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Research Paper

EFFECTS OF ORGANIC MANURE ON SOIL PROPERTIES AND HEALTH AND GROWTH PERFORMANCE OF CACAO (*Theobroma cacao* L) IN SOUTHWESTERN NIGERIA

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Abstract

Soil fertility status and activities of soil borne microbes are major factors influencing cacao growth and establishment on the field; in this study, the manurial potentials of some organic wastes and their effects on soil health and growth of cacao in Ibadan and Owena soils of Southwestern Nigeria were investigated in 2012. The experiments were Randomized Complete Block Design (RCBD) with 3 replications. The fertilizer types were: Goat Dung (GD), Sunshine Organic and Organo-Mineral Fertilizers (OF and OMF) and NPK (15:15:15) which were applied at 0, 200, 400 and 600 kg/ha. Plantain suckers were established at 3 x 3 m in June, 2011 as shade crop and cacao seedlings were transplanted into the plots one year later (June, 2012). The fertilizers were applied to treatment plots one month after cacao seedlings were transplanted using ring method. Data were collected on cacao growth parameters such as plant height, stem diameter, number of leaves and number of branches, monthly commencing at 3 months after transplanting (MAT) and for 96 weeks after transplanting (WAT). Biological properties (arbuscular myccorhizal spores, nematodes, moulds, yeast and bacteria) were also determined on the soil samples using standard procedures. Goat dung (GD), organo-mineral fertilizer (OMF) and organic fertilizers (OF) had pH ranged between 7.00 and 8.17 while they contained in similar order 2.86, 0.63 and 3.64 g/kg organic carbon (OC), 4.80, 1.09 and 6.27 % organic matter (OM), 1.26, 0.06 and 2.16% Nitrogen (N), 113.24, 138.06 and 7.08 cmol/kg Phosphorus (P), 2.60, 2.00 and 13.10 cmol/kg Calcium (Ca) respectively. All fertilizer types and rates at Ibadan and Owena significantly increased cacao seedlings' growth relative to the control. Application of GD, OMF and OF increased albuscular mycorizal spores

relative to NPK and the control. The population of other soil microbes (bacteria yeast and moulds) was also influenced by fertilizer types and rates. Application of 600 kg/ha NPK significantly reduced soil nematode population when compared with other fertilizer types, rates and the control plots in Ibadan, while 400 kg/ha OF and 200 kg/ha OMF significantly reduced nematode population relative to the control in Owena.
Key words: cacao, fertilizers, biological properties, growth.

INTRODUCTION

Cacao was classified as a member of the family Sterculiaceae until recently when Alverson et al. (1999) through the application of molecular marking technology, reclassified the crop as a member of the family Malvaceae. Its natural habitat is the lower middle storey of the evergreen rain forest. The importance of cocoa in the Nigeria economy remains high. Cocoa exports have been and will continue to be a significant factor in the economic growth of Nigeria. For instance, in 1969, cocoa alone earned N106 million which accounted for 40% of all agricultural exports for the year (Federal Office of Statistics 1972).

Despite negative effect of government policy, cocoa remains the highest foreign exchange earner of all agricultural export crops which the Structural Adjustment Programme (SAP) sought to promote as a development policy objective in Nigeria (Tijani et al. 2001). However, the discovery of crude oil in large quantities has brought a downward trend in Nigeria cocoa production and position in the world market (Ayoola et al. 2000). Côte d'lvoire, which was third in Africa with 143,000 tonnes behind Nigeria's l96, 000 tonnes in 1970, is now the largest producer in the world with 1.2 million tonnes annually accounting for about 40 % of total world production. Nigeria is now the fourth largest producer after Côte d'lvoire, Ghana and Indonesia (ICCO 2005). The practice of planting cacao seedlings under banana or plantain widely preferred with increase risk of *Meloidogyne incognita* infection on cacao seedlings, as the nematode is frequently associated with the shade crop and parasitic nematodes have become common pests of cacao in West Africa (Fademi et al. 2006). Plant-parasitic nematodes as pests, pose threat to young cacao seedlings at both establishment and fruiting stages of cocoa. *Meloidogyne incognita* appears to be the most common nematode of cacao in Nigeria. The symptoms observed were, die back, stunting, wilting, chlorosis, reduction in the size of the leaves and galling of the root or complete death of the seedlings (Orisajo and Dongo 2005). Hence there is an urgent need to develop new management tools that are affordable, accessible, environmentally and toxicologically safe in controlling the pests. Conveniently, the use of resistant planting materials is the most economic approach to nematode control in plants. However, cacao was not listed among the eight nut/fruit trees which have locally available nematode – resistant or nematode-tolerant seeds or rootstocks (Sasser and Frekman 1987) Nematodes, hence, pose a threat to cacao that can not be addressed through resistance or tolerance of planting materials (Sasser and Frekman 1987). Therefore other methods of control have to be investigated. Furthermore, it is well documented that the increase in plant growth resulting from Arbuscular Mycorrhizal Fungus (AMF) association is usually due to increased mineral elements uptake by the hyphae from the soil and increased uptake of macro-elements other than P has also been reported (Hodge *et al*., 2001) as well as increased uptake of some micro-elements (Kothari et al. 1991a). In addition Mycorrhizae have been shown to play an important role in maintaining soil physical

properties (Degens et al. 1996). It has also been observed that soil organic amendments foster beneficial soil micro organisms (Doran 1995). Limited field studies have been conducted to determine the impact of soil amendments on microbial communities in organic and conventional production systems (Gunapala and Scow 1998). However, it has been shown that microbial activity and biomass is higher in fields when organic manures are applied compared with application of synthetic fertilizers (Doran 1995).
The aim of this study therefore is to examine the influence of fertilizers (organic and inorganic) on the diversity of soil microbes, soil chemical properties and growth of cacao seedlings on the field.

MATERIALS AND METHODS

Field experiments were conducted at Ibadan, headquarters of the Cocoa Research Institute of Nigeria (CRIN), and in Owena, a CRIN Sub-station in Ondo State.

