0c	0c	0c	0b
Mean	*9.45 a*	*4.82 b*	*3.76 b*	*3.76 b*

[a]Agro-ecological zones (HF = Humid Forest, GS = Guinea Savannah, SHS = Sub-Humid Savannah, SS = Sudan Savannah). Values in parentheses represent total number of soil or root samples collected in the given agro-ecological zone. [b]Each nematode population density (untransformed data) represents mean value per 1 g roots + 1 g soil. Statistical analysis and mean separations were undertaken on $\log_{10}$ (x+1) transformed data per g roots and per g soil.

(SS) were, respectively, 90, 42, 22 and 17. The humid forest was found to be the agro-ecological zone harbouring the greatest diversity of plant parasitic nematodes (mean of 9 nematodes/g of sample, calculated across nematode species) (Table IV). In the GS, HF and SHS agro-ecological zones, *Meloidogyne* spp. had the largest population densities (86, 39 and 20 nematodes/g of sample, respectively) followed by *H. dihystera* (10, 11 and 11 nematodes/g of sample, respectively). Population densities of other nematode species were similar and significantly smaller than those of *Meloidogyne* spp. or *H. dihystera*. In the SS zone, *Meloidogyne* spp. also had the largest population density (35 nematodes/g of sample); but population densities of all other nematode species were similar and significantly lower than that of *Meloidogyne* spp. (Table IV).

Occurrence and incidence of plant-parasitic nematodes. *Meloidogyne* spp. were the most prevalent nematodes in the surveyed sites (79.1% of total 171 root or soil samples collected) followed by *H. dihystera* (73.6%); each of the other nematode species was observed in less than 20% of the samples. *Ditylenchus* sp. was observed only in samples from tomato and carrot and *Xiphinema* sp. in samples from carrot and celosia (prevalence = 1.6% for each of the two nematode species) (Table V). The relative abundance for *Meloidogyne* spp., *H. dihystera* and *P. coffeae* was 62.1, 9.5 and 1.9, respectively, and less than 1 for all other nematode species. Though *Tylenchulus* sp. was less prevalent or abundant compared to *H. dihystera* and *P. coffeae*, the mean intensity of *Tylenchulus* sp. was second (33.4), behind *Meloidogyne* spp. (99.3), and larger

Table V. Prevalence, relative abundance and mean intensity of plant-parasitic nematodes in 26 vegetable crops sampled in eighteen communes of intensive vegetable production in Benin between May 2004 and November 2005.

Nematode species	Prevalence[a] (%)	Relative abundance[b]	Mean intensity[c]
Aphelenchoides bicaudatus	9.9	0.6	12.3
Ditylenchus sp.	1.6	<0.5	5.8
Helicotylenchus dihystera	73.6	9.5	20.9
Hemicriconemoides strictathecatus	5.3	<0.5	17.1
Meloidogyne spp.	79.1	62.1	99.3
Pratylenchus coffeae	16.5	1.9	23.3
Rotylenchulus reniformis	9.3	0.5	13.2
Scutellonema clathricaudatum	15.4	0.8	10.5
Quinisulcius capitatus	8.2	0.6	15.3
Tylenchulus sp.	3.3	0.6	33.4
Xiphinema sp.	1.6	<0.5	6.3

[a](Number of samples having a particular nematode species/number of samples examined) x 100. [b]Total number of individuals of a particular species per g soil and root sample in all the samples/number of samples including those with zero counts for that species. [c]Mean number of individuals of a particular nematode species per g soil and roots in the positive samples/number of positive samples

Table VI. Physico-chemical properties of soils under vegetable production in Benin.

