Olabiyi et al., *Cogent Food & Agriculture* (2016), 2: 1207843
http://dx.doi.org/10.1080/23311932.2016.1207843

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SOIL & CROP SCIENCES | RESEARCH ARTICLE

Impact assessment of neem compost and *Trichoderma harzianum* solution in the control of root knot nematode disease on cowpea

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Received: 19 April 2016
Accepted: 27 June 2016
First Published: 04 July 2016

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Reviewing editor:
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Abstract: Efficacy of neem leaf based compost and *Trichoderma harzianum* solution were assessed on root knot nematode pest of two cowpea varieties at the screen house of the International Institute of Tropical Agriculture (IITA), Ibadan, Nigeria during 2012 planting season. The two cowpea varieties which have been confirmed to be susceptible to root knot nematode, IT96D-610 and IT84S-2246-4 were the test crops. The experimental pots (10 kg soil) were inoculated with 5,000 root knot nematode eggs. Treatments were neem compost and *T. harzianum* solution with application rates of 2 ton/ha and 1×10^{-7} spores respectively. The pots that were not treated served as the control. Each treatment was replicated 4 times while the statistical design was a complete randomized design. Data collected included growth and yield of cowpea, as well as the nematode numbers in the soil and root. The results indicated that neem compost and *T. harzianum* solution significantly increased growth and yield of IT96D-610 and IT84S-2246-4 cowpea varieties grown in the root knot nematode inoculated soil, while root knot nematode population in soil and root were significantly reduced. Gall index was also significantly lower in the neem compost and *T. harzianum* treated cowpea compared to the untreated control.

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Dr Timothy Ipoola Olabiyi is an Associate Professor of Nematology in the Department of Crop and Environmental Protection, Ladoke Akintola University of Technology, Ogbomoso, Nigeria. He obtained B. Agric.; MSc Crop Production and PhD Crop Protection from the University of Ilorin in 1990, 1994 and 2004 respectively. He did Post-Doctoral program at Coventry University, UK (2007–2008) and Short courses on Organic Agriculture at Coventry University, UK (2007) and on IPM and Food Security at Wageningen University, The Netherlands (2014). He has 70 research publications and 1 book to his credit. He has held many Administrative positions including Headship of his Department. His research interests are crop protection, organic agriculture, nematology, climate change and bio-pesticide technology. Olabiyi has won many awards based on his excellent academic performances. He had supervised many Undergraduate and Post-Graduate students. He is a member of many professional Societies.

PUBLIC INTEREST STATEMENT

Disease on crop is inevitable. Disease-causing organisms are naturally present in the soil, water, air and plant parts. They are capable of causing 100% crop loss if appropriate control is not put in place. In like manner, cowpea is seriously affected by root knot nematode. Over the years, this notorious micro-organism had been controlled effectively using industrially manufactured chemicals. However, the chemicals are very expensive and hazard to the environment. Researchers have now spurred other alternatives that are cheap and environment friendly. Amongst suggested alternatives are application of compost and biological agents. The current research assessed neem compost and biological agent as measures of control of root knot nematode disease of cowpea. Our results showed that Neem compost and the biological agent effectively controlled the nematode. The compost and biological agent are recommended for farmers' use in the control of nematode disease.

Subjects: Agriculture; Agriculture & Environmental Sciences; Bioscience; Environment & Agriculture

Keywords: *Trichoderma harzianum*; root knot nematode; neem compost; cowpea

1. Introduction

Cowpea (*Vigna unguiculata*) is an important food legume and essential component of cropping systems in the drier region of the tropics and subtropics (Singh, Ehlers, Sharma, & Freirefilho, 2003), and it is an important livelihood of millions of people (Quin, 1997). The grain contains between 20 and 25% proteins (Kay, 1979). Cowpea is a valuable and dependable commodity that produces income for many smallholder farmers and traders in sub-saharan Africa (Langyintuo, Lowenberg-DeBoer, & Faye, 2003). It is a deep rooted crop which, and does well in sandy soil and is more tolerant to drought than soybean (Dadson, Hashem, Javaid, Joshi, & Allen, 2003). It forms a major component of the tropical farming system because of its ability to improve soil fertility through nitrogen fixation (Abayomi, Ajibade, Sammuel, & Saadudeen, 2008). Cowpea can fix about 240 kg ha^{-1} atmospheric nitrogen and makes about 60–70 kg ha^{-1} nitrogen available for succeeding crops grown in rotation (Aikins & Afuakwa, 2008).