Ibadan experimental location

Ibadan, Oyo State, is located on latitude 07^0 10' N and longitude 03^0 52'E, and an altitude of about 122 meters above sea level in the humid tropical rain forest zone of Nigeria. Ibadan is characteristically hot and humid, reputed for seasonal rainfall, high temperatures and high humidity and distinct dry and rainy seasons. The dry season extends from early November to March and is characterized by dry atmosphere and intense scorching sun. The rainy season which is characterized by high humidity and cloudy sky, runs from end of March to early November. There is usually a two-week dry spell in August. The rainy season has an average of 8 rainy days in a month at the commencement of the rains, this increase from April to about 15 rainy days in September. The annual rainfall is between 1200-1500 mm. The maximum temperature ranges between 26 to 35 ^{o} C with an average of about 30.1^{o}C while the minimum temperature ranges from 15 to 24^o C with an average of 19.5^{o}C. Relative humidity is high throughout the year and ranges from of 50 to 89 % with an average of 79 %. There are seasonal variations in the values of relative humidity, which varies from 65 to 89 % during the rainy season and 46 – 70 % during the dry season.

Owena experimental location

The Owena Substation of the cocoa Research Institute of Nigeria, located in Ondo State, lies on latitude 07^o N and longitude 05^o, 7^1E and at an altitude of about 22.5 meters above sea level. Owena is situated at about 21 kilometres south–east of Akure between Akure and Ondo. There are two seasons: rainy (wet) season which spans late March to November of the same year and a dry season that runs from December of one year to late March of the following year. There are 15 to 17 rainy days per month during the rainy season. The dry season on the contrary to the situation in Ibadan` has characteristic of scanty rainfall, the average ranges between 1500 – 1600 mm per annum. The maximum temperature in Owena is usually between 28 and 34^o C while the daily minimum temperature ranges between 18 and 23^o C. The relative humidity during the rainy season ranges from 69 to 80 %, and between 56 and 64 % during the dry season.

Acquisition and preparation of experimental materials

Seedlings cacao F3 Amazon genotype was collected from CRIN Seed Garden, while plantain suckers were collected from experimental plots in both Ibadan and Owena. Experimental plots of about 30 by 120 m were mapped out and the experiment was laid out in rows of 3 x 3 m. Sunshine organic and organo-minerals fertilizers used for the experiments were obtained from the Ministry of Agriculture, Ondo State, while the N.P.K. 15:15:15 was obtained at Ayedaade Local Government Gbongan, Osun State. Goat

dung manure was obtained from Ilesha Garage Akure, Ondo State. The goat dung was collected dried and carefully sorted to remove foreign materials and packed in 50 kg bags for application on the field.

Treatments and experimental design

Field experiments were conducted in two stations of the Cocoa Research Institute of Nigeria (CRIN), Ibadan Headquarters and Owena Sub-Station, Ondo State. The experiments were conducted between June, 2012 and June, 2014. The experiments were Randomized Complete Block Design (RCBD) with 3 replicates. The 4 fertilizers were: Goat Dung (GD), Sunshine Organic and Organo-Mineral Fertilizers (OF and OMF) and NPK 15:15:15 and the rates of application were: 0, 200, 400 and 600 kg/ha. Lay-out of each experimental site (Measurement, pegging and holing) was carried out before planting. Four hundred and thirty two (432) plantain suckers were planted at the spacing of 3 x 3 m in each of the experimental sites in Ibadan and Owena between second and third week of June, 2011, while Four hundred and thirty two (432) five months old cacao seedlings (F3 Amazon) of average height of 46 cm (already raised in the nursery) were transplanted in to one year old plantains in 2012 at the spacing of 3 x 3 m on each of the sites (Ibadan and Owena). Four plants were randomly tagged for data collection. Top soil samples were collected randomly from each experimental sites (Ibadan and Owena) using soil auger. The samples were bulked and air dried before being subjected to routine laboratory analysis of particle size analysis was determined by the hydrometer method (Kettler et al. 2001) and organic cabon content (OC) by the potassium dichromate oxidation method (Zhang et al. 2001). Soil pH was read on pH meter (1:1 water). Organic matter was determined by the Murphy blue coloration and determined on a spectronic 20 at 882um (Murphy and Riley, 1962). Soil potassium (K), calcium (Ca) and Magnesium (Mg) were extracted with lMNH_4 OAC, PH_7 and were determined with flame photometer; Mg was determined with an atomic absorption spectrophotometer. The total nitrogen (N) was determined by the Microkjedahl method (AOAC, 1990). Two grammes (2 g) each of the organic fertilizers used were also analyzed for nutrient composition. The fertilizers were applied to treatment plots one month after transplanting using ring method of fertilizer application at 5cm away from the base of cacao (July, 2012). Monthly Data collection on growth parameters (plant height, stem diameter, number of leaves and number of branches) commenced 3 months after transplanting. The experiments were monitored for 24 months (96 weeks after planting). At 15 months after transplanting, soil samples were collected from treatment plots and were processed and analyzed for and soil biological properties (Arbuscular myccorhizal colonization, soil nematodes, moulds, yeasts and bacteria) were determined using standard procedures.

Data were collected on the growth parameters of cacao seedlings such as: Plant height measured in centimetre using a meter rule on the surface and the tip of the main stem; Number of leaves was counted. Stem diameter was measured in centimetre with the use of Vernier'Caliper 30 cm above the ground level. Number of branches was also counted. These growth parameters were taken monthly for 24 months commencing from 3 months after transplanting (3 MAT).

Soil samples collection and analysis

Top soil samples were collected randomly at the depth of 15 cm from each of the experimental sites at both locations (Ibadan and Owena) with the aid of soil auger at 30cm depth. For the pre-cropping analysis, the samples were bulked together and mixed thoroughly, air dried at room temperature and analysed for various elements.

Particle analysis was carried out using the hydrometer method as described by Bouyoucos (1951). The pH was determined in water (1:1 soil: water ratio) using a pH meter with glass electrode as described by Jackson (1965). Total Nitrogen was determined using kjeldahi procedure as described by Jackson (1965). Organic carbon content was determined using the Walkey-Black method (Nelson and Sommers 1982). Phosphorus determination was done by the Bray method as described by Bray and Kurt (1945).

Exchangeable K, Ca, Mg and Na were determined by extraction with ammonium acetate and the amounts of K, Ca and Na in the filtrate were determined using flame photometer with appropriate filter while Mg was determined using a perking Elmer Atomic Absorption Spectro photometer (AAS).

