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Full Length Research Paper

Pathogenicity and reproduction of *Meloidogyne incognita* (Kofoid and White) chitwood on African yam bean, *Sphenostylis stenocarpa* (Hochst *Ex*. A. Rich) Harms accessions

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Studies were carried out to determine the host status of 12 African yam bean (*Sphenostylis stenocarpa*) accessions to *Meloidogyne incognita* infection. The experiments were laid out in a completely randomized design (CRD) with six replications. Inoculated plants received 4,000 eggs of *M. incognita*, while those without eggs served as controls. Data were collected on growth, yield and yield parameters of the African yam bean accessions. Data collected were subjected to statistical analyses using Genstat for windows, version 3.2. Results showed that growth, yield and yield parameters were significantly (P≤0.05) reduced in inoculated accessions when compared with the controls. Four accessions, TSS 63, Eha-Amufu, TSS 56 and Ugbokolo, were tolerant to *M. incognita* infection, while the rest of the accessions, TSS 3, TSS 4, TSS 22, TSS 5, TSS 10, TSS 11, TSS 112 and TSS 7, were susceptible to *M. incognita* infection. Percentage yield reduction as a result of *M. incognita* infection ranged from 0.21 to 74.3%. It was observed that none of the accessions was resistant to *M. incognita* infection. The tolerant accessions are therefore recommended for use by African yam bean farmers as they would check yield losses caused by *M. incognita.*

Key words: African yam bean, *Meloidogyne incognita*, pathogenicity, *Sphenostylis stenocarpa*.

INTRODUCTION

African yam bean, *Sphenostylis stenocarpa* (Hochst *Ex* A. Rich) Harms belongs to the family Fabaceae (Syn. Leguminosae) and classified under the sub-family Papilionoideae (Heywood, 1971). African yam bean is widely distributed and found growing wild or cultivated throughout the west of tropical Africa (Zohary, 1972; Okigbo, 1973; Anonymous, 1979). The reported food values for total seed protein content varied from 19.5 to 29% on a dry weight basis. Value similar to these was reported for cowpea (*Vigna unguiculata* (L.) Walp; 24%), but low compared to those of winged bean (*Psophocarpus tetragonolobus* (L.) D.C.; 32.8%) and soybean (*Glycine max* (L.) Merril; 35.1%) (Duke, 1981). Uguru and Madukaife (2001) reported a crude protein range of 18.1 to 25.8% for African yam bean, which compares well with 16.9 to 25.4%, 21.2 to 22.5% and 24.4 to 28.0% for Bambara groundnut, pigeon pea and cowpea, respectively. African yam bean has also been rated as equal or superior to pigeon pea and cowpea in methionine and lysine content (Uguru and Madukaife, 2001; Ene-Obong, 2008). It contains about 50% carbohydrate and 5 to 6% fibre (Anonymous, 1979; Enwere, 1998). In addition, a highly insecticidal substance, *lectin*, has been isolated from African yam bean seed and this could be utilized in future for gene cloning with the aim of imparting pest resistance to plants (Omitogun, et al., 1999). Although, African yam bean is

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cited in the list of 400 under-exploited tropical legumes (Rachie and Roberts, 1974; Anonymous, 1979), it has attracted research interest in recent times because of its nutrient composition (Nwokolo, 1987; Edem et al., 1990; Uguru and Madukaife, 2001). In fact, given the nutritional status of *S. stenocarpa*, it has the potential of replacing animal proteins in the diets of many poor Africans who cannot afford the exorbitant cost of proteins from animals.

In addition to the nutritional potentials of African yam bean it is wise to depend on a broader range of plant species as against a few major staple crops to ensure agro-biodiversity and food security (Naylor et al., 2004). African yam bean is a legume and its production has been reported to be adversely hampered by plant parasitic root-knot nematodes, *Meloidogyne* spp. (J. Eze, Nsukka, Nigeria, personal communication). The root-knot nematodes have been implicated in disease, yield reduction and failures in crops (Bridge, 1972; Amosu, 1974; Ogunfowora, 1977; Olowe, 1981). Yield losses of 33.83 and 40.07% in pea and grains, respectively have been reported and attributed to root-knot nematode, *Meloidogyne incognita* (Sasser, 1989). An estimated 40% yield reduction in tomato, garden egg, soybean, cucumber and melon heavily infested with root-knot nematodes in northern Nigeria have also been reported (Bridge, 1972). Hence, some measures aimed at reducing the adverse effects of root knot nematodes in host plants have been adopted by many researchers (Singh and Sitaramaaiah, 1970; Asmus and Ferraz, 1988; Sterling, 1991; Akhtar and Alam, 1993). but the use of host resistance/tolerance-strategy remains cheapest, easiest in application and safest environmentally (Olowe, 2009).

Afolami and Orisajo (2003) reported five varieties of rice (WAB 450-24-3-2-P-18-HB, WAB 450-24-3-1-P-37-B, WAB 450-1-B-P-33-HB, WAB 181-18 and FARO 48) that are tolerant to *M. incognita* infection. Ogaraku and Akueshi (2005b) reported that four cowpea cultivars (IT89KD-288, IT91K-118-20, Tvnu 72 and IT845-2246) are resistant to both *M. javanica* and *M. incognita* infections. In addition, Mbah et al. (2006) reported tomato varieties, VF Roma and UC-82-B, to be resistant to *M. incognita* infection in southeastern Nigeria. The present study was carried out to assess the pathogenicity of *M. incognita* on 12 African yam bean accessions and the host status of the various accessions to *M. incognita* infection.

MATERIALS AND METHODS

Germplasm collection

A total of 12 African yam bean accessions were collected for this study. Ten accessions (TSS 56; TSS 22; TSS 7; TSS 10; TSS 11; TSS 112; TSS 63; TSS 4; TSS 5 and TSS 3) were collected from the collections of the International Institute for Tropical Agriculture (IITA), Ibadan, Nigeria. The other two accessions were collected from two African yam bean producing localities. They included Ugbokolo in Benue State and Eha-Amufu in Enugu State, Nigeria.

Screen house experiment

Top garden soil collected from fallow land was mixed with well-dried pig manure at a ratio of 4:1 by volume according to a modified method of Ene-Obong and Okoye (1992). The soil-manure mixture was sterilized by heating in a large drum to a temperature of 100°C for 3 h and later air-dried for 4 weeks before the experiment to avoid any possible side effects of heating (Anderson and Ingram, 1989). The physical and chemical properties of the soil were determined according to standard methods (IITA, 1989). The textural class of the soil was loamy (56% coarse sand, 23% fine sand, 15% silt, 6% clay) pH (in H_20) 6.3, pH (in KCl) 5.6, OM 3.98%, OC 2.31%, BS 90.87%, N 0.0654%, CEC 10.4%, Na 0.1, K 0.15, Ca 7.2, Mg 2.0, exchangeable acidity 1.0 and P (in ppm) 47.76. Plastic planting buckets (5 L capacity) were filled with the heat-sterilized soil-manure mixture, after which seeds of each of the 12 African yam bean accessions were planted and replicated six times. The buckets were arranged in a completely randomized design (CRD) at the Department of Botany Screen House, University of Nigeria, Nsukka (Latitude 06° 86′ 39.6″ N, Longitude 007°41′ 20.4″ E and altitude 433 m above sea level).