Plant parasitic nematodes are a serious pest and constitutes a major production constraint to cowpea in most growing areas of the world (Sikora & Greco, 1990; Sikora, Greco, & Silva, 2005). Symptoms of nematode damage on cowpea include stunted growth, yellowing, presence of galls, excessive root branching and reduced functioning of root systems. Poor germination and death of seedlings may be observed in case of heavy infestations (Mishra, 1992). Several nematode species are known to cause losses to cowpea throughout the world. Caveness and Ogunfowora (1985) listed 55 species of plant parasitic nematode associated with cowpea production. The root knot nematode *Meloidogyne incognita* and *Meloidogyne javanica*, are documented to cause major losses, with *M. incognita* indicated to be the most detrimental species to cowpea (Sarmah & Sinha, 1995). A comprehensive survey of cowpea growing areas in Nigeria revealed root knot nematode present in all the 248 farms infested with *M. incognita*, *M. javanica* and *Meloidogyne arenaria* present in 52, 44 and 4% of the soil samples respectively (Olowe, 2004).

Compost have been used with varying levels of success to suppress many soil–borne plant pathogens and the diseases they cause (Litterick, Harrier, Wallace, Watson & Wood, 2004). The disease suppressive properties of compost rely on a number of factors, which include microbial activities, microbial population dynamics, nutrient concentration and attendant chemical and physical factors. *Trichoderma* spp. are widely distributed all over the world and occur in nearly all soils and other natural habitats, especially in those containing organic matter (Attitslla & Salleh, 2010).

More recently concerns for the environment, high cost of nematicides, the non- availability of nematicides in time of need and the hazard they bring as well as the fear of nematodes developing resistance to synthetic nematicides have motivated researchers to exploit alternative way of controlling nematode. In this regard nematologists, especially in the developing world tend to look for indigenous, non-expensive but effective method of nematode control and with less or no hazard to plant, soil and livestock (Jada, 1993). Adegbite and Adesiyan (2005) had advocated for the need to develop naturally occurring nematicides which are harmless to man and livestock, but effectively controlled nematodes. The objective of this research work was therefore to determine the effectiveness of neem compost and *Trichoderma harzianum* in the performance and control of root knot nematode disease of cowpea.

2. Materials and methods

The research was conducted in the screen house at the International Institute of Tropical Agriculture (IITA), Ibadan, Oyo State, Nigeria between March and November, 2012. Sandy loamy soil was collected from a depth of 25 cm from IITA farm, soil collected was sieved with 2 mm sieve to remove plant debris and stones, steam sterilized and used to fill plastic 24 plastic buckets of 30 cm diameter.

The inoculum used consisted of root knot (*M. incognita*) nematode egg which was extracted from infected *Celosia* root in the laboratory using the method described by Hussey and Baker (1973).

The two cowpea varieties which have been confirmed to be susceptible to root knot nematode, IT96D-610 and IT84S-2246-4 were planted (IITA, 1984). Three seeds were planted per pot and later thinned to one healthy plant per pot. The experimental plastic pots with 10 kg steam sterilized soil were inoculated with approximately 5,000 root knot nematode egg, within the cowpea rhizosphere, using a 5 ml syringe at three weeks after planting.

A windrow method was used for the preparation of neem compost at Teaching and Research Farm, Ladoke Akintola University of Technology, Ogbomoso, Nigeria. Freshly harvested neem leaves (100 kg) was chopped (5–10 cm sizes), wrapped with plastic and properly covered up for a period of 5 months with regular turning (monthly interval) with garden spade (Olabiyi, Ogunniran, Ojo, Atungwu, & Abolusoro, 2013). The fully decomposed materials were spread and air-dried for 3 weeks. The compost, in air-dried form, was grind into powder form using attrition mill before application. However, pure culture of *T. harziarium* was obtained from the Agronomy Laboratory, Ladoke Akintola University of Technology, Ogbomoso, Nigeria. The fungus was cultured on Potato Dextrose Agar (PDA) under sterile condition using autoclave and lamina flow. The *T. harziarium* conidia on the PDA were harvested with sterile water for immediate use.