Determination of diversity of soil microbes

Soil samples were collected from 15 cm depth in each experimental plot around cacao seedlings, placed in nylon bags, thoroughly mixed, and air-dried for 48 hours. The samples were sieved through 2 mm sieve and stored in the shade at room temperature until they were analyzed.

Soil assay for nematodes

The soil was assayed to confirm the presence and the initial population density of the nematodes. A 100g sub-sample soil was assayed for nematodes using the White-head and Hemming (1965) tray modification of Baermann (1917) technique as described below:

One hundred grams soil was put into a set up that has two plastic sieves with extractor tissue sandwiched in between. The plastic sieves with the soil were thereafter placed in a plastic bowl, and water was added to the extraction bowl just enough to wet the soil. The set-up was left undisturbed for 48 hours. Thereafter, the plastic sieve containing the soil was removed briskly, and the nematode suspension in the bowl was poured into a nalgene wash bottle and allowed to settle (Caveness 1975). The supernatant was siphoned out, and the suspension containing nematodes was then poured into a labelled beaker, and adjusted to 10 ml by adding water. This was homogenized and 1ml of the suspension was taken with the use of pipette, dispensed into the Doncaster (1962) nematode counting dish and examined under a stereomicroscope. Nematodes was transferred with a sharpened broom stick to a slide with a drop of water, covered (with a cover slip) and examined under a compound microscope with a 40, 60 and 100 X objective for identification using taxonomic keys (Luc and de Guiran 1990) and counted. The identification and counting were repeated three times and mean population of nematodes per sample were calculated. The various procedures followed for soil assayed were outlined by Coyne et al. (2007).

Extraction of vesicular *arbuscular mycorrhiza* (VAM) fungal spores from soil

The wet-sieving method (Gerdamann and Nicholson 1963) was employed. A 100 g sub-sample soil was suspended in 750 ml tap water in 1000 ml capacity plastic container for sedimentation after which the suspension was mixed vigorously. The soil suspension was strained through four sieves of 710, 500, 250 and 53µm mesh sizes (Model Endecotts UK) arranged in the order, 710, 500, 250 µm, to remove rock fragments and coarse woody and root debris. The soil suspension was finally strained through sieve 53 µm to collect the spores mixed with the solid matter; thereafter the solid matter collected was transferred to 50 ml centrifuge tube. Water was added to the tube and the soil sample was re-suspended. The tubes were centrifuged at 1800 rpm for 5 minutes. The supernatant which contained floating organic materials including dead spores was

discarded. The pellet was re-suspended in sucrose solution (440 g/l). Carefully, the tubes were balanced and centrifuged up to 1800 rpm for 2 minutes and stopped immediately. Rapidly, the supernatant was sieved (32 micron) and washed thoroughly (at least 1minute) to replace the sucrose and alleviate osmotic stress on spores. The pellet left in centrifuge tube was discarded. Carefully, all of the solid materials from the sieve were washed into the petri dish marked with a grid to facilitate spore counting and collection under a stereo- zoom microscope. The spores in suspension were filtered, counted and identified using Stereo-zoom microscope (Model Leica $MZ12_5$) at magnification 40 X and Compound microscope

Enumeration of Bacteria, Yeast and Moulds in the soil in soil sample

Nutrient agar (NA) and Potato Dextrose Agar (PDA) were prepared according to manufacturers specification (BIOMARK) by dissolving 28 g of NA in a litre of distilled water in a conical flask while 39 g of PDA was dispersed in a litre of distilled water separately, allowed to soak for few minutes, thoroughly mixed, cotton plugged, covered with aluminium foil and then sterilised by autoclaving at 15 lbs pressure (121^o C) for 15 minutes. The media were allowed to cool and mixed well before dispensing. The standard procedures for determining the total number of soil microbes were adopted for bacteria, yeast and moulds culturing. Suspension of the soil samples was prepared with sterile distils water and serial dilution of 5-7 factors was made for accurate counting. Then 1ml portion of the sample was aseptically pipetted into different sterile Petri dishes with the aid of sterile needle and syringe. About 20ml of the cool (40^o C) sterile molten agar media was added, swirled gently for even distribution of inoculums within the plate, then allowed to set and incubated. Nutrient agar plates for bacteria were incubated at 37^{oC} for 24hrs while PDA plates for fungi and yeast were incubated at 27^o C for 48 – 72 hrs. At the end of incubation period, the bacteria that grew into colonies were counted using colouring counters while yeast and fungi colonies were squarely counted and recorded appropriately per gram of the soil samples investigated respectively.

STATISTICAL ANALYSES

Analysis of variance was performed on all data to test the treatment effect on different parameters and significant means were separated using Tukey's Honest Significant Difference (HSD) ($P < 0.05$).

RESULTS AND DISCUSSION

The pre- planting soil physical and chemical properties of the organic wastes used for the experiments are presented in Tables 1 and 2.The results of the particle size analysis of Ibadan and Owena soils showed that the soils were sandy loam and Alfisols (Soil Survey Staff 1999). The silt + clay contents of the soils at Ibadan and Owena (24.98 % and 25.96 %) respectively were below the 32 % estimated as adequate soils ideal for tree crops especially cacao (Egbe et al. 1989). Based on the established critical levels for soils in south western Nigeria, the soils at Ibadan and Owena were acidic with pH ranging between 4.56 – 5.70 and low in organic matter (1.26 – 1.61 %) compared to the reported critical levels of 3 % organic matter (Agboola and Corey 1973). The total nitrogen of Owena soil was less than 0.15 % which is considered optimal for most crops including cacao and the soil also had low CEC (Ogunwale et al. 2002). This suggests the need to improve on the soil organic matter and hence the CEC for enhanced nutrient retention and the release of same to crops upon external fertilizer application (Agboola and Omueti 1982). The improvement of soil organic matter (SOM) can be achieved by

