Journal of Agriculture and Ecology Research International
6(3): 1-6, 2016; Article no.JAERI.23382
ISSN: 2394-1073

SCIENCEDOMAIN ***international***
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The Impact of Organic Soil Amendments on Population Densities of Plant Parasitic Nematodes and Okra Yield

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Authors' contributions

This work was carried out in close collaboration with all authors. Author DY initiated the study, devised the protocol and drafted the first manuscript. Author OK perfected the statistical analyses and managed the final manuscript. Author AJ collected and managed the field data. Authors SAJ and AB extracted, preserved, quantified and identified the plant parasitic nematodes. All authors read and approved the final manuscript.

Article Information

DOI: 10.9734/JAERI/2016/23382
Editor(s):
(1) Małgorzata Pietrowska-Borek, Department of Biochemistry and Biotechnology, University of Life Sciences, Poznan, Poland.
(2) Krzysztof Skowron, Department of Microbiology, Nicolaus Copernicus University in Torun, Collegium Medicum of L. Rydygier in Bydgoszcz, Poland.
Reviewers:
(1) Montaser Fawzy Abdel-Monaim, Agricultural Research Center, Giza, Egypt.
(2) Tunira Bhadauria, Kanpur University, India.
(3) Anonymous, Embrapa Research Station, Brazil.
(4) Mahmoud M. A. Youssef, Nematology Laboratory, National Research Centre, Cairo, Egypt.
Complete Peer review History: http://sciencedomain.org/review-history/13159

Original Research Article

Received 28th November 2015
Accepted 18th January 2016
Published 3rd February 2016

ABSTRACT

Damaged okra plant roots as a result of plant parasitic nematodes infection leads to a reduced water and nutrients intake. The impact of some organic soil amendments on population densities of plant parasitic nematodes and okra yield was investigated between March and December 2013 in Ghana under natural field conditions. Neem (*Azadirachta indica*) seed and cocoa (*Theobroma cacao*) bean testa powders suppressed population densities of *Meloidogyne incognita*, *Pratylenchus coffeae*, *Rotylenchulus reniformis* and *Helicotylenhus multicintus* in 100 cm^3 rhizosphere soils and 5 g roots weight. Suppression in population densities of the plant parasitic nematodes did not reflex into yield improvement.

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Keywords: Okra yield; organic amendments; plant parasitic nematodes; population densities.

1. INTRODUCTION

Okra (*Abelmoschus esculetnus*) is found in nearly every market in Ghana. It is an important constituent of most local dishes and one of the important vegetables with nutritional values. It is successfully grown across the forest, transitional and savanna zones of the sub region. In Ghana, okra is among the non-traditional export crops of importance, constituting approximately 0.02% of gross domestic product [1] and the fourth most popular vegetable crop after tomatoes, pepper, and garden eggs [2].

Okra is however, extremely susceptible to plant parasitic nematodes (PPN) infection. *Meloidogyne* species especially excite conspicuous galls on the roots and the roots succumb to other microbial infections leading to rotting [3]. The damaged roots result in a reduced water and nutrients uptake. The leaves would suffer and the plants struggle to put on marketable fruits.

Most okra growers employ crop rotation and synthetic nematicides to combat the plant parasitic nematodes menace. Crop rotation has been used with a measure of success, but the extensive host range of the soil-borne pests makes selection of rotational crops difficult [4]. Export markets are also increasingly concerned about chemical residues in vegetable products from African markets [5]. Environmental hazards and consumers health as a result of abuse of chemical nematicides demands alternative methods of managing plant parasitic nematodes [6].

Some organic soil amendments (OSA) have been useful for the management of soil-borne pests. They are harmless to humans, livestock and the environment. They are also used for improving soils' physical conditions and adding to soil nutrients content. OSA promotes the growth of saprophytic nematodes and nematode trapping fungi which attack larvae of some genera of PPN.

In this work, the impact of neem seed and cocoa bean testa powder on population densities of plant parasitic nematodes and okra yield was investigated.

2. METHODOLOGY

2.1 Study Site

The experiment was sited at Fumesua (6° 43 ′N, 1° 36 ′W) in the Ejisu-Juaben Municipality of the Ashanti Region, Ghana. The area falls in the semi-deciduous forest zone and experiences a bi-modal rainfall regime. Fumesua soils belong to the ''Bomso series'' Ferric Acrisol with an average pH of 4.7 [7]. Soil had been cropped to *Zea mays* in the last two cropping seasons. Common weed species at the site were *Imperata cylindrica* and *Euphorbia heterophyla*. The major rainfall season is usually from April to August whilst the minor from September to November annually. The entire study was under rain-fed conditions. Agro-meteorological data from the nearest weather station (6° 41 ′N, 1° 33 ′W; altitude: 261.4 m) over the study period has been presented below (Table 1).

