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Effect of Poultry Manure on Growth, Nutrients Composition and Heavy Metal Content of *Amaranthus caudatus* L.

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ABSTRACT: The aim of this study was to evaluate the effect of poultry manure on the growth of *Amaranthus caudatus* with a view to establishing the optimum poultry manure treatment for its growth. The study was carried out between the months of February and May 2020, in a portion beside the botanical garden in the University of Benin, consisted of three (03) treatments namely PM1 (10%), PM2 (20 %), and PM3 (30 %) with poultry manure and control (PM0) in a randomized complete block design (RCBD) and replicated three times. Data for growth parameters (height, number of leaves, leaf area, and number of branches) were collected from 1 to 6 weeks after transplanting (WAT). At 6 WAT, PM1 recorded the highest values for growth. At that time, PM1, PM2 and PM3 recorded leaf area values of 111.66 ± 24.11 cm², 74.33 ± 3.17 cm², 64.66 ± 22.34 cm², respectively compared to the control with 5.33 ± 0.33 cm². PM1 showed enhancing effect on proximate composition of ash, lipid, crude protein, crude fibre and dry matter compared to control. Carbohydrate and moisture content were higher in PM0. *A. caudatus* grown in 10 % poultry manure treatment (PM1) elicited high levels of Ca, Mg, K, Na, P, Zn and Fe. Total lead in *A. caudatus* plants derived from PM1 was higher than PM0. The results of this study shows that PM1 elicited higher yield in *A. caudatus* when compared with other treatments and is therefore recommended for growing the plant.

Keywords: Poultry manure, Heavy metal, *Amaranthus caudatus*, Performance.

Introduction

The demand for food has doubled as the world's population has grown, putting a strain on limited land resources. Vegetables, in addition to other food options, are regarded as a low-cost source of nutrition (Hussain *et al.*, 2009). Vegetables are high in vital biochemicals and nutrients including sugars, carotene, protein, vitamins, calcium, iron, ascorbic acid and trace mineral concentrations (Salunkhe and Kadam, 1998). Samkeliso *et al.*, (2020) reported that *Amaranthus*, also known as African spinach, is an excellent source of carotene, folic acid, vitamin C, calcium, iron and micronutrients. Amaranth is among the main vegetable in the Amaranthaceae family. Amaranth has widely been cultivated in central Asia, probably Iran (Kawazu *et al.*, 2003), and has a cultivation period of over 2000 years (Daneshvar, 2000), but also commonly cultivated in several regions in Nigeria. Fertilizers, both organic and inorganic, are important for normal plant growth by providing the nutrients that plants need to grow at their best (Erisman *et al.*, 2008). Organic fertilizers have been used for decades, while chemically synthesized inorganic fertilizers were produced just during the industrial revolution. Inorganic fertilizer has aided global population growth significantly; it is estimated that about half of the world's population is now fed as a result of artificial nitrogen fertilizer use (Erisman *et al.*, 2008).

Fertilizer usage at optimum results in higher yield and improved crop quality. The proper utilization of plant nutrients is critical in deciding the consistency of the harvest. Fertilizer application up to an optimum level often improves efficiency, while applications well in excess of this which result in poorer quality, either due to a simple nutrient excess or a mismatch between nutrients (Marschner, 1986).

According to Oyedeji *et al.*, (2014), NPK and poultry manure improved the growth and yield of three different species of amaranth (*Amaranthus hybridus*, *Amaranthus deflexus*, and *Amaranthus cruentus*), but had different effects on proximate composition, with NPK showing the highest crude fibre, protein, and fat content while poultry manure elicited the highest fat content. According to Emede *et al.* (2012), poultry manure positively influenced the plant growth and yield of *Amaranthus cruentus L.* According to Abdelrazzag (2002), increasing the rate of sheep and chicken manure increased the N content of onion significantly, while P and K content remained low. A mixture of chicken manure and biofertilizer increases onion yield and nutrient content in tuber (Fatma *et al.*, 2007). When compared to chemical fertilizers, the application of organic manures significantly increased levels of organic C and N, as well as the formation of water stable aggregates (N'Dayegamiye, 2006). According to Samkeliso *et al.* (2020), *Amaranthus* growth and yield increased significantly ($p < 0.05$) when fertilized with stillage, kraal manure, and compost. Several studies have focused on the effect of organic, inorganic, or combined fertilizer on soil properties, nutrient uptake, growth, yield, and some mineral contents. However, research on the effect of organic fertilizers on the nutritional composition and heavy metal contents of *Amaranthus caudatus* is limited.