organic fertilizer application, either as sole or in combination with inorganic fertilizers as organo-mineral fertilizer (OMF). The application of inorganic fertilizer to a soil with low organic matter content is a waste of resources and time (Agboola and Obigbesan 1975). Hence there is need for proper SOM management on Ibadan and Owena soils to reduce the deleterious effects on soil physical, chemical and biological properties. Although, available P was also low in Ibadan, this level of available P is considered inadequate for cacao (Wessel 1971; Egbe et al. 1989). Only Ibadan soil gave exchangeable potassium above the critical value of 0.3 cmol/kg required for cacao. The exchangeable Ca^{+} of Owena soil fell below the critical value of 5 cmol/kg required for cacao growth. At both locations, the exchangeable Mg^{+} was adequate for cacao production. Obatolu (1991) earlier observed the general low Mg^{+} nutrient contents of these soils. The low nutrient contents of the soils implied the need for external input of nutrients in order to meet the requirements for optimal cacao growth. It is obvious that the soils of both Ibadan and were inherently low in fertility and were therefore expected to show positive response to soil amendment. The insufficient levels of the major nutrients in the soils in both locations showed that the soils were depleted in nutrients and would not be able to meet the nutritional needs of cacao plants unless external nutrients supply is made to support optimum growth of cacao plants.

Among the organic fertilizers applied, goat dung (GD) produced the highest pH, though the organic and organo-mineral fertilizers had pH above 5 (acidity levels) which indicated that they could be effective as liming materials. The organo-mineral fertilizer (OMF) had the highest available P followed by GD and organic fertilizer (OF) produced highest percentage N. The results were in agreement with the works of Adejobi et al (2011a) who reported that GD, OMF and OF were as effective as NPK fertilizer as sources of plant nutrients. In particular, OF had the highest OM, K, Mg, Ca and Na concentrations relative to others, this implied fact that OF could be a good source of these nutrients for plant growth. The organic manure types and rates applied increased the abundance of Verscular Abuscular Mychorizal Spores (VAMS) in the soil relative to NPK rates and the control in both Ibadan and Owena (Table 3). The presence or availability of mychorizal fungi is necessary for proper growth of seedlings in forestry nurseries and for successful start off on the field after transplanting (Mohammand et al. 2003). This result implied that organic sources of nutrients such as farmyard manure (FYM), compost, crop residues and slow release mineral fertilizers do not suppress AMF but stimulate them (Alloush and Clark 2001). However, it has been observed that over use of organic fertilizers especially those high in P, such as poultry manure, may impact negatively on AMF. However, the precise effect of organic amendments is unpredictable on any given soil or with any particular amendment (Alloush and Clack 2001). Jordan, (2000) found that AMF in an organically manured soil were effective at increasing crop available P comparable to application of super-phosphate. However, this does not always translate into higher yields even when phosphorous use efficiency is higher (Galvez et al. 2001). At Ibadan, application of 400 kg/ha OF enhanced abundance of AMF than other fertilizer types, rates and the control. The lower number of AMF count recorded under NPK rates was in agreement with the findings of Burrow and Pfeger (2002) who stated that use of readily soluble fertilizers, particularly N fertilizer, had negative impact on AMF colonization, population and diversity. Again, the result of this study showed that plots amended with the fertilizers suppressed the population of soil nematodes that are associated with cacao in both Ibadan and Owena experiments (Table 4). Soil treated with NPK fertilizer at 600 kg/ha and organo-mineral fertilizer (OMF) at 200 kg/ha gave the most effective nematode control at Ibadan and

Owena experiments. In both Ibadan and Owena soils, highest nematode populations were recorded on the control plots. Several studies have shown the advantages of using organic manures in controlling plant parasitic nematodes (Dias et al. 2000; Nagesh 2002 and Salgado et al. 2003). This study has shown that fertilizers like GD, OF, OMF and NPK have nematicidal effects on soil parasitic nematodes: *Meloidogyne incognita*, *Heterodera sahatti*, *Ditylenchus dispsaci*, *Paralongidorus sali*, and *Psilenchus sp* identified in this study. This result is consistent with the report of Riegel and Noe (2000) that application of poultry litter 14 days before planting was optimal for reduction of 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 and Matute 2003; Yeates and Boag 2004). In general, amending the soil with organic materials having low C: N ratio (less than 1:20) resulted in an abundance and enrichment of opportunist antagonistic microbes (Ferris and Matute 2003; Wang et al. 2004 and 2006) and rapid mineralization of N in the form of NH_4^+ or NO_3^- for absorption and uptake by plant roots (Powers and Mcsorley 2000). The fertilizers used in these experiments have low C: N (1:4, 1:1, 1:6) and this appeared to have resulted in the suppression of nematode population on cacao seedlings. It has been established that organic amendments released some chemicals into the soil that are directly responsible for nematode control. Ricin, a proprein derived from castor bean has nemato-toxic potential (Rich et al. 1998). The neem tree (*Azadirachta indica*) contains a group of chemicals known as limonoids and these compounds have proven highly effective in nematode control. Phenols and tannins are nematicidal at certain concentrations, and since some organic amendments added to soil contain high levels of these compounds, they may have a direct effect on nematode mortality (Badra and Eligindi 1979). Differences in soil microbial counts (Bacteria, Yeasts and Moulds) as obtained under the types and rates of fertilizers applied suggest their use for enhancement of fertility and crop yield (Table 5). The results showed that GD applied at 200 and 600 kg/ha NPK increased soil bacteria count significantly at both Ibadan and Owena 2012 experiments, 400 kg/ha OMF, 600 kg/ha OF and 200 kg/ha GD significantly enhanced yeast population at both Ibadan and Owena relative to the control and 200 kg/ha NPK and 400 kg/ha OF increased soil fungi at Ibadan and Owena experiments relative to other fertilizer treatments and the control. Stimulating effects of fertilizers on soil microbes was observed in the locations when compared with the control.

