HOST-PARASITE RELATIONSHIP OF OKRA AND SINGLE AND COMBINED INFECTION OF *MELOIDOGYNE JAVANICA* AND *FUSARIUM OXYSPORUM*

C.C. Iheukwumere[1]* C.I. Iheukwumere and C.U. Aguoru***

*Department of Biological Sciences, University of Agriculture, Makurdi, Benue State, Nigeria
**Department of Food Science and Technology, University of Agriculture, Makurdi, Benue State, Nigeria

Summary. An outdoor pot and a field experiments were conducted to investigate the host-parasite relationships of okra and *Meloidogyne javanica* in the presence and absence of *Fusarium oxysporum* f.sp. *vasinfectum*. Inoculation with the two pathogens, singly and in combination, significantly reduced all growth and yield components of okra. Single inoculation with *M. javanica* reduced plant growth more than inoculation with *F. oxysporum* alone. Reductions in plant growth caused by combined inoculation of the two pathogens were greater than those of single inoculation with either pathogen. In combined inoculations, damage was more severe when the inoculation of the nematode preceded that of the fungus in both the pot and field trials. Severity of root galling and nematode population in soil and roots were greater when the nematode was inoculated singly. Mixed infection reduced the severity of *M. javanica* in pot and field experiments, especially when *F. oxysporum* was inoculated prior to the nematode in the pot experiment.

Keywords: *Abelmoschus esculentus*, *Fusarium* wilt, pathogenicity, root-knot nematode.

Okra, *Abelmoschus esculentus* (L.) Moench, is an annual plant of the family Malvaceae. It is a tender vegetable (Kemble *et al*., 2007) that is especially important in West Africa, India, Brazil and the United States (ECHO, 2003; Alimi, 2004; Farinde *et al*., 2006) and can serve as a protein source (Vanlommel *et al*., 1996). Young fruits, harvested before differentiation of fibres and full seed developments, are consumed either alone or in salads, and after cooking in salty water are added to stews. They are also used in the preparation of certain African sauces (Farinde *et al.*, 2006). Leaves, buds and flowers are also edible. Dried seeds are nutritious and can be used to prepare vegetable curds or roasted and ground for use as a coffee additive or substitute (Farinde *et al*., 2006). Leaves are considered good cattle feed and okra mucilage is suitable for medicinal and industrial applications (Farinde *et al*., 2006).

In Nigeria, okra is grown in all states of the federation and suffers attacks from several viruses, insect pests and other disease agents, especially plant parasitic nematodes and fungal pathogens (Vanlommel *et al*., 1996; Farinde *et al*., 2006; Kemble *et al*., 2007). Mixed infections of the crop by root-knot nematodes and several fungal pathogens, especially *Fusarium oxysporum* f. sp. *vasinfectum* (Secht) Snyder *et* Hansen (Oyekan and Naik, 1987), are frequently found in okra fields. A preliminary survey of okra fields in Benue State, which is the "food basket of the nation", has shown several cases of mixed infection by *Meloidogyne javanica* (Treub) Chitw. and other fungal pathogens, including *F. oxysporum* f. sp. *vasinfectum* (Oyekan and Naik, 1987). *Meloidogyne javanica* is a serious pest of vegetables (Hashmi and Hashmi, 1990; Kemble *et al*., 2007) and is the predominant root-knot nematode species in Benue State (Adesiyan *et al*., 1990). Also, okra is reported to be highly susceptible to *F. oxysporum* in Nigeria (Marley *et al*., 2006).

Though there are reports on single and mixed infection effects of a root-knot nematode and *Fusarium* spp. on some crops (Sharma, 1990), literature on mixed infection of any *Meloidogyne* and *Fusarium* species on okra is presently lacking in Nigeria. Therefore, the aim of this study was to investigate the effects of the infection complex of *M. javanica* and *F. oxysporum* in okra and to generate basic information on this aspect of plant pathology, which has suffered long neglect in Nigeria.