2.2 Field Layout and Soil Sampling

The experimental field was manually slashed with cutlass and all above-ground biomass removed until the soil surface was revealed. Randomized Complete Block Design (RCBD) with three replications separated by 2 m alleys was used. Planting distance used was (1 x 1 m). Each plot measured (3 x 4 m) with 2 test and 2 border rows. Each row contained 5 hills and a total of 20 hills per plot. Total land area was 499 m^2 (26 x 19 m). Representative soil samples were taken from each plot to determine the initial and final nematode population densities. Soil sampling was done along the Z-plane up to about 20 cm deep with a soil augur.

Table 1. Agro-meteorological data from nearest weather station (2013)

0900 hrs					1500 hrs			
Temperature (℃)				RH	Temperature (℃)		Rainfall† (mm)	Vapour Pressure (mbs)
Max.	Min.	Dry bulb	Wet bulb	(%)	Dry bulb	Wet bulb		
31.8	22.0	25.2	23.0	82	31.0	24.2	1,379.1	25.7

Max. = Maximum temperature, Min. = Minimum temperature, RH = Relative humidity; † = Total rainfall

2.3 Treatments

Air-dried neem seeds and cocoa bean testa were pounded separately into nearly powder. They were measured into; 80 g neem seed powder (NSP), 40 g NSP, 80 g cocoa bean testa powder (CBTP), 40 g CBTP per plant and control (unamended plot) to make up the experimental treatments. The treatments were applied 3 weeks before okra seeds were sown as spot application at about 2 inches away from each hill.

2.4 Cultural Practices and Data Collection

Three seeds were sown per hill and thinned out to one after germination. The plot was kept weed-free throughout the study with hoe and cutlass. Insect pests were managed with Cypercal 50 (Cypermethrin 50 g/l; EC) insecticide. Morphological data were collected on six tagged plants randomly selected from the test rows on each plot. Plant height and stem girth were taken at fortnightly intervals. Dry shoot biomass was taken after fresh shoot samples were oven-dried at 80℃ for 48 h. Second, third and fourth stage nematodes per 100 cm^3 rhizosphere soil and 5 g root weight were determined. Mature okra pods were harvested and quantified accordingly every other day. Yield of okra fruits was expressed as weight of production per plot and subsequently converted to tons/ha.

Determination of nematodes from soil samples followed the modified Baermann funnel extraction method [8]. Eggs were extracted from 5 g root weight subsamples [9] and incubated [10] for juveniles. The nematodes in the samples were heat-killed in an oven at 60℃ for about 3 minutes and few drops (2 or 3) of 4% formaldehyde (formalin) added. Microscopic examination and counting were done on counting tray [11] with the aid of a tally counter.

Nematode counts were log transformed (ln (x + 1)) to improve homogeneity of variance before ANOVA was performed using GenStat (12.0). Significant mean separation was done with Least Significance Difference (LSD) test (*P*=.05).

3. RESULTS AND DISCUSSION

Plant parasitic nematodes recovered from initial soil samples in the order of abundance were; *Rotylenchulus reniformis, Pratylenchus coffeae, Meloidogyne incognita* and *Helicotylenchus multicintus* (Fig. 1).

Differences in okra plant height, number of leaves, stem girth and dry shoot weight in both the major and minor seasons (Figs. 2 and 3) were not clear among the treatments.

Various products prepared from the leaves and seeds of neem plants have been reported as effective protectants against nematode pests (12). In this work, the control (unamended plot) recorded high population densities of *Meloidogyne incognita*, *Rotylenchulus reniformis* and *Helicotylenchus multicintus* in final rhizosphere soil. These population densities were significantly (*P*=.05) different from those of the amended plots (Table 2).

Control effect on *Pratylenchus coffeae* population density was significant (*P*=.05). It was 71%, 67%, 71% and 52% over the 80 g NSP, 40 g NSP, 80 g CBTP and 40 g CBTP amended plots, respectively. Treatments effect on population densities on the amended plot was not different except the 40 g CBTP which significantly suppressed *Meloidogyne incognita, Pratylenchus coffeae and Rotylenchulus reniformis* in rhizosphere soils.