The enhancement of soil biological parameters by application of organic fertilizers on soil at 15 MAT has been reported by other authors (Bian et al. 2008; Emitsev et al. 2010). Additionally, as compared to other fertilizers and the control, the fungal populations at both Ibadan and Owena were significantly enhanced when 200 kg/ha NPK and 400 kg/ha OF were applied. The decline in the population of this group of micro-organisms in the control plots was an expected occurrence (Table 5). The lower fertilizer rates (200 and 400 kg/ha,) stimulated the development of the bacteria, yeasts and moulds in the two locations with the exception of bacteria in Owena (Table 5). This finding was in agreement with the results of Barabas et al. (2002), who reported an increase in the count and diversity of bacteria, actinomycete and fungal species under lower mineral nitrogen application. The decreased rates of application of mineral fertilizer (200 kg/ha) led to increase in the count of the fungal populations at both Ibadan and Owena. This tendency is frequently associated with changes in the soil physical and chemical characteristics (Stark et al., 2007) as well as with the alterations in the structure of soil microbial cenosis expressed through the predominance of

toxinogenic and phytopathogenic fungi. Also, Barabasz et al. (2002) has cautioned that inadequate application of N fertilizers can result in the production of toxic metabolites (nitrosamines, etc) that can have not only depressing effects on most soil micro-organisms but also cause teratogenic, carcinogenic and allergic effects in higher organisms (plants, animals and humans) through the food chain. In general the dynamics of microbial population during the growing season for cacao seedlings on the field, apart from effects of fertilizer types and rates can be attributed to the effects of climatic factors and the excretory function of the root and moisture regimes during the growing season (Bolton et al. 1992). The fertilizer treatments enhanced height of cacao seedlings than control in both locations at 3, 4 and 5 months after transplanting (Table 6). Cacao plants were taller at Ibadan than Owena at 5 MAT. The differences in height between the both locations were attributed to the differences in the nutrient- releasing pattern and growing environmental conditions between the two locations. The significant increases in the growth parameters of cacao seedlings at 3, 4 and 5 MAT in Ibadan and Owena relative to the control under the fertilizer treatments can be attributed to the nutrient contents of organic fertilizers applied which enhanced cacao seedlings growth. This finding that the manure (GD, OMF and OF) improved the growth of cacao is consistent with earlier findings of Adeniyan and Ojeniyi (2005) and Moyin-Jesu (2007) who reported that organic manures supported the growth and development of maize and coffee. Poor growth of cacao seedlings as a result of low nutrient status of soil was generally observed in the unfertilized treatment plots in the two locations in 2012 indicating that the soils of both locations were low in fertility and not supportive of good cacao growth. The higher plant height observed in Ibadan 2012 experiment at 3, 4 and 5 MAT, where the highest rate of NPK was applied (600 kg/ha) could be as a result of the rapid release of nutrients following mineralization and the consequent absorption for cacao growth. The number of branches recorded at control was lower but not significantly different from plots where the fertilizer types were applied in Ibadan and Owena at 3 MAT (Table 8). The similar performance of cacao seedlings with regards to number of branches under control plots (Ibadan and Owena 3 MAT) compared to organic fertilizer treatments might be due to the high initial nutrient contents of the soils.

The values of growth parameters (plant height, number of leaves and stem diameter) at 15, 16 and 17 MAT due to manures of both animal and plant origins were higher compared to that of inorganic origin (NPK 15: 15: 15 fertilizer, tables 9, 10 and 11) . This might be due to presence of other vital nutrient elements like Ca, Mg, OC and other micro-nutrients that are required for cacao seedlings growth which are absent in the NPK 15: 15:15 fertilizer.(Ipinmoroti et al. 2002) The relatively taller plant under 600 kg/ha OF in Ibadan at 15, 16 and 17 MAT could have stemmed from the nutrient contents of the organic fertilizers (essential soil nutrients) which though released slowly, last longer in the soil for optimum crop performance (Lombin 1981). Titiloye et al. (1985) have reported a survey of 45 waste materials which were found to be rich in the nutrient elements (N, P, K, Ca, Mg, Zn, Cu, Fe and Mn contents). The farm wastes therefore represent a potential source of nutrients that could be harnessed to boost crop growth and productivity (Solomon and Ogeh 1995).

Table 1: Physical and chemical characteristics of the soils before planting in Ibadan and Owena (2012 experiments)

Soil Properties	Ibadan 2012 Experiment	Owena 2012 Experiment
Sand (%)	75.1	74.1
Silt (Silt (%)	17.4	16.3
Clay (%)	7.5	9.6
Textural class	Sandy loam	Sandy loam
pH (water)	4.56	5.70
Organic carbon (g/kg)	0.43	1.51
Organ Organic matter (%)	1.26	1.61
Total I Nitrogen (%)	1.26	0.14
Available P (cmol/kg)	8.56	13.52
K^+ (cmol/kg)	3.26	0.24
$Ca^{++}Ca^{++}$ (cmol/kg)	6.00	2.60
Mg^{++} (cmol/kg)	3.00	1.00
Na^+ (cmol/kg)	2.52	0.15
Al^{+++}	1.21	1.86
H+(cmol/kg)	6.89	8.12
ECEC	19.88	13.97

Table 2: Chemical composition of the organic materials used

Properties	Goat dung (GD)	Organo-mineral fertilizer (OMF)	Organic fertilizer (OF)
pH (water)	8.17	7.00	7.30
Organic carbon (g/kg)	2.86	0.63	3.64
Organic matter (%)	4.80	1.09	6.27
Total nitrogen (%)	1.26	0.06	2.16
Available P (cmol/kg)	113.24	138.06	7.08
K^+ (cmol/kg)	0.41	0.19	5.56
Mg^{++} (cmol/kg)	1.20	1.00	6.00
Ca^{++} (c Ca^{++} cmol/kg)	2.60	2.00	13.10
Na^+ (cmol/kg)	0.38	0.18	2.30
C:N	1:4	1:1	1:6

Table 3: Effects of organic and inorganic fertilizer types and rates on albuscular mycchorizal spore count (Ibadan and Owena 2012 experiments)

Treatments	Ibadan 2012	Owena 2012
GD 600	28.00de	35.00bc
GD 400	47.33ab	54. 00a
GD 200	42.00abc	35.00ab
Control	26.87de	33.65bcd
OMF 600	37.66bcd	47.33a
OMF 400	20.00e	31.33bcd
OMF 200	41.00abcd	36.33ab
Control	28.21de	32.99bcd
OF 600	28.33cde	44.66ab
OF 400	52.00a	51.33a
OF 200	38.33bcd	36.33ab
Control	26.98de	32.87bcd
NPK 600	36.00bcd	34.00bc
NPK 400	33.66cd	33.00bcd
NPK 200	30.33cde	32.66bcd
Control	27.33de	33.66bcd

