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Evaluation of poultry manure application rate and plant population on growth, dry matter partitioning and nutrient uptake of Cock's comb (*Celosia argentea. L*)

Makinde, E. A.; Salau, A.W. and Odeyemi, O.M

Department of Horticulture, Federal University of Agriculture, Abeokuta. P.M.B. 2240, Abeokuta, 110001, Nigeria.

ABSTRACT

Cock's comb (*Celosia argentea* L.) is an important leaf vegetable in the tropics grown for its succulent leaves that are rich sources of protein, vitamins and minerals essential for combating malnutrition, especially in the rural communities. Two field experiments were conducted to evaluate the growth, dry matter partitioning and nutrient uptake of Cock's comb in response to different application rates of poultry manure (PM) and plant population between August 2013 and July 2014. The experiment was a split plot arrangement fitted into a randomized complete block design, with four replicates. PM was applied at 0, 5 and 10 t/ha while Cock's comb was established at plant populations of 200,000 and 400,000 plants/ha in the late season of 2013 and early season of 2014 concurrently. Data were taken on plant growth, dry matter partitioning, yield and nutrient contents. Application of 5 t/ha PM significantly ($P < 0.05$) increased plant height, number of leaves, leaf area, total fresh yield of stem and leaves, leaf dry weight, stem dry weight, root dry weight and total dry weight relative to plants in control plots in the early and late seasons. Plant population had no significant effect on growth and dry matter partitioning of Cock's comb. Interaction of PM and plant population on nitrogen uptake was significantly high when 10 t/ha PM was applied at 400,000 plants/ha. Phosphorus and Potassium uptake increased in plots fertilized with 5 t/ha PM at 400,000 plants/ha. For optimum growth, dry matter yield and nutrient uptake, Cock's comb could be planted at a population up to 400,000 plants/ha with 5 t/ha PM.

Keywords: Cock's comb, *Celosia argentea*, performance, plant nutrients, manure.

INTRODUCTION

Cock's comb, *Celosia argentea* (L) is a popular leaf vegetable in the tropics. The leaves are very succulent and serve as an excellent source of protein, calcium, iron, vitamins A and C. It is majorly cultivated on small and scattered plots in home gardens, farmland and urban and peri-urban areas, making it difficult to estimate the cultivated area, but it amounts to thousands of hectares (Denton, 2004). In the tropics, Cock's comb is produced by small holder farmers, solely or intercropped with arable starchy staples to produce enough food to satisfy their dietary and cash requirements (Gbadamosi and Adeoluwa, 2014). The leaves and succulent stem are consumed as vegetable because they constitute a cheap and rich nutrient sources for the low income earners, and the seeds could also be processed into food items, supplement and additives (Schippers, 2000). It is also used for medicinal purposes and the treatment of ailments such as abscesses, cough, diabetes, diarrhoea, dysentery, eczema, eye problems, gonorrhoea, infected sores, liver ailments, menstruation problems, muscle troubles, skin eruptions, snakebites and wounds (Schippers, 2000). Cock's comb thus has high potentials for reducing malnutrition which is rampant in the tropics (Adeyeye *et al.*, 2013)

Cock's comb grows rapidly from seed and it is easy to plant in most climates and thrives in well-drained soils with pH 6.0 - 6.4 (Grubben, 2004). It could either be cultivated during the wet or dry season but irrigation is usually required for the dry season cultivation (Schippers 2000). However, there has been rapid decline in tropical soil fertility and crop productivity has become a major concern and indeed a great hindrance to achieving food sufficiency in the tropics (Babajide and Olla, 2014) Achieving food security in its totality has therefore been a major challenge. Farmers are often faced with the challenge of producing Cock's comb on poor soil conditions resulting from continuous cultivation.

Cock's comb could however, be cultivated using either organic or inorganic fertilizers but the use of synthetic, inorganic fertilizers has been on a decline, resulting from high cost and scarcity in Nigeria. Comparing the growth, dry matter accumulation and shoot yield of Cock's comb with poultry manure and urea inorganic fertilizer, Adeyeye *et al.* (2013) has recommended either 140 kg N/ha Urea or 6 t/ha poultry manure. However, an optimal rate for fresh shoot weight of 90 kg/ha NPK 15-15-15 was recommended by Ojo (1998). According to Adedokun and Aiyelaagbe (2008), synthetic, inorganic fertilizers provide readily- available nutrients for plants; and their uses in Nigerian agriculture is hampered by problems of high cost, scarcity and lack of established soil testing programmes. Some of the chemical fertilizers, like Urea and Sulphate of ammonia aggravate soil acidity, as a result of H⁺ released into the soil. Also, Nigerian farmers have limited access to synthetic fertilizers because of its low production, availability, procurement and poor distribution (NISER, 2003). The undesirable pollution of the soil and water by inorganic fertilizers is also responsible for increasing rate of the use of organic manures that are cheaper, readily available, and less harmful (Eghareva and Ogbe, 2002). Organic manures however, increase soil nutrient status and enhance the soil biological, chemical and physical properties (FAO 2005; Ibeawuchi *et al.*, 2006; Odeyemi *et al.*, 2015) and support crop performance and yield (Adebayo and Akoun, 2002). Poultry manure is an organic waste material consisting of faeces and urine from poultry. It is an excellent fertilizer material because of its high nutrient content, especially nitrogen, phosphorus and potassium (Hochmuth *et al.*, 2009). It decomposes in the soil and releases nutrients for crop uptake. It also serves as a soil amendment to increase the organic matter content of the soil which increases the soil moisture holding capacity; improves overall soil structure; lowers soil bulk density and thus increasing the efficiency of the crop production and irrigation. Poultry manure application has been reported to increase N, P, K uptake in fluted pumpkin leaves (Awodun, 2007).

Plant population affects competition for growth resources. Plant spacing, planting density and spatial arrangement make up the population. Same population can be achieved with different spacings at different spatial arrangements. Using the same spacing with varying planting densities results in varying populations. Increased plant population decreases plant yield and yield components. Decreased plant growth and yield due to competition may result from high plant populations. The increased population may however compensate for decreases in growth and yield. High planting density can be sustained with adequate nutrient supplies. Optimum population of Cock's comb needs to be investigated with nutrient supplies. This study was conducted to determine the poultry manure application rate and plant population that supports optimum growth, nutrient uptake and dry matter yield of Cock's comb, *Celosia argentea*.

MATERIALS AND METHODS

Location of the Experiment

Two field trials were conducted at the Federal University of Agriculture, Abeokuta, Nigeria (7° 15' N, 3° 20' E; 100 m above sea level) between August and October, 2013 and between May and July, 2014. The area lies within the forest savanna transition zone (Aiboni, 2001). The wet season usually extends from March to October while the dry season starts in November and ends in February. Total rainfall recorded was 266.6 mm during the 2013 planting period but was 321.0 mm during the 2014 planting period. Monthly maximum temperature range was between 28.6°C and 31.7°C in 2013 but was between 29.9°C and 32.1°C in 2014. The monthly minimum temperature range was between 21.1°C and 23.1°C in 2013 but was between 23.3°C and 23.4°C in 2014. Relative humidity was between 67.2 and 71.4 % in 2013 but was between 64.4 and 68.8 % in 2014 (Table 1).

Cropping History

A pre-cropping visual estimation of the vegetation of the site indicated that broad leaf weeds were more common than grasses. Prominent weeds were: *Aspilia africana* (Pers) C. and *Eleusine indica* Gartn. Other weeds present were: *Alternanthera pungens* H.B.K.; *Mariscus flabelliformis* Kunth and *Aneilema beninense* L. The site had been cropped with cereals and fruit vegetables for two years, using crop residues to re-cycle nutrients and left un-cropped for two years prior the establishment of this trial.

Pre Cropping Soil and Poultry Manure Analysis

The soil of the experimental plot was a sandy - loam with a pH of 6.9. Nitrogen was very low at 0.08 %, available P and the exchangeable K were also marginal (Table 2). The poultry manure (PM) contained 2.5, 3.9 and 2.2 % N, P and K, respectively with an organic matter of 25.5 % (Table 2). The 5 t/ha PM was applied to supply 127, 196 and 110 kg/ha of N, P and K, respectively while the 10 t/ha PM was applied to supply 225, 392 and 221 kg/ha of N, P and K, respectively.

Experimental design and treatments

The experiment was a split plot arrangement fitted into a randomized complete block design, replicated four times. Poultry manure was applied at 0, 5 or 10 t/ha at 2 weeks before sowing on sub-plots of 3 m by 1.5 m with border spacing of 1m with intra row spacings of 10 or 5 cm to give the populations of 200,000 and 400,000 plants/ha, respectively.

Crop Husbandry

Seeds of Cock's comb (var. TLV 8) were planted in August, 2013 for the late season and in May, 2014 for the early season. Weeding was carried out at 3 weeks after sowing (WAS), manually with a hoe. Harvesting was at 4 and 6 WAS by total plant removal method. The experiment was repeated

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between May and July, 2014 with the same procedures as with earlier cultivation but without a re-application of poultry manure. The plots were maintained as in the previous year, with new crops established.

Soil and Plant analysis

Prior to planting, the initial nutrient content of the soil was determined by taking ten core samples from the top 20 cm of the soil. At the end of both late and early planting seasons, three core samples were taken per plot, bulked to have one composite sample per plot that was analysed. Leaves from five tagged sample plants from the inner rows were detached at 4 weeks after sowing in both seasons to determine the foliar nutrient content. The leaf samples were placed in paper bags and air dried at room temperature (28°C) for 48 hours.

Data collection

Data collected include: plant shoot height, number of leaves/plant, average leaf area (cm²), fresh plant weight (g), dry plant weight (g/plant). Leaf area was estimated from its relationship with the midrib length. One hundred leaves of varying sizes from the upper, middle and lower portions of plants from the border rows were detached at 4 weeks after sowing. The mid rib lengths and width of each leaf was measured. The area of each leaf was determined by graph paper tracing. The measurements of the lengths and the widths were each regressed with the graph paper tracing values to generate a regression relationship.

The resulting regression relationship for the leaf mid rib length was:

$$Y = 7.49X - 35.66 (R^2 = 0.93)$$

For the leaf breadth, the resulting regression relationship was:

$$Y = 8.51X - 11.67 (R^2 = 0.83)$$

The relationship with the midrib length was adopted, as a result of the better relationship.

Mid rib lengths of five leaves/plant from the upper, middle and lower portions of the foliage of ten plants per plot sampled from the inner rows were measured at 4 weeks after sowing. The average per plot was used to estimate the leaf area, using the generated regression equation:

$$Y = 7.49X - 35.66 (R^2 = 0.93)$$

Where: Y = Estimated Leaf Area

X = Average mid rib length of leaves from 10 plants/plot.

The sampled plants were separated into leaves, stem and root to determine the dry matter contents. Plant dry weight was from the fresh samples dried in the oven at 105°C for 48 hours.

Plant nutrient uptake (kg/ha) was calculated as the product of plant sample concentration and the biomass generated per treatment (Drakopoulos *et al.*, 2015)

Nutrient Uptake = Nutrient Concentration x Plant Biomass/treatment.

Apparent Nitrogen recovery (ANR) was calculated as:

$$ANR = \frac{(N_{trt} - N_{ctl})}{N_{app}} \times 100$$

Where: N_{trt} = Nitrogen Uptake in treated plant, averaged over the replicates.

N_{ctl} = Nitrogen Uptake in untreated plant

N_{app} = Nitrogen applied

Same procedure for Apparent Nitrogen Recovery (ANR) was applied for Apparent Phosphorous Recovery (APR).

Data Analysis

Data obtained were subjected to analysis of variance (ANOVA) using procedures of GenStat statistical software package (2011). Treatment means were separated using Least Significant Difference (LSD) p 0.05.