A widespread misunderstanding is that organic fertilizers are safer than inorganic chemicals for plants and the environment. Improper application of organic fertilizers can also lead to contamination of the surface and groundwater, may cause deficiency of nutrient or toxicity of plants. Manure as well as inorganic fertilizers are good for plants and the environment if properly used (Koenig and Johnson, 1999). Hence, it is important to evaluate the effect of different concentrations of organic fertilizer (poultry manure) on the growth, nutrients composition and heavy metals content of *Amaranthus caudatus*.

The aim of this study is to evaluate the effect of poultry manure at different rates on growth, nutrients composition and heavy metals content of *Amaranthus caudatus* with a view to establishing the optimum poultry manure treatment for growing *A. caudatus*.

Materials and methods

Study area: This study was carried out in a portion beside the botanical garden in the University of Benin, Benin City, Nigeria. Benin metropolis is located in the humid tropical rainforest belt of Nigeria and belongs to Af category of Koppen's climatic classification marked by two distinct seasons namely the raining and dry seasons. Benin's rainy season begins in March/April and ends in October/November. Rainfalls are heavy and usually double upper limits, with a brief dry spell in August known as the "August Break. The annual mean rainfall value is 1339 mm, and based on the figures above, plenty of rain is available for the production of a variety of agricultural crops. The University of Benin is located at Latitude 6°20'1.32" N, north of the equator, and Longitude 5° 36' 0.54" E, East of the Greenwich meridian, with an annual mean temperature ranging from 19.44 °C to 31.11 °C. (Odemerho, 1988). The soil is loamy sand, acidic in reaction with little nutrient pool and classified as an ultisol.

Chemicals: Chemicals and reagents used were purchased from (Sigma chemical Company St. Louis, U.S.A) and were of analytical grade.

Seeds: The seeds of *Amaranthus caudatus* were obtained from Oba market in ring road, Benin City, Edo State. *Amaranthus caudatus* sample collected was authenticated at the herbarium unit of Plant Biology and Biotechnology Department, University of Benin, Benin City, Edo State.

Collection of soil sample: Soil was taken from Capitol area in the University of Benin at a depth of 0 – 15 cm using the zigzag method (Brady and Weil, 2008). The sample was collected from different points and bulked to form a composite sample. The composite sample was air-dried, crushed and sieved through a 2mm mesh sieve and part of it was stored for chemical analysis.

Poultry manure: The poultry manure was collected at Uwaifo's poultry farm in Oluku, Benin City, Edo State.

Experimental design and fertilizer treatment: The experiment included ten (10) fertilizer treatments for the Amaranth variety with factorial arrangement fitted into randomized complete block design (RCBD) and replicated three (3) times. Hence the experiment had a total of 30 experimental pots. The treatments are as follows:

- i. Control (no manure) PM0
- ii. 10 % poultry manure PM1
- iii. 20 % poultry manure PM2
- iv. 30 % poultry manure PM3

Planting and nursery management: Prior to planting, the amaranth seeds were soaked in water for about 24 h in order to enhance germination. The soaked seeds were first sown in the nursery of about 1.5 cm deep and were watered twice daily. Appropriate nursery management practices were carried out as at when needed to obtain

healthy and uniform seedlings. During soil preparation, poultry, cow, and compost manures were incorporated into specific pots based on treatment level and thoroughly mixed with the soil before being left for one week to allow for mineralization. After 2 weeks in the nursery, seedlings were transplanted at random to well-prepared beds (pots). Watering cans were used to water the seedlings twice daily, and the surrounding areas were weeded on a regular basis. To prevent harbouring pests, the experimental area and its surroundings were kept clean.