Treatment means within each column followed by the same letters are not significantly different from each other using Tukey's HSD at 5% level

Table 4: Total population of nematodes as affected by fertilizer types and rates 15 MAT at Ibadan and Owena 2012 experiments

Fertilizers Types	Rate (kg/ha)	Ibadan 2012	Owena 2012
GD	600	63.99j	129.66b
	400	252.31b	98.32f
	200	244.32c	102.99d
	Control	355.87a	195.43a
OMF	600	52.31k	81.32h
	400	83.32h	117.65c
	200	88.65g	55.65m
	Control	365.67a	194.00a
OF	600	105.64f	89.32g
	400	50.98l	101.98e
	200	165.65d	72.31j
	Control	357.12a	193.23a
NPK	600	14.00m	65.32k
	400	138.64e	73.97i
	200	80.31i	64.98i
	Control	358.31a	195.32a

Treatment means within each column followed by the same letters are not significantly different from each other using Tukey's HSD at 5% level.

Table 5: Effects of fertilizer types and rates on colonies of bacteria, yeasts and moulds in Ibadan and Owena (2012 experiments).

	Ibadan 2012			Owena 2012		
	Bacteria (cfu/g)	Yeasts (cfu/g)	Moulds (cfu/g)	Bacteria (cfu/g)	Yeasts (cfu/g)	Moulds (cfu/g)
GD 600	183.33b	100.00bc	25.00c	38.33e	30.66c	10.66bc
GD 400	200.00b	75.00de	20.00cd	121.00b	60.00ab	10.00bc
GD 200	295.00a	120.00b	26.66c	71.00cd	65.00a	11.00bc
Control	71.43d	44.32g	15.43cd	31.54e	27.54cd	9.00c
OMF 600	196.67b	100.00bc	25.00c	60.00d	55.00ab	15.66bc
OMF 400	196.00b	203.33a	19.66cd	134.66b	55.66ab	9.00bc
OMF 200	100.00cd	95.00cd	22.00cd	100.00c	55.66.ab	20.66b
Control	68.76d	40.99g	16.98ed	30.76e	26.76cd	7.99c
OF 600	170.00b	190.00a	16.00ed	42.00e	56.66ab	8.66c
OF 400	120.00c	70.00e	20.00cd	92.00c	50.00b	50.00a
OF 200	176.67b	50.00ef	24.00c	64.66d	30.66c	9.33bc
Control	70.98d	42.23g	16.12ed	30.11e	25.32cd	7.89c
NPK 600	160.00b	54.67ef	37.33b	270.00a	25.00cd	10.66bc
NPK 400	176.67b	61.67ef	22.00cd	30.00e	20.00d	6.00c
NPK 200	180.00b	110.00cd	45.33a	35.33e	50.00b	16.66bc
CONTR	70.00d	43.33fg	15.00ed	30.00e	25.00cd	8.00c

Treatment means within each column followed by the same letters are not significantly different from each other using Tukey's HSD at 5% level

Table 6: Effects of organic and inorganic fertilizer types and rates on plant height of cacao seedlings in Ibadan and Owena (2012 Experiments)

Treatments		Ibadan			Owena		
		Months after transplanting (MAT)			Months after transplanting (MAT)		
Fertilizers	Rates (kg/ha)	3	4	5	3	4	5
Goat Dung	600	40.20ab	44.80abc	5066abc.	35.89ab	45.18abc	51.23ab
	400	42.83ab	49.50ab	53.33ab	36.22ab	53.97ab	63.09a
	200	38.86ab	43.14bc	41.33bc	34.89ab	47.55abc	54.24ab
	Control	34.89b	37.32c	38.12c	23.99b	34.67c	32.41c
Organo-Mineral Fertilizer	600	43.70ab	49.27ab	52.33ab	30.33ab	42.09abc	46.95abc
	400	46.40ab	51.67ab	55.00a	25.67b	42.32abc	46.88abc
	200	43.70ab	50.17ab	54.33ab	33.00ab	53.11ab	58.58ab
	Control	35.43b	37.87c	37.98c	24.99b	34.56c	32.43c
Organic manure	600	43.33ab	49.41ab	54.33ab	31.28ab	43.56abc	46.04abc
	400	40.97ab	45.50abc	48.67abc	32.22ab	48.01abc	53.61ab

	200	42.53ab	47.35abc	52.33ab	39.53a	57.63a	62.52a
	Control	36.00b	36.99c	39.00c	25.00b	35.00c	31.98c
NPK	600	48.07a	55.21a	56.00a	33.22ab	51.08ab	51.76ab
15:15:15	400	47.23ab	50.82ab	53.00ab	32.61ab	44.39abc	45.88abc
	200	44.47ab	51.62ab	52.66ab	28.55ab	40.72bc	42.69bc
	Control	35.84b	37.47c	38.66c	24.89b	34.75c	32.41c

Treatment means within each column followed by the same letters are not significantly different from each other using Turkey's HSD at 5% level.

Table 7: Effects of organic and inorganic fertilizer types and rates on number of leaves of cacao seedlings in Ibadan and Owena (2012 experiments)

Treatments		Ibadan			Owena		
		Months After Transplanting			Months After Transplanting (MAT)		
Fertilizers	Rates	3	4	5	3	4	5
	(kg/ha)						
Goat	600	14.50a	17.17abc	20.00ab	17.00ab	20.17ab	26.35a
Dung	400	12.60ab	14.47bcde	17.00bcd	17.79ab	20.47a	25.67a
	200	18.83a	22.88a	25.67a	18.00a	19.13ab	21.77abc
	Control	7.45bc	8.98c	9.65e	11.11ab	12.43b	13.00d
Organo-	600	15.60a	19.64ab	21.67ab	14.22ab	17.06ab	22.73abc
Mineral	400	15.30a	19.11ab	21.67ab	9.33b	13.12ab	15.70cd
Fertilizer	200	15.60a	19.11ab	21.67ab	14.89ab	19.55ab	26.40a
	Control	7.89c	7.99e	10.00e	10.99ab	12.98b	13.11d
Organic	600	12.60ab	16.50abcd	19.00abc	14.33ab	16.81ab	23.76ab
manure	400	13.93a	17.73ab	21.33ab	15.66ab	20.29a	25.80a
	200	15.70a	19.23ab	22.33ab	14.54ab	18.17ab	23.68ab
	Control	6.99bc	7.98e	9.95e	11.00ab	12.55b	12.56d
NPK	600	6.43c	10.33cde	11.67cde	14.33ab	17.42ab	19.61abcd
15:15:15	400	6.53c	9.87de	11.33de	14.05ab	13.75ab	16.54bcd
	200	13.03ab	16.33abcd	17.33bcd	10.89ab	16.65ab	15.27cd
	Control	7.50bc	8.33e	9.67e	11.03ab	12.43b	12.89d