Four weeks after planting (WAP), plastic buckets containing African yam bean seedlings were inoculated with suspension containing 4,000 eggs of *M. incognita*. The suspension was poured into a shallow trench created around the base of seedlings and covered immediately with top soil (Goswami and Chenulu, 1974; Hussey and Boerma, 1981). The buckets containing seedlings in six replications but without root-knot nematode (RKN) inoculum served as the controls. At the onset of flowering, six plants per accession were selected and used for the determination of the following parameters: number of leaves/plant, number of branches on main vine/plant, fresh shoot weight/plant, dry shoot weight/plant, fresh root weight/plant, dry root weight/plant, days to 50% flowering and number of galls/plant, number of nodules/plant, number of branches/plant and number of unfilled pods/plant. At maturity, pod length/plant, number of pods/plant, seed yield/plant, number of seeds/pod, 100-seed weight/plant, number of nematodes in roots/plant, number of *M. incognita* eggs/root system, number of *M. incognita* juveniles per 500 g rhizosphere soil/plant and number of galls/plant were determined. Data collected were subjected to statistical analyses using Genstat for windows, version 3.2. Treatment means were separated using least significant difference (LSD) at P=0.05.

Identification of root-knot nematode

To identify the *Meloidogyne* sp., 10 to 20 single galls containing mature females from the African yam bean field were teased to remove adult females under a dissecting microscope (Southey, 1970). The adult females were used for the preparation of the perineal patterns for the identification of the species of root-knot nematode according to the procedures described by Hartman and Sasser (1985). The *Meloidogyne* sp. was identified by comparing the perineal patterns with those described by Eisenback et al. (1981).

Maintenance and population build-up of *M. incognita* inoculum

Pure culture of *M. incognita* was maintained in susceptible African yam bean accession (TSS 112) planted in heat-sterilized garden soil. The plants were watered as and when due for about eight weeks to allow for reasonable nematode reproduction.

Extraction of nematode eggs used for inoculation from galled roots.

The galled roots of the African yam bean were carefully uprooted, washed free of soil and cut into 1 to 2 cm segments. The eggs were extracted from the galls using the NaOCl-extraction method (Barker, 1985). The cut galls were placed in 200 ml of 1.0% NaOCl solution, covered and shaken vigorously for 4 min. The NaOCl solution containing the galled segments was immediately poured through a 200-mesh (75-µM) sieve and nested over a 500-mesh sieve to collect the freed eggs. The 500-mesh sieve containing the eggs was placed under a stream of cold water to wash off residual NaOCl from the eggs. The remaining roots were rinsed with water to remove additional eggs that were also collected by sieving. The extracted eggs collected on the 500-mesh sieve were washed into a graduated beaker and the volume made up to 1,000 ml with chlorine-free tap water. The egg suspension was stirred for even distribution with a magnetic stirrer. Five aliquots of 1 ml each of the egg suspension were pipetted into a counting dish and the number of eggs counted under a stereo-microscope. The egg suspension was regulated such that 1 ml contained 200 eggs.

Nematode inoculation

An estimated total of 4,000 eggs of *M. incognita* were used as the standard inoculum strength for all the experiments. Since 1 ml of egg suspension contained 200 eggs of *M. incognita,* 20 ml would contain 4,000 eggs. Hence, 20 ml of the egg suspension was poured into a shallow trench created around the root tips of each of the test plants (Hussey and Boerma, 1981) and covered immediately with top soil (Goswami and Chenulu, 1974).

Extraction of nematode juveniles from rhizosphere soil

Soil samples (500 g each) were collected from the rhizosphere of *Meloidogyne*-infected plants with polyethylene bags. Samples were immediately taken to the laboratory for subsequent extraction and counting of *M. incognita* juveniles present. The method of extraction adopted here was the modified Baermann funnel method of Flegg and Hooper (1970). A glass funnel was placed on a retort stand and plastic mesh was shaped to fit inside the top of the funnel and held tightly with a rubber band. Rubber tubing was attached to the stem of the funnel and the tip closed with a pinch clamp. The funnel was filled with water to the level of the plastic mesh. Air bubbles were avoided by slightly opening the pinch and allowing some water to drop. Cotton wool layer was placed on the mesh and water was added to bring the level just above the cotton wool. The soil sample was placed on top of the cotton wool. The funnel was left undisturbed for 48 h, after which the nematode juveniles that have settled in the rubber tubing were collected by slightly opening the pinch clamp and allowing 10 to 30 ml of water to collect into a 100 ml beaker. The juvenile suspension was homogenized using a magnetic stirrer and the estimated number of juveniles per kilogram soil was made from the average of four counts.

Estimation of number of nematode eggs, juveniles and females in roots

The method of estimation used here is similar to that of Siddiqui et al. (2001). One gram sub-sample of galled root was macerated for 30 to 40 s in a Waring blender and counting was done using the suspension obtained. The total numbers of nematodes present in the roots were calculated by multiplying the number of nematodes present in 1 g of root by the total weight of root.

RESULTS

Results on the effects of *M. incognita* infection on growth parameters of 12 African yam bean accessions showed significant differences (P≤0.05) between *M. incognita*-infected accessions and the uninfected controls in most of the accessions (Table 1). The growth parameters also differed significantly (P≤0.05) among the different accessions. Generally, growth parameters were higher in the non-infected controls when compared with the nematode-infected accessions except for the dry root and fresh root weights. Seed yield was negatively correlated with dry root (r = -0.054) and fresh root (r = -0.134) weights (Table 3).

Results on the effects of *M. incognita*-infection on yield and yield parameters of 12 African yam bean accessions showed significant differences (P≤0.05) between *M. incognita*-infected and the uninfected controls in some of the accessions (Table 2). However, there was no significant difference (P≤0.05) between the infected and uninfected controls in seed yield and days to 50% flowering for the tolerant accessions (TSS 63, Eha-Amufu, TSS 56 and Ugbokolo). Also, yield and yield parameters differed significantly (P≤0.05) among the different accessions. The least percentage yield losses of 0.26, 0.21, 2.43 and 0.46 as a result of *M. incognita*-infection were produced by the tolerant accessions, TSS 63, Eha-Amufu, TSS 56 and Ugbokolo, respectively. Conversely, the highest percentage yield losses of 65.3, 74.3, 47.3, 42.0, 47.2, 56.5, 42.9 and 62.3 were recorded by the susceptible accessions, TSS 3, TSS 4, TSS 22, TSS 5, TSS 10, TSS 11, TSS 112 and TSS 7, respectively. Seed yield was positively correlated with pod length (r = 0.590, P≤0.01), number of pods (r = 0.577, P≤0.01), 100-seed weight (r = 0.400, P≤0.01), number of nodules (r = 0.537, P≤0.01) and number of seeds (r = 0.577, P≤0.01). However, number of unfilled pods (r = -0.162) and days to 50% flowering (r = -0.298) were negatively correlated with seed yield (Table 3).