MATERIALS AND METHODS

Pot experiment

Planting material. Seeds of okra cv. Annie Oakley, one of the most common cultivars of okra grown in Nigeria, were supplied by the National Cereals Research Institute (NCRI) substation at Yandev in Gboko, Benue State, Nigeria. They were surface-sterilized according to the methods of Koenning and McClure (1981) by dipping in 1.05% sodium hypochlorite solution for 5 minutes and were subsequently rinsed in six changes of sterile distilled water for 5 more minutes before planting. Seeds were sown at the rate of six per pot in 2.2 kg of autoclaved sandy-loam soil that was poured into 16-cm-diameter plastic pots placed on a concrete

[1] Corresponding author e-mail: ceceiheukwumere@yahoo.com

floor outside the University laboratory at Makurdi, Nigeria. Seedlings were thinned to one per pot 7 days after sowing.

Fungus isolation and identification. Diseased tissues from okra roots infected with *F. oxysporum* f. sp. *vasinfectum* under field conditions were cut into 1-2 mm diameter fragments and sterilized in 0.1% mercuric chloride for 2 minutes. They were rinsed in three changes of sterile distilled water before plating on water agar. The plates (9.3-cm-diameter) were incubated in the laboratory at room temperature (25 ± 2 °C) for 3 days and subsequently sub-cultured on Potato Dextrose Agar (PDA) plates (9.3-cm) (Chiejina, 2006). The PDA plates were then incubated for 7 days (Ataga and Ota-Ibe, 2006) and the cultures sub-cultured repeatedly on PDA plates until pure cultures of the isolate were established. At the conclusion of the incubation period, the fungus was identified based on growth-habit characteristics using a stereomicroscope (6-50×). Identification was made by following the descriptions of Alexopoulos *et al*. (2002). Discs cut from the 7-day-old cultures of the fungus with a 0.5-cm-diameter sterile cork borer (Ataga and Ota-Ibe, 2006) were repeatedly taken and placed on a Triple Beam Balance until a 1 g sample of the fungus culture was obtained. This amount of the fungus was used for the inoculation of each okra plant.

Nematode source and maintenance. The population of *M. javanica* was derived from a single egg mass taken from an infected tomato plant (*Solanum lycopersicum* L.) growing in a field previously planted to egg plant. The identity of the nematode was confirmed by observation of the perineal patterns prepared according to Hartman and Sasser (1985) and compared with the illustrations by Eisenback *et al*. (1981). The nematode was reared on tomato plants grown for 8 weeks in 20-21 cm-diameter polyethylene bags placed on a concrete floor outdoors. The tomato plants were carefully uprooted and their galled roots thoroughly washed under tap water. The roots were cut into 1-2 cm fragments and placed on modified Baermann's funnels (Whitehead and Hemming, 1965) for 18 hr to extract the nematodes. A millilitre of the nematode juvenile suspension was placed in a counting dish and the number of juveniles estimated under a stereomicroscope at a magnification of ×40. The J2 suspension was diluted with water to 500 J2s per ml and used for inoculation of the okra seedlings.

Ten-day-old okra seedlings were not watered the day preceding inoculation to avoid over-wetting of the soil during inoculation, which could result in draining of excess water and loss of some of the juveniles in the process. Inoculation was done by pouring a suspension of the required number of juveniles into a 0.5-1-cm-deep trench made around the root rhizosphere of the test plants, which was then re-closed with the excavated soil (Iheukwumere *et al*., 1995). *Fusarium oxysporum* was inoculated by inserting 1 g of the fungus culture into shallow holes made in the plant root rhizosphere at same depth as for the nematodes. Treatments consisted of plants inoculated with: *i*) 500 J2s only (N); *ii*) 1 g of the fungus culture only (F); *iii*) 500 J2s and 1 g of the fungus culture simultaneously (N + F); *iv*) 500 J2s followed by 1 g of the fungus culture 7 days later (N + f); *v*) 1 g of fungus culture followed by 500 J2s 7 days later (F + n); *vi*) no nematode or fungus, which served as control (C).

