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Management of root-knot nematode (*Meloidogyne incognita*) on cowpea (*Vigna unguiculata* L. Walp.) with oil cakes

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Abstract

Nematodes pose a threat to cowpea (*Vigna unguiculata* L. Walp.) production. Since management of phytopathogens by plant extracts are safer than agrochemicals, the efficacy of oil cakes of Indian almond (*Terminalia catappa*), Palm kernel (*Elaes guineensis*) and Shea nut (*Vitelleria paradoxa*) against root-knot nematode (*Meloidogyne incognita*) was tested in a Completely Randomised Design with four replications. Cowpea plants were inoculated with 1000 root-knot-nematode eggs/100g of soil two weeks after sowing before measuring plant height, number of leaves, canopy spread, number of pods/plant, number of seeds/pod, root galling and root-knot nematode population. Indian almond cake-treated plants had a significantly lower ($P<0.05$) root-knot nematode population and infection index (root gall) and a significantly higher ($P<0.05$) yield than the other treatments and the control. Since Indian almond cake was the most effective in reducing galling and nematode population, farmers can use it as a soil amendment.

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Introduction

Cowpea (*Vigna unguiculata* (L.) Walp.) is a popular and nutritionally important legume crop (Singh *et al*., 1997) which is now widely cultivated throughout the tropics and subtropics (Duke, 1990). In Africa, the average cowpea grain yields vary from 50 to 550 kg per ha, typically being only about 10-20% of cowpea yield potential under optimal growing conditions (Hall *et al*., 2003). Addition of cowpea in the traditional farming system is most appropriate as it supports sustainable land use by improving soil fertility and serving as rotation crop. However, tropical conditions meditated by high temperatures as found in Ghana, favour high reproduction rates of plant parasitic nematodes particularly *Meloidogyne incognita* (Bridge *et al*., 2005) and resulting high yield losses may be incurred. Yield losses of between 73-100% were reported in Northern Ghana due to root-knot nematode (Hemeng, 1981) while Sawadogo *et al*. (2009) reported that in the humid southwestern part of Burkina Faso, at least 90% of cowpea fields were infested with root-knot nematodes. In most of West Africa, pests are reported to be the single most important constraint to cowpea production (Singh *et al.,* 1990). Symptoms of damage induced by root-knot nematode (*Meloidogyne* spp.) include development of chlorosis on leaves, stunted growth, root galling, and excessive branching of the root (Adegbite, 2011). Heavy infestation of cowpea by *Meloidogyne* spp. leads to early senescence of the crop (Olowe, 2004). Nematode infection impairs the function of the root system (Mishra, 1992). When nematodes build up on the cowpea, they can cause damage leading to yield loss (Gallaher and McSorley, 1993). Rose *et al*. (1989) reported yield losses of more than 90 % in high population of root-knot nematodes.

Several investigators have reported a reduction in the population level of root-knot nematodes following the addition of soil amendments, such as oil seed cake of mung bean (*Vigna radiata*) (Tiyagi and Alam, 1994; Kolade *et al*., 2005). Generally nematicides are recommended for the control of nematodes but they are harmful to beneficial flora and fauna of the soil. The environment is also affected as a result of continuous nematicide application (Thomson, 1992). There are instances where nematicides have been found to contaminate the underground water and thus they have potential to be toxic to human and farm animals (Alam and Jairajpuri, 1990). Plant products such as oil seed cakes have shown success in plant disease control and are known to be harmless, eco-friendly, non-phytotoxic and readily available (McSorley *et al.,* 1999). The study aimed to determine the effectiveness of oil cakes of Indian almond (*Terminalia catappa*), Palm kernel (*Elaes guineensis*) and Shea nut (*Vitelleria paradoxa*) against root-knot nematode of cowpea.

Materials and methods

Experimental site

Laboratory experiments were conducted at the Spanish laboratory of the University for Development Studies, Nyankpala, Ghana while other experiments were conducted during the 2011 cropping season at the screen house of the Faculty of Agriculture, University for Development Studies, Nyankpala Campus. The area is located 16km West of Tamale with latitude of 9°25'N and longitude 0°58'W (SARI, 2006). The screen house constructed with plant table net operates under natural conditions of 12 hours of daylight. No artificial light was used.