RESULTS

Plant height, number of leaves/plant and leaf area of Cock's comb as affected by poultry manure rate and plant population: Application of poultry manure significantly increased plant height, number of leaves/plant and the leaf area of *Celosia argentea* both at the late and early seasons (Table 3). At 4 and 6 WAS in both late and early seasons, plants from plots fertilized with 5 t/ha grew taller than plants on 0 t/ha plots that were not fertilized but were comparable with plots fertilized with 10 t/ha. The number of leaves/plant recorded a similar trend as the plant height, with 5 t/ha application resulting in comparable number of leaves/plant as 10 t/ha but were significantly ($P < 0.05$) higher than from the untreated control plots. In the late season, the unfertilized plots could not develop fully-expanded leaves, even with 6 weeks of growth. Plant leaf area with 5 t/ha application were comparable with 10 t/ha application but were higher than from the untreated control plots (Table 3). Plant population had no significant effect on plant height, number of leaves/plant and leaf area in both seasons (Table 3).

Yield of *Celosia argentea* as affected by poultry manure rate and plant population: In both seasons, application of 5 t/ha of PM significantly influenced the fresh leaf, stem and root weights of *Celosia argentea* (Table 4). The fresh leaf weight was higher in both seasons at either 4 or 6 WAS, relative to the control plants. In the late season, 5 t/ha had higher root weights than 10 t/ha at 4 WAS but were similar at 6 WAS. The unfertilized plots had lower root weights at either 4 or 6 WAS. In the early season, application of 10 t/ha PM resulted in comparable leaf weights, except at 6 WAS. The fresh stem weight recorded a similar trend as the fresh leaf weight. Application of 10 t/ha also resulted in comparable stem weights except at 4 WAS in the early season. By 6 WAS, fresh root weight with the unfertilized plots were similar with root weights from both manure levels. (Table 4). Plant population did not significantly affect *Celosia* plant fresh leaf weight, fresh stem weight, fresh root weight and total fresh weight (Table 4).

Biomass accumulation of *Celosia argentea* as affected by poultry manure rate and plant population: Total plant fresh weight was higher with 5 t/ha than with 10 t/ha PM at both 4 and 6 WAS in the two seasons. The unfertilized plots accumulated a lower fresh weight than the two manure levels in the both seasons (Table 5). At 6 WAS, with 5 t/ha, total plants fresh weight was reduced from 1206 to 371 kg/ha in the late season while at 10 t/ha, it was reduced from 807 to 244 kg/ha. Total plants dry weights were generally about 8% of the fresh weight at 4 WAS in the early season and were all similar. At 6 WAS, dry plant weight from 5 t/ha PM was about 8.8 % of the fresh weight and was higher than the 8.0 % dry plant weight of plants fertilized with 10 t/ha. The unfertilized plots had lower dry plant weights of about 8.2 % of their fresh weight. In the late season at 4 WAS, application of 5 t/ha PM had a plant dry weight that was 6.9 % of the fresh weight while 10 t/ha had a weight which was 7.5 % of the fresh weight. By 6 WAS, total plant weight from 5 and 10 t/ha PM were both 12.6 % of their fresh weights (Table 5). Plant population had a similar range of dry weight proportions over the seasons. There was no interaction effect of the poultry manure and plant population on plant biomass accumulation (Table 5).

Biomass partitioning of *Celosia argentea* as affected by poultry manure rate and plant population: Leaf dry weight of *celosia* was not influenced by the rate of poultry manure applied in the early season at 4 WAS. However, at 6 WAS, plots fertilized with 5 t/ha produced higher leaf dry weight, compared with 0 and 10 t/ha that had lower but similar dry leaf weights (Table 6). The trend was similar in the late season, with 5 t/ha having higher leaf weight at 4 WAS. At 6 WAS, 10 t/ha had comparable leaf weight. The unfertilized plots had lower leaf dry weights. Both the dry stem weight

International Journal of Organic Agriculture Research & Development Volume 13, Sept. (2016) and the dry root weights had a similar trend as the dry leaf weight. They were similar at 4 WAS in the early season but 5 t/ha had higher weight at 6 WAS in the early season and throughout the late season. They were however, comparable with weights from 10 t/ha. The unfertilized plots generally had lower weights than from 5 t/ha PM. The leaf, stem and root dry weights were generally higher at 6 WAS, relative to 4 WAS in both seasons but were all comparable with both populations at each period. About 80 % of the plant parts were generally consumable, with plant roots less than 20 % (Fig.1). The interaction of poultry manure and plant population on biomass partitioning was not significant (Table 6).

Table 1: Meteorological data during the experiment in 2013 and 2014 at Abeokuta

	Rainfall (mm)	Relative Humidity (%)	Temperature (°C)	
			Maximum	Minimum
2013				
August	35.2	71.7	28.6	21.1
September	136.0	69.7	28.9	22.4
October	94.4	67.2	31.7	23.1
Total	265.6			
2014				
May	113.8	67.7	32.1	23.4
June	116.5	64.4	31.5	23.4
July	90.7	68.8	29.9	23.3
Total	321.0			

Source: Department of Agrometeorology and Water Resources Management, University of Agriculture, Abeokuta, Nigeria (2013-2014)

Table 2: Initial soil chemical and physical properties and poultry manure analysis.

Parameter	Soil	Poultry Manure
pH (H ₂ O)	6.9	7.48
Organic matter	1.15 %	25.5 %
Organic Carbon	0.67 %	14.83 %
Total N	0.08 %	2.55 %
Available P	5.88 mg kg ⁻¹	3.92 %
Exchangeable K	0.28 cmol kg ⁻¹	2.21 %
Exchangeable Ca	8.68 cmol kg ⁻¹	8.45 %
Exchangeable Mg	2.01 cmol kg ⁻¹	0.36 %
Exchangeable Na	0.77 cmol kg ⁻¹	1.23 %
ECEC	11.53	- ^a
Base Saturation	996.00 g.kg ⁻¹	- ^a
Sand	932.00 g.kg ⁻¹	- ^a
Silt	60.00 g.kg ⁻¹	- ^a
Clay	8.00 g.kg ⁻¹	- ^a
Textural class	Sandy loam	- ^a

-^a = not determined.

Table 3: Plant height, number of leaves/plant and leaf area of *Celosia argentea* with poultry manure rate and plant population

Treatment	Plant height (cm) *				Number of leaves/plant*				Leaf area (cm ²) *			
	Late season		Early season		Late season		Early season		Late season		Early season	
	4*	6	4	6	4	6	4	6	4	6	4	6
Poultry manure (PM)												
0 t/ha	0.0	0.0	10.3	31.0	0.0	0.0	9.1	18.9	0.0	0.0	25.7	102.0
5 t/ha	11.6	19.5	17.0	56.7	12.9	16.8	14.1	34.1	15.2	14.0	50.4	196.0
10 t/ha	11.5	18.5	13.5	48.6	12.3	17.4	13.1	36.3	12.5	13.4	37.4	166.0
LSD (5%)	2.4	3.6	6.1	14.5	1.0	1.2	4.1	10.0	3.1	2.5	22.3	53.6
Plant Population (PP)												
200,000 plants/ha	7.7	12.8	12.3	39.8	8.5	11.8	11.1	28.6	9.7	9.6	33.0	133.0
400,000 plants/ha	7.7	12.5	14.9	51.2	8.3	11.1	13.0	30.9	8.8	8.7	42.7	175.0
LSD (5%)	ns	ns	ns	ns	Ns	ns	ns	ns	ns	ns	ns	ns
PM x PP	ns	ns	ns	ns	Ns	ns	ns	ns	ns	ns	ns	ns

*WAS: Weeks after sowing

Table 4: Fresh weight of *Celosia argentea* as influenced by poultry manure rates and plant population

Treatment	Fresh leaf weight (g/plant)				Fresh stem weight (g/plant)				Fresh root weight (g/plant)			
	Late season		Early season		Late season		Early season		Late season		Early season	
	4*	6	4	6	4	6	4	6	4	6	4	6
Poultry manure (PM)												
0 t/ha	0.0	0.0	34.0	87.0	0.0	0.0	21.0	68.0	0.0	0.0	7.9	30.0
5 t/ha	44.6	58.6	112.0	209.0	28.2	79.6	86.0	219.0	3.5	29.1	29.4	114.0
10 t/ha	32.2	43.1	66.0	137.0	16.0	47.2	56.0	168.0	2.0	19.2	23.8	58.0
LSD 5%	17.6	24.4	56.3	56.9	18.4	46.5	58.5	107.6	0.8	15.5	20.4	58.8
Plant Population (PP)												
200,000 plants/ha	24.2	40.7	65.0	160.0	13.8	54.9	47.0	166.0	102.7	259.9	18.8	86.0
400,000 plants/ha	27.0	27.2	76.0	128.0	15.7	29.7	61.0	138.0	115.6	151.1	21.9	48.0
LSD 5%	ns	ns	Ns	ns	Ns	ns	ns	ns	ns	ns	ns	ns
PM x PP	ns	ns	Ns	ns	Ns	ns	ns	ns	ns	ns	ns	ns

*WAS Week after sowing

Table 5: Biomass accumulation of *Celosia argentea* with poultry manure rate and plant population

Treatment	Plant fresh weight (kg/ha)				Plant dry weight (kg/ha)			
	Late season		Early season		Late season		Early season	
	4*	6	4	6	4	6	4	6
Poultry manure (PM)								
0 t/ha	0.0	0.0	62.9	185.0	0.0	0.0	5.2	15.1
5 t/ha	76.3	167.3	227.4	542.0	6.2	21.0	18.4	47.8
10 t/ha	50.2	198.5	145.8	363.0	4.4	14.0	11.7	29.0
LSD (5%)	15.7	28.7	55.4	36.3	0.3	4.8	9.6	4.7
Plant Population (PP)								
200,000 plants/ha	140.7	355.5	130.8	312.0	4.0	8.4	10.6	33.3
400,000 plants/ha	158.3	208.0	158.9	314.0	3.1	8.4	12.9	28.0
LSD (5%)	Ns	Ns	ns	Ns	ns	ns	ns	Ns
PM x PP	Ns	Ns	ns	Ns	ns	ns	ns	Ns

* WAS: Weeks after sowing

Table 6: Biomass partitioning of *Celosia argentea* with poultry manure rate and plant population

Treatment	Dry Leaf Weight (g/plant)				Dry Stem Weight (g/plant)				Dry Root Weight (g/plant)			
	Late season		Early season		Late season		Early season		Late season		Early season	
	4*	6	4	6	4	6	4	6	4	6	4	6
Poultry manure (PM)												
0 t/ha	0.0	0.0	3.1	7.9	0.0	0.0	1.3	4.2	0.0	0.0	0.8	3.0
5 t/ha	3.5	9.9	10.8	20.5	1.3	5.6	5.0	16.5	1.4	5.5	2.6	10.8
10 t/ha	2.0	7.8	7.8	12.9	1.4	3.3	2.8	9.6	1.0	2.9	1.1	6.5
LSD (5%)	0.8	4.4	Ns	6.4	0.7	3.4	3.8	7.2	0.4	3.5	ns	4.8
Plant Population (PP)												
200,000 plants/ha	2.1	7.3	5.8	14.7	1.0	3.6	3.0	10.7	0.9	3.8	1.8	7.9
400,000 plants/ha	1.6	4.4	8.6	12.8	0.8	2.2	3.0	9.6	0.7	1.8	1.3	5.6
LSD (5%)	ns	ns	Ns	ns	ns	ns	ns	ns	ns	ns	ns	ns
PM x PP	ns	ns	Ns	ns	ns	ns	ns	ns	ns	ns	ns	ns