The experiment was arranged in a completely randomized design with five replicates per treatment on a concrete floor at 28 ± 3 °C. Plants were watered as needed and examined weekly for symptoms of wilting. The experiment was terminated 56 days after inoculation, when the plants had matured. Plant growth was determined by measuring shoot length, dry foliar weight, root length, dry root weight, and dry pod weight. Nematode infection was assessed by scoring root galling on a 0-5 rating index following the method of Taylor and Sasser (1978), as follows: 0 = no galls; 1 = 1-2 galls; 2 = 3-10 galls; 3 = 11-30 galls; 4 = 31-100 galls; 5 = more than 100 galls. The number of second stage juveniles of the nematode per treatment was determined by first carefully mixing the soil in each pot with a hand trowel. Then, nematodes from a 250 cm^3 soil sub-sample from each of the pots were extracted by combining the Cobb's sieving and decanting technique with the Baermann's funnel (Barker, 1985; Alam *et al*., 1990). Eggs and J2s of the nematode in 5 g of roots were extracted with sodium hypochlorite solution, according to the method of Hussey and Barker (1973).

All data were subjected to analysis of variance and means compared with Least Significant Difference (LSD). Data on root gall and nematode population were square root transformed before being subjected to analysis of variance.

Field experiment

The field trial was conducted in a sandy loam farmer's field at Makurdi, during the growth season June-August 2008. The field had previously been cropped to tomato and was naturally infested with *M. javanica*, but had not shown incidence of *F. oxysporum* or any other fungus infestation. The trial was arranged according to a randomized block design with each of the four blocks, of 56 m × 19.6 m, spaced 2 m from each other. Every block was of six plots, which measured 7 m × 2.4 m each, and each plot consisted of 6 rows of plants spaced 40 cm apart. Five seeds were sown per planting hole, 2-3 cm deep; plants were thinned to one per planting position 5 days after sowing and were spaced 70 cm apart along the rows (10 plants per row) (Atiri and Ibidapo, 1989). Inoculation with fungus was done 7 or 14 days after sowing, whereas nematode infection was by the naturally occurring nematode population in the soil. The outer rows of each plot and the last two plants on either end of each row were

not observed to avoid edge effects. The treatments were: *i*) okra seedlings exposed to natural infection by the nematode in the soil at planting (N); *ii*) seedlings inoculated with 1 g of the fungus culture 7 days after sowing on carbofuran (Furadan 10G) treated plots (F_7); *iii*) seedlings inoculated with 1 g of fungus culture 14 days after sowing on carbofuran treated plots (F_{14}); *iv*) seedlings exposed to natural infection by the nematode in the soil, followed by inoculation with 1 g of fungus culture 7 days after sowing ($N + f_7$); *v*) seedlings exposed to natural infection by the nematode in the soil, followed by inoculation with 1 g of fungus culture 14 days after sowing ($N + f_{14}$); *vi*) seedlings with no nematode or fungus treatment in plots treated with carbofuran, which served as control (C).

The fungus was inoculated as in the pot experiment. The nematicide carbofuran, as a granular formulation, was incorporated into the top 10-15 cm of soil at the rate of 3 kg a.i. per hectare (Atungwu and Kehinde, 2008) prior to sowing.

Nematode population (second stage juveniles: J2) was assessed before treating and sowing and at the end of the experiment (at harvest). At each of the sampling times, soil samples were collected from each plot by random collection along a zig-zag path of 50 2.5-cm-diameter soil cores taken to a depth of 15-20 cm within each plot (Windham and Barker, 1986), using a soil auger. Soil samples were immediately bulked in polyethylene bags, securely tied and labelled. In the laboratory, each sample was carefully and thoroughly mixed and nematodes from two 250 cm^3 sub-samples were extracted by combining Cobb's sieving and decanting technique with Baermann's funnels and counted under a stereomicroscope.

At the end of the experiment, 56 days after sowing, 24 plants per plot from the central four rows were carefully uprooted. Growth, yield components and root galling indices were evaluated. All data were statistically analyzed and means separated with LSD.