Sites	Physical properties				Chemical properties										
	Clay	Loam	Sand	Texture	OM	N	C/N	pH_{H2O}	Ca^{++}	Mg^{++}	K^{+}	Na^{+}	S	CEC*	P total
	(%)				(%)	(cmol/kg)									(mg/kg)
Heavier soils															
Cocotomey	4.4 *(1.8)*[§]	1.8 *(0.4)*	94.2 *(3.0)*		2.2 *(1.1)*	0.107 *(0.041)*	11.3 *(3.1)*	6.8 *(0.3)*	7.0 *(3.6)*	4.5 *(2.1)*	0.7 *(0.4)*	0.8 *(0.0)*	12.5 *(5.7)*	8.5 *(2.7)*	2001.7 *(582.0)*
Grand-Popo	5.1 *(4.6)*	3.2 *(2.3)*	91.6 *(6.1)*		1.4 *(0.8)*	0.082 *(0.041)*	9.8 *(1.0)*	6.6 *(0.9)*	6.1 *(3.6)*	4.0 *(2.4)*	0.4 *(0.4)*	1.3 *(0.4)*	11.7 *(6.2)*	6.2 *(2.7)*	1703.8 *(896.6)*
Ouidah	1.5 *(0.8)*	3.4 *(1.6)*	94.8 *(0.5)*	Sand	1.8 *(0.6)*	0.104 *(0.004)*	10.1 *(2.8)*	7.2 *(0.6)*	7.0 *(0.6)*	4.5 *(1.0)*	0.9 *(0.1)*	1.5 *(0.3)*	13.7 *(0.3)*	7.2 *(1.7)*	3065.0 *(1371.8)*
Porto-Novo	4.1 *3.1*	3.2 *(2.9)*	92.7 *(6.0)*		2.1 *(1.1)*	0.164 *(0.161)*	11.2 *(1.8)*	6.9 *(0.4)*	7.7 *(3.8)*	4.9 *(1.8)*	0.8 *(0.1)*	1.0 *(0.2)*	14.2 *(5.7)*	7.6 *(3.4)*	1801.9 *(875.1)*
Bohicon	14.4 *(2.8)*	14.1 *(1.2)*	70.6 *(3.3)*		7.4 *(0.8)*	0.310 *(0.037)*	13.8 *(0.1)*	7.4 *(0.2)*	17.0 *(2.8)*	6.9 *(1.3)*	0.9 *(0.0)*	0.6 *(0.0)*	25.4 *(4.1)*	15.5 *(0.1)*	3500.0 *(1468.0)*
Djougou	10.2 *(7.9)*	15.9 *(10.0)*	73.5 *(17.0)*		2.8 *(1.5)*	0.153 *(0.074)*	10.5 *(0.8)*	7.6 *(0.6)*	9.6 *(2.9)*	6.3 *(1.7)*	0.8 *(0.2)*	0.8 *(0.1)*	17.3 *(4.8)*	13.1 *(6.7)*	1999.1 *(1044.4)*
Kandi	10.0 *(2.9)*	17.3 *(3.3)*	72.1 *(5.7)*		2.2 *(0.9)*	0.121 *(0.042)*	10.5 *(0.9)*	7.3 *(0.2)*	6.4 *(3.2)*	4.5 *(2.2)*	0.8 *(0.2)*	0.7 *(0.1)*	12.1 *(5.3)*	12.4 *(1.6)*	951.9 *(340.7)*
Malanville	10.0 *(3.6)*	19.4 *(4.5)*	70.0 *(3.8)*	Loam Sandy	1.0 *(0.2)*	0.064 *(0.012)*	9.3 *(0.2)*	5.6 *(0.9)*	4.7 *(2.2)*	3.2 *(1.4)*	0.4 *(0.2)*	0.7 *(0.1)*	9.0 *(3.3)*	10.0 *(1.2)*	304.6 *(64.6)*
Natitingou	11.5 *(2.0)*	15.7 *(3.8)*	72.3 *(4.3)*		2.2 *(2.7)*	0.071 *(0.018)*	9.9 *(0.4)*	7.0 *(1.1)*	5.3 *(3.1)*	3.2 *(1.6)*	0.7 *(0.3)*	1.8 *(3.5)*	9.4 *(4.7)*	9.4 *(1.6)*	223.6 *(72.3)*
Parakou	10.4 *(2.0)*	12.8 *(3.2)*	76.6 *(3.3)*		2.3 *(0.7)*	0.132 *(0.039)*	10.3 *(0.3)*	7.7 *(1.0)*	11.0 *(4.3)*	7.5 *(1.9)*	0.9 *(0.0)*	0.8 *(0.2)*	19.5 *(6.6)*	12.6 *(3.3)*	1454.9 *(755.7)*
Save	11.1 *(2.5)*	19.8 *(10.9)*	69.1 *(10.9)*		3.3 *(0.6)*	0.162 *(0.030)*	11.9 *(0.4)*	8.3 *(0.3)*	15.4 *(4.6)*	8.9 *(2.7)*	0.9 *(0.0)*	1.2 *(0.7)*	26.5 *(7.3)*	15.0 *(2.2)*	1747.7 *(622.2)*
Lighter soils															
Cotonou	5.8 *(3.5)*	6.2 *(6.4)*	87.7 *(9.0)*		2.0 *(1.0)*	0.106 *(0.051)*	10.8 *(1.9)*	6.9 *(0.4)*	6.8 *(3.7)*	4.7 *(1.8)*	0.8 *(0.2)*	1.3 *(0.3)*	13.5 *(5.3)*	6.9 *(2.6)*	2587.3 *(1626.6)*
Dassa	4.4 *(2.0)*	17.4 *(5.7)*	77.5 *(7.7)*	Sand Loamy	1.5 *(0.6)*	0.082 *(0.032)*	10.7 *(0.2)*	6.7 *(0.8)*	6.3 *(3.0)*	3.7 *(1.8)*	0.9 *(0.0)*	0.5 *(0.2)*	10.7 *(4.7)*	6.8 *(1.2)*	1517.3 *(1272.5)*
Seme	10.1 *(4.7)*	5.6 *(5.2)*	84.5 *(8.7)*		1.2 *(0.5)*	0.076 *(0.027)*	9.4 *(0.9)*	6.5 *(06)*	5.7 *(1.6)*	3.8 *(0.9)*	0.5 *(0.3)*	1.3 *(0.2)*	11.1 *(2.7)*	7.0 *(1.9)*	2707.3 *(2538.7)*