* WAS: Weeks after sowing

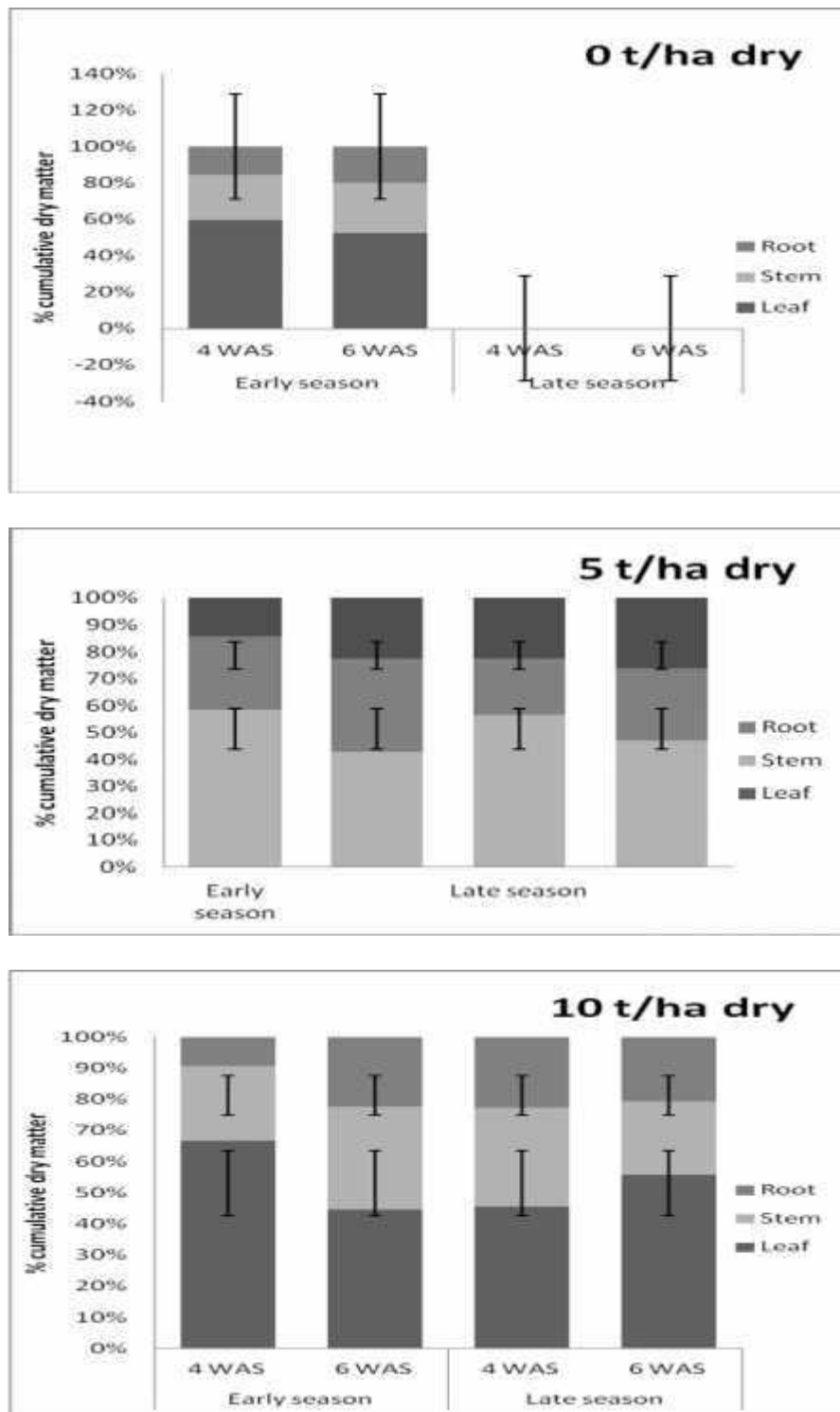


Fig. 1 Cummulative dry matter proportions of *Celosia argentea* with poultry manure rate

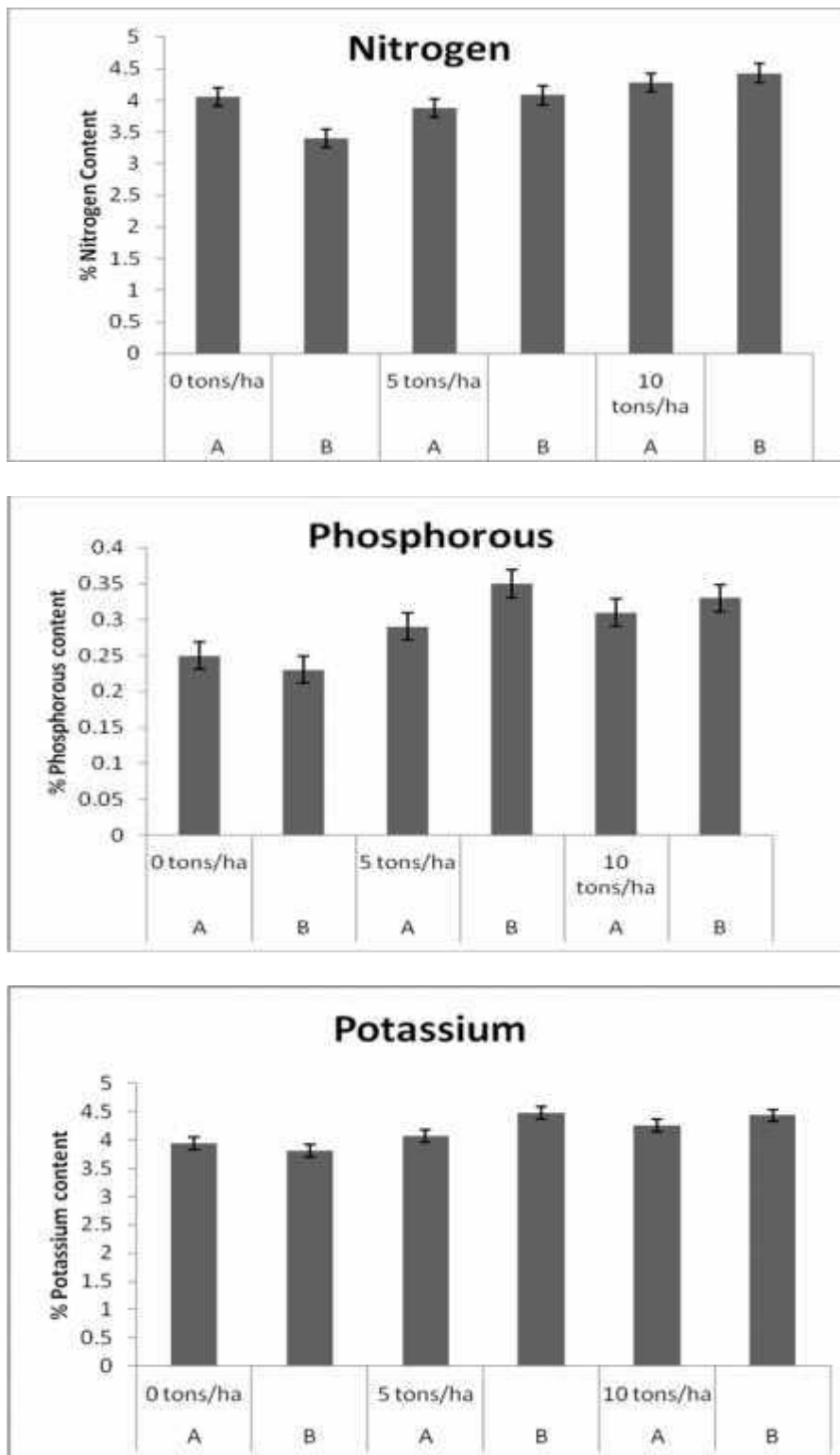
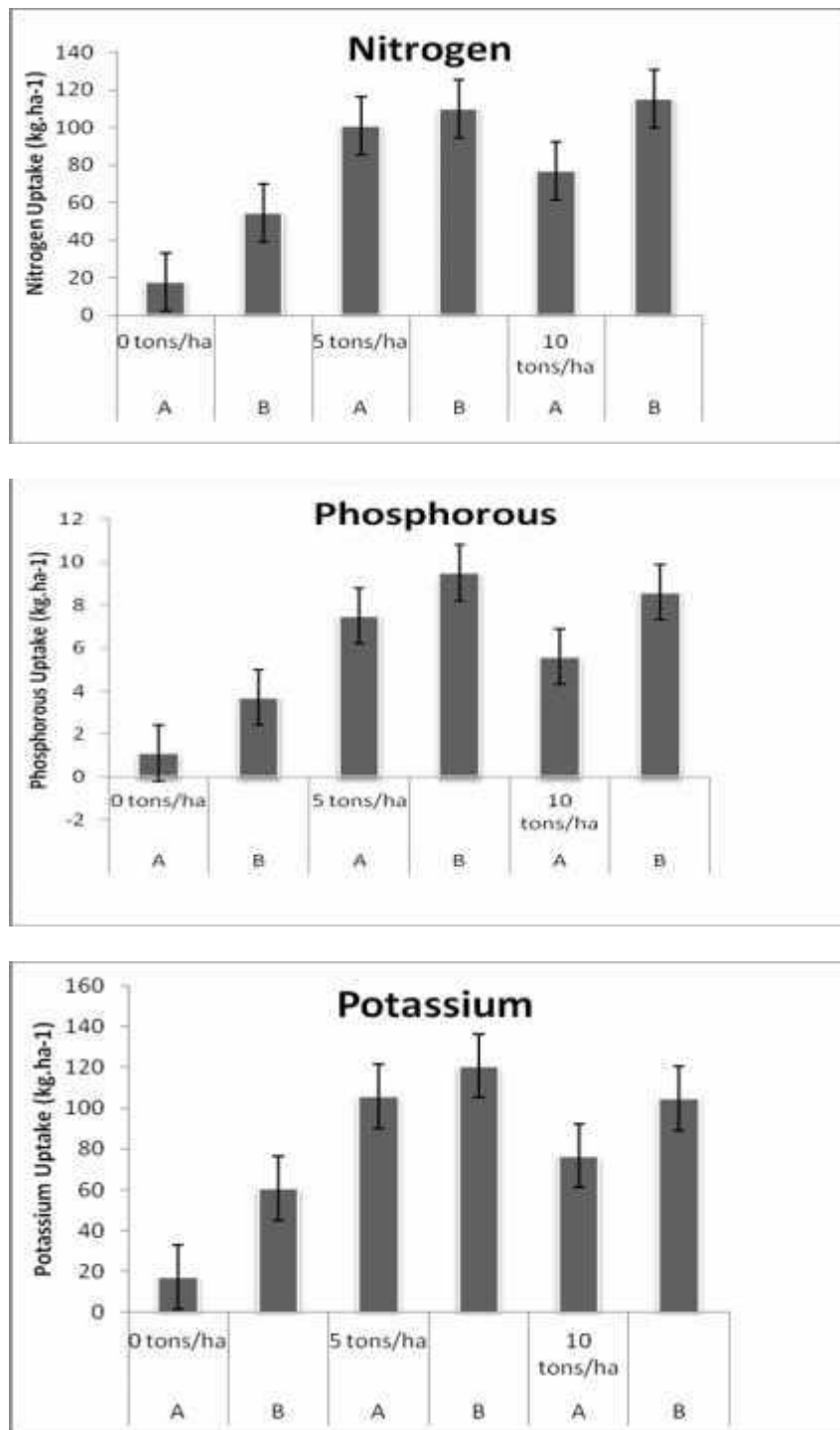