The treatments, neem compost and *T. harzianum* solution, were applied at the rates of 2 ton/ha (Renco, Sasanelli, D'Addabo, & Papajova, 2010) and 1×10^{-7} spores respectively, at four weeks after planting (Olabiyi, 2009). The pots that were not treated served as the control. Each treatment was replicated 4 times while the statistical design was a complete randomized design. Watering, manual weeding and insect control using Cypermethrin 10% EC were done whenever necessary.

Data collected included plant height, number of leaves, seed weight, fresh and dry root weights, gall index, final nematode populations in roots and soil. Gall index was determined using standard rating scheme 0-5 (Sasser, 1984), where 0 = no infection, 1 = 1–5% of root galled, 2 = 6–25% root galled, 3 = 26–50% root galled, 4 = 50–75% root galled, 5 = 76–100% root galled. At final harvest of the two cowpea varieties, nematode populations in 1.0 g root were extracted and counted following the method described by Hussey and Baker (1973), while the nematode population in 200 ml soil were assessed using the pie-pan extraction method as described by Whitehead and Hemming (1965). All data collected were subjected to analysis of variance and differences between the means were separated using Duncan's multiple range test at 5% probability level.

3. Results

The efficacy of neem compost and *T. harzianum* on the growth of two cowpea varieties, IT96D-610 and IT84S-2246-4, planted on root knot nematode infested soil is shown in Table 1. There was a significant difference between the treated and untreated pots. The cowpea varieties treated with neem compost had the highest plant height and number of leaves, closely followed by the *T. harzianum* treatments except for number of leaves in IT96D-610 where the number was similar in both treatments. Both treatments performed significantly better than the untreated control.

The efficacy of neem compost and *T. harzianum* on the yield of cowpea varieties (IT96D-610 and IT84S-2246-4) is shown in Table 2. The results obtained from the application of both treatments (neem compost *and T. harzianum)* showed significant differences with the untreated control for all parameter measured. The plants treated with neem compost performed overall better than the *T. harzianum* treatment with significant differences for number of pods for both varieties and seed weight per plant for IT84S-2246-4.

The efficacy of neem compost and *T. harzianum* on the soil root knot nematode population and root damage on two cowpea varieties (IT96D-610 and IT84S-2246-4) is shown in Table 3. Significant differences were visible between the treated and control plants on fresh and dry root weight in both

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Table 1. Effect of neem compost and *T. harzianum* on the growth of cowpea varieties planted in root knot nematode infested soil

Treatment	Plant height		Number of leaves	
	IT96D-610	IT84S-2246-4	IT96D-610	IT84S-2246-4
Neem compost	40.3a	45.7a	50a	57a
T. harzianum	36.6a	38.4c	51a	53a
Control	23.4b	24.2c	34b	35b

Note: Figures having the same letters along the same column are not significantly different from one another.

Table 2. Effect of neem compost and *T. harzianum* on the yield of cowpea varieties planted in root knot nematode infested soil

Treatment	Number of pod per plant		Number of seed per pod		Weight of seed per plant	
	IT96D-610	IT84S-2246-4	IT96D-610	IT84S-2246-4	IT96D-610	IT84S-2246-4
Neem compost	9a	11a	8a	9a	19.1a	35a
T. harzianum	6b	8b	8a	8a	17.1a	13b
Control	1c	2c	2b	2b	3b	3c

Note: Figures having the same letters along the same column are not significantly different from one another.