Data collection for performance: Data were first collected one week after transplanting (WAT) and subsequently at one-week interval for up to six weeks after transplanting. Data collected include: number of leaves, plant height, number of branches, leaf length and leaf width, which was used to compute the leaf area. Chemical analysis for proximate and mineral compositions as well as heavy metals content was determined after harvesting.

Performance: The number of leaves was counted from the plants and the average computed (Masarirambi *et al.*, 2012). Plant height is the length of the plant from the stem (surface of the soil) to the apex or tip of the plant. Plant height was measured using a measuring tape for plants per pot and the average computed (Masarirambi *et al.*, 2012). The leaf length and width were determined by measuring the length and the width of the leaves using a meter rule and the average was computed. The leaf length was measured from the leaf base to the tip whereas the leaf width was measured at the broadest part of the leaf blade (Masarirambi *et al.*, 2012). The number of branches was physically counted from the plants and the average computed. The leaf area (LA) was computed by multiplying the leaf length (LL) by the leaf width (LW) and the product multiplied by the correction factor (Samkeliso *et al.*, 2020).

Harvesting of the vegetables: The leaves of the spinach grown in the field experiment in control and treated soil were harvested at vegetative phase (market maturity) of plant development and were used for the analysis.

Preparation of plant for analysis: The leaves of each treatment were uniformly mixed before sampling and prepared for appropriate analysis as below.

Preparation of plant for chemical analysis: Upon arrival at the laboratory, the fresh and healthy vegetable were immediately washed under tap water and excessive water dripped off. Edible portions of the vegetables were cut into small pieces and homogenized using a Commercial Blender for two minutes. The homogenized sample was used for analysis immediately, but otherwise were transferred into an air-tight container and kept at 20 °C before analysis (Abuye *et al.*, 2003).

Preparation of plant for mineral and heavy metals analysis: Upon arrival at the laboratory, the fresh and healthy vegetables were immediately washed under tap water to remove all soil particles and then spread on newspaper for drying. After sun drying, the plants were weighed and oven dried at 70 °C for 72 h. The herbage was grounded. The powdered form was kept for mineral and heavy metals analysis (Abuye *et al.*, 2003). The minerals that were analyzed are calcium (Ca), magnesium (Mg), potassium (K), sodium (Na), phosphorus (P), while heavy metals were iron (Fe), copper (Cu), zinc (Zn), manganese (Mn), lead (Pb) and cadmium (Cd).

Estimation for proximate composition

Determination of moisture: A washed and dried platinum dish was weighed as W1 with an analytical balance, and then 3g of the sample was added and weighed as W2. The dish and its content was then transferred into a desiccator, then to an oven and left for three hours at 105 °C until the water completely dried. The platinum dish was transferred and cooled in the desiccator and weighed as W3. (A.O.A.C., 1990).

Calculation:

$$\% \text{ Moisture content} = \frac{(W2 - W3)}{(W2 - W1)} \times 100\%$$

Determination of ash content: A cool dry platinum dish was weighed as W1 using an analytical balance and 3 g of the sample was added and weighed W2. Then it was pre-ash on open flame and then transferred using a tong into a muffle furnace at 550 °C until it fully ashed (the colour changes to grey) and then weighed as W3.

Calculation:

$$\% \text{ Ash content} = \frac{(W3 - W1)}{(W2 - W1)} \times 100\%$$

Determination of total fat: 2 g of the sample was weighed into a clean dry test tube. Then 10ml of distilled water and 10 ml of concentrated HCl were added and placed in a boiling water bath for 30 min for hydrolysis. The hydrolysed sample was cooled and transferred to the separating funnel. The test tube was rinsed with 10 ml ethanol and added to the experimental separating funnel. Then to the experimental separating funnel, 30 ml diethyl ether was added and shaken then allowed to settle for separation to occur. A clean dry empty conical flask was weighed as W1. The ether layer was collected in a pre-weighed conical flask. The ethanol layer was re-extracted twice with 25 ml diethyl ether and the ether layer was added to the pre-weighed flask and the combined ether extract was evaporated over a boiling water bath. After the evaporation, the evaporated conical