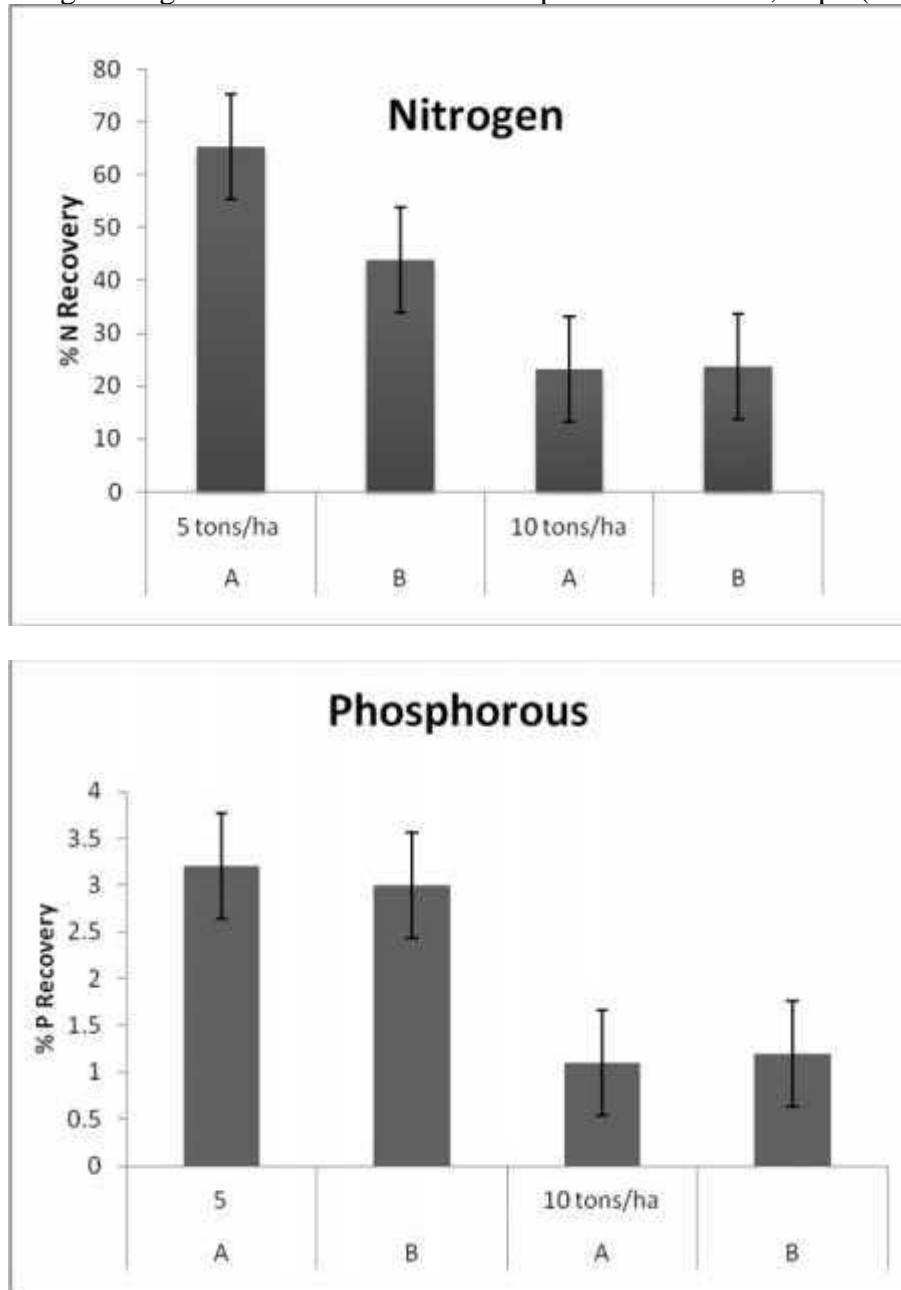
Fig. 2: Nitrogen, Phosphorous and Potassium contents (%) of *Celosia argentea* with interaction of poultry manure rate and plant population.
 A – 200,000 plants/ha; B – 400,000 plants/ha



A – 200,000 plants/ha; B – 400,000 plants/ha

Fig. 3: Nitrogen, Phosphorous and Potassium uptake (Kg/ha) of *Celosia argentea* with interaction of poultry manure rate and plant population.

A – 200,000 plants/ha; B – 400,000 plants/ha



A – 200,000 plants/ha; B – 400,000 plants/ha

Fig.4: Average Nutrient recovery (%) of *Celosia argentea* with interaction of poultry manure rate and plant population

Nutrient concentration of *Celosia argentea* as affected by poultry manure rate and plant population: The nitrogen, phosphorous and the potassium contents of the *Celosia* plants were affected by the interaction of poultry manure and plant population (Fig.2). The lower population of 200,000 plants/ha contained a higher nitrogen concentration of 4.05 % when 400,000 plants/ha population had 3.4 %. With 5 t/ha PM application, plant nitrogen concentration was higher with 400,000 plants/ha, when they were similar with 10 t/ha application. A similar trend as nitrogen was observed for P and K concentrations. The lower population of 200,000 plants/ha had higher P and K concentration. With 5 and 10 t/ha PM, the population of 400,000 plants/ha had higher values than 200,000 plants/ha (Fig.2).

Nutrient uptake of Celosia argentea as affected by poultry manure rate and plant population:

Nutrient uptake by *Celosia argentea* was significantly influenced by combined application of poultry manure and plant population (Fig.3). Planting at 200,000 plants/ha had a lower uptake relative to uptake with 400,000 plants/ha. With 200,000 plants/ha, plant nitrogen uptake was lower with 0 t/ha than with 5 t/ha. With 400,000 plants/ha, nitrogen uptake increased with increase in poultry manure application. The P and K uptake showed a similar trend as N uptake (Fig.3). The highest P and K uptake from application of 10 t/ha PM with 400,000 plants/ha was followed by application from 10 t/ha with 200,000 plants/ha population. The lowest uptake was from the unfertilized plants at 200,000 plants/ha (Fig.3).

Apparent Nutrient Recovery: Apparent Nitrogen Recovery (ANR) was generally higher with 5 t/ha than with 10 t/ha. The highest was from 5 t/ha with 200,000 plants/ha. It was reduced with increased population to 400,000 plants/ha (Fig 4). The ANR was 24 % at 10 t/ha with 400,000 plants/ha but was 23 % with a population of 200,000 plants/ha. The Apparent Phosphorous Recovery (APR) was highest at 5 t/ha with 200,000 plants/ha followed by 400,000 plants/ha of 5 t/ha. The 10 t/ha application had an APR of 1.10 and 1.20 %, at 200,000 and 400,000 plants/ha, respectively (Fig 4).

DISCUSSION

Cock's comb, *Celosia argentea* is a crop of the warm, moist environment and was supported by the rainfall, the temperature and the relative humidity of the study area over the two years. The pre cropping nutrient status of the experimental site was a reflection of previous cropping of the land, with no records of optimum fertilizer application other than crop residues that were recycled. This accounts for the soil's low contents of organic carbon, total N, available P and exchangeable K. The total N content was below the critical level of 1.5 g/kg (Aduayi et al. 2002). Available phosphorus (Bray 1) was very low based on the 8-12 mg/kg critical level reported by Udo *et al.* (2009) or the 10 – 16 mg/kg critical level reported by Adeoye and Agboola (1985). The exchangeable K content was also lower than the critical 0.6-0.8 cmol/kg content recommended in the region by Adeoye and Agboola (1985).

Cock's comb growth and biomass accumulation was generally more favoured in the early season due to the higher total rainfall, relative to the late season. The weather data showed that rainfall was low in the first 4 weeks of growth in the late season and was inadequate to establish the crops, unlike in the early season, when the rainfall was much higher and plant establishment was achieved early. Manure application resulted in a better growth due to the supply of nutrients required for optimum Cock's comb growth as earlier reported (Senjobi *et al.*, 2010). Application of 5 t/ha poultry manure, to supply 127 kg N/ha, on a tropical soil, with a native content of 0.08 % was optimum for cultivation of celosia. Higher rate of application has shown a reduced performance, with plant height, number of leaves per plant and leaf area, indicating an antagonistic effect of the high rates of nutrients supplied. The leaf production of *Amaranthus* has been reported enhanced by manure application (Adewole and Dedeke, 2012; Seeiso and Materechera, 2014). On a tropical soil, with a native content of 0.08 % N, establishing Cock's comb up to a population of 400,000 plants/ha is still supported. The season of cultivation, as well as the plant age at harvest affected the growth and biomass accumulation of *Celosia argentea*. The better growth of the early season generally translated to higher biomass yields. Plants generally had about 8% dry matter in the early season, indicating the need for a high fresh intake to meet a nutritional target. A reduced fresh weight intake to meet a nutritional target can only be met by producing Cock's comb in the late season and harvesting late when the fresh weight is lower and the plant dry weight is higher. With delayed harvesting till 6 weeks after sowing, plant biomass was increased. A higher biomass yields of Cock's comb with more frequent cuttings have been reported (Seeiso and Materechera, 2014) and also with age of transplants in *Solanum melongena* (Lawal *et al.*, 2015).

Fertilizer application increased the plant N, P and K contents due to supply of nutrients for plant uptake with the applied fertilizers. The rate of application however, did not affect the plant N, P and K contents but rather plant population, indicating the contribution of the plant population, as a result of the increased number of plants considered to determine the uptake. Individual plants had higher concentrations of N, P and K with the lower population of 200,000 plants/ha due to the lower total nutrient demand. The concentrations were lower with 400,000 plants/ha due to limitation in available nutrients but the high population amounted to a higher total nutrient uptake. Increased uptake of N, P and K with increased fertilizer rate was an indication of release of more nutrients with higher rates of fertilizer. The 5 t/ha poultry manure however, supplied enough nutrients for optimal N, P and K uptake and nutrient recovery. This agrees with an earlier observation with poultry manure, compared with Cowdung application (Makinde and Ayoola, 2012).

CONCLUSION

Application of 5 t/ha of poultry manure to *Celosia argentea* resulted in optimum growth and nutrient uptake in both early and late seasons. A population of 400,000 plants/ha can be adopted for cultivating Cock's comb, *Celosia argentea* on a tropical soil with a native 0.08 % Nitrogen content.

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Analysis of Selected Heavy Metals in Fresh Cow milk, drinking water, grass and soil of dairy farms in Saki East local Government area of Oyo State, Nigeria

Sansi, J.A.A.¹; Sansi, O.O.² and Ojezele, O.J.¹

¹ Federal College of Animal Health and Production Technology, Moor Plantation, Ibadan. Oyo State.

² Centre for Sustainable Industrial and Integrated Development, Ibadan, Oyo State.

ABSTRACT

Urbanization and industrialization have increased in recent times and these have caused increase in the inorganic contaminants present in the air, food items, water and soil. Soundness of agricultural processes is a matter of concern and safety of the food produced for both animal and human consumption. Twenty-nine fresh milk samples, composite soil, grass and drinking water samples were collected from three dairy farms in Saki East Local Government area of Oyo state, Nigeria. These were analyzed for the concentration of selected heavy metals (copper, cadmium and lead) using Atomic Absorption Spectrometry. The mean concentrations of copper in milk, drinking water, soil and grass were 1.33 mg/L, 0.03 mg/L, 0.11 mg/Kg and 0.16 mg/Kg respectively. The mean concentrations of cadmium in milk, drinking water, soil and grass were 0.12mg/L, 0.13mg/L, 0.11mg/Kg, and 0.10mg/Kg respectively. The mean concentrations of lead in milk, drinking water, soil and grass were 0.77 mg/L, 0.85 mg/L, 0.5 mg/Kg and 0.20 mg/Kg respectively. Mean concentrations of cadmium in water, lead in milk and lead in water were above the permissible standard limits and therefore were not fit for human or animal consumption. Advocacy for organic dairy is highly recommended as one of the standards required for certification is proper analysis of the soil and drinking water sources for heavy metal contaminants. The result of such analyses would guide the prospective farmers in the choice of farm sites and paddocks. Routine analysis for heavy metal concentration in fresh milk, drinking water, grass/feed and soil of dairy farms is strongly recommended.