The host status of the 12 African yam bean accessions is shown in Table 4. Results indicated that four accessions, TSS 63, Eha-Amufu, TSS 56 and Ugbokolo, were tolerant as they did not suffer any significant (P≤0.05) yield loss from nematode infection. These tolerant accessions also recorded the least gall indices and reproduction factors. The rest of the accessions, TSS 3, TSS 4, TSS 22, TSS 5, TSS 10, TSS 11, TSS 112 and TSS 7, were categorized as susceptible as they recorded significant (P≤0.05) yield loss as a result of nematode infection. They also presented the highest gall indices and reproduction factors. In addition, TSS 4 and 5 in the control experiments were the highest-yielding accession, although their susceptibility to nematode-infection was very high (Tables 3 and 4).

DISCUSSION

Results obtained from this study showed that *M.*

Table 1. The effects of *M. incognita* infection on growth parameters of 12 African yam bean accessions, *S. stenocarpa*, in a screen house experiment.

Accession	Health condition	Fresh shoot weight	Dry root weight	Dry shoot weight	Vine length	Number of branches	Number of leaves	Fresh root weight
TSS 3	Control	142.5 ± 17	1.5 ± 0.3	33.7 ± 1.0	278.8 ± 15.4	22.5 ± 2.6	150.0 ± 21.6	6.7 ± 2.3
	Infected	68.3 ± 20.8	4.2 ± 0.8	16.7 ± 1.6	223.3 ± 9.7	12.3 ± 1.7	53.8 ± 12.5	40.4 ± 10.6
TSS 63	Control	524.0 ± 4.9	2.7 ± 0.2	154.5 ± 4.1	338.3 ± 14.1	21.0 ± 0.8	226.8 ± 14.2	12.0 ± 5.5
	Infected	511.3 ± 8.5	7.4 ± 0.7	108.7 ± 2.5	336.5 ± 19.6	18.5 ± 1.2	222.3 ± 14.1	65.6 ± 4.7
TSS 4	Control	354.0 ± 38.5	3.5 ± 0.5	89.9 ± 1.7	380.0 ± 10.8	30.3 ± 5.8	306.8 ± 14.2	24.5 ± 8.7
	Infected	130.0 ± 61.2	8.6 ± 0.9	35.0 ± 3.8	311.3 ± 8.4	16.3 ± 3.5	59.5 ± 16.5	43.9 ± 11.6
TSS 22	Control	81.7 ± 34.7	2.5 ± 0.3	31.0 ± 1.4	287.8 ± 3.3	19.3 ± 3.7	106.0 ± 13.9	6.5 ± 1.5
	Infected	48.4 ± 18.4	2.7 ± 0.6	16.2 ± 2.8	196.3 ± 31.9	8.3 ± 3.1	55.5 ± 11.8	20.0 ± 3.8
TSS 5	Control	790.0 ± 29.4	5.5 ± 0.6	187.8 ± 6.5	527.5 ± 22.1	31.3 ± 0.9	296.0 ± 31.7	23.1 ± 8.9
	Infected	381.0 ± 21.9	8.9 ± 1.1	103.2 ± 9.8	330.8 ± 14.9	20.0 ± 1.4	136.5 ± 12.4	64.4 ± 16.5
EHA-AMUFU	Control	225.0 ± 11.8	2.9 ± 0.2	62.9 ± 4.6	365.0 ± 12.9	22.0 ± 0.8	154.0 ± 25.5	11.8 ± 4.2
	Infected	213.5 ± 10.9	7.6 ± 0.9	55.4 ± 1.7	364.3 ± 19.8	19.3 ± 0.9	145.8 ± 25.5	55.9 ± 8.2
TSS 10	Control	578.8 ± 43.0	1.8 ± 0.2	141.9 ± 2.3	455.0 ± 34.5	31.0 ± 4.8	183.3 ± 17.5	9.2 ± 1.2
	Infected	355.3 ± 29.2	6.0 ± 0.8	79.5 ± 1.9	331.3 ± 30.6	14.0 ± 2.1	136.5 ± 22.8	46.5 ± 4.4
TSS 56	Control	111.3 ± 14.4	2.7 ± 0.3	40.3 ± 1.1	366.8 ± 5.3	14.5 ± 0.6	147.8 ± 21.3	9.6 ± 2.2
	Infected	102.8 ± 13.6	6.2 ± 1.6	35.0 ± 1.5	363.5 ± 21.7	12.0 ± 1.4	140.0 ± 21.6	50.0 ± 9.2
TSS 11	Control	622.5 ± 65.2	1.8 ± 0.1	163.4 ± 4.5	403.3 ± 5.3	26.0 ± 2.1	260.0 ± 10.9	13.5 ± 3.8
	Infected	181.3 ± 13.1	11.9 ± 1.0	45.6 ± 3.9	300.0 ± 21.6	19.0 ± 1.4	120.3 ± 9.7	106.4 ± 21.7
TSS 112	Control	276.3 ± 38.1	4.4 ± 0.2	97.5 ± 3.0	336.3 ± 22.1	25.8 ± 1.7	188.5 ± 9.1	25.2 ± 6.4
	Infected	190.5 ± 28.1	22.7 ± 2.1	53.9 ± 2.3	281.3 ± 21.7	10.3 ± 0.5	141.5 ± 9.6	148.1 ± 28.3
UGBOKOLO	Control	547.5 ± 37.0	2.6 ± 0.3	134.8 ± 0.8	360.8 ± 8.6	19.5 ± 1.2	131.3 ± 19.0	22.0 ± 13.8
	Infected	530.3 ± 25.9	7.7 ± 0.8	132.1 ± 1.3	345.0 ± 36.9	17.8 ± 1.7	122.8 ± 19.6	39.7 ± 11.7
TSS 7	Control	251.3 ± 23.2	3.3 ± 0.4	63.5 ± 4.6	441.3 ± 30.1	21.5 ± 5.8	175.0 ± 31.0	15.9 ± 3.8
	Infected	125.3 ± 12.5	11.9 ± 1.2	26.7 ± 1.3	293.8 ± 47.5	12.5 ± 3.6	75.0 ± 12.9	87.8 ± 10.7
LSD(0.05)INTERACTION		42.43	1.20	23.30	38.50	3.84	31.41	15.09

Table 2. The effects of *M. incognita* infection on yield and yield parameters of 12 African yam bean accessions, *S. stenocarpa*, in a screen house experiment.