flask was placed in an oven maintained at 105 °C for 2 h after which it was cooled in the desiccator and weighed again as W2. % Fat was calculated thus:

$$\% \text{ Fat} = \frac{W2 - W1}{W} \times 100\%$$

Determination of crude protein using Kjeldahl method

Digestion: One gram of the sample was weighed into a distillation flask, 25 ml of concentrated sulfuric acid (H₂SO₄) was added and a pinch of copper (II) sulfate (CuSO₄) and anti-bumps were added. The resulting mixture was digested till the mixture becomes colourless. It was then removed from heating and allowed to cool.

Distillation: Exactly 200 ml of distilled water was poured into the mixture followed by 85 ml of 50 % sodium hydroxide (NaOH) and then rinsed with another 50 ml of distilled water for neutralization. Then it was distilled in the upper layer of the Kjeldahl apparatus, where 50 ml of 2 % boric acid with 3 drops of screened methyl red in a 250 ml conical flask was used to collect the released ammonia gas. For the titration, about 200 ml of the distillate was collected in the conical flask and titrated using 0.05 M of H₂SO₄ till colour changed from blue light red. The titre value was recorded as (V). The % N was calculated thus:

$$\% \text{ N} = \frac{V \times 0.0014 \times 100}{\text{Weight of sample (W)}}$$

where: W = weight of sample taken

$$\% \text{ Protein} = \% \text{ N} \times F$$

F = factor equal to 6.25

Determination of crude fibre: 2 g of the sample was put into a round bottom flask, 100 ml of (0.23M) sulfuric acid solution was added and the mixture boiled under reflux for 30 min. The hot solution was quickly filtered under suction. The insoluble matter was washed several times with hot water until it was acid free. This was quantitatively transferred into the flask and 100 ml of hot (0.312M) sodium hydroxide solution was added and the mixture boiled again under reflux for 30 min and quickly filtered under suction. The insoluble residue was washed with boiling water until it was alkaline free. It was dried to constant weight in the oven set at 105 °C, cooled in a desiccator and weighed (C2). The weighed residue was then incinerated in a muffle furnace at 550 °C for 30 min, cooled in the desiccators and reweighed (C3). % Crude Fibre was calculated thus:

$$\% \text{ Crude Fibre} = \frac{C2}{W} \times 100$$

where: W = Weight of original sample.

C2-C3 = the loss in weight on ashing (incineration).

Determination of available carbohydrate: Available carbohydrate was determined by calculation thus:

$$\text{Available Carbohydrate (\%)} = 100 - (\text{Ash} + \text{Moisture} + \text{Protein} + \text{Fat} + \text{Crude fibre})$$

Digestion of plant samples for mineral analysis: 1 g of the defatted sample was weighed into ashing beaker and was ashed in the muffle furnace at 550 °C for 3 h. After cooling, the ashed sample was dissolved in 10 ml of 20 % nitric acid. Thereafter, it was filtered through the Whatman filter paper into a 100 ml volumetric flask and made up to the mark with distilled water. This digest was subsequently used for the determination of Ca, Mg, Na, K, P, using AAS Perkin Elmer Analyst 400 (Soil Survey Staff, 2014).