Key words: fresh cow milk, soil, drinking water, grass, organic

Corresponding author: rsansi17@gmail.com

INTRODUCTION

Increased urbanization and industrialization have caused an upsurge in the input of inorganic contaminants in air, water and soil. Drinking contaminated water or eating food grown on contaminated soil may pose serious health risks to both humans and animals (Bushra *et al.*, 2014). Consumers are therefore taking into account qualitative aspects that include health and safety as well as societal benefits which include environmental impact and animal welfare ethics (Kaditi and Swinnen, 2007).

Heavy metals are defined as metallic elements that have a relatively high density compared to water (Fergusson,1990) and could cause both environmental and health hazards when they persist in the environment (Tassew Belete *et al.*, 2014). Lead as a soil contaminant is a widespread issue; It accumulates with age in bones aorta, and kidney, liver and spleen. It can enter the human body through uptake of food (65%), water (20%) and air (15%) (Nazir *et al.*,2015). Natural and human related activities are responsible for the release of heavy metals into the environment (Abdul Khaliq *et al.*,2012). Milk and its products are main constituents of the daily food consumed by infants, school age children and the aged (Enb. *et al.*,2009). It is an excellent source of calcium, magnesium, zinc, vitamin D, vitamin A and vitamin B-12. Atomic Absorption Spectrometry (AAS) is an analytical technique that measures the concentration of elements. It is sensitive and detects the wavelength of light specially absorbed by an element. In the practice of good agricultural system, especially organic agriculture, certain standards are required.

Scientific assessment of soil quality is essential to monitor the sustainability of agricultural systems (Franzluebbers and Haney,2006). Soil quality is the capability of soil to produce safe and nutritious crops in a sustained manner over time and to enhance human and animal health without impairing the natural resource base or harming the environment (Parr *et al.*,1992). According to the IFOAM (2005), accumulation of heavy metals should be reduced and the appropriate remedial measures should be put in place where necessary. It is required that the analysis of the relevant products and possible sources of pollution (soil, water, air, inputs) is undertaken to determine the level of contamination. Appropriate responses, such as detection of contamination sources, consideration of background contamination and other relevant factors are required. This study was done to determine the presence of selected toxic heavy metals namely, cadmium, lead and copper in fresh cow milk, grass, soil and drinking water of cattle in three farm settlements of Saki East Local Government area of Oyo state.

MATERIALS AND METHODS

Sample collection

The samples were collected from three different farms in Ogboro, one of the five major communities in Saki East Local Government area of Oyo State, Nigeria. Saki East is located on latitude 8.704675 and longitude 3.589439. The area was selected based on the fact that one of the main occupation of the inhabitants is cattle rearing, production and sale of fresh milk and local cheese. The area also has a history of blacksmiths, goldsmiths and clay pot molder. A total of twenty- nine cows of different indigenous breeds were used for the collection of milk sample from three farms A, B and C at which 10, 11 and 8 samples were collected respectively. The samples were collected by convenience method as some farmers refused to cooperate and others

did not have milking cows at the time the sample collection was done. They were either dry or too young.

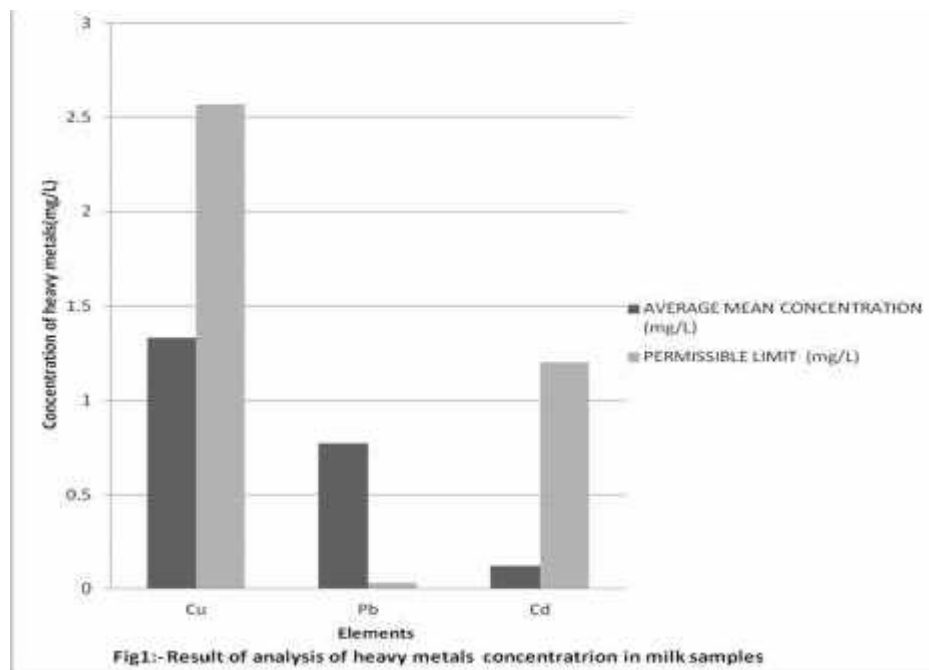
Each sample of milk was collected into polyethylene bottles of 100ml which had been previously soaked in 20% HNO₃ for 24 hours and rinsed with de-ionized water. These were sealed, labeled and stored in the refrigerator at about 5°C or less. Composite grass and soil samples were taken from the cattle grazing area. Water samples were also taken from the river and wells which were the drinking water sources.

Sample analysis

The milk samples were prepared by wet digestion method and analyzed by Atomic Absorption Spectrometry as described by AOAC (2000), Qin *et al.*(2009) and Bushra Iftikhar *et al.*, (2014). Exactly 2 grammes of each grass sample was washed three times with distilled water to remove adhering particles. This was oven dried at 100°C to remove the moisture. It was ground with mortar and pestle. The dried sample was placed in a muffle furnace at 550°C for 4hrs till the sample was converted to ash. It was put in a desiccator to cool and to prevent moisture after which 5ml of already prepared 10% Nitric acid and perchloric acid with a tablet of selenium catalyst added to it. The tube was placed inside a digestion block, and slowly digested and filtered. Mixture was filtered, poured into 50ml volumetric flask and topped up with distilled water. It was taken to the laboratory for heavy metal analysis using atomic absorption spectrometry. The soil sample was digested using 2grammes of the sample as was done with the grass. It was then taken to the laboratory for atomic absorption spectrometry. The water sample was put in a beaker, 5ml of nitric and perchloric acid was added and put on a heating mantle to boil for an hour until it gave clear solution. It was poured into a 50ml volumetric flask topped up. With distilled water and taken to the laboratory for heavy metal analysis using the AAS.

RESULTS

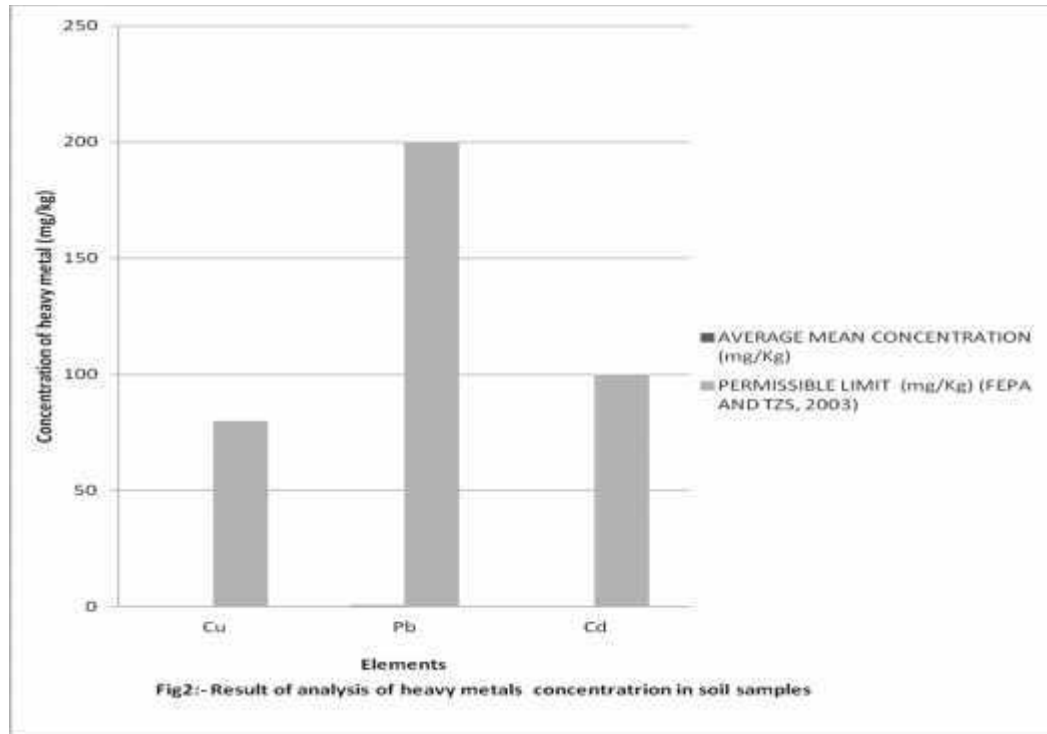
The average mean concentration of each element, copper (Cu), lead (Pb) and cadmium (Cd) was determined and compared with the permissible standard in literature for milk, soil, grass and water.