Source of seed and oil cakes

Cowpea seeds of the 'Paditua' variety released by Savannah Agricultural Research Institute (SARI), Nyankpala, Ghana were used for the study. Shea nut cake was obtained from Nyankpala market in the Northern Region. Indian almond cake and palm kernel cakes were obtained from Kwahu-Tafo in the Eastern Region.

Soil preparation and sterilization

Top soil was collected from the experimental site and sieved to remove large particles, stones, plastic materials and plant debris. It was then mixed with river sand in the ratio 3:1. The mixing was done on a concrete floor to obtain a uniform mixture. The soil mixture was moistened, and then put into a cut drum before heating at 85°C for 20 minutes and then cooling overnight on a large metal sheet.

After cooling, four 5 kg heaps of the sterilized soil were separately placed on thirty-two trays. Twenty-four of the heaps were each thoroughly mixed with 15 g of oil seed cakes whilst the remaining eight control pots received no oil seed cake. Five kilograms (5 kg) of each of the resulting mixtures was placed into cylindrical plastic pots of 18 cm radius. To allow proper decomposition before the cowpea seeds were sown, the mixtures were watered every 5 days for 3 weeks.

Cultural practices

Four seeds of cowpea variety 'Paditua' were sown per pot. One week after sowing, the seedlings were thinned out to two healthy seedlings per pot. The pots were arranged in a completely randomized design (CRD) on benches in a screen house at a mean temperature of 28°C. Two weeks after sowing, each of the treated cowpea seedlings was inoculated with 1000 eggs/ml egg suspension of *Meloidogyne incognita*. All plants of the various treatments and control received one liter of tap water daily for 3 months.

Extraction of nematode eggs

Extraction of *Meloidogyne incognita* from infested cowpea roots was based on the method described by Hussey and Barker (1973). Galled root pieces of cowpea containing egg masses were cut into small pieces, and placed in a 500 ml beaker and mixed with 200 ml of 0.5% chlorox (NaOCl) solution by shaking vigorously for 3 minutes. This was done in order to digest the gelatinous matrix encasing the eggs. The solution was poured through two nested sieves; 200 mesh (75 μm) and 500 mesh (25 μm). Eggs in the 200 mesh sieves were washed free of NaOCl solution with cold tap water into a one liter beaker. The cut roots in the original beaker were washed twice with tap water to obtain additional eggs. The number of eggs per 2 ml of water was estimated by counting 3 samples of 2 ml each using a Doncaster counting dish under a stereo microscope resulting in an average of 199.6 eggs/ml.

Counting of root-knot nematode eggs

The suspension of eggs was stirred continuously to ensure uniform distribution before aliquots were taken. Aliquots of 1 ml of suspension were drawn into the Doncaster counting tray to determine the number of eggs per unit volume of the suspension. With the help of a hand tally counter and a stereo microscope, the eggs were counted. The counting was done three times and recorded. The total number of eggs in 1 ml was multiplied by the total egg suspension to obtain the total number of nematode eggs in the suspension.

Inoculation of cowpea plants

Two weeks after sowing, each of the cowpea plants was inoculated with 1000 eggs/1ml of egg suspension of *Meloidogyne* spp. The inoculum was introduced into the soil with a pipette through three holes which were 2 cm from the point of sowing. The holes were covered lightly with soil after the inoculation. A final application was carried out at four weeks after the first application. Watering was done during evenings.

Experimental design

The pots were arranged in a completely randomized design (CRD). There were four treatments each replicated four times. The treatments were the application of 15 g of Indian almond cake, palm kernel cake, Shea nut cake, and a control (with no oil seed cake).

Data collection

Data were collected on the following parameters: plant height, total number of branches, canopy size, number of pods per plant, number of seeds per pod, and estimation of root-knot nematode population in the soil. To determine galling, at sixty days after inoculation, the plants were uprooted from the soil gently and pods harvested. The plants were then washed in a bucket of water and dipped into a beaker containing water. Galls and number of eggs in egg masses were observed through the beaker and rated with Bridge and Page (1980) gall rating chart.