Accession	Health condition	Pod length	Number of unfilled pods	Number of pods	Seed weight	Seed yield	Number of nodules	Number of seeds	Days to 50% Flowering
TSS 3	Control	24.7 ± 1.6	3.3 ± 0.4	12.4 ± 1.8	26.8 ± 0.7	49.3 ± 10	20.8 ± 1.7	16.3 ± 0.8	96.3 ± 5.9
	Infected	21.3 ± 0.9	13.5 ± 5.2	6.3 ± 0.5	24.6 ± 1.2	17.1 ± 0.8	7.3 ± 1.7	10.9 ± 1.6	103.3 ± 1.7
TSS 63	Control	21.5 ± 1.4	7.3 ± 3.3	8.8 ± 0.8	29.1 ± 0.7	37.2 ± 0.5	25.5 ± 1.2	14.6 ± 0.9	97.7 ± 6.1
	Infected	22.5 ± 1.6	7.5 ± 1.7	8.6 ± 0.8	28.7 ± 0.2	37.1 ± 1.6	20.3 ± 1.7	15.1 ± 0.8	97.7 ± 4.0
TSS 4	Control	26.9 ± 1.6	5.0 ± 4.2	24.6 ± 0.8	34.5 ± 1.3	153.3 ± 1.6	48.3 ± 5.6	18.1 ± 0.8	99.7 ± 1.2
	Infected	25.0 ± 2.7	12.0 ± 5.4	9.0 ± 0.6	29.1 ± 0.2	39.4 ± 1.0	7.8 ± 2.7	15.4 ± 0.6	105.0 ± 1.6
TSS 22	Control	23.6 ± 1.5	10.0 ± 8.2	13.8 ± 0.8	34.3 ± 1.6	73.6 ± 1.6	21.8 ± 2.7	15.6 ± 2.3	99.7 ± 1.2
	Infected	22.5 ± 0.6	16.5 ± 10.7	8.5 ± 0.5	30.2 ± 0.6	38.8 ± 0.8	6.0 ± 2.1	15.2 ± 0.8	106.0 ± 3.5
TSS 5	Control	25.9 ± 2.9	10.0 ± 9.9	25.8 ± 2.4	35.7 ± 3.3	155.9 ± 3.2	69.8 ± 1.7	16.9 ± 1.6	95.7 ± 2.0
	Infected	24.8 ± 0.4	16.5 ± 11.0	20.0 ± 1.6	27.8 ± 1.6	90.3 ± 1.3	14.5 ± 3.4	16.0 ± 1.4	105.7 ± 1.2
EHA-AMUFU	Control	25.5 ± 1.5	7.5 ± 8.0	18.6 ± 1.6	31.3 ± 1.4	93.1 ± 0.8	39.5 ± 1.2	16.0 ± 1.6	92.3 ± 3.4
	Infected	25.3 ± 1.2	8.5 ± 5.0	18.0 ± 1.6	30.9 ± 1.6	92.9 ± 1.6	33.0 ± 2.9	16.7 ± 0.8	92.3 ± 2.6
TSS 10	Control	25.7 ± 1.9	5.0 ± 2.1	21.7 ± 0.8	26.5 ± 0.7	96.8 ± 2.8	46.5 ± 3.1	16.9 ± 0.8	91.0 ± 2.1
	Infected	23.6 ± 0.3	13.0 ± 4.7	15.4 ± 0.4	22.3 ± 1.6	51.1 ± 0.7	19.3 ± 2.5	14.9 ± 1.6	96.3 ± 11.8
TSS 56	Control	25.2 ± 2.3	6.8 ± 3.8	8.3 ± 0.5	35.8 ± 3.7	45.2 ± 0.8	37.0 ± 2.1	15.3 ± 0.8	96.7 ± 0.9
	Infected	24.0 ± 0.8	7.5 ± 1.9	8.5 ± 0.1	34.7 ± 1.6	44.1 ± 3.0	32.5 ± 1.2	15.0 ± 1.6	96.7 ± 0.5
TSS 11	Control	25.1 ± 2.8	5.7 ± 3.5	24.6 ± 2.4	26.7 ± 1.6	99.8 ± 0.8	49.8 ± 2.5	15.1 ± 1.5	96.3 ± 3.0
	Infected	21.9 ± 0.5	14.0 ± 9.9	11.8 ± 0.8	25.1 ± 1.6	43.4 ± 2.3	22.8 ± 6.6	14.7 ± 0.8	102.7 ± 2.4
TSS 112	Control	23.7 ± 2.4	9.7 ± 4.5	23.8 ± 2.4	29.6 ± 1.9	95.8 ± 0.7	34.0 ± 5.8	13.6 ± 1.5	100.7 ± 1.8
	Infected	22.2 ± 0.5	18.5 ± 7.0	11.8 ± 2.4	23.1 ± 1.6	37.6 ± 2.4	11.0 ± 2.9	13.9 ± 1.6	104.0 ± 2.9
UGBOKOLO	Control	30.0 ± 5.9	6.0 ± 2.1	12.0 ± 0.8	42.4 ± 1.5	87.0 ± 0.8	31.3 ± 2.6	17.1 ± 1.6	99.0 ± 2.1
	Infected	32.1 ± 3.3	6.2 ± 3.3	16.5 ± 0.8	34.3 ± 1.6	86.6 ± 2.4	26.3 ± 3.5	15.3 ± 0.5	99.0 ± 1.6
TSS 7	Control	25.3 ± 1.2	8.7 ± 2.8	19.6 ± 0.9	28.8 ± 3.2	95.6 ± 0.8	41.8 ± 3.1	16.9 ± 1.6	93.7 ± 2.0
	Infected	25.0 ± 1.6	12.3 ± 5.1	9.6 ± 0.2	26.8 ± 1.6	36.0 ± 0.8	7.5 ± 2.3	13.9 ± 0.8	101.0 ± 0.8
Lsd (0.05) interaction		3.01	8.45	1.88	2.50	3.71	4.43	0.513	5.14

Table 3. Relationship among *M. incognita* infection parameters and parameters of growth and yield of twelve African yam bean accessions in a pot experiment.

