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Effects of poultry litter and carbofuran soil amendments on *Meloidogyne incognita* attacks on cacao

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ABSTRACT

Objective: Growth retardation, galling of the root or complete death of cacao seedlings are the symptoms expressed by root-knot nematode, *M. incognita*, infestation in the nursery. This experiment was conducted in the nursery to evaluate the effect of poultry litter and carbofuran incorporated into the soil on the population dynamics of *M. incognita* and cacao growth.

Methodology and results: The experiment was set as a randomized complete block design of a 4-by-3 factorial arrangement with four rates of poultry litter (0, 5, 7, 10g/pot) and three rates of carbofuran (0, 1, 2g/pot). Lower population densities of *M. incognita* were observed in soils treated with poultry litter and carbofuran compared to the control. Poultry litters at high rates alone or combined with carbofuran consistently stimulated growth of cacao seedlings and reduced root galling and the nematode population densities.

Conclusion and applications of findings: Poultry litter can be used effectively as an organic soil amendment to supply nutrients to the crop and suppress *M. incognita* populations to reduce damage to seedlings in the nursery. This finding will contribute to reducing the current level of frustration that is faced by resource-poor farmers due to poor establishment of cacao seedlings and high cost of nematicides.

Key words: Organic amendments, *Meloidogyne incognita*, cacao seedlings, poultry litter, carbofuran.

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INTRODUCTION

Cacao is one of the most important tropical crops (FAOSTAT, 2006). About 70% of the world's cocoa is produced in West Africa where it contributes significantly to the economies of countries in this sub-region, as well as many other countries in Central America and South East Asia (Taylor & Taylor, 2006). Nigeria is the fourth largest producer of cocoa in the world with an estimated production of 485,000 metric tons in 2006 (FAOSTAT, 2006). The production of cocoa in Nigeria has witnessed a downward trend since the early 1970s due to numerous factors, e.g. ageing trees, shortcomings in applying recommended agronomic techniques by farmers, and the effects of pests and diseases.

Root-knot disease caused by root-knot nematodes, *Meloidogyne* spp, is a well-known disease of many tropical and sub-tropical plants. *Meloidogyne* spp are the most important nematodes of cacao due to their pathogenicity and wide distribution in cocoa producing regions (Campos & Villain, 2005). It is a common pest of cacao in West Africa (Whitehead, 1969; Asare-Nyako & Owusu, 1979; Fademi *et al*., 2006). Symptoms of *M. incognita* damage on cacao seedlings are dieback, stunting, wilting, chlorosis and reduction in size of the leaves, galling of the root or complete death of the seedlings (Afolami & Caveness, 1983; Orisajo & Fademi, 2005; Orisajo *et al*., 2007). Although control strategies have been based on the use of chemical nematicides (Afolami, 1993), the chemicals are usually too expensive for resource-poor farmers and their use often results in a noticeable decrease of many soil biological processes. There is need to develop new management tools that are environmentally and toxicologically safe (Gullino *et al*., 2003).

The need for alternatives to nematicides has stimulated research focusing on sustainable tactics for management of plant parasitic nematodes (McSorley & Poranzinska, 2001). The addition of organic amendments, including animal wastes and leaf extracts, has been shown to have a suppressive effect on plant parasitic nematodes (Widner & Abawi, 2002; Salgado & Campos, 2003; Sundararaju & Kumar, 2003; Abubakar *et al*., 2004; Walker, 2004; Orisajo & Dongo, 2005; Orisajo & Fademi, 2005; Orisajo *et al*., 2007). Application of poultry litter supplies nutrients to the crop, impacts communities of soil organisms and may stimulate organisms that are antagonistic to nematodes (Riegel & Noe, 2000; Koenning *et al*., 2003). The objectives of this research were to assess the effect of poultry litter and carbofuran on the population dynamics of *M. incognita* and subsequent cacao growth.