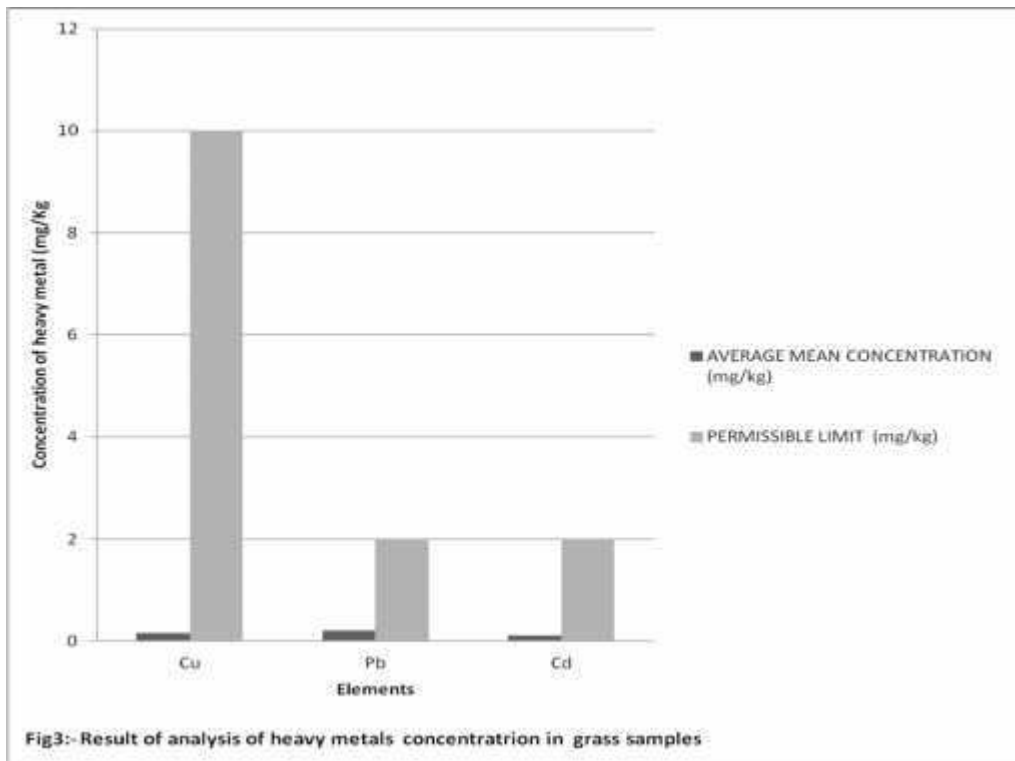
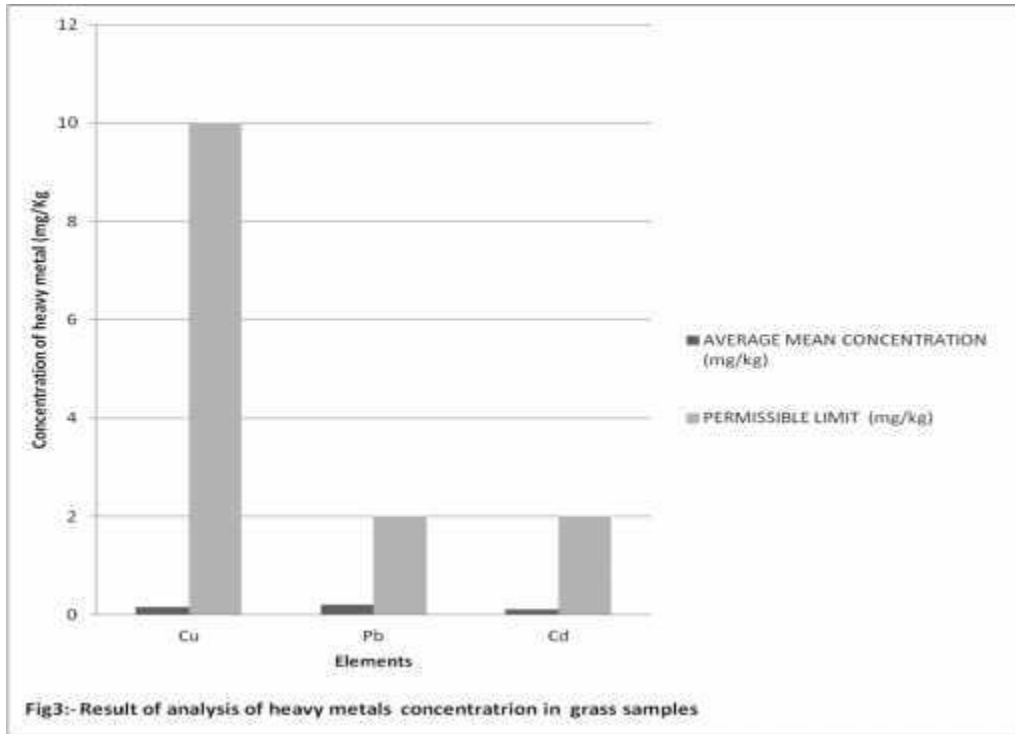
Copper (Cu): The permissible limit of copper for cow milk is 2.57mg/L as recommended by WHO (2003). The average concentration of Cu from all collected milk samples from the three farms (Farm A, Farm B and Farm C) in Saki east was 1.33mg/L (Fig. 1) and was below the permissible limit.

Cadmium (Cd): The permissible limit of Cadmium in cow milk, as recommended by WHO (2003), is 1.20mg/L. The result of the average concentration of Cadmium (Cd) from the collected fresh cow milk samples from the three farms all at Saki east was 0.12mg/L which was below the permissible limit (Fig. 1)

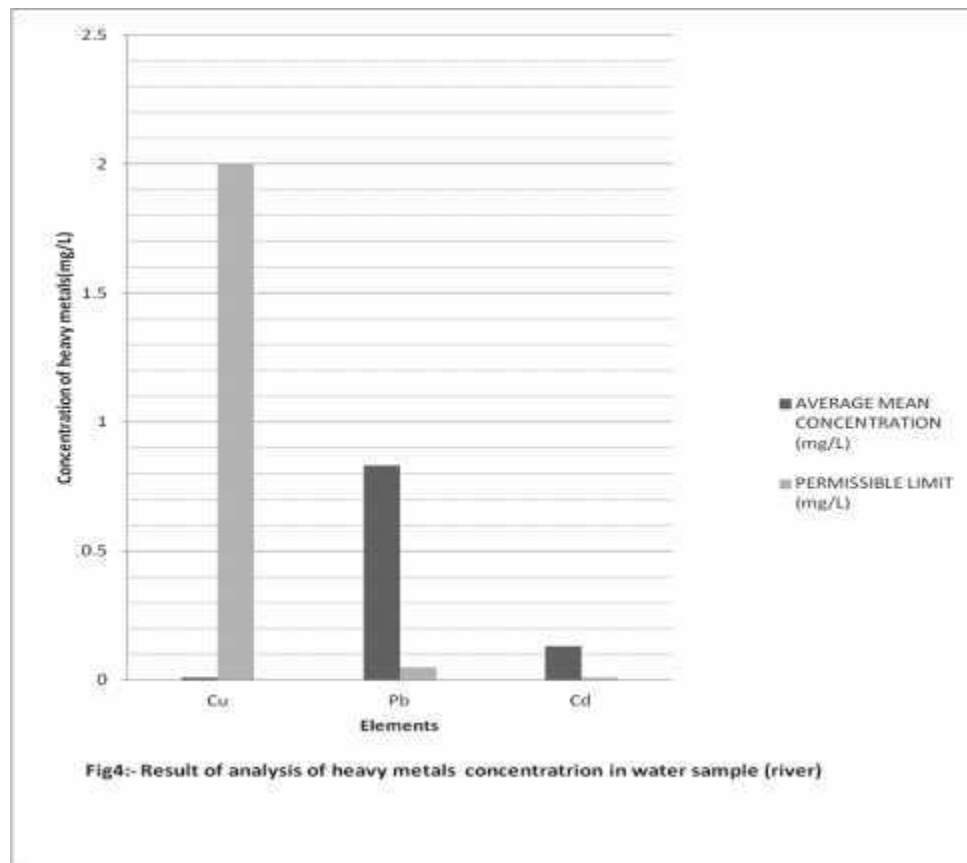
Lead (Pb): The permissible limit in cow milk sample recommended was 0.03mg/L. (WHO, 2003). All the collected milk samples from the three farms (Farm A, Farm B and Farm C) in Saki east had average concentration value of 0.77mg/L. The value was above the permissible limit (Fig.1).

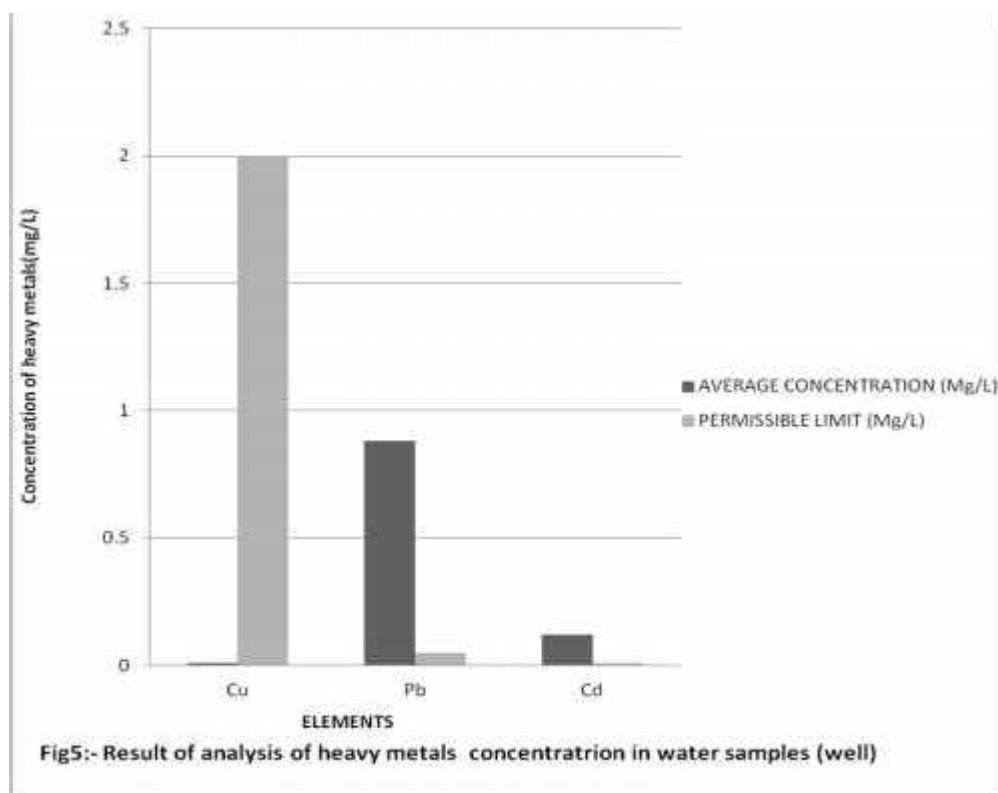


The average Concentration of copper in all the soil samples From Farm A, Farm B and Farm C was 0.11mg/Kg (Fig. 2) which was below EPA (2002)/TZS (2003) guidelines for heavy metals in soil (70-80mg/kg). The average concentrations of cadmium in the soil samples collected from Farm A, Farm B and Farm C was 0.11mg/kg which did not exceed the (TZS, 2003) maximum limit of 200mg/kg for soil. Concentration of lead in all the soil samples: Average concentration of lead in soil samples from Farm A, Farm B and Farm C was recorded to be 0.5mg/kg (Fig. 2) which was below EPA (2002) guidelines for heavy metals in soil (1.6mg/kg).



The permissible limit of copper for plants is 10mg/kg recommended by WHO (1996), Hassan *et al.*,(2012). In all the collected grass samples from Farm A, Farm B, and Farm C the average concentration of the copper was 0.16mg/kg (Fig. 3) the concentration of copper was recorded below the permissible limit. The permissible limit of Cadmium in plants, recommended by WHO (1996) is 2.00mg/kg. In the grass samples from farm A, Farm B and farm C the Average concentration of cadmium was recorded to be 0.10mg/Kg limit which was far below the permissible limit (Fig. 3). The permissible limit in plants as recommended by WHO (1996) was 2.0mg/kg. The average concentration of Lead (Pb) in the grass samples collected from Farm A, Farm B and Farm C was 0.20mg/kg (Fig. 3). Therefore, the concentration of lead was recorded below the permissible limit of WHO (1996).





The maximum permissible limit for Copper in water was 2mg/L as recommended by WHO (1996), in water samples from Farm A, Farm B and Farm C the average concentration of copper in water was 0.03 mg/L (Fig. 5). Concentration of copper was recorded below the permissible limit. The maximum permissible limit for cadmium in water was 0.05 mg/L (Hassan *et al.*, 2012). The average concentration of cadmium in water samples from Farm A, Farm B and Farm C was 0.13mg/L (Fig. 4) which was recorded above the permissible limit of 0.05mg/L (Hassan *et al.*, 2012). Standards permissible limit of lead in water was 0.05mg/L and the average concentration in water samples collected from Farm A, Farm B and Farm C was 0.85mg/L (Fig. 6) the concentration of lead was above the permissible limit.