Parameter	FSHINT	PODLT	DYRTWT	DYRSHWT	NUFLP	NPODS	SDWT	MNYD	MNVLT	NNDLS	NBRCH	NLVS	FRTWT	NSD/PD	50%FLR	NEGSRT	NJ2SOL	NJ2RT	NGLS	MNGID
FSHWT	1.000																			
PODLT	0.421**	1.000																		
DYRTWT	0.016	-0.096	1.000																	
DYRSHWT	0.863**	0.444**	0.072	1.000																
NUFLP	-0.218	-0.276	0.183	-0.089	1.000															
NPODS	0.536**	0.407**	0.104	0.504**	-0.051	1.000														
SDWT	0.122	0.565**	-0.354	0.170	-0.396**	0.054	1.000													
MNYD	0.524**	0.590**	-0.054	0.546**	-0.162	0.898**	0.400**	1.000												
MNVLT	0.535**	0.500**	00.90	0.533**	-0.344*	0.465**	0.378**	0.579**	1.000											
NNDLS	0.359*	0.366*	-0.115	0.367*	-0.440**	0.410**	0.486**	0.537**	0.708**	1.000										
NBRCH	0.572**	0.276	-0.020	0.482**	-0.224	0.519**	0.118	0.567**	0.586**	0.429**	1.000									
NLVS	0.671**	0.031	0.188	0.535**	-0.227	0.343*	0.086	0.316*	0.561**	0.581**	0.416**	1.000								
FRTWT	-0.001	-0.252	0.853**	0.010	0.259	0.075	-0.481**	-0.134	0.037	-0.054	0.028	0.305*	1.000							
NSD/PD	0.286*	0.327*	-0.012	0.398**	-0.049	0.412**	0.382**	0.577**	0.492**	0.404**	0.367*	0.321*	-0.124	1.000						
50%FLR	-0.233	-0.202	0.149	-0.238	0.332*	-0.258	-0.200	-0.298*	-0.453**	-0.577**	-0.154	-0.409**	0.080	-0.190	1.000					
NEGSRT	-0.093	-0.344*	0.706**	-0.036	0.398**	0.177	-0.671**	-0.122	-0.112	-0.216	00.009	0.010	0.706**	-0.048	0.301*	1.000				
NJ2SOL	-0.162	-0.347*	0.663**	-0.122	0.404**	0.141	-0.686**	-0.161	-0.180	-0.360*	-0.142	-0.068	0.610**	-0.103	0.331*	0.925**	1.000			
NJ2RT	-0.314*	-0.441**	0.509**	-0.257	0.455**	0.029	-0.699**	-0.265	-0.330*	-0.474**	-0.209	-0.277	0.508**	-0.161	0.363*	0.855**	0.856**	1.000		
NGLS	-0.201	-0.363*	0.623**	-0.188	0.367*	0.154	-0.662**	-0.162	-0.195	-0.312*	-0.029	-0.110	0.587**	-0.034	0.295*	0.896**	0.873**	0.802**	1.000	
MNGID	-0.407**	-0.314*	0.270	-0.367**	0.225	0.006	-0.428**	-0.233	-0.310*	-0.411**	-0.153	-0.340*	0.251	-0.108	0.347*	0.521**	0.566**	0.596**	0.691**	1.000

*Significant at 0.05 level of probability. **Significant at 0.01 level of probability. FSHWT = Fresh shoot weight/plant; PODLT = Pod length (cm)/plant; DYRTWT = Dry root weight/plant; DYSHWT = Dry shoot weight/plant; NUFLP = Number of unfilled pods/plant; SDWT = 100-seed weight/plant; MNYD = Mean seed yield/plant; MNVLT = Main vine length (cm)/plant; NNDLS = Number of nodules/plant; NBRCH = Number of branches on main vine/plant; NLVS = Number of leaves/plant; FRTWT = Fresh root weight/plant; NSD/PD = Number of seeds/pod/plant; 50%FLR = Days to 50% flowering; NEGST = Number of eggs/root/plant; NJ2S0L = Number of juveniles in 500 g rhizosphere soil; NJ2RT= Number of juveniles/root/plant; NGLS= Number of galls/root/plant; MNGID = Mean gall index/root/plant.

incognita-infection caused significant (P≤0.05) reduction in almost all growth and yield parameters of the African yam bean. These results were similar to the findings of Ogbuji and Ezekwesili (1976), Nwanguma and Fawole (1998) and Ogaraku and Akueshi (2005b). The reduction in number of branches, number of leaves, number of nodules, pod length, shoot weight, main vine length, number of pods, 100-seed weight and number of seeds in *Meloidogyne*-infected plants, contributed immensely to the observed reduction in seed yield. An estimated yield loss of between 5.2 and 59.1% in rice (Afolami and Orisajo, 2003); and between 20 and 49% in cowpea (Ogaraku and Akueshi, 2005b) have been reported as a result of *Meloidogyne* spp. infection. Root-knot nematodes are known to interfere with carbon dioxide assimilation and the partitioning and translocation of photo-assimilates (Melakeberhan and Webster, 1993). The total amount of carbon dioxide available for assimilation is reduced grossly in *Meloidogyne*-infected plant and this diminishes photosynthesis which ultimately results in decreased biomass and crop yield (Loveys and

Table 4. Host status rating of twelve African yam bean accessions in a screen house experiment (after Afolami, 2000).

Accession	Mean Seed yield /plant (g)					
	GI	R	Control (B)	Inoculated (A)	Yield Difference in g/plant (A-B)	Host status rating
TSS 3	3.8	2.1	49.3	16.3	-33.0*	Susceptible
TSS 63	2.8	1.3	37.2	37.1	-0.1	Tolerant
TSS 4	4.0	3.1	153.3	38.1	-115.2*	Susceptible
TSS 22	3.8	1.9	73.6	38.6	-35.0*	Susceptible
TSS 5	4.0	3.6	155.9	90.3	-65.6*	Susceptible
EHA-AMUFU	3.0	1.5	93.1	92.8	-0.3	Tolerant
TSS 10	4.0	3.9	96.8	50.7	-46.1*	Susceptible
TSS 56	3.0	1.5	45.2	44.2	-1.0	Tolerant
TSS 11	4.0	5.1	99.8	42.2	-57.6*	Susceptible
TSS112	4.0	5.7	95.8	33.1	-62.7*	Susceptible
UGBOKOLO	3.0	1.4	87.0	86.5	-0.5	Tolerant
TSS 7	4.0	2.4	95.6	36.0	-59.6*	Susceptible

GI = Gall Index (after Taylor and Sasser, 1978): 1 = 1 to 2 galls; 2 = 3 to10 galls; 3 = 11 to 30 galls; 4 = 31 to 100 galls; 5 = > 100 galls. R = Nematode reproduction factor (after Oostenbrink, 1966): R = Pf/Pi, where Pf is the estimated final population of RKN in both soil and roots (juveniles and eggs in roots + juveniles in soil), Pi is the initial standard inoculum of 4,000 eggs). *= Statistically significant yield difference (P≤0.05); - = yield loss over control. Host status rating after Afolami (2000): **Resistant** means R ≤ 1, GI ≤ 2 (no significant yield loss). **Tolerant** means R > 1, GI > 2 (no significant yield loss); **susceptible** means R > 1, GI > 2 (significant yield loss); **hyper-susceptible** means R > 1, GI > 2 (significant yield loss). LSD (0.05) for comparing accession means x health condition means interaction = 3.71.