Treatment means within each column followed by the same letters are not significantly different from each other using Tukey's HSD at 5% level

Table 8: Effects of organic and inorganic fertilizer types and rates on number of branches of cacao seedlings in Ibadan and Owena (2012 experiments)

Treatments		Ibadan			Owena		
		Months after transplanting			Months after transplanting (MAT)		
Fertilizers	Rates	3	4	5	3	4	5
	(kg/ha)						
Goat	600	1.50a	2.06ab	2.38ab	0.00a	1.22ab	2.24ab
Dung	400	1.00a	1.85ab	2.53ab	0.00a	0.53abc	1.24ab
	200	1.00a	1.84ab	2.15ab	0.22a	0.42abc	0.63ab
	Control	0.23a	0.34b	0.56b	0.00a	0.00c	0.17b
Organo-	600	1.00a	2.15ab	2.48ab	0.00a	0.12c	1.18ab
Mineral	400	1.67a	2.59ab	3.60ab	0.00a	0.17c	0.63ab
Fertilizer	200	1.00a	1.66ab	2.07ab	0.53a	0.78abc	1.52ab
	Control	0.24a	0.40b	0.60b	0.00a	0.00c	0.19b
Organic	600	1.33a	2.36ab	2.97ab	0.00a	1.31a	2.27a
fertilizer	400	2.33a	3.68a	4.10a	0.00a	0.83c	1.72ab
	200	2.33a	3.75a	4.17a	0.44a	0.86abc	1.43ab
	Control	0.27a	0.43b	0.76b	0.00a	0.01c	0.19b
NPK	600	1.00a	2.02ab	2.15ab	0.22a	0.61abc	1.05ab

15:15:15	400	0.90a	1.80ab	2.03ab	0.00a	0.00c	0.59ab
	200	2.17a	3.47a	3.57ab	0.00a	0.22bc	0.44b
	Control	0.25a	0.34b	0.52b	0.00a	0.00c	0.18b

Treatment means within each column followed by the same letters are not significantly different from each other using Tukey's HSD at 5% level

Table 9: Effects of organic and inorganic fertilizer types and rates on plant height of cacao seedlings in Ibadan and Owena (2012 experiments)

Treatments		Ibadan			Owena		
	Rates	Months After Transplanting (MAT)			Months After Transplanting (MAT)		
	(kg/ha)	15	16	17	15	16	17
Goat	600	71.67abc	73.00abcd	80.33abc	157.00ab	196.00ab	205.67a
Dung	400	67.67abc	69.00abcd	72.33bcd	159.67ab	192.33abc	196.33ab
	200	77.67abc	81.00abcd	86.67abc	186.67a	191.33abc	192.67ab
	Control	45.43c	47.43d	50.87d	108.32b	87.54f	90.65e
Organo-	600	70.33abc	77.67abcd	85.67abc	160.67ab	205.67a	207.33a
Mineral	400	86.33ab	91.33ab	95.33ab	142.00ab	137.33bcd	140.67bcde
Fertilizer	200	78.33abc	83.33abc	91.67abc	129.67ab	171.67abcd	175.00abc
	Control	44.76c	47.76d	51,98d	104.98b	86.21f	88.00e
Organic	600	93.33a	99.67a	105.67a	152.67ab	179.00abc	184.33ab
Manure	400	84.67ab	90.33abc	96.67ab	149.00ab	161.00abcd	165.00abcd
	200	87.67ab	90.33abc	96.67ab	146.67ab	160.33abcd	163.67abcd
	Control	47.98c	48.56d	50.78d	105.98b	84.98f	85.99e
NPK	600	67.00abc	70.00abcd	80.00abc	113.50b	130.00cdef	136.00bcde
15:15:15	400	54.73bc	56.33cd	63.33cd	111.00b	137.33bcde	144.00abcd
	200	69.33abc	73.67abcd	78.67abc	139.67ab	111.00def	115.33cde
Control		46.00c	48.00d	50.00d	106.00b	85.33f	87.00e

Treatment means within each column followed by the same letters are not significantly different from each other using Tukey's HSD at 5% level

Table 10: Effects of organic and inorganic fertilizer types and rates on number of leaves of cacao seedlings in Ibadan and Owena (2012 experiments)

Treatments		Ibadan			Owena		
	Rates	Months After Transplanting (MAT)			Months After Transplanting (MAT)		
Fertilizers	(kg/ha)	15	16	17	15	16	17
Goat	600	93.33a	93.33a	101.00a	77.00ab	101.33b	106.00b
Dung	400	42.67bc	43.67bcd	47.67bc	57.67abc	135.00ab	137.67ab
	200	44.00bc	48.33bcd	53.33bc	83.00a	156.67ab	161.00ab
	Control	21.33c	23.21d	24.54c	24.87bc	76.65b	82.00b
Organo-	600	60.00b	61.00abc	69.33ab	67.33abc	171.67ab	176.00ab
Mineral	400	45.67bc	47.33bcd	51.67bc	40.67abc	70.00b	72.33b
Fertilizer	200	32.67bc	35.67bcd	39.67bc	93.00a	306.00a	310.00a
	Control	19.99c	22.98d	23.67c	24.98bc	79.09b	87.09b
Organic	600	51.33bc	58.00bcd	65.67b	56.67abc	114.33ab	121.00ab
Manure	400	33.67bc	35.33bcd	44.67bc	47.67abc	110.33ab	111.67ab
	200	34.67bc	38.33bcd	43.67bc	38.33abc	114.33ab	118.00ab
	Control	20.87c	24.98d	23.78c	25.00bc	79.78b	80.98b
NPK	600	60.00b	65.00ab	70.00ab	35.50abc	120.00ab	125.00ab
15:15:15	400	26.00c	29.67cd	37.33bc	50.67abc	48.00b	52.00b
	200	37.00bc	40.00bcd	48.67bc	22.00bc	80.67b	83.00b
	Control	20.33c	23.33d	24.67c	24.33bc	78.67b	81.00b