MATERIALS AND METHODS

Experimental site: Nursery experiments were carried out at the research farm of Cocoa Research Institute of Nigeria (CRIN) at Ibadan, Nigeria (Latitude 7.26°, Longitude 3.54° and 122m above sea level). The annual rainfall ranges between 1200-2500mm, distributed over 5-7 months from April to October. The average daily temperature range is 26-30°C.

Soil analysis and assay: Sandy-loam top soil normally used for raising cacao seedlings was collected in bulk from the CRIN research field naturally infested with the root-knot nematodes. It was thereafter thoroughly mixed and distributed into 2-litre black polyethylene bags (pots) - the type used for raising cocoa seedlings commercially. The initial physico-chemical properties of the soil are presented in Table1. The soil was assayed to confirm the presence and population density of root-knot nematodes (*Coyne et al*., 2007).

Poultry litter: Poultry litter (poultry excrement and saw dust bedding) was collected from a commercial broiler house in Ibadan. Litter was collected from coops, which were cleaned approximately once a month and air-dried. The nutrient content of the litter is presented below.

Experiment set-up: The experiment was set as a randomized complete block design of a 4-by-3 factorial arrangement with four rates of poultry litter (0, 5, 7, 10g/pot) and three rates of carbofuran (0, 1, 2g/pot). The different treatment combinations were mixed with sandy-loam top soil and kept moist 14 days before cacao seeds were planted. Three seeds of *Theobroma cacao* cv. F_3 Amazon were planted in each pot and later thinned to one per pot a week after emergence. Pots were arranged on nursery benches in a randomized complete block design with six replications.

After sowing, regular observations were made to record data on phytotoxic effects of any treatment on plants and disease symptoms. The experiment was terminated 26 weeks after planting. The growth parameters such as plant height, stem girth, and numbers of leaves were recorded. Plant areas were determined with the aid of electronic leaf meter.

To assess infection the roots were carefully freed of soil, washed under a gentle stream of tap water, mopped and galls counted using a hand lens at 3-5 X magnification. Root galling was assessed using the 0-5 gall index (Sasser *et al*., 1984). Nematode eggs were collected from each root system using sodium hypochlorite method (NaOCl) of Hussey and Barker (1973) and counted. Aliquots of 100-cm^3 soil samples from each pot were assayed for juveniles of *M. incognita* using the modified Baermann technique (*Coyne et al*., 2007).

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Table 1: Initial physico-chemical properties of soil at experimental site at CRIN, Ibadan, Nigeria.

Properties	Value
Physical properties	
a. Sand (%)	72
b. Silt (%)	15
c. Clay (%)	13
d. Textural Class	Sandy loam
Chemical properties	
a. pH in H_2O 1:1	6.0
b. Organic carbon (gkg^{-1})	3.3
c. Total nitrogen ((gkg^{-1}))	0.4
d. Available phosphorus ($mgkg^{-1}$)	24.35
e. Exchangeable potassium ($cmolkg^{-1}$)	0.2
f. Exchangeable magnesium ($cmolkg^{-1}$)	0.85
g. Exchangeable calcium ($cmolkg^{-1}$)	2.66
h. Exchangeable sodium ($cmolkg^{-1}$)	0.6
i. Exchangeable manganese ($cmolkg^{-1}$)	0.34

Data analysis: Prior to statistical analyses, data were checked for normality and homogeneity of variances, and transformed where necessary. A log transformation [$\log_{10}(x + 1)$] was applied to the data on nematodes (densities per 100-cm^3 soil, densities per gram root and gall index). Analyses of variance (ANOVA) were carried out to test for main effects and interactions. Preplanned comparisons between treatment combinations were tested with linear contrasts. Regression analyses were used to develop linear models relating nematode numbers and rate of poultry litter application to cacao seedling growth. All analyses were performed using GENSTAT (version 7.1, VSN International Ltd., Lawes Agricultural Trust, Hempstead, UK).