RESULTS

Pot experiment

Less than a week after the second inoculation (i.e. 5 days after 2nd inoculation; 22 days after sowing, DAS), infected plants showed mild signs of wilting. The severity of these symptoms was greatest in pots inoculated with N+F, N+f and F+n and were most evident at noontime, when the intensity of the sunlight was greatest. The severity of the plant wilting increased with time and by the 2nd week after inoculation (31 DAS) mild symptoms of leaf curling and chlorosis were evident. These symptoms were strongest on plants treated with N+F, N+f and F+n, followed by those infected with nematode only (N). The plants infected with only fungus (F) showed little or no signs of chlorosis.

By the 3rd week after inoculation (38 DAS), the severity of symptoms of the disease complex on plants with N+F, N+f and F+n inoculations had reached a level at which the plants could barely recover from the wilting. Similar symptoms, but less severe, were shown by plants inoculated with only nematodes (N). By this time, the plants infected with the fungus only (F) had begun to show clear but mild symptoms of chlorosis on their leaves. Leaves on some plants with N+F, N+f and F+n treatments had begun to senesce.

Senescence was observed during the 4th week (45 DAS) for some of the plants inoculated with N alone. This effect was not observed on plants inoculated with F alone. Visual inspections of the leaves showed that the size of the leaf lamina appeared to be reduced most in N+F, N+f and F+n treatments, followed by those inoculated with only N and, to a much less extent, by those inoculated with F only.

Examination of the roots, showed that single infection with N considerably reduced root growth and branching but no root decay was observed. Infection with F alone also led to reduction in root growth coupled with decay of some of the roots. When both pathogens were present, root growth was greatly reduced and root decay increased when N was inoculated

Table I. Effects of single and mixed inoculation of *Meloidogyne javanica* and *Fusarium oxysporum* in pots on shoot length, dry foliar weight, root length, dry root and dry pod weight of okra[1].

Treatment[2]	Shoot length (cm)	Leaf dry weight (g)	Root length (cm)	Root dry weight (g)	Pod dry weight (g)
C	47.7a	45.5a	39.4[a]	6.8a	42.1a
F	40.1b	38.2b	32.1b	4.9b	36.3b
N	31.8c	30.2c	23.9c	2.4c	29.9c
N + F	22.7d	19.2d	14.9d	2.0c	22.9d
N + f	17.2d	4.9f	7.0e	0.5e	10.8f
F + n	18.3d	10.4e	10.7e	1.2d	17.0e
LSD at 5%	7.44	4.09	4.10	0.61	5.09

[1]Each value is a mean of five replicates. Means followed by the same letter in each vertical column are not significantly different at $P \leq 0.05$.

[2]C = control no nematode or fungus inoculated; F = fungus; N = nematode; N + F = nematode and fungus inoculated simultaneously; N + f = nematode inoculation followed by the fungus 7 days later; F + n = fungus inoculation followed by the nematode 7 days later.

Table II. Effects of single and mixed inoculation of okra with *M. javanica* and *F. oxysporum* in pots on root galling, soil and root population densities of the nematode[1].

Treatment[2]	Gall index[3]	Juveniles[4] in 250 cm[3] soil	Eggs and juveniles[4] in 5 g root
C	-	-	-
F	-	-	-
N	4.8a	255[a]	85[a]
N + F	3.5c	91c	30c
N + f	4.3b	152b	51b
F + n	2.2d	50d	17d
LSD at 5%	0.4	20	11

[1]Each value is a mean of five replicates. Means followed by different letters in each vertical column are significantly different at $P \leq 0.05$.
[2]C = control no nematode or fungus inoculated; F = fungus; N = nematode; N + F = nematode and fungus inoculated simultaneously; N + f = nematode inoculation followed by the fungus 7 days later; F + n = fungus inoculation followed by the nematode 7 days later.
[3]Gall index: 0 = no gall; 1 = 1-2 galls; 2 = 2-3 galls; 3 = 11-30 galls; 4 = 31-100 galls; 5 = more than 100 galls (Taylor and Sasser, 1978)
[4]J2 = Second stage juveniles

prior to F. The reduction in root growth and the decay of roots of plants inoculated with N+F was less severe than in plants with N+f and F+n inoculations.