Determination of Ca, Mg, K and Na: AAS Perkin Elmer Analyst 400 was used for the determination of Ca, Mg, K and Na. The standard readings for the different concentrations of each cation were used to plot a graph of Absorbance vs Concentration (ppm) (Soil Survey Staff, 2014). The operational conditions for the metals are:

- i. Wavelengths at 422.7 nm, 285.2 nm, 766.5 nm, 589.0 nm, 882 nm for Ca, Mg, K, Na, P, respectively.
- ii. Slit of 0.7 nm, 0.7 nm, 0.7 nm and 0.2 nm for Ca, Mg, K and Na respectively.
- iii. Relative noise at 1.0, 1.0, 1.0 and 1.0 for Ca, Mg, K and Na respectively.
- iv. Linear range of 5.0 mg/L, 0.5 mg/L, 2.0 mg/L and 1.0 mg/L for Ca, Mg, K and Na respectively.

$$\text{Exchangeable bases (meq/100g)} = \frac{\text{ppm graph} \times \text{V.E.} \times \text{DF} \times 100}{1000 \times W \times \text{Eq Wt. of element}}$$

where: V.E. = Volume of extractant

DF = Dilution factor

W = Wt of sample used

Determination phosphorus (Bray and Kurtz – Method): 1 g of the sample was weighed into a 25 ml bottle with screw cap. Then 7 ml of the extraction solution was added (with pipette). The container was shaken for exactly 1 minute and the suspension was filtered into a dry bottle or beaker, after which 2 ml of the filtrate (pipette) was transferred to a dry 25 ml beaker. Five (5) ml of ammonium molybdate reagent and 1ml stannous chloride dilute solute were added to the filtrate in the beaker and mixed. It was then made up to the 10 cm³ mark. The colour development was measured after 10 min at 890 nm on a Spectronic 20. The absorption was read. The standards were measured in the same way (Soil Survey Staff, 2014).

Soil analysis: The sampled soil was analyzed at the Benin Owinna River Basin Laboratory, Faculty of Life Sciences, University of Benin, Nigeria. The following parameters were analyzed in the sampled soil: particle size, pH (in water), organic carbon, organic matter, available phosphorus, total nitrogen, cation exchange capacity (CEC) and exchangeable bases (Soil Survey Staff, 2014).

Determination of soil particle size: 50 g of 2 mm sieved soil was weighed into a 250 ml plastic beaker. Then 100 ml of 50 % calgon was added into the beaker and stirred with a glass rod. Distilled water of 100 ml was also added and stirred. It was left to stand for about 30 min with occasional stirring. The sample was transferred into a mixing cup and the content of the beaker was washed into the cup and stirred for 5 min. The mixture was then transferred completely into a 1000 cm³ measuring cylinder and made up to 1000 cm³ mark with water. The suspension was thoroughly mixed using a long-handle plunger to ensure that the sediment at the bottom of the cylinder is completely disturbed. Hydrometer readings were taken at 40 sec and 2 h. Before reading is due, the hydrometer was lowered gently into the suspension. Readings were taken at the top of the meniscus. A blank cylinder was prepared by making 100 ml of 50 % calgon solution up to one (1) litre mark with water. Hydrometer readings were also taken for the blank at 40 sec and 2 h. Temperatures of both the suspension and the blank were taken. After the last reading, the suspension was poured through a 0.2 mm mesh sieve. Sand grains retained on the sieve were washed with water into a beaker and dried in an oven at 105 °C (Soil Survey Staff, 2014). The corrected hydrometer readings [C (g/L)] were obtained by subtracting the blank reading RL(g/L) from the hydrometer readings in the soil suspension R(g/L) and adding 0.36 g/L for every degree above 20 °C

$$C = R - RL + (0.36T)$$

where T=Room temperature minus 20

The percentages by weight of the Silt, Clay and Sand are given by:

$$\% \text{ clay} = \frac{\text{Corrected 2h reading} - \text{Blank} \times 100}{\text{Weight of soil taken}}$$

$$\% \text{ silt} = \frac{\text{Corrected 40 sec reading} - \text{Blank} \times 100 - \% \text{ clay}}{\text{Weight of soil taken}}$$

$$\% \text{ soil} = 100 - (\% \text{ clay} + \% \text{ silt})$$

The sample was then classified according to the USDA or International Systems of Textural Classification

Determination of soil pH (in water): Twenty grams of air-dry soil (passed 2 mm sieve) was weighed into a 50 ml beaker and 20 ml of distilled water was added. The suspension was stirred several times during the next 30 