The physico-chemical analyses were undertaken on a total of 92 soil samples collected from urban and peri-urban vegetable production sites located in fourteen communes of Benin. The soil sampling was undertaken between May 2004 and November 2005 and analyses undertaken by the "Laboratoire des Sciences du Sol, Eaux et Environnement" of the Benin National Agricultural Research Institute (INRAB), Agonkanmey Station. * = exchangeable cation capacity. Numbers in parentheses represent standard deviations of means.

Table VII. Correlation coefficients (r) between soil physico-chemical properties and population density of nematode species associated with vegetable crops in urban and peri-urban vegetable production sites of Benin.

Nematode	Physical properties					Chemical properties										
	Clay (≤2 µm)	Fine alluvium (2-20 µm)	Coarse alluvium (20-50 µm)	Fine sand (50-200 µm)	Coarse sand (200-2000 µm)	OM	N	C/N	pH_{H2O}	Ca^{++}	Mg^{++}	K^{+}	Na^{+}	S	CEC	P total
	(%)					(%)	(cmol/kg)									(mg/kg)
Meloidogyne spp.	-0.16[a]	-0.19	-0.21	0.22	-0.21	0.10	0.05	0.01	0.00	0.11	0.08	0.01	0.16	0.11	-0.21	0.27
	ns[c]	ns	0.04[b]	0.04	0.04	ns	Ns	ns	ns	ns	ns	ns	ns	ns	0.04	0.01
Helicotylenchus dihystera	0.09	0.21	0.01	-0.01	-0.07	0.19	0.21	0.07	0.35	0.38	0.41	0.23	-0.08	0.39	0.24	0.08
	ns	0.04	ns	ns	Ns	ns	0.04	ns	<0.001	<0.001	<0.001	0.04	ns	<0.001	0.02	ns
Pratylenchus coffeae	0.02	-0.02	0.16	0.03	-0.08	0.00	0.04	-0.16	0.02	-0.09	-0.06	0.05	-0.02	-0.08	0.08	0.14
	ns	ns	ns	ns	Ns	ns	Ns	ns	ns	ns	ns	ns	ns	ns	ns	ns
Combined nematodes[d]	-0.15	-0.09	-0.16	0.21	-0.22	0.12	0.08	0.001	0.12	0.17	0.17	0.07	0.12	0.18	-0.12	0.28
	ns	ns	ns	0.04	0.04	ns	Ns	ns	ns	ns	ns	ns	ns	ns	ns	0.01

The soil samples (total of 92) used for the physico-chemical analyses were collected from vegetable production sites located in fourteen communes of Benin. Only the four nematode species for which population density calculated across crops is higher than 1 nematode/g sample are presented in the Table.
In each cell, the number above represents the correlation coefficient[a] and the number below the value of the probability[b], P. [c]Non significant P. [d]Values for "combined nematodes" in the table represent r and P values obtained from nematode population density of all the 11 nematodes species and the soil physico-chemical properties' data. ns = non significant.

than that of *P. coffeae* or *H. dihystera* (23.3 and 20.9, respectively).