All treatments significantly reduced shoot and root lengths, and dry leaf, pod and root weights (Table I). In the single inoculations, *M. javanica* reduced plant growth more severely than the single inoculation with *F. oxysporum*. The combined inoculations N+F, N+f, and F+n caused greater plant growth reductions than the single inoculations with N or F. Moreover, with the exception of the effect on shoot length, the differences in the effects on dried foliar weight, root dry weight and pod dry weight were generally highly significant. Inoculation of the nematode 7 days prior to the fungus (N+f) reduced growth components more than in the N+F or F+n treatments. The only exceptions to this are for the effect on shoot length, for which the N+f treatment was similar to those of N+F and F+n, and for the effect on root length, where the effects were similar for the N+f and F+n treatments.

Root galling was more severe and nematode populations in soil were greatest when *M. javanica* was inoculated singly (Table II). *Fusarium oxysporum* clearly reduced the severity of damage from the nematode with the reduction being greater with F+n than N+f or N+F inoculations (Table II).

Field experiment. Symptoms on okra were similar to those observed in the pot experiment. However, visual observations after germination showed that, in the N, $N+f_7$ and $N+f_{14}$ treatments, plant vigour was hampered more than in the single infections with F_7 and F_{14}. Plant growth was visibly faster in the F_7 and F_{14} treatments and fastest in the control (C) plants. Again, disease severity also increased with time and was more evident in mixed than in single infections. In the N plots, plants had considerably more reduced and extensively decayed roots than in the F_7 or F_{14} treatments. Visual inspection of foliar and root systems showed that disease severity was in increasing order of magnitude for C, F_{14}, F_7, N, $N+f_{14}$ and $N+f_7$. On control plants, which had only a few galls on the roots, there were no visible foliar symptoms; generally, these plants grew well and better than the rest of the treated plants.

There were significant differences in shoot length among C, F_{14}, F_7 and N, in decreasing order of magnitude. However, no significant difference in shoot length was detected between $N+f_{14}$ and $N+f_7$, but plants in these treatments were significantly shorter than those in the C, F_{14}, F_7 and N treatments (Table III). Foliar dry weight also showed significant differences among treatment means, which were, in decreasing order of magnitude, for C, F_{14}, F_7, N, $N+f_{14}$ and $N+f_7$ at $P \leq 0.05$ (Table III). Data for root length and pod dry weight followed the same trend (Table III).

The nematode population before treating was evenly spread in the field and in the range 184-186 juveniles per 250 cm^3 of soil. But nematode population at harvest showed significant ($P \leq 0.05$) differences among the treatments and in decreasing order for the N, $N+f_{14}$ and $N+f_7$ treated plots. No significant differences in nematode population were detected among C, F_7 and F_{14} but each of these treatments had nematode populations significantly lower than those of N, $N+f_{14}$ and $N+f_7$ (Table IV). The root gall indices also differed significantly among the treatments: they were negligible in C, F_7 and F_{14}, but severe and in order of severity in $N+f_7$, $N+f_{14}$ and N (Table IV).

DISCUSSION

These studies demonstrate the vulnerability of okra to attack by *M. javanica* and *F. oxysporum* both individ-

Table III. Effects of single and mixed inoculation of *M. javanica and F. oxysporum* on shoot length, dry foliar weight, root length, root and pod dry weight of okra in the field[1].