Table 3. Effect of neem compost and *T. harzianum* on nematode population density, reproductive factor and gall index on cowpea at 18 weeks after planting

Treatment	Initial nematode population (Pi)		Fresh root weight (g)		Dry root weight (g)		Gall index		Final nematode population (Pf)		Root extraction		Reproduction factor (Pf/Pi×100)		% nematode reduction			
															Soil		Root	
	A	B	A	B	A	B	A	B	A	B	A	B	A	B	A	B	A	B
Neem compost	5,000	5,000	0.38c	0.27b	0.14b	0.09b	1b	2b	388b	473b	1,004b	1,045b	0.07	0.09	93.4	91.8	88.4	87.8
T. harzianum	5,000	5,000	0.63b	0.86b	0.18b	0.10b	1b	1b	428b	336b	1,458b	1,030b	0.09	0.06	92.7	94.2	83.2	88.0
Control	5,000	5,000	4.58a	3.47a	2.41a	1.86a	5a	4a	5,876a	5,783a	8,654a	8,558a	1.75	1.16	0.0	0.0	0.0	0.0

Notes: A = IT96D-610; B = IT84S-2246–4.

Figures having the same letters along the same column are not significantly different from one another.

varieties (Table 3). The control plants had the highest root weight due to the galls caused by nematode infestations. Gall index can be seen in Table 3 and it is much higher in the control than in the treatments. Galls often cause the roots to be much heavier than non-infested roots. Nematode population in soil and roots were significantly lower in the treated plants. Both treatments seemed to control root knot nematodes very well which is confirmed with the reproduction factor which is 0.06–0.09 for the treatments in both varieties compared to 1.16–1.17 in the untreated plants. Both varieties reacted similarly to the bio-nematicide treatment as nematode numbers were similar.

4. Discussion

The effectiveness of neem compost and *T. harzianum* solution in controlling root knot nematode, in cowpea cultivation has been established in this study. The treatments were effective in reducing root galling, nematode population in the root and soil. Application of neem compost as soil amendment and *Trichoderma* spp. as bio-control agents in crop production have been reported to be effective by numerous researchers (Atungwu, Ademola, & Aiyelaagbe, 2009; Chimbekujwo & Bukar, 2013; Neher, 2001; Radwan, Farrag, Abu-Elamayem, & Ahmed, 2012). Al-Hazmi and TariqJaveed (2015) reported significant reduction in root galling, egg production and soil juveniles treated with isolates of *T*. harzianum and *T*. viride. Atungwu et al. (2009) reported the effectiveness of neem leaf powder and a novel organic based fertilizer in managing soil nematode population. Findings in this study

also indicated that neem compost and *T. harzianum* solution can be used to improve the growth and yield of cowpea grown on nematode infested soil (Al-Hazmi & TariqJaveed, 2015; Raddy et al., 2013; Sharon et al., 2001). The result of this study confirmed that neem compost and *T. harzianum* solution can be used as natural amendment to reduce dependence on synthetic fungicides (Claudius-Cole, Aminu, & Fawole, 2010; Radwan et al., 2012).

Many fungi agents have been tested for their potential as bio-control agents (Al-Hazmi & TariqJaveed, 2015; Sharma & Pandey, 2009). Secondary metabolites from fungi also contain compounds that can be toxic to plant parasitic nematodes (Dababat & Sikora, 2007). Many attempts have been made to use *Trichoderma* species to control plant parasitic nematode (Neher, 2001; Windham, Windham, & Williams, 1989). *Trichoderma* spp. have been used as a bio control agent against plant parasitic nematode and this fungus may also promote plant and have the ability to colonize root surface and the cortex (Sharon et al., 2001). *Trichoderma* spp. led to inhibition of the nematode activities and movement. *Trichoderma viride* in combination with organic amendment was also known to produce growth hormones, which have observed to have added response in boosting the plant vigour. It has been reported that *Trichoderma* has not only be produced to parasitize nematode but also help in tolerance to stress condition by enhanced root development (Chimbekujwo & Bukar, 2013; Neher, 2001).

5. Conclusion

In conclusion neem compost and *T. harzianum* (2.0 t/ha and 1×10^{-7} spores respectively) were effective promising measure for controlling root-knot nematode on cowpea. It can therefore be recommended to the farmers for use in areas where root knot nematodes are a problem.

Funding
The authors received no direct funding for this research.

Competing Interests
The authors declare no competing interest.

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Citation information
Cite this article as: Impact assessment of neem compost and Trichoderma harzianum solution in the control of root knot nematode disease on cowpea, T.I. Olabiyi, O.J. Ojo & B.O. Adewuyi, *Cogent Food & Agriculture* (2016), 2: 1207843.

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