DISCUSSION

High values of lead and cadmium in this result agrees with Malhat *et al.*(2012) that global population was exposed to lead from air and from major sources including paint, pesticides, smoking, automobile emission, batteries, mining, burning of coal, chemical industries e.t.c. The result of this work shows that rural areas like Ogboro community are not exempted from this challenge. This is a great contrast to the report of Tassew Belete *et al.* (2014) carried out in Ethiopia where lead and cadmium were reported to be absent. The concentration of lead in the drinking water was high and this might be one of the causes of high lead concentration in milk. The selected metal (Pb) concentration in milk was found to be higher than the permissible level even though the concentration in grass they fed on was acceptable. A similar report was given by Bushra Iftikhar *et al.* (2014) in Pakistan. This is probably because of bio-accumulation. Miranda *et al.*, in 2005 similarly stated that the food chain is an important source of Cd and Pb

accumulation, especially for plants grown on polluted soils, also that significant amounts of Cd and Pb can be transferred from contaminated soil to plants and grass, causing accumulation over time, of these potentially toxic metals in grazing ruminants, particularly in cattle. Accumulation of Cd and Pb in ruminants causes toxic effects in cattle, but also in humans consuming meat and milk contaminated with toxic metals (Vromman *et al.*, 2008; Cai *et al.*, 2009). Metal residues in milk are of particular concern because milk is largely consumed by infants and children (Tripathi *et al.*, 1999). Cd and Pb are amongst the elements that have caused the most concern in terms of adverse effects on human health. The results of this work agree with Jen, *et al.* (1994) that concentration of Cd and Pb in cow milk should be monitored to ensure the consumers' health .

CONCLUSION

The concentrations of lead (Pb) contaminant in milk and water samples and cadmium in water samples were above the permissible limits. It could be concluded that the milk samples from the study area were not fit and wholesome for human consumption, as they were not free from hazardous heavy metal contaminants. Further investigation should be carried out on the causes of high lead (Pb) in milk samples and high lead (Pb) and cadmium (Cd) in water samples in (Ogboro community) Saki East local Government of Oyo State. The history of metal workers such as blacksmiths and goldsmiths, some of who may still be present could have contaminated the land and water over the years.

In order to minimize the hazardous effects of these pollutants and protect human health, it is hereby recommended that strict and routine monitoring of milk supply should be undertaken by regulatory bodies. Farmers should be warned not to graze their animals in the areas where there is possibility of heavy metal contamination e.g. where mining, metal smelting, battery charging or similar activities are done. Further research should be carried out on samples from areas where there are activities like smelting, welding, painting etc. Rural areas should not be assumed to be 'organic by default' but all necessary analyses should be done, especially soil and water analysis to ensure that they are suitable for organic and even conventional farming.

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Comparative effects of effective microorganism activated solution, leaf extracts and carbofuran on the growth and yield of cucumber planted on *Meloidogyne incognita* infested soil.

S. A. Abolusoro¹, N.B. Izuogu³, O. O. Ogunbeku¹, J. Ezenyeogbara¹, G.O. Agbaje¹,
P.F. Abolusoro² and S. A. Hinmikaiye⁴.

¹ Landmark University, Omu-Aran, College of Agricultural Sciences,

² Kogi State College of Education Technical, Kabba, Kogi State, Nigeria

³ Faculty of Agriculture, Dept. of Crop Protection, University of Ilorin Nigeria

⁴ College of Agriculture Ahmadu Bello University Kabba Kogi State Nigeria

ABSTRACT

Two field experiments were conducted between 2014 and 2016 on the effects of effective microorganism activated solution (EMAS), carbofuran and some plant leaf extracts (*Azadirachta indica* and *Tithonia diversifolia*) on the performance of cucumber planted on root-knot nematode (RKN) infested soil. The trials were carried out on two separate fields in 2014/2015 and 2015/2016, each measuring about 15 m x 10 m. The experimental designs were randomized complete block, comprising of five treatments (EMAS, *Azadirachta indica*, *Tithonia diversifolia* Carbofuran and control). Each treatment was randomly assigned to a plot in a block and replicated three times. After land preparation, Cucumber seed *cv.* Monalisa F1 which is nematode susceptible was planted and inoculated with 2000 J2 *M. incognita* two weeks after planting while treatments were applied one week after inoculation. The result from the experiment showed that all the cucumber treated with EMAS, plant extract and carbofuran resulted into significantly ($P \leq 0.05$) increased vegetative growth and fruit yield compared with the control. Nematode soil population and root gall index were reduced in plant extract and carbofuran treated soil while it remained high in EMAS and not significantly ($P \leq 0.05$) different from the control. Phytochemical screening of the plant extracts revealed the presence of alkaloid, tannin, flavonoid and saponin. EMAS did not bring about reduction in soil nematode population as it was not significantly ($P \leq 0.05$) different from the control but recorded the highest yield showing that it strengthened the tolerance level of cucumber to nematode attack.

Key words: *M. incognita*; nematode; cucumber; *Azadirachta indica*; *Tithonia diversifolia*; EMAS; carbofuran

Corresponding author: stevabolusoro2005@yahoo.co.uk

INTRODUCTION

Cucumber (*Cucumis sativus* L.) is a member of Cucurbitaceae family. It is an important vegetable and one of the most popular member of the Cucurbitaceae family (Lower and Edwards 1986; Thoa, 1998). Cucumber have not received as much attention as other vegetables in terms of health benefits, but this widely cultivated food provides us with a unique combination of nutrients. At the top of the phytonutrients list for cucumbers are its Cucurbitacins, Lignans and Flavonoids. These three types of phytonutrients found in cucumber provides us with valuable antioxidant, anti-inflammatory and anticancer benefits. cucumbers are mostly eaten raw. Sliced cucumber with the peel contains only 8 calories per serving, with only 0.6 gram total fats and less than 1 gram sugar. Cucumber is a nutrition choice, considering that it contains almost half a gram of dietary fiber which is essential for slowing down body's digestion of food giving body enough time to absorb essential nutrients (Dannie, 2013). Cucumber is grown for its immature fruits that are used as salad vegetable and for pickles.

Cucumber, like other vegetable crops, is attacked by wide range of pests and diseases, resulting in great yield losses. Prominent among them is root-knot nematode, *Meloidogyne incognita*, which brings about a great yield reduction (Aminu-Taiwo and Fawole, 2012; Rivera and Abballay, 2008). Management of root-knot nematode with synthetic nematicides can be very effective (Sikora and Feanadeq, 2005; Adegbite and Agbaje, 2007; Dubery and Triberdi, 2010). However, scarcity, high cost, environment safety and global restriction on importation of synthetic nematicides have spurred scientists to search for alternative control measure against nematode pests of economic food crops (Anonymous, 2004). The use of plant extract has been suggested (Hoan and David, 1979).

In the present study, extracts from the leaf of *T. diversifolia* and *A. indica* as well as carbofuran were compared with EMAS (Effective microorganism activated solution). EMAS comprises of a wide variety or multi-culture of effective, beneficial and non-pathogenic microorganisms coexisting together. Essentially, it is a combination of aerobic and anaerobic species commonly found in all ecosystems. EMAS is not a pesticide and contains no inorganic chemical. It is a microbial inoculant that works as bio-control measure in suppressing and or controlling pest through the introduction of beneficial microorganism to soils and plant. Pests and pathogens are suppressed or controlled through natural processes by enhancing the competitive and antagonistic activities of the microorganisms in EMAS inoculants (Higa, 1998).

The research on the use of EMAS for nematode control in nematode infested crop is yet to be exploited, if at all is available. It is against this background that this research work is conducted with the following objectives:

- To compare the performance of root-knot nematode infested cucumber treated with EMAS, *T. diversifolia* (wild sunflower) *A. indica* (neem) extracts and carbofuran
- To compare the soil nematode population and level of root damage on cucumber due to treatment with EMAS, *T. diversifolia*, *A. indica* extracts and Carbofuran
- To access if treatments have effect on the nutrient value of cucumber
- To investigate the bio-active chemical components present in the leaf extracts of *T. diversifolia* and *A. indica*.

MATERIALS AND METHODS

Sources of Seeds and Control Agents

Seed of cucumber (*Cucumis sativus*) Manolisa F1 which is susceptible to *M. incognita* infection were obtained from Shongai Centre Benin Republic, Cotonou. The leaves of *T. diversifolia* (Mexcan sunflower) and *A. indica* (neem) were obtained from Omu-Aran town, Kwara State. Furadan (3G), a brand of Carbofuran, was purchased from agrochemical store in Ilorin, kwara State, Nigeria.

Experimental Site

Two field trials were carried out at Landmark University, Teaching and Research farm, Omu-Aran, Kwara State, Nigeria. Two separate plots, 15 m x 10 m each, were prepared for the experiment between the months of September, 2014 to January, 2015 and at the same period in 2015/ 2016.

Experimental Design and Treatment Preparation

The experimental designs were randomized complete block, comprising of five treatments (*Tithonia*, *Azadirachta*, EMAS, Carbofuran and Control) and each treatment was replicated three times. Plant extracts were from *T. diversification* and *A. indica* leaf. The leaves were collected from each plant, air-dried for 3 – 4 weeks on laboratory benches and then ground into powder using blender, and soaked in an aspirator separately over a period of 24 hours. This was done by pouring 1 kg each of the powder plant material into one litre of distilled water separately and it remains in the solution for 24 hours. The solution obtained thereafter was 100% concentrated or 1,000,000 ppm (Stock solution) which was diluted to 500,000 ppm (50%). EMAS was applied at 20 mls into knapsack sprayer load (15 litres) and Carbofuran was applied at the rate of 3kg ai/ha (Recommended rates).

Crop Establishment

The seeds of cucumber *cv* Nomalisa F1 which had been confirmed to be susceptible to nematode attack were planted into each of the experimental plots (Aminu-Taiwo and Fawole, 2012). Two seeds were planted per hole, at a depth of 5-6 cm and 60 cm spacing between and within rows. Two weeks after planting, seedlings were thinned to one healthy plant per stand.

Extraction and Inoculation of Cucumber

Plant parasitic nematodes were extracted from previously infested tomato grown in pure culture of *M. incognita* in previous experiment using standard method (Whitehead and Hemmings, 1965). *M. incognita*, (2000 J2) were introduced into the root of each cucumber. Meanwhile the initial soil nematode population was recorded.

Treatment Application

A week after inoculation, *Tithonia*, *Azadirachta* and EMAS, carbofuran were applied. *Tithonia* and *Azadirachta* extracts were applied at the rate of 500,000 ppm (50%), this is by pouring 100ml of the solution within the plant rhizosphere. EMAS was applied at the rate of 1,000 ppm (0.1%) by direct spray on the plant biomass (recommended application rate and method) but it was applied four times over a period of four weeks at weekly interval while carbofuran was applied at the rate of 3kg ai/ha. Untreated plot served as the control.

Assessment of Crop Growth and Yield

Four weeks after planting, data on growth parameters (numbers of leaves, average vine length, stem girth) and yield components (number of fruits and fruit weight) were recorded.

Assessment of fruit for mineral nutrient composition

Fruits were harvested at maturity, and samples were analysed for total mineral matter. Analysis was carried out according to the method described by Kacar (1995).

Phytochemical screening of test plants (*Azadirachta indica* and *Tithonia diversifolia*)

Test for alkaloids

To the extracts, dilute hydrochloric acid was added, shaken well and filtered. With the filtrate, the following test was performed:

Mayer's reagent test

To 3ml of filtrate, few drop of Mayer's reagent were added along sides of tube. Formation of creamy precipitate indicates the presence of alkaloids.

Test of flavonoids

Alkaline reagent test

The extract was treated with few drop of sodium hydroxide solution separately in a test tube. Formation of intense yellow colour, which becomes colourless on addition of few, drops of dilute acid indicates the presence of flavonoids.

Test for Saponin

Froth test

The extract was diluted with diluted water and shaken in a graduated cylinder for 15 minutes. The formation of layer of foam indicates the presence of saponins.