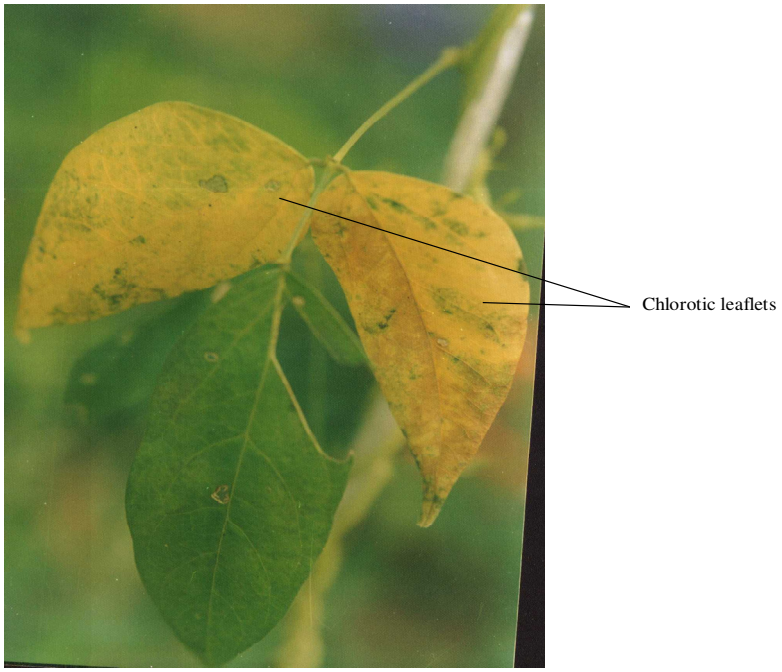

Figure 1. The African yam bean leaflets showing chlorosis (yellowing) as a result of *M. incognita* infection.

Bird, 1973; Melakeberhan and Ferris, 1989; Melakeberhan et al., 1990).

The noticeable yellowing (chlorosis) of leaflets (Figure 1) and stunted growth in *M. incognita*-infected plants could be attributed to decreased leaf nitrogen, chlorophyll and potassium concentration with subsequent decreases in photosynthetic ability of the host plants (Melakeberhan et al., 1986, 1987). Decreases in potassium concentration is particularly important because of its effect on photosynthesis, either by affecting carbon dioxide uptake

or by altering other key physiological processes such as osmotic potential. Deformations and damages done to roots of host plants by root-knot nematodes can lead to reduced absorption of nutrients and impair nutrient availability in infected host plants (Price and Sanderson, 1984; Rawthorne and Hague, 1986). The observed significant reduction in the number of nodules in *M. incognita*-infected plants could be another explanation for the significant reduction in seed yield since nodules harbor nitrogen-fixing bacteria in legumes. The total inhibition of nodulation by cyst-nematodes has also been reported in soybean (Huang and Barker, 1983). The presence of nematodes in the rhizosphere soil can lead to competition for space and a reduction in root hairs through which infection by nitrogen-fixing bacteria can take place (Epps and Chambers, 1962; Malek and Jenkins, 1964). Huang et al. (1984) reported that nematodes interfere with soybean lectin metabolism, thereby leading to a reduction in the rate of binding of rhizobia to *Heterodera glycine*-infected soybean roots. These phenomena subsequently suppressed the formation of nodules and perhaps yield.

Furthermore, the prolongation of the transition from vegetative growth phase to reproductive phase (flowering) in most of the African yam bean accessions could be attributed to the presence of nematodes in the roots, which disrupted the physiological activities of root xylem and phloem. Impaired supplies of nitrogen, phosphorus and potassium in plants have been reported to cause delayed flowering and flower abortion (Takahashi et al., 1973; Besford and Maw, 1975). Higher root weights in *M. incognita*-infected African yam bean compared with the uninoculated control could be as a result of giant cell formation which led to root galling. This result was similar to the findings of Ononuju and Fawole (2000), Afolami and Orisajo (2003) and Ogaraku and Chhangani (2010). Giant cell formation is triggered-off by enzymatic secretions from root-knot nematodes in host plants, which induce re-differentiation process that ultimately leads to the formation of multinucleated feeding cells called giant cells (Davis and Mitchum, 2005). Giant cell formation progresses through the processes of hyperplasia and hypertrophy of surrounding cortical cells (Williamson and Hussey, 1996). Galls serve as nutrient sinks in infected plants and cause nutrients to be redirected from shoot to roots (Melakeberhan et al., 1990). It has also been reported that the presence of nematode infection in plants induces an increase in plant hormones (indole-acetic acid, cytokinins, abscisic acid) concentrations which can lead to accelerated growth around the nematode feeding sites (Viglierchio and Yu, 1968; Glazer et al., 1983, 1984; Volmar, 1991).

The host status rating showed that four accessions (TSS 63, Eha-Amufu, TSS 56 and Ugbokolo) were tolerant. Eight other accessions (TSS 3, TSS 4, TSS 22, TSS 5, TSS 10, TSS 11, TSS 112 and TSS 7) were assigned the susceptible status. Tolerance in this context denotes a situation where *Meloidogyne* spp. reproduced freely and caused galling in the roots of African yam bean accessions ($R > 1$, $GI > 2$), but failed to cause any statistically significant ($P \leq 0.05$) yield reduction at harvest (Canto-Saenz, 1983; Afolami, 2000). In fact, none of the African yam bean accessions qualified for the resistant status in the actual sense. This is because a *Meloidogyne*-resistant crop variety disallows free reproduction ($R \leq 1$), suppresses formation of galls in roots ($GI \leq 2$) and ultimately suffers no statistically significant ($P \leq 0.05$) yield reduction (Canto-Saenz, 1983; Cook and Evans, 1987; Afolami, 2000). The susceptible accessions, though with higher gall index (GI), differed essentially from the tolerant accessions by the extent of yield losses they sustained from *M. incognita*-infection. Although, TSS 4 and 5 had higher gall indices and sustained significant ($P \leq 0.05$) yield losses as a result of *M. incognita*-infection, they ranked highest in terms of seed yield in the control experiments. These accessions are high yielding but for their susceptibility to *Meloidogyne* infection. Even though susceptible to *Meloidogyne* infection, they also compared favorably with the tolerant accessions in terms of yield.