Treatment[2]	Shoot length (cm)	Leaf dry weight (g)	Root length (cm)	Root dry weight (g)	Pod dry weight (g)
C	49.4a	233.4a	42.3a	20.6a	308.6a
F_7	32.1c	179.4c	25.7c	14.8c	233.9c
F_{14}	40.8b	206.4b	35.5b	17.7b	271.6b
N	23.1d	151.4d	20.2d	11.7d	195.3d
$N+f_7$	10.1e	93.0f	11.0f	8.7e	116.6f
$N+f_{14}$	13.0e	123.1e	16.0e	9.1e	156.6e
LSD at 5%	8.56	26.64	4.01	2.51	36.52

[1]Each value is a mean of four replicates with 24 plants sampled per treatment. Means followed by the same letter in each vertical column are not significantly different at $P \leq 0.05$.
[2]C = control, no nematode or fungus inoculated; F_7 = fungus inoculated 7 days after sowing; F_{14} = fungus inoculated 14 days after planting; N = natural inoculation with nematode at sowing; $N+f_7$ = plants naturally inoculated with nematode followed by inoculation with fungus 7 days later; $N+f_{14}$ = plants naturally inoculated with nematode followed by inoculation with fungus 14 days later.

Table IV. Effects of single and mixed inoculation of *M. javanica* and *F. oxysporum* on soil population densities and root gall index of the nematode in the field[1].

Treatment[2]	Mean pre-plant population ($J2^3$)	Mean final population ($J2^3$/250 cm^3)	Gall index[4]
C	186a	20d	1d
F_7	186a	15d	1d
F_{14}	185a	17d	1d
N	184a	378a	5a
$N+f_7$	186a	282c	3c
$N+f_{14}$	186a	320b	4b
LSD at 5%	12	27	0.9

[1]Each value is a mean of duplicate 250 cm^3 soil samples per each of the four replicates. Means followed by the same letter in each vertical column are not significantly different at $P \leq 0.05$.
[2]C = control, no nematode or fungus inoculated; F_7 = fungus inoculated 7days after sowing; F_{14} = fungus inoculated 14 days after planting; N = natural inoculation with nematode at sowing; $N+f_7$ = plants naturally inoculated with nematode followed by inoculation with fungus 7days later; $N+f_{14}$ = plants naturally inoculated with nematode followed by inoculation with fungus 14 days later.
[3]J2 = Second stage juveniles.
[4]Gall index: 0 = no gall; 1 = 1-2 galls; 2 = 2-3 galls; 3 = 11-30 galls; 4 = 31-100 galls; 5 = more than 100 galls (Taylor and Sasser, 1978).

ually and in combination. The data support work of others indicating that both pests are highly destructive pathogens of vegetable crops (Hashmi and Hashmi, 1990; Marley *et al.*, 2006; Kemble *et al.*, 2007).

In pots, the severity of the disease on okra was greatest in N+F or N+f and F+n inoculations. The fungus appeared to have had an inhibitory effect on the development and pathogenicity of the nematode, especially in the F+n treatment. Reduction of the nematode population in the presence of the fungus also occurred in the field. However, the methods used to extract nematodes from the soil do not extract eggs and, therefore, data on the final nematode soil population densities reported in Table IV must be considered underestimated. The reduction of the nematode population in the presence of the fungus could be attributed to root damage by the fungus, which possibly deprived the juveniles of the necessary feeding sites (Sakhuja and Sethi, 1986). Additionally, it is known that feeding sites of sedentary endoparasitic nematodes are preferable substrates for plant parasitic fungi (Sakhuja and Sethi, 1986). Abdel-Momen and Starr (1998) showed that giant cells were disrupted and damaged by fungal colonization of the root tissues, essentially because giant cells are nutrient-rich and thus suitable for fungal colonization. Further reasons for the inhibition of the nematode may be competition for nutrients (Jorgenson, 1970) and root space (Ketudat, 1969). In contrast, in pots in which the nematode was inoculated before the fungus (N + f), nematode pathogenicity and development was greater than in the (F + n) pots, but less than in those inoculated with the nematode only (N). This is probably because the nematode had time to establish before the fungus could grow and develop to a stage at which it could hinder its growth and pathogenicity. The inhibition of nematode reproduction in a host by a fungus has been demonstrated by other workers, such as Sharma (1990) on soybean [(Glycine max (L.) Merr.)] in the disease complex

caused by *M. incognita* (Kofoid *et* White) Chitw. and the soil-inhabiting fungi, *Fusarium* sp. and *Pythium* sp.