RESULTS

The nutrient content (%) of poultry litter that was used to amend soil at the CRIN experimental site was Nitrogen 1.75; Carbon 27.9; Phosphorus 1.53; Potassium 1.85; Calcium 4.26 and Magnesium 0.8 %. Significant interactions were observed between main effects and experimental runs for nematode population data. Lower population densities of *M. incognita* were observed in soils treated with poultry litter and carbofuran compared to the control (Fig. 1A). Poultry litter at high rates alone or combined with carbofuran consistently reduced root galling and the nematode population densities (Fig. 1B).

Increasing rate of poultry litter applied resulted in a linear increase in plant height of the cacao seedlings in nematode-infested soil (Fig. 2A). The highest plant heights were with litter at 7 and 10g/pot. There was a positive linear relationship between leaf area and litter rate (Fig. 2B). Significant differences in leaf area with litter at 5, 7, and 10g (linear contrast) were not observed (Table 2). Carbofuran treatment had no significant effect on leaf area (Table 2).

Dry shoot weights of *M. incognita*-infested cacao seedlings had a positive response to increasing litter rates (data not shown). Also, there was a significant effect on dry shoot weights of the cacao seedlings in carbofuran treated soil compared to the control (Table 3). However, the effect is more pronounced in soil amended with litter. In most cases, stunted growth, chlorosis, reduction in leaf size, and wilting of cacao seedlings were observed in the control (Plate 1).

Soil amendments with poultry litter alone or combined with carbofuran increased significantly ($P \leq 0.05$) the dry root weight and stem girth of cacao seedlings compared to untreated soils (Table 4).

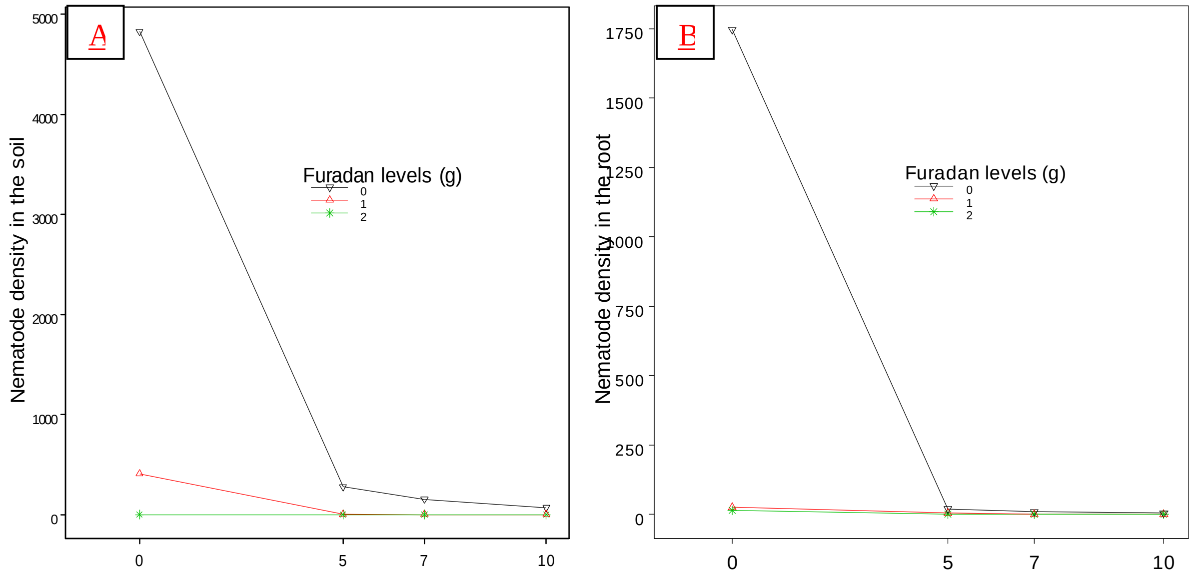

Figure 1: Effects of poultry litter and carbofuran soil amendments on the soil densities of *M. incognita* (A) and root densities (B) in infested cacao seedlings. Transformed data [$\log_{10}(x+1)$] used in statistical analysis; back-converted means of *M. incognita* second-stage juveniles/100cm^3 of soil and eggs/g of root shown.