Data analysis

Data were subjected to analysis of variance with

Genstat (Discovery Edition 4), and treatment means were separated using the least significant difference (LSD) at 5%.

Results

Effect of soil amendment with oil cakes on growth and yield of cowpea

Plant height

There were significant differences (P<0.05) in plant height at 2, 4 and 6 weeks after planting (WAP) (Fig. 1). The palm kernel cake treatment produced the tallest plants at 2, 4 and 6 WAP. However, at 8 WAP, Indian almond cake produced the tallest plants followed by those treated with palm kernel cake, Shea nut cake and the control.

Table 1. Effect of oil cake on number of pods per plant and number of seeds per pods.

Treatments	Pods/plant	Seeds/Pod
Control	2.67	9.33
Indian almond cake	3.67	15.67
Palm kernel cake	3.33	11
Shea nut cake	3.67	14
LSD (0.05)	*0.99*	*2.28*

Number of leaves

Plants grown in soil treated with palm kernel cake had a significantly higher (P<0.05) mean leaf count than the other treatments and the control from 2 – 8 WAP (Fig. 2).

Canopy spread

Palm kernel cake – treated plants produced wider canopies than the other treatments and the control but the differences were not significant (P>0.05) (Fig. 3).

Table 2. Effect of oil cake treatments on root galling in cowpea.

Treatments	Root galling index	Egg mass
Control	5.25	3.43
Indian almond cake	0.00	0.00
Palm kernel cake	4.50	3.00
Shea nut cake	3.25	0.75
LSD (0.05)	*1.57*	*0.80*
CV%	*31.4*	*25.39*

Scale: 0-10, where 0 = no gall, 10 = severely galled and plant usually died.

Number of pods per plant and number of seeds per pod

Plants treated with Indian almond and Shea nut cakes produced the same number of pods per plant (3.67) which was higher than those of other treatments and the control but the differences were not significant (P>0.05). Plants treated with Indian almond cake produced a higher number of seeds per pod (15.67) which was significantly higher (P<0.05) than those produced by other treatments and the control except Shea nut cake (Table 1).

Effect of soil amendment on root nematode infestation of cowpea:

Root galling and egg mass

Plants treated with Indian almond cake produced the least number of galls and egg masses followed by those treated with Shea nut cake, palm kernel cake and the control and the differences were significant (P<0.05) (Table 2).

Effect of oil cake on root-knot nematode population

Indian almond cake-treated plants recorded a significantly lower (P<0.05) population of nematodes followed by Shea nut cake (1.89), palm kernel cake (2.43) and the control (3) (Table 3).

Table 3. Effect of oil cake treatments on root-knot nematode population of cowpea.

Treatments	Untransformed mean	Transformed mean
Control	1005	3.00
Indian almond cake	22	1.36
Palm kernel cake	267	2.43
Shea nut cake	77	1.89
LSD (0.05)	*92.6*	*1.97*
CV (%)	*17.5*	*1.27*

Log (x+1) transformed, where x = mean count.

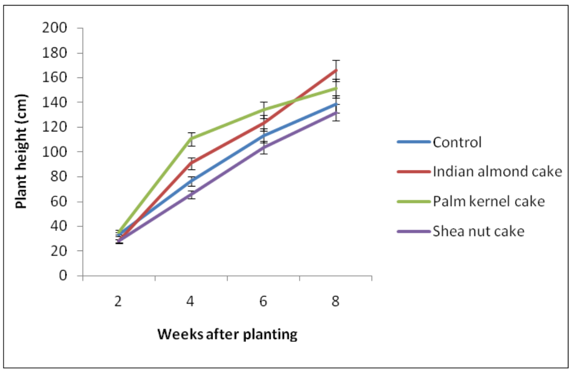

Fig. 1. Effect of oil cake treatments on plant height.