DATA COLLECTION AND ANALYSIS

In both experiments, data were collected on the growth and yield components of cucumber. Data was also collected on the soil nematode population and gall index. The data were pooled together, means were subjected to analysis of variance (ANOVA) and means separated using Duncan's multiple range text at 5% probability level.

RESULTS

Effect of leaf extracts of wild sunflower leaf, neem leaf, EMAS and carbofuran on the growth and yield of cucumber.

The effects of different leaf extracts (wild sunflower and neem), EMAS and Carbofuran on the leaf number of Cucumber was presented in Table 1. Cucumber treated with leaf extract (wild sunflower and neem), EMAS and Carbofuran produces more leaves than the control. The number of leaves produced by the plants that were treated with leaf extract, EMAS and carbofuran recoded significantly greater number of leaves from week 6 of the experiment. EMAS and carbofuran treatments were significantly better than the leaf extract treatment at week 8.

Table 1: Effect of EMAS, leaf extracts and carbofuran on the mean number of cucumber infected with *M. incognita*

Treatments	4WAP	5WAP	6WAP	7WAP	8WAP
Neem	7.67 ^a	13.43 ^{ab}	17.67 ^{ab}	24.43 ^b	26.10 ^b
Mexican sunflower	9.66 ^a	14.17 ^{bb}	17.43 ^{ab}	24.60 ^b	26.19 ^b
EMAS	9.455 ^a	16.20 ^a	27.43 ^{ab}	29.30 ^a	35.90 ^a
Carbofuran	9.47 ^a	15.53 ^a	27.70 ^a	28.43 ^a	33.53 ^a
Control	8.43 ^a	13.43 ^{ab}	15.10	18.43 ^c	19.90 ^c

Means within the same column followed by the same letters(s) are not significantly different ($P \leq 0.05$)

Table 2: Effects of EMAS, leaf extracts of plant and carbofuran on the vine length of cucumber infected with *M. incognita*

Treatments	4WAP	5WAP	6WAP	7WAP	8WAP
Neem	23.50 ^a	46.00 ^{ab}	69.77 ^{ab}	87.13 ^{ab}	88.00 ^b
Mexican sunflower	32.20 ^a	50.67 ^{ab}	67.10 ^{ab}	89.90 ^b	89.97 ^b
EMAS	35.70 ^a	58.63 ^{ab}	80.23 ^a	111.60 ^a	127.77 ^a
Carbofuran	38.20 ^a	61.10 ^{ab}	80.03 ^a	113.67 ^a	123.33 ^a
Control	27.20 ^a	34.00 ^a	64.30 ^b	80.23 ^c	82.23 ^b

Means within the same column followed by the same letters(s) are not significantly different ($P \leq 0.05$)

Table 3: Effects of EMAS, leaf extracts of plant and Carbofuran on the stem girth of cucumber infected with *M. incognita*

Treatments	4WAP	5WAP	6WAP	7WAP	8WAP
Neem	0.47 ^a	0.67 ^a	0.70 ^a	0.80 ^a	0.80 ^a
Mexican sunflower	0.50 ^a	0.70 ^a	0.70 ^a	0.76 ^a	0.76 ^a
EMAS	0.53 ^a	0.73 ^a	0.76 ^a	0.80 ^a	0.83 ^a
Carbofuran	0.47 ^a	0.68 ^a	0.75 ^a	0.80 ^a	0.82 ^a
Control	0.40 ^a	0.60 ^a	0.66 ^a	0.76 ^a	0.76 ^a

Means within the same column followed by the same letters(s) are not significantly different ($P \leq 0.05$)

Table 4: Effects of EMAS, leaf extracts of plant and Carbofuran on the flowering of cucumber infected with *M. incognita*

Treatments	Average number of days to flowering		
	Flower initiation	50% flowering	100% flowering
Neem	20.66 ^a	24.66 ^a	29.60 ^a
Mexican sunflower	20.67 ^a	24.50 ^a	29.32 ^a
EMAS	19.95 ^a	24.50 ^a	28.30 ^a
Carbofuran	19.60 ^a	24.33 ^a	28.60 ^a
Control	20.60 ^a	24.56 ^a	29.68 ^a

Means within the same column followed by the same letters(s) are not significantly different ($P \leq 0.05$)

Table 5: Effects of EMAS, leaf extracts of plant and carbofuran on the yield of cucumber infected with *M. incognita*

Treatments	Yield Component		
	Average fruit no / plot	Average fruit weight (kg)/plot	Yield (kg/.ha)
Neem	19.30 ^b	5.17 ^b	7274.60 ^{bc}
Mexican sunflower	20.00 ^b	4.99 ^b	6991.20 ^{bc}
EMAS	34.30 ^a	7.00 ^a	9362.00 ^a
Carbofuran	33.21 ^a	6.90 ^a	8641.20 ^a
Control	16.90 ^c	3.20 ^c	5291.40 ^b

Means within the same column followed by the same letters(s) are not significantly different ($P \leq 0.05$)

Table 6: Effects of EMAS, leaf extracts of plant and carbofuran on the nematode Population and Root gall index of cucumber infected with *M. incognita*

Nematode population			
Treatments	Initial nematode population	Final nematode population	Root gall index
Neem	2005	562 ^b	3 ^b
Mexican sunflower	2007	537 ^b	3 ^b
EMAS	2011	2239 ^c	4 ^c
Carbofuran	2068	409 ^a	2 ^a
Control	2009	2540 ^c	4 ^c

Means within the same column followed by the same letters(s) are not significantly different ($P \leq 0.05$)

Table 2 shows the leaf extract, EMAS and Carbofuran treatments brought about variation in the length of cucumber vine. There was no significant difference in the vine length in the week 4 and 5 of the experiment. Significant differences were observed between week 6 and 8. EMAS and Carbofuran treatments recorded longer vine and significantly longer than other treatments in week 8. Table 3 shows that there was no significant difference in the stem girth of all the treatments tested. Table 4 shows that the leaf extract, EMAS and Carbofuran treatments do not affect the flowering time of cucumber. There was no significant difference in the number of days to first flower initiation, number of days to 50 and 100 % flowering respectively.

Table 5 showed that different treatments brought about variation in the yield component of cucumber plant. EMAS and carbofuran treatments recorded significantly greater yield among the various treatments. The number of fruit and average fruit weight was higher in EMAS and Carbofuran treatments compared to the leaf extract as well as control treatments and they were significantly higher than the other treatments in both the fruit weight and fruit number. EMAS treated plot recorded over 9 t/ha followed by Carbofuran with 8t/ha. Neem and wild sunflower extracts recorded over 5 and 7 t/ha respectively while the control treatment recorded about 5 t/ha.

Table 6 shows that initial and final nematode population and the gall index for all the treatments. There was a significant reduction in the soil final nematode population in plant extracts and Carbofuran treatments. There was no significant reduction in the soil final nematode population and root gall index for EMAS and control treatments indicating that the EMAS treatment has no effect on soil nematode, there were multiplication of soil nematode and high gall index which is not different from the control.

Table 7 shows that the nutrients composition of treated cucumber fruits was not affected by the various treatments. Table 8 shows that the plant extracts contained various bioactive chemical components which are capable of controlling the root-knot nematode. The bioactive chemical components observed in the plant extracts were: Alkaloid, Tannin, Flavonoid and saponin.

Table 7: Effects of EMAS, leaf extracts of plant and Cabofuran on the mineral nutrient composition of cucumber infected with *M. incognita*

Mineral nutrient composition of <i>M. incognita</i> infected cucumber						
Treatments	Na	Ca	K	P(ppm)	N(%)	CP(%)
Neem	0.221	0.1	1.03	53.2	2.69	16.81
Mexican sunflower	0.22	0.1	1.03	52.8	2.67	16.68
EMAS	0.44	0.28	1.28	53.2	2.71	16.93
Carbofuran	0.22	0.53	1.54	53.0	2.73	17.06
Control	2.22	1.02	1.03	52.8	2.66	16.63

Means within the same column followed by the same letters(s) are not significantly different ($P \leq 0.05$)

Table 8: Phyto active Chemical components present in neem and Mexican sunflower

Treatments	<i>Azadirachta indica</i>	<i>Tithonia diversifolia</i>
Alkaloid	+	+
Tannin	+	+
Flavonoid	+	+
Saponin	+	+

+ Chemical components detected

- Chemical components not detected

DISCUSSION

Application of neem and wild sunflower extracts as well as EMAS and Carbofuran brought about significant improvement in the growth and yield of nematode infected cucumber compared with untreated control. Plant extracts (neem and wild sunflower leaf) and carbofuran brought about significant reduction in the soil population and root gall index of *M. incognita* infested cucumber leading to improved growth and yield compared with the untreated control. The observed improvement on the growth and yield of *M. incognita* infected cucumber treated with plant extracts and carbofuran may be due the negative effects of the extracts and carbofuran on *M. incognita*. This negative effect of extract and carbofuran on nematode brought about positive growth and yield index on *M. incognita* infected cucumber. These type of control effects of Carbofuran and plant extracts on nematode pest of plants have been reported by many workers including Oyedunmade (2004); Abolusoro (2006); Olabiyi *et al* (2008); Oyedunmade *et al* (2009); Abolusoro *et al* (2010).

EMAS did not reduce the soil population and root gall index of *M. incognita* infected cucumber but performed better than other treatments with respect to growth and yield parameters. This observation is a clear indication that EMAS has no toxic effect on nematode yet it recorded better growth and yield compare to other treatments. The reason for this paradox might be due to the fact that EMAS is capable of strengthening the tolerance level of *M. incognita* infected cucumber to nematode attack, hence promoting growth and yield of such plant. This observation is similar to that of Afolami and Adigbo 1999 who observed heavily galled roots of okra by *M. incognita* did not negatively correlate with yield as a result of the tolerance conferred by the okra plant to nematode attack.

The result of this study indicated that the leaf extracts of neem and wild sunflower contained alkaloid, tannin, flavonoid, and saponin. This might suggest the presence of nematicidal components in the plants. Neem leaf extracts was found to possess nematicidal property (Abid and Magbool, 1991; Siddiquu and Alam, 1989; 1999; Abolusoro 2006). Olabiyi *et al* 2013 reported the presence of tannins, saponin, flavonoids, alkaloids as some of the bioactive chemical components of wild sun flower. There was a marginal rise in some of the mineral nutrient elements of Cucumber fruits in all the treatments (extracts, EMAS and carbofuran) compare with the untreated control. This observation was in agreement with Shwetha (2008) who, reported similar trends.

CONCLUSION AND RECOMMENDATION

The result of these experiments showed that plant extracts are capable of reducing the soil population as well as root gall index of *M. incognita* infected plant and thereby promoting growth and yield. EMAS does not reduce soil population on nematode and root gall index as it was not significantly different from the untreated control, yet it brought about better growth and yield. It therefore means that EMAS strengthens tolerance level of *M. incognita* infected plant and can be used in synergy with other control or bio-control agents so as to enhance the growth and yield of crop. The concentration of the plant extracts utilized should be increase in another experiment to monitor the level of toxicity of the extract to nematode on the crop under investigation.

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Olabiyi, T.I.^{1*}; Oyedunmade, E.E.A.² and G. J. Ibikunle³
¹ Faculty of Business, Environment and Society, Coventry University, Priory Street, CV1 5FB, Coventry, U.K.
² Department of Crop Protection, Faculty of Agriculture, University of Ilorin, P.M.B. 1515, Ilorin, Nigeria.
³ Department of Science Laboratory Technology, Ladoke Akintola University of Technology, P.M.B. 4000. Ogbomoso, Nigeria
*Corresponding author. E-mail: t.olabiyi@yahoo.co.uk
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