DISCUSSION

Results from this experiment showed that *M. incognita* population densities decreased in response to increasing rates of poultry litter in amended soil. This result was consistent with the report of Riegel and Noe (2000) showing that the application of litter 14 days before planting was optimal for effects on nematode population densities. Many factors could affect the response of nematode communities to nutrient sources. Most importantly, nematode communities were often affected by the nutrient composition, particularly the C:N ratio, of the organic amendments (Ferris & Matute, 2003; Yeates & Boag, 2004). In general, amending the soil with a low C:N ratio (less than 20:1) substrate resulted in an abundance of enrichment-opportunist antagonistic microbes (Ferris & Matute, 2003; Wang *et al*., 2004 & 2006) and rapid mineralization of N in the form of NH_4^+ or NO_3^- for absorption and uptake by plant roots (Powers & McSorley, 2000). The poultry litter used in this experiment has a low C:N ratio (16:1) and this resulted in the suppression of nematode population on cacao seedlings.

Table 2: Analysis of variance and linear contrasts for leaf area of *M. incognita* infested cacao seedlings.

Source of variation	df	MS[a]	F pr[b]
Total	35		
Blocks	2	0.092	
Poultry litter (A)	3	15.149	<0.001
Carbofuran (B)	2	0.177	0.128
A x B	6	0.037	0.826
Error	22	0.078	
Linear contrasts			
Control versus A	1	45.008	<0.001
A at rate 7g versus A at rate 10g	1	0.284	0.070
A at rate 7g versus A at rate 5g	1	0.005	0.797
Control versus A at rate 5 g	1	27.578	<0.001

[a] Mean square; [b] Probabilities associated with individual F tests.

Table 3: Analysis of variance for dry shoot weight of *M. incognita* infested cacao seedlings.

Source of variation	df	MS[a]	F pr[b]
Total	35		
Blocks	2	0.007	
Poultry litter (A)	3	15.967	<0.001
Carbofuran (B)	2	0.037	0.015
A x B	6	0.037	0.278
Error	22	0.007	

[a] Mean square; [b] Probabilities associated with individual F tests

Table 4: Analyses of variance for dry root weight and stem girth of *M. incognita*-infested cacao seedlings.

Source of variation		Dry root weight		Stem girth	
	df	MS[a]	Fpr [b]	MS	Fpr
Total	35				
Blocks	2	0.00031		0.00001	
Poultry litter (A)	3	0.49069	<0.001	0.00421	<0.001
Carbofuran (B)	2	0.15811	<0.001	0.00061	<0.001
A x B	6	0.09374	<0.001	0.00104	<0.001
Error	22	0.00025			

[a] Mean square; [b] Probabilities associated with individual F tests

Plant height, leaf area, dry shoot and root weights were all stimulated by the addition of litter in *M. incognita*-infested soil. Agronomic studies on the plant growth-enhancing effects of poultry manure have shown an increase in the stem and leaf biomass of plants (Mohanty *et al*., 2006). Phosphorus uptake is enhanced by the application of poultry manure, and this could be attributed to the greater leaf biomass yield in poultry manure-treated soil (Mohanty *et al*., 2006). A common form of poultry manure is poultry litter, which consists of poultry droppings and sawdust (Gullino *et al*., 2003). Poultry litter contains significant quantities of N, P, K, Ca, Mg and micronutrients and can be used as a substitute for commercial fertilizers (Sims & Wolf, 1994).

The addition of poultry litter to soil leads to a better environment for the growth of plant roots. This enhances the utilization of soil nutrients, as a consequence of which the nematode damage might have been markedly reduced (Van Den Boogert *et al.,* 1994). A significant increase in root mass of plants in soil amended with poultry litter and subsequent decrease in number of nematodes observed in this experiment may be responsible for the increased growth of the cacao seedlings. This is in agreement with previous report (Van Den Boogert *et al.,* 1994) that such decrease means fewer disturbances to the seedlings resulting in an unhindered growth.