Treatment means within each column followed by the same letters are not significantly different from each other using Tukey's HSD at 5% level

Table 11: Effects of organic and inorganic fertilizer types and rates on number of branches of cacao seedlings at 15, 16 and 17 MAT in Ibadan and Owena (2012 experiments)

Treatments		Ibadan Months After Transplanting (MAT)			Owena Months After Transplanting (MAT)		
Fertilizers	Rates (kg/ha)	15	16	17	15	16	17
Goat Dung	600	12.86a	13.33a	15.33a	11.16ab	17.33abc	17.33abcd
	400	9.33abc	10.00ab	12.66ab	8.00abc	24.00abc	26.33abcd
	200	5.33abc	7.00ab	7.83ab	7.66abc	30.33ab	34.33a
	Control	2.43c	3.55b	3.56b	3.51c	5.11c	6.56d
Organo-Mineral Fertilizer	600	7.00abc	8.00ab	7.00ab	12.06a	30.00ab	31.00abc
	400	6.33abc	8.00ab	8.00ab	6.00abc	13.66abc	16.66abcd
	200	5.00abc	5.66ab	7.00ab	9.50abc	25.66abc	27.66abcd
	Control	2.33c	3.56b	3.67b	3.53c	4.99c	6.65d
Organic Fertilizer	600	9.66abc	11.66ab	12.66ab	9.16abc	24.66abc	33.00ab
	400	6.86abc	7.66ab	9.00ab	6.50abc	34.00a	38.00a
	200	9.66abc	11.33ab	13.33ab	7.16abc	30.66ab	35.00a
	Control	2.33c	3.76b	3.32b	3.56c	5.00c	6.67d
NPK 15:15:15	600	12.00ab	12.50ab	13.50ab	6.75abc	12.50abc	11.00bcd
	400	3.53bc	3.90ab	3.90ab	6.00abc	9.00bc	10.66bcd
	200	9.20abc	9.66ab	13.33ab	5.66abc	11.66abc	9.66cd
	Control	2.33c	3.66b	3.66b	3.50c	5.00c	6.66d

Treatment means within each column followed by the same letters are not significantly different from each other using Tukey's HSD at 5% level

CONCLUSION

This research work explored the effects of Goat Dung (GD), Sunshine Organic and Organo-Mineral Fertilizers (OF and OMF) and NPK 15:15:15 on soil biological properties and growth of cacao between 2012 and 2014 in two cacao growing ecologies of south western, Nigeria. The results showed that organic manures applied increased the abundance of Verscular Abuscular Mychorizal Spores (VAM) in the soil relative to NPK and the control in Ibadan and Owena. Soil application of 400 kg/ha OF enhanced AMF compared with other fertilizer types and the control in Ibadan. Application of 600 kg/ha NPK and 200 kg/ha OMF produced the lowest nematode count in Ibadan and Owena. Similar trends were observed in Ibadan and Owena, where highest nematode populations were recorded for the control plots.

The total count of soil bacteria was highest for 200 kg/ha GD and 600 kg/ha NPK at Ibadan and Owena. Fungal populations at Ibadan and Owena were significantly enhanced when 200 kg/ha NPK and 400 kg/ha OF were applied. Lower rates of application of NPK fertilizer at 200 kg/ha led to increased fungal populations at both Ibadan and Owena. In Ibadan (2012 experiment), application of NPK at 600 kg/ha gave significant ($P<0.05$) plant height at both 3,4 and 5 MAT of cacao relative to the control while 600 kg/ha OF gave significant ($P<0.0.5$) higher plant higher at both 15, 16 and 17 MAT relative to the control. Again, in Owena, 200 kg/ha OF enhanced plant height at 3, 4 and 5 MAT compared with the control. However, 200 kg GD enhanced higher plant height at 15 MAT compared with the control and 600 kg/ha OMF significantly increased height of cacao at 16 and 17 MAT respectively. Similar trends were obtained in Owena

at application of 200 kg/ha OMF produced significantly higher number of leaves at 15, 16 and 17 MAT over the control. The effect of 200 kg/ha GD was significant on number of leaves in Ibadan at 3, 4 and 5 MAT. Also, 600 kg/ha GD produced marked effect on number of cacao leaves relative to the control at 15, 16 and 17 MAT.

Application of 400 and 200 kg/ha OF had profound effect on number of branches in Ibadan at 3, 4 and 5 MAT compared with other treatments and the control. 600 kg/ha GD had better influence on number of branches in Ibadan at 15, 16 and 17 MAT compared with other treatments and the control.

In Owena at 4 and 5 MAT, the number of branches produced under 600 kg/ha OF was higher compared with other treatments and the control. At 15 MAT, the number of branches produced under 600 kg/ha OMF was greater than what obtained under other treatments and the control followed by 600 kg/ha GD, while application of 400 kg/ha OF produced higher number of branches at 16 and 17 MAT which was about 85 and 82 % increases over the control respectively. In order to suppress nematode population at Ibadan, 600 kg/ha NPK is recommended. Adoption of application of 400 kg/ha OF and 200 kg/ha OMF is recommended for reducing soil nematode population at Owena.

Higher rate of GD (600 kg/ha) is recommended to promote bacterial population. 200 kg/ha OMF is recommended for increase in yeast population and 400 kg/ha OF for fungi at Ibadan, while 400 kg/ha NPK is recommended for optimum fungi population at Owena. In order to increase soil bacteria and yeast population at Ibadan and Owena to attain rapid decomposition of organic matter, 200 kg/ha GD is recommended. The fertilizer treatments enhanced the growth parameters, soil and leaf chemical composition of cacao seedlings than the control in both locations, organic manures performed better than the organo-minerals (OMF), while the organo-mineral fertilizers were better than NPK.

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