Physico-chemical properties of soils under vegetable production in surveyed sites and their relationship with nematode population density. The physico-chemical analyses of soil samples indicated three types of soil texture in the surveyed vegetable production areas: sandy soil with 92-95% of sand, loam sandy soil with 69-77% of sand and 13-20% of loam, sand loamy soil with 78-88% of sand and 6-10% of clay (Table VI). Heavier soils (sand and loam sandy soils) and lighter soils (sand loamy soil) represent, respectively, 60.7% and 39.3% of total soil samples collected. Organic matter content ranged from critical (1.0%) to good (3.3%) levels across surveyed communes. Exceptionally, high organic matter content (7.4%) occurred in samples collected from Bohicon. Soil N content ranged from 0.1 to 0.3%. Meanly to weakly acid soils were collected from Seme and Malanville, respectively, moderately from Parakou and Save, weakly basic from Bohicon and Djougou and neutral from other communes. C/N ratios ranged between 9 and 14 indicating a very good mineralization of organic matter. Soil potassium content was average (0.4 cmol/kg) in Grand-Popo and Malanville, very rich (>0.8 cmol/kg) in Ouidah, Bohicon, Parakou, Savè and Dassa, and rich (0.5 -0.8 cmol/kg) in other communes. The base saturation was very weakly leached on all sites. The exchangeable cation capacity (ECC) was average (10-16 cmol/kg) in Bohicon, Djougou, Kandi, Malanville, Parakou and Savè and low (7-9 cmol/kg) in others communes. The largest values for all assessed properties (with the exception of pH, $Mg^{++}$and Na^{+}) were obtained with samples collected from Bohicon, and the smallest values with samples collected from Malanville (with the exception of Na^{+} and ECC). Both Bohicon and Malanville have loam sandy soil (Table VI).

Table VII indicates the coefficients of correlation between physico-chemical soil properties and population density of the three most important nematodes (in terms of population density, prevalence and relative abundance) recovered from root and soil samples. In general, when it exists, the correlation between soil physical and chemical properties and nematode population density is weak (maximum value of r = 0.41). No significant correlation was observed between population density of *Meloidogyne* spp. and clay or fine alluvium soil. But for soil with particle size larger than 20 µm, *Meloidogyne* spp. population density was correlated with soil physical properties (r = -0.21 or 0.22; P = 0.04). The population density of *Meloidogyne* spp. was not correlated with soil chemical properties except with ECC and P_{total} (r = -0.21 and 0.27, respectively; P = 0.04 and 0.01, respectively). The population density of *H. dihystera* was mostly correlated with soil chemical properties with positive correlation with soil N, pH, Ca^{++}, Mg^{++}, K^{+}, S, and ECC, with r ranging from 0.21 to 0.41 and P from less than 0.01 to 0.04. The population den-

sity for this nematode was correlated only with fine alluvium soil (r = 0.21 and P = 0.04). *Pratylenchus coffeae* population density was not correlated with any physical or chemical properties of the collected soil samples. The combined nematode population density (of all eleven nematode genera) was correlated only with fine or coarse sandy soil and P_{total} (r = 0.21, -0.22 and 0.28; and P ranging from 0.01 to 0.04). Organic matter and C/N ratio were not correlated with the population densities of any of the three most important nematode species, or with the combined nematode population density.

DISCUSSION

This study provides the most detailed information on PPN occurring on vegetable crops in Benin to date; very limited studies had previously been conducted. The study of Dodego (1979) focused on cowpea and tomato, while that of Sikora (1991) focused on aubergine, carrot, celosia, crin-crin, lettuce, okra and tomato; both studies covered limited vegetable production areas.

It is generally assumed that the presence of the host plant is the main determinant of the population densities of PPN (Yeates, 1987). Taylor (1976) observed that, in general, most vegetable crops have been recorded as hosts for at least one nematode species. The current study supports this observation and shows not only the polyphagous status of many of the recorded nematode species, but also the variability in host susceptibility of the sampled crops (expressed as percentage infected crops and nematode population density, respectively). The most cultivated and consumed vegetable crops in Benin (James *et al.*, 2006) were good hosts for the two most prevalent and relatively abundant nematode species, i.e. *Meloidogyne* spp. and *H. dihystera*, explaining to some degree their countrywide distribution. Among these species, *Meloidogyne* spp. were identified as major system pests in the agro-ecosystems, affecting 96.1% of sampled crops and with a population density of up to 174 nematodes/g of samples observed in the current study. The dominance of *Meloidogyne* spp. on vegetable crops has also been commented on by numerous researchers (Sikora and Fernandez, 2005), who observed that these nematode species are so common in sub-tropical and tropical vegetable production that they are frequently accepted to represent "nematodes" in general. Such a situation presents a serious risk to vegetable production worldwide and helps explain the extensive use of chemical pesticides on vegetable crops (James *et al.*, 2006).