Organic amendments to soil have been shown to have beneficial effects on soil nutrients, soil physical conditions, soil biological activity and crop viability. Organic materials represent a very important resource for the improvement of soil fertility because decomposed materials ultimately serve as nutrients for plant growth. The observed increase in growth of cacao seedlings in poultry litter-amended soils compared to both the carbofuran treated and untreated soils may be attributed to, among others, the increase in nutrients supply to the soil, resulting from the addition of organic amendments.

Organic soil amendment is a nematode management option, and numerous aspects of research would have practical applications in commercial agriculture for the amelioration of pest problems. Relationships between parasitic nematodes, the soil environment, organic amendments and plant-host health are obviously complex and make it difficult to assess the activities occurring in soil. However, use of poultry litter as soil amendment has been shown in this experiment to reduce *M. incognita* infection on cacao seedlings in the nursery.

This finding will contribute to reducing the current level of frustration that is faced by resource-poor farmers due to poor establishment of cacao seedlings and high cost of nematicides. The use of poultry litter as a nematode management tool would be most efficient when used in combination with other management practices that are currently available. This machinery, if put into practice, may be the beginning of a switch to unconventional, less competitive and cheap alternative source of preventing cacao seedlings from nematode attacks and thus enhancing plant performance.

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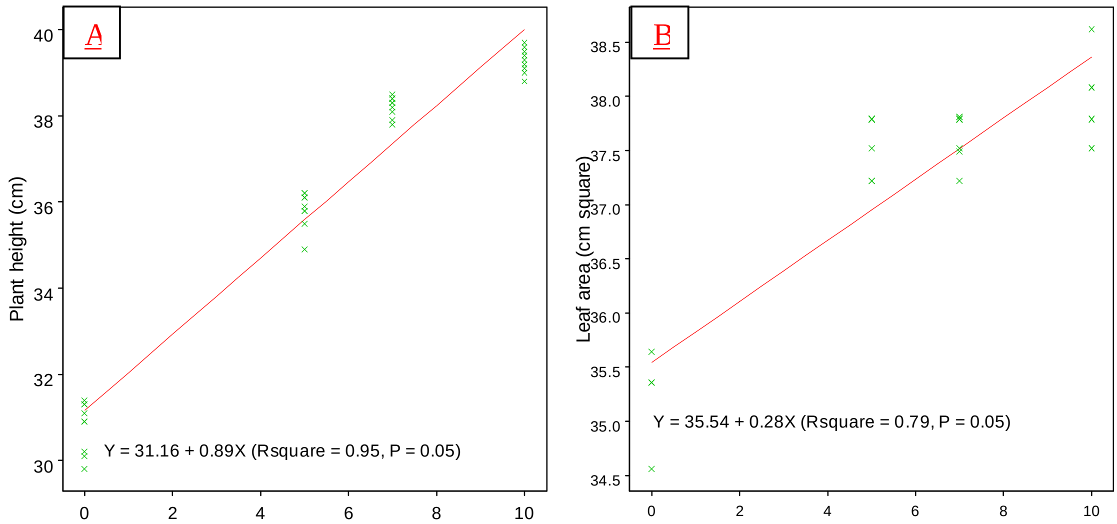

Figure 2 A & B: Influence of poultry litter applications at different rates (0, 5, 7, or 10g/pot) on plant height (A) and leaf area (B) of *M. incognita* infested cacao seedlings. Each asterisk is the mean of 6 replications. Regression equation is based on analysis of all data. Similar relationship was observed between rates and dry shoot weight (data not shown).

Plate 1: Stunted growth, chlorosis, reduction in leaf size, and wilting of *M. incognita*–infested cacao seedlings (left) compared to similar age plant (right) in soil amended with poultry litter.

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