In the field, the results followed the same trend observed in the pot study, thereby corroborating the findings in that experiment. The field data further confirmed the negative impacts that single and combined infections of N, F or $N+f_7$ and $N+f_{14}$ could have on okra, with disease severity and growth reductions being higher in combined inoculations with $N+f_7$ and $N+f_{14}$. However, the better plant growth and yield performance of F_7 and F_{14} treatments compared to $N+f_7$ and $N+f_{14}$ treatments may partly be due to the insecticide effect of Furadan on other soil borne insects. Although okra plants treated with only N in pots had considerably reduced root growth with no decayed roots, those from the field were not only reduced but also extensively decayed. This was probably the consequence of interactions between the nematode and other biotic agents that probably were present in the field and not in the pot experiment, where sterilized soil was used.

In conclusion, the results verify and extend field observations on the single and combined effects of root-knot nematodes and the *Fusarium* wilt fungus on growth of okra in Nigeria. Therefore, further investigation should be undertaken under field conditions to obtain insights on the control of these serious diseases of okra and to formulate sound management measures to be used by farmers.

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LITERATURE CITED

Abdel-Momen S.M. and Starr J.L., 1998. *Meloidogyne javanica-Rhizoctonia solani* disease complex of peanut. *Fundamentals of Applied Nematology, 21*: 611-616.

Adesiyan S.O., Caveness F.E., Adeniji M.O. and Fawole B., 1990. *Nematode Pests of Tropical Crops*. Heinemann Educational Books, Nigeria Limited, Ibadan, Nigeria, 114 pp.

Alam M.M., Samad A. and Anver S., 1990. Interaction between *Tomato mosaic virus* and *Meloidogyne incognita*. *Nematologia Mediterranea*, *18*: 131-133.

Alexopoulos C.J., Mims C.W. and Blackwell M., 2002. *Introductory Mycology*, 4th Edition. John Wiley & Sons Inc., Singapore, 869 pp.

Alimi T., 2004. Use of cultural practices and economic impact of insecticide use, awareness and practices of insecticide safety precaution, on okra production. *Journal of Vegetable Crop Production, 10*: 23-36.

Ataga A.E. and Ota-Ibe G., 2006. Seed-borne fungi of the wild mango (Ogbono) (*Irvingia gabonensis* (Aubry-Leconte Ex O'Rorke) Bail) and their effects on food composition. *Nigerian Journal of Botany, 19*: 54-60.

Atiri G.I. and Ibidapo B., 1989. Effects of combined and single infections of mosaic and *Leaf curl viruses* on okra (*Hibiscus esculentus*) growth and yield. *Journal of Agricultural Science* Cambridge, *112*: 413-418.

Atungwu J.J. and Kehinde L.O., 2008. Evaluation of organic based fertilizer as an alternative to Furadan in the management of *Meloidogyne incognita* on soybeans in Nigeria. *International Journal of Nematology*, *18*: 61-65.

Barker K.R., 1985. Nematode extraction and bioassays. Pp. 19-35. *In*: An Advanced Treatise on *Meloidogyne* - Volume II Methodology (Barker K.R., Carter C.C. and Sasser J.N., eds). International *Meloidogyne* Project. North Carolina State University Graphics, Raleigh, N.C., USA.

Chiejina N.V., 2006. Potentials of the leaf extracts of *Azadirachta indica* A. Juss and *Ocimum gratissimum* L. for the control of some potato (*Solanum tuberosum* L.) fungal diseases. *Nigerian Journal of Botany, 19*: 68-73.

ECHO, 2003. *Plant information sheet*, N.F.T. Meyers, USA. *http://www.echonet.org*.

Eisenback J.D., Hirschmann H., Sasser J.N. and Triantaphyllou A.C., 1981. *A Guide to the Four Most Common Species of Root-knot Nematodes* (Meloidogyne spp.) *with Pictorial Key*. North Carolina State University Graphics, Raleigh, N.C., USA, 48 pp.