When soils were amended with organic matter, some plant pathogens were hindered in their development or their antagonists increased in number. Nemato-toxic compounds of neem plants, especially azadirachtins were released through volatilization, exudation, leaching and decomposition [12,13]. Neem leaf amended soil recorded a significant reduction of *M. incognita* over the untreated control plot in okra under field conditions [14]. *Meloidogyne incognita* and *Pratylenchus coffeae* were recorded from roots and their densities in the control plot were higher. Eighty (80) g NSP amended plot recorded the least in *M. incognita* population density per 5 g root weight but was not different from those of the 40 g NSP and 80 g CBTP amended plots. *Pratylenchus coffeae* recovered from 40 g CBTP amended plot was the least in population density per 5 g root weight which was 98.5% less than that of the control plot (Table 3).

Differences in fruit yield in both seasons were not clear. It implied that, the treatments did not have any significant (*P*=.05) effect on fruit yield. In the major season, 80 g NSP and 80 g CBTP amended plots recorded the highest (1.37 t/ha) and least (0.67 t/ha), respectively. In the minor season however, 40 g CBTP amended plot recorded the highest (1.58 t/ha) in fruit yield whilst 80 g CBTP recorded the least (1.11 t/ha).

The control plots yielded 0.90 and 1.42 t/ha over the two seasons, respectively (Table 4).

Okra fruit yield was generally higher in the minor season than the major. This could be attributed to the cumulative beneficial effects of the treatments. Lower fruit yield realized was partly due to the wider planting distance used which resulted in a lower plant population density. This is because higher plant population density could lead to increased yield resulting from greater number of fruits per unit area.

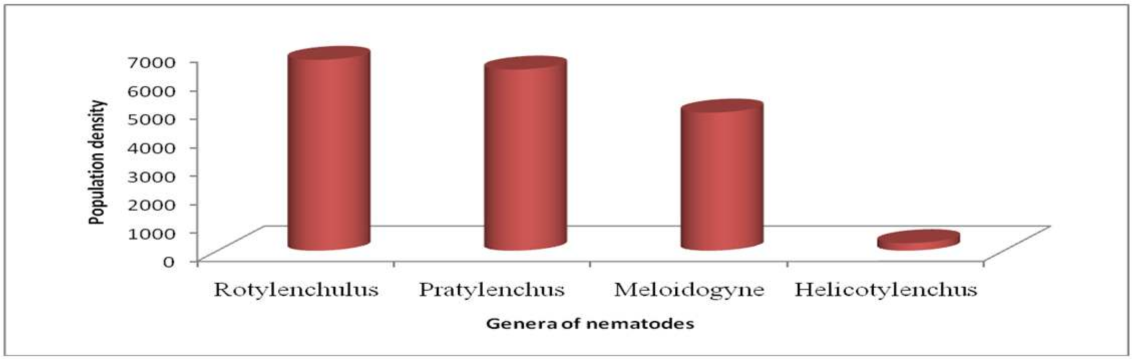

Fig. 1. Initial plant parasitic nematodes population densities; *Rotylenchulus reniformis*, *Pratylenchus coffeae*, *Meloidogyne incognita* and *Helicotylenchus multicintus*

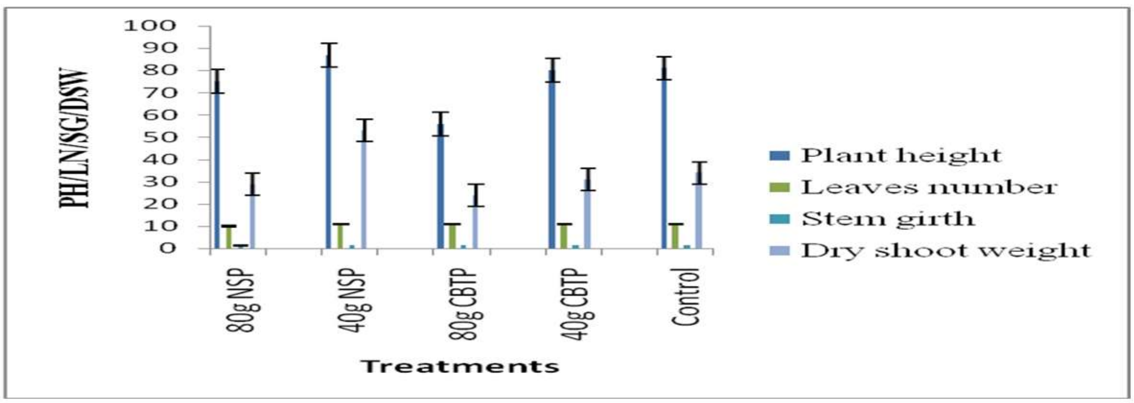

Fig. 2. Morphological parameters recorded in the major season; PH (Plant height), LN (Leaves number), SG (Stem girth), DSW (Dry shoot weight)