It is therefore recommended that the high yield 'factor' in TSS 4 and 5 be investigated by plant geneticists/ breeders and incorporated into the low-yielding tolerant accessions to improve on their yields. Also, cultivation of susceptible accessions with other crops (mixed cropping) is recommended, as this will reduce serious multiplication of *Meloidogyne* species in the soil. Okigbo (1973) and Ezueh (1984) observed highest yields in African yam bean when planted with maize, yams or vegetables without linking the implication in plant nematode control. It is also known that mixed cropping discourages the rapid build-up of a single nematode species in the field environment (Ogbuji, 1979a). The locally sourced accessions (Eha-Amufu and Ugbokolo) might have attained their tolerant status through long years of cultivation and selection in their various local environments. Meanwhile, the tolerant accessions are recommended for African yam bean farmers to check yield losses since most of the farmlands are infested with *Meloidogyne* species.

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REFERENCES

Afolami SO, (2000). Suggestions for the Improvement of Current Methods of Studying and Reporting Resistance to root-knot nematodes. Int. J. Nematol. 10(1): 94-100.

Afolami SO, Orisajo SB(2003). Effects of *Meloidogyne incognita* on growth and yield of some NERICA and Asian upland rice varieties. Nig. J. Plt. Prot. 20: 25-40.
Akhtar M, Alam MM (1993). Utilization of waste materials in nematode control: A review. Bio. Technol. 45: 1-7.
Amosu JO(1974). Reaction of cowpea to root-knot nematode, *Meloidogyne incognita* in western Nigeria. Nig. Agric. J. 2: 165-169.
Anderson JM , Ingram JSI (1989).Tropical Soil Biology and Fertility: A Handbook of Methods. Wallingford, United Kingdom: CAB International, p. 177.
Anonymous (1979). *Tropical Legumes: Resources for the future.* Washington, D.C.: National Academy of Science, p. 332.
Asmus RMF, Ferraz S (1988). Antagonismo de algumase species vegetais, principalmente leguminosas, a *Meloidogyne javanica*. Fitopatol. Brasil 13: 20-24.
Barker KR (1985). Nematode Extraction and Bioassays. In: *An Advanced Treatise on Meloidogyne. Volume II Methodology*. Barker K R Carter CC Sasser J N (Eds). pp.19-35. Raleigh, N. C.: North Carolina State University Graphics,
Besford RT, Maw G A (1975). Effect of potassium nutrition on tomato plant growth and fruit development. Plt. Soil. 42: 395-412.
Bridge J (1972). Plant-parasitic nematodes of irrigated crops in the Northern States of Nigeria. Sam. Misc. Papers, pp. 42:17.
Canto-saenz M (1983). The nature of resistance to *Meloidogyne incognita* (Kofoid & White) Chitwood. Proceedings , third research & planning conference on root-knot nematodes, *Meloidogyne* spp. Carter, C.C. (Ed). International *Meloidogyne* Project. Lima, Peru. p. 233.
Cook R , Evans K (1987). Resistance and tolerance. In: Principles and practices of nematode control in crops . Brown RH Kerry BR (Eds.). Academic Press, Sydney. p. 179-231.
Davis EL, Mitchum MG (2005). Nematodes. Sophisticated parasites of legumes. Plt. Physiol. 137: 1182-1188.
Duke JA (1981). Handbook of legumes of world economic importance. New York: Plenum Press, p. 287.
Edem DO, Amugo CI , Eka OU (1990). Chemical composition of yam beans (*Sphenostylis stenocarpa*). Trop. Sci. 30: 59-63.
Eisenback JD, Hirshchmann H, Sasser JN, Traintaphyllou AC(1981). *A guide to the four common species of root-knot nematodes (Meloidogyne spp.) with a pictorial key.* Raleigh:North Carolina State Graphics, p. 48.
Ene-Obong EE, Okoye FI (1992). Interrelationship between yield and yield components in the African yam bean, *Sphenostylis stenocarpa* (Hochst Ex. A. Rich.) Harms. Beitr. Trop. Landwirtschaft Vet. Med. 30: 283-290.
Ene-Obong HN (2008). Nutrition science and practice: Emerging issues and problems in food composition, diet quality and health. An inaugural lecture of the University of Nigeria, Nsukka. p. 83.
Enwere NJ (1998). Food of plant origin. Processing and technology profiles. Nsukka: Afro-orbis publishers, 76pp.
Epps JM , Chambers AY (1962). Effect of seed inoculation, soil fumigation and cropping sequence on soybean nodulation in soybean-cyst nematode infested soil. Plant. Dis. Rep. 46: 48-51.
Ezueh MI (1984). African yam bean as a crop in Nigeria. World Crops, 36(6): 199-200.
Flegg FFM, Hooper DF (1970). Baermann funnel method In*: Laboratory methods for work with plant and soil nematodes.* Southey, J. F. (Ed.) pp .5-22. Her Majesty`s stationary office, London.
Glazer I, Orion D, Apelbaum A (1983). Interrelationships between ethylene production, gall formation, and root-knot nematode development in tomato plants infected with *Meloidogyne javanica*. J. Nematol. 15: 539-544.
Glazer I, Apelbaum A, Orion D (1984). Reversal of nematode-induced growth retardation in tomato plants, *Lycopersicon esculentum*, by inhibition of ethylene action.J. Am. Soc. Hort. Sci. 109: 886-889.
Goswami BK , Chenulu VV (1974). Interaction of root knot- nematode, *Meloidogyne incognita* and tobacco mosaic virus in tomato. Indian J. Nematol. 4: 69-80.
Hartman KM , Sasser JN (1985). Identification of *Meloidogyne* species on the basis of differential host test and perineal- pattern morphology. In: An advanced treatise on Meloidogyne. Volume II: Methodology. Barker KR, Carter CC, Sasser JN. (Eds.), North Carolina State University Graphics, Raleigh. pp.69-77.
Heywood VH (1971). The leguminosals systematic purview. In: Chemotaxonomy of legminosae Harborne, J.B. Bontler, D. and Turner, R.L. (Eds), pp.1-23. Academic press Inc., .London.
Huang JS ,Barker KR (1983). Influence of *Heterodera glycines* on leghemglobins of soybean nodule. Phytopathology. 73: 1002-1004.
Huang JS, Barker KR ,Van Dyke CG (1984). Suppression of binding between rhizobia and soybean roots by *Heterodera glycines*. Phytopathology, 74: 1381-1384.
Hussey RS, Boerma RH (1981). A green-house screening procedure for root-knot nematode resistance in soybeans. Crop Sci. 21: 794-796.
International Institute of Tropical Agriculture (1989).Automated and semi- automated methods for soil analysis. Manual series No. 7 .Ibadan, Nigeria.
Loveys BR , Bird AF (1973). The influence of nematode on photosynthesis in tomato plants. Physiol. Plant Pathol. 11: 11-21.
Malek RB , Jenkins WR (1964). Aspects of the host- parasite relationships of nematodes and hairy vetch. New Jersy Agric. Expt. Sta. Bull. 813: 31.
Mbah MC, Aniekwe NI, Nwafor SO (2006). Relative resistance of five tomato varieties to root-knot nematodes, *Meloidogyne incognita* (Kofoid and White) Chitwood in Abakiliki, southeastern Nigeria. Nig. J. Bot. 19(2): 236-241.
Melakeberhan H,Ferris H (1989). Impact of *Meloidogyne incognita* on physiological efficiency of *Vitis vnifera*. J. Nematol. 21: 74-80.
Melakeberhan H, Ferris H, Dias J (1990). Physiological response of resistant susceptible *Vitis vinifera* cultivars to *Meloidogyne incognita* J. Nematol. 22: 224-30.
Melakeberhan H, Webster JM , Brooke RC (1986). Relationship between physiological response and yield loss of different age French bean to *Meloidogyne incognita*. Plant Pathol. 35: 205-13.
Melakeberhan H, Webster JM, Brooke RC, D`Auria JM, Cackett MC (1987) Effect of *M. incognita* on plant nutrient concentration and its influence on the physiology of beans. J. Nematol. 19: 324-30.
Melakeberhan H , Webster JM (1993). The phenology of plant-nematode interaction and yield loss. In:, *Nematode interactions*. Khan, M.W. (Ed.).Chapman and Hall, London. pp. 26-41.
Naylor RL, Falcon WP, Goodman RM, Jahn MM, Sengooba T, Tefera H, Nelson R J (2004).Biotechnology in the developing world: a case for orphan crops. Food Pol. 29: 15-44.
Nwanguma EE, Fawole B (1998).Effect of population densities of *Meloidogyne incognita* on plant biomass of susceptible and resistant tomatoes. Nig. J. Hort. Sci. 3: 111-116.
Ogaraku AO, Akueshi CO (2005b).Effects of *Meloidogyne javanica* Treub on seven varieties of Cowpea, *Vigna unguiculata* (L.)Walp, in a pot experiment. Nig.
J. Bot. 18: 273-279.
Ogaraku AO ,Chhangani S (2010). Pathogenicity of *Meloidogyne javanica* (Treub) Chitwood on cowpea, *Vigna unguiculata* (L.) Walp and nematicidal effect of *Tagetes erecta* (L.) root extracts. Nig. J. Bot. 23: 225-234.
Ogbuji RO , Ezekwesili NO (1976). Some measured effects of root-knot nematode injury on tomato plants. Nig. J. Plant Prot. 4: 27-29.
Ogbuji RO (1979a). The status of root-knot nematodes in the Eastern States of Nigeria. In: Proceedings on the research planning conference on root-knot nematodes. Ibadan, Nigeria. pp.44-46.
Ogunfowora AO(1977). The effects of population levels of *Meloidogyne incognita* on yield of tomato in southwestern Nigeria. Nig. J. Plant Prot. 5: 61-67.
Okigbo BN (1973).Introducing the African yam bean: *Sphenostylis stenocarpa* (Hochst. *Ex.* Rich) Harms. Proc. 1st IITA Grain Legume Improvement Workshop, London. 29th Oct-2nd Nov. pp. 224-337.
Olowe T (1981). Importance of root-knot nematodes on cowpea, *Vigna unguiculata* (L.) Walp in Nigeria. In: proceedings of *International Meloidogyne Project research and planning conference on root-knot nematodes*, *Meloidogyne* spp.,Region IV, Ibadan, Nigeria. p. 85-109.
Olowe T (2009). Cowpea Germplasm resistant to *Meloidogyne arenaria* race 1, *Meloidogyne incognita* race 4 and *Meloidogyne javanica*. Eur. J. Sci. Res. 28(3): 338-350.
Omitogun GO, Jackai LEN , Thottappilly G (1999). Isolation of insecticidal lectin-enriched extracts from African yam bean