The most important nematodes (*Meloidogyne* spp.) were not recorded from onion and were in low densities on amaranth, bell pepper, green bean, green pepper, pumpkin and vernonia. The low susceptibility of some vegetable crops (especially amaranth and vernonia) to *Meloidogyne* spp. appears to have been subconsciously exploited by vegetable growers in Benin through their use in rotation and intercropping systems as management strategies against this nematode. Nevertheless, at some surveyed sites, amaranth was severely infected by *Meloidogyne* spp., confirming the existence of different *Meloidogyne* species and leading to the need for an alternative option for *Meloidogyne* spp. management complementary to intercropping and rotation systems (Afouda *et al.*, 2008).

In Benin, cyst nematodes, *Heterodera* sp. (Dodego, 1979) and *Heterodera schachtii* Schmidt and *Globodera rostochiensis* (Woll.) Skarbilovich (Sikora, 1991), were found associated with vegetable crops. These nematode species were not detected in the current study, one of the reasons perhaps being the difference in the extraction methods used and the surveyed sites. Also, six nematode species (*A. bicaudatus*, *Ditylenchus* sp., *H. strictathecatus*, *S. clathricaudatum*, *Q. capitatus* and *Xiphinema* sp.) were identified as new records from seven vegetable crops previously sampled in Benin between 1979 and 1991. This reinforces the importance of diagnostic services in the development and implementation of IPM systems for vegetable production.

Meloidogyne spp. were observed in all the surveyed communes and AEZ, indicating their wide distribution on vegetable crops in Benin. However, the population densities varied with the commune. *Meloidogyne* spp. were less abundant in Hilla-Condji, Glazoue and Bohicon, where vegetable production is less intensive than in Cocotomey, Cotonou and Seme (James *et al.*, 2006). Similarly, the population density of *H. dihystera* was largest in samples from Parakou and Save, which are among the most intensive vegetable production areas in the country. The highest numbers of PPN species were observed in the intensive vegetable production areas of Cotonou, Djougou, Malanville, Parakou and Porto-Novo. In intensive production systems, the frequent use of pesticides and resistant or non-host plants for a certain nematode species may lead to an imbalance between the compositions of nematode communities, favouring the proliferation of other species (Baujard *et al.*, 1995). Nematodes are generally less important under more extensive and varied growing systems typical of shifting cultivation and multiple-crop farming in subsistence agriculture, or in widely spaced rotations of commercial farming systems, than in more intensive production, where mono-cropping and narrow rotations are practiced (Netscher, 1978). Our results on diversity and distribution of nematode species according to AEZ support these observations and those from other studies (Waliullah, 1992; McSorley *et al.*, 1994).

Several studies carried out with soils differing in texture and chemical composition have demonstrated the influence of soil physical and chemical properties on nematode population density, distribution and community structure. This validates the potential of nematodes as bio-indicator organisms of soil status (Kandji *et al.*, 2001). But, in general, the influence of soil texture on

nematode population density was limited in our survey, which additionally indicates that the influence of soil texture on nematodes is nematode species dependent. The population density of *H. dihystera* was influenced by most (seven out of eleven) of the soil chemical components analysed and by one physical property, indicating that this nematode is very sensitive to soil chemical properties. In contrast, no relationship exists between *P. coffeae* population density and soil characteristics. A general observation shows that, in the current study, all soil properties that have a relationship with combined nematode population density are also related to the population density of *Meloidogyne* spp. This is probably a result of the significant predominance of *Meloidogyne* spp. in terms of population density over other nematode species in the statistical analysis of the data.

The results confirm the importance of *Meloidogyne* spp. and indicate the harmful potential of *P. coffeae* and *H. dihystera* in vegetable production in Benin. Attention should be more focused on these nematodes in order to increase productivity of most vegetable crops grown in the country. The knowledge achieved on the nematological situation of vegetable crops in Benin will help producers to establish appropriate integrated management strategies.