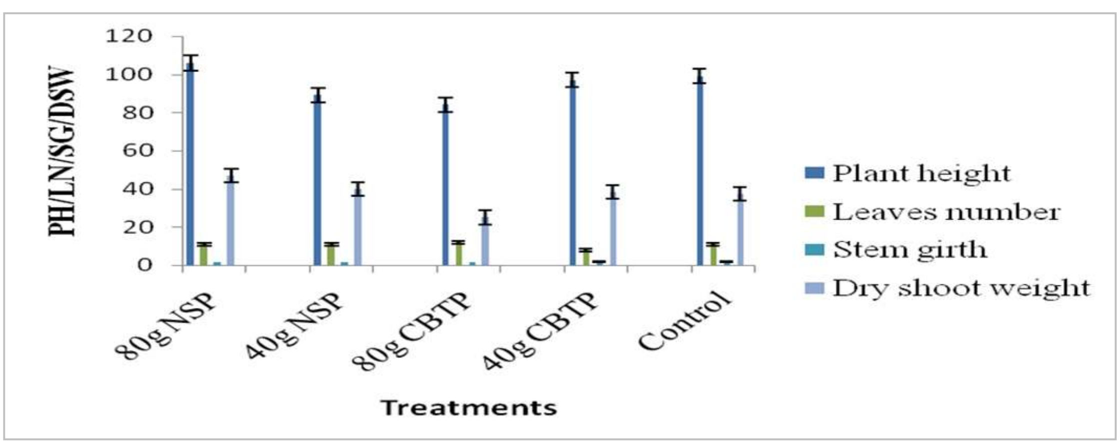

Fig. 3. Morphological parameters recorded in the minor season; PH (Plant height), LN (Leaves number), SG (Stem girth), DSW (Dry shoot weight)

Table 2. Nematodes densities/100 cm^3 soil at final harvest

Treatment	*Meloidogyne incognita*	*Pratylenchus coffeae*	*Rotylenchulus reniformis*	*Helicotylenchus multicintus*
80g NSP	24 (5)*	37 (6)	561 (20)	25 (5)
40g NSP	69 (8)	50 (7)	757 (23)	24 (5)
80g CBTP	59 (7)	35 (6)	579 (18)	23 (5)
40g CBTP	158 (11)	103 (10)	1752 (42)	25 (5)
Control	535 (23)	455 (21)	5536 (74)	165 (13)
LSD	(3.3)‡	(1.2) ‡	(10.8) ‡	(1.1) ‡

**Log transformed (ln (x + 1)) data used in analysis in parentheses, ‡ = Significant at 5% level*

Table 3. Nematodes population densities/5 g root weight

Treatments	*Meloidogyne incognita*	*Pratylenchus coffeae*
80g NSP	3 (2)*	25 (5)
40g NSP	19 (4)	50 (7)
80g CBTP	22 (5)	42 (6)
40g CBTP	72 (8)	4 (2)
Control	3629 (59)	166 (33)
LSD	(1.2)‡	(1.3) ‡

**Log transformed (ln (x + 1)) data used in analysis in parentheses, ‡ = Significant at 5% level*

Table 4. Yield of okra (t/ha)

Treatment	Major season	Relative yield	Minor season	Relative yield
80g NSP	1.37	2.04	1.49	1.34
40g NSP	0.80	1.19	1.28	1.15
80g CBTP	0.67	1.00	1.11	1.00
40g CBTP	0.89	1.32	1.58	1.42
Control	0.90	1.34	1.42	1.27
LSD 5%	(0.21)NS		(0.37)NS	

Relative yield = Ratio of a particular treatment on the least yielding treatment, NS = Not significant

Organic soil amendments have nematicidal effect on plant parasitic nematodes. Plant parasitic nematode population levels may change due to toxic metabolites release from organic amendments breakdown [15]. Evaluating the potential of cocoa bean testa powder to control plant parasitic nematodes in *Dioscorea rotundata,* nematode population densities were suppressed in rhizosphere soils and yam tubers but there were no differences in tuber yield [16]. In this work, there was significant treatments effect on plant parasitic nematodes population densities but no differences in fruit yield were observed.

4. CONCLUSION

The organic soil amendments evaluated suppressed population densities of plant parasitic nematodes in rhizosphere soils and roots but did not significantly improve okra fruit yield, plant height, number of leaves, dry shoot weight and stem girth.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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Peer-review history:
The peer review history for this paper can be accessed here:
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