(*Sphenostylis stenocarpa*) and other legume species. Entomol. Exp. Appl. 90: 301-311.
Ononuju CC, Fawole B (2000). Influence of *Meloidogyne* sp. on nodule development and nitrogen accumulation in soybean. J. Agric. Envt. 1: 73-77.
Oostenbrink M (1966).Major characteristics of the relation between nematodes and Plants. Wagenigen: Meded. Landbouwhoghese, p. 48.
Price NS , Sanderson J (1984). The translocation of calcium from oat roots infected by the cereal cyst nematode *Heterodera avenae* (Woll.). Revue de Nematol. 7: 239-243.
Rachie KO, Roberts LM (1974). Grain legumes of low land tropics. Adv. in Agron. 26: 1-132.
Rawthorne D ,Hague NGM (1986). The effect of *Heterodera avenae* on the root system of susceptible and resistant oat seedlings. Ann. Appl. Biol. 108:89-98.
Sasser JN (1989). *Plant parasitic nematodes: the farmer's hidden enemy*. Raleigh:North Carolina State University Graphics, p. 115.
Siddiqui ZA, Iqbal A , Mahmoud I (2001). Effects of Pseudomonas fluorescens and fertilizers on the reproduction of *Meloidogyne incognita* and growth of tomato. Appl. Soil Ecol. 16: 179-185.
Singh RS , Sitaramaiah K (1970). Control of plant parasitic nematodes with organic amendments. PANS 16: 287-297.
Southey JF (1970). Laboratory methods for work with plant and soil nematodes.London: Her Majesty's stationery office, p. 148.
Stirling GR (1991). Biological control of plant parasitic nematodes. Progress, problems and prospects.London: CAB International, p. 282.
Takahashi B, Eguchi T ,Yoneda K (1973).Studies on the flower formation in tomatoes and egg-plants I: Effect of temperature regimes and fertilizer levels on the flower bud differentiation in tomato. J. Jpn. Soc. Hort. Scl. 42: 147-157.
Taylor AL, Sasser JW (1978). Biology, Identification and Control of Root-knot Nematodes. Raleigh: North Carolina State Graphics, p. 111.
Uguru MI, Madukaife SO (2001).Studies on the variability in agronomic and nutritive characteristics of African yam bean, *Sphenostylis stenocarpa* (Hochst *Ex*. A. Rich.)Harms.Plant Prod. Res. J. 6(2): 10-19.
Viglierchio DR ,Yu PK (1968).Plant growth substances and plant parasitic nematodes. II: Host influence on auxin content. Exptal. Parasitol. 23: 88-95.
Volmar KM (1991).Abscisic acid and ethylene increase in *Heterodera avenae*-infected tolerant or intolerant oat cultivars.J. Nematol. 23: 425-431.
Williamson VM , Hussey RS(1996). Nematode pathogenesis and resistance in plants. Plant Cell, 8: 1735-1745.
Zohary M (1972). Flora Palaestina. Part two. Platanaceae to Umbelliferae. Jerusalem: The Israel Academy of sciences and Humanities, pp. 1-6.