Farinde A.J., Owolarafe O.K. and Ogungbemi O.I., 2006. Assessment of production, processing, marketing and utilization of okra in Egbedere local government area of Osun State, Nigeria. *Journal of Agronomy, 5*: 342-349.

Hartman K.M. and Sasser J.N., 1985. Identification of *Meloidogyne* species on the basis of differential host test and perineal pattern morphology. Pp. 69-78. *In*: An Advanced Treatise on *Meloidogyne*, Volume II, Methodology (Barker K.R., Carter C.C. and Sasser J.N., eds). International *Meloidogyne* Project, North Carolina State University Graphics, Raleigh, NC, USA.

Hashmi S. and Hashmi G., 1990. Crop rotation and *Meloidogyne javanica* in vegetable production. *International Nematology Network Newsletter, 7*: 19-21.

Hussey R.S. and Barker K.R., 1973. A comparison of methods of collecting inocula of *Meloidogyne* spp. including a new technique. *Plant Disease Reporter, 57*: 1025-1028.

Iheukwumere C.C., Atiri G.L., Fawole B. and Dashiell K.E., 1995. Evaluation of some commonly grown soybean cultivars for resistance to the root-knot nematode and soybean mosaic virus in Nigeria. *Fitopatologia Brasileira, 20*: 190-193.

Jorgenson E.C., 1970. Antagonistic interaction of *Heterodera schachtii* (Schmidt) and *Fusarium oxysporum* (Woll.) on sugarbeet. *Journal of Nematology, 2*: 393-398.

Kemble J.M., Sikora E.J., Zehnder G.W. and Patterson M.G., 2007. Guide to commercial okra production. *http://www.aces.edu/pubs/docs/A/ANR-0959*

Ketudat U., 1969. The effects of some soil-borne fungi on the sex ratio of *Heterodera rostochiensis* on tomato. *Nematologica, 15*: 229-233.

Koenning S.R. and McClure M.A., 1981. Interaction of two potyviruses and *Meloidogyne incognita* in chilli pepper. *Phytopathology, 71*: 404-408.

Marley P.S., Kroschel J. and Elzein A., 2006. Host range of *Fusarium oxysporum* (PSM 197) to be used as a mycoherbicide for the control of striga *Hermonthica* in West Africa. *Nigerian Journal of Botany, 19*: 17-28.

Oyekan P.O. and Naik D.M., 1987. Fungal and bacterial diseases of soybean in the tropics. Pp. 47-52. *In*: Soybean for the Tropics: Research, Production and Utilization (Singh S.R., Rachie K.O. and Dashiell K.E., eds). John Wiley and Sons Limited, New York, USA.

Sakhuja P.K. and Sethi C.L., 1986. Multiplication of *Meloidogyne* as affected by *Fusarium solani* and *Rhizoctonia bataticola* on groundnut. *Indian Journal of Nematology, 16*: 1-3.

Sharma N., 1990. A disease complex of soybean involving nematode, *Meloidogyne incognita,* and the soil-inhabiting fungi, *Fusarium* sp. and *Pythium* sp. *International Nematology Network Newsletter*, *7*: 17-19.

Taylor A.L. and Sasser J.N., 1978. *Biology, Identification and Control of Root-knot Nematodes* (*Meloidogyne* spp.). North Carolina State Graphics, Raleigh, NC., USA, 111 pp.

Vanlommel S., Duchateau L. and Coosemans J., 1996. The effect of okra mosaic virus and beetle damage on yield of four okra cultivars. *African Crop Science Journal, 4*: 71-77.

Whitehead A.G. and Hemming J.R., 1965. A comparison of some quantitative methods of extracting small vermiform nematodes from soil. *Annals of Applied Biology, 55*: 25-38.

Windham G.L. and Barker K.R., 1986. Relative virulence of *Meloidogyne incognita* host races on soybean. *Journal of Nematology*, *18*: 327-331.

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