Discussion

Effect of oil cakes on plant growth and yield

Plants treated with palm kernel cake produced the tallest plants from 2 to 6 WAP but Indian almond cake treated plants were the tallest at 8 WAP. Plants treated with palm kernel cake produced more leaves and wider canopies than the other treatments and the control. Additions of oil cakes to the soil could have increased the organic matter content, thus favoring growth of the plants and also increased the water holding capacity of the soil. The promotion of growth by palm kernel cake confirms the findings of Kolade *et al.* (2005) who reported that palm kernel waste promoted plant growth parameters such as plant height, stem girth, canopy spread and number of leaves. The relatively poor performance of Shea nut cake treatment with respect to growth could be due to the presence of some anti-nutritive factors such as oxalate, tannin and phytate in the leaves and other parts of the tree (AOAC, 1999). A similar report was made by Sikora (1992) that poor performance of Shea nut cake in terms of growth could be due to the presence of oxalate, tannin and phytate, which are anti-nutritional factors that can impede the absorption of minerals like iron, magnesium, potassium and calcium, which are essential for metabolism of the plant.

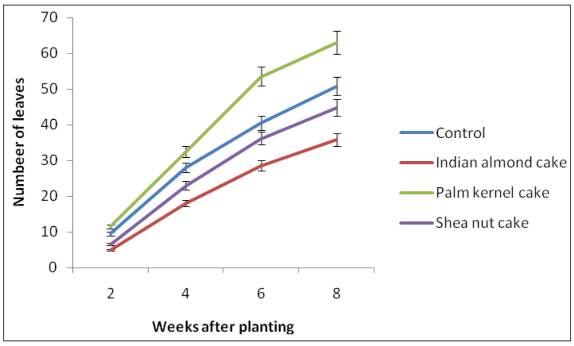

Fig. 2. Effect of oil cake treatments on number of leaves.

Plants treated with Indian almond and Shea nut cakes produced a higher number of pods per plant but they were not significantly different (P>0.05) from the other treatments and the control. Indian almond treated plants produced a significantly higher (P<0.05) number of seeds per pod than the other treatments and the control except Shea nut cake. The control plants developed poorly probably due to heavy infestation which could have resulted in lower numbers of pods per plant and numbers of seeds per pod, a confirmation of Tiyagi and Alam (1994) report that nematode infestation can result in 50-80% yield loss. Rachie and Lawal (1975) also reported that *Meloidogyne* spp. is the most economically important plant parasitic nematode attacking cowpea plants. The attack can be very severe due to fast development of the nematode population in the soil which is confirmed by the higher population of nematodes recorded in control treatments. Gallaher and McSorley (1993) also confirmed that *Meloidogyne* spp. build up on cowpea plants eventually hampers normal development and causes yield loss.

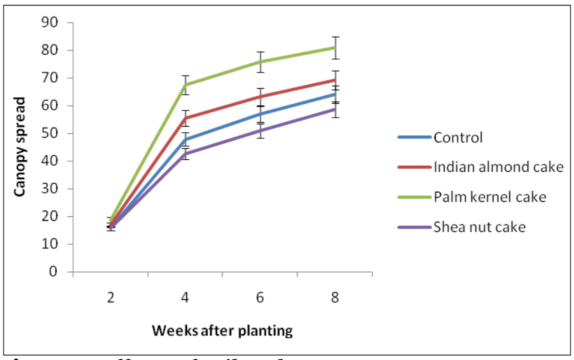

Fig. 3. Effect of oil cake treatments on canopy spread.

Effect of oil cakes on root-knot nematode infestation

Indian almond cake treated plants had the least number of galls and egg masses thus making it the most effective suppressant of gall formation. Although palm kernel cake promoted growth to a greater extent, Indian almond cake inhibited galling best and had the lowest nematode population in treated soil. The lower incidence of galling in oil cake-treated plants could be due to the production of toxic metabolites during microbial degradation of organic matter as reported by Sikora (1992). It could also be attributed to an increase in the number and density of antagonist of root-knot nematodes resulting in an increase in the overall antagonistic potentials in the soil (Fernandez *et al.*, 1989). The lower incidence of root-knot nematode in oil cake-treated plants could also be due the generation of toxic amounts of ammonia (NH_3).

Conclusion

Although palm kernel cake promoted growth, Indian almond cake completely inhibited galling. Since Indian almond cake completely inhibited galling, its use by farmers as a soil amendment should be encouraged.

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