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LITERATURE CITED

Adam K.S. and Boko M., 1993. *Le Benin*. Agence de Cooperation Culturelle et Technique. Les Editions du Flamboyant/EDICEF, Cotonou, Benin, 95 pp.

Afouda L., Baimey H. and Fanou H., 2008. Evaluation of *Amaranthus* sp. and *Vernonia amygdalina*, and soil amendments with poultry manure for the management of root-knot nematodes on eggplant. *Phytoparasitica*, *36*: 368-376.

Baujard P., Bour E. and Martiny, B., 1995. Incidence des nematodes phytoparasites sur la culture du sorgho dans la zone Sahélienne du Sénégal, Afrique de l'Ouest. *Afro-Asian Journal of Nematology, 5*: 1-10.

Boag B., 1993. Standardization of ecological terms in Nematology. *Fundamental and Applied Nematology, 16*: 190-191.

Coyne D., Nicol M.J. and Claudius-Cole B., 2007. *Practical plant nematology: A field and laboratory guide*. SP-IPM Secretariat, International Institute of Tropical Agriculture (IITA), Cotonou, Benin, 82 pp.

Curran J., McClure M.A. and Webster J.M., 1986. Genotypic differentiation of *Meloidogyne* populations by detection of Restriction Fragment Length Difference in Total DNA. *Journal of Nematology, 18*: 83-86.

Dodego H.E., 1979. Les nématodes phytoparasites en RPB. Cas de: tomate, riz, niébé, agrumes. Extraction, Recensement, Nuisibilité. Mémoire de DEAT. Lycée Agricole Médji de Sékou (LAMS). Allada, Bénin, 78 pp.

Gomez K.A. and Gomez A.A., 1984. *Statistical Procedures for Agricultural Research*. John Wiley and Sons. Inc. New York, USA. 680 pp.

James B., Godonou I., Atcha C. and Baimey H., 2006. Healthy vegetables through participatory IPM in peri-urban areas of Benin. Summary of activities and achievements, 2003-2006. IITA Benin, Abomey-Calavi, Benin. 134 pp.

Kandji S.T., Ogol Callistus K.P.O. and Albrecht A., 2001. Diversity of plant-parasitic nematodes and their relationships with some soil physico-hemical characteristics in improved fallows in western Kenya. *Applied Soil Ecology, 18*: 143-154.

McSorley R., Dickson D.W., Brito J.A. and Hochmuth R.C., 1994. Tropical rotation crops influence nematode densities and vegetable yields. *Journal of Nematology*, *26*: 308-314.

Netscher C. and Sikora R.A., 1990. Nematode parasites of vegetables. Pp. 237-283. *In*: Plant Parasitic Nematodes in Subtropical and Tropical Agriculture (Luc M., Sikora R.A. and Bridge J., eds). CAB International, Wallingford, UK.

Netscher C., 1978. *Morphological and physiological variability of species of Meloidogyne in West Africa and implication for their control*. Mededelingen Landbouwhogeschool Wageningen, The netherlands, 46 pp.

Popovici I. and Ciobanu M., 2000. Diversity and distribution of nematode communities in grasslands from Romania in relation to vegetation and soil characteristics. *Applied Soil Ecology*, *14*: 27-36.

Quénéhervé P., 1988. Population of nematodes in soils under banana cv. Poyo in the Ivory Coast. 2. Influence of soil texture, pH and organic matter on nematode populations. *Revue de Nématologie*, *11*: 245-251.

Sikora R.A., 1991. Presence, distribution and importance of plant parasitic nematodes in vegetables in Benin with special reference to soil antagonistic potentials. Rapport d'étude, Porto-Novo. SPV. 32 pp.

Sikora R.A. and Fernandez E., 2005. Nematode parasites of vegetables. Pp 319-392. *In*: Plant-Parasitic Nematodes in Subtropical and Tropical Agriculture, Second Edition (Luc M., Sikora R.A. and Bridge J., eds). CABI Publishing, Wallingford, UK.

Taylor D.P., 1976. Plant nematology problems in tropical Africa. *Helminthological Abstracts Series B, Plant Nematology*, *45*: 269-284.

Waliullah M.I.S., 1992. Nematodes associated with vegetable crops in the Kashmir valley, India. *Nematologia Mediterranea*, *20*: 47-48.

Yeates G.W., 1987. How plants affect nematodes. *Advances in Ecological Research, 17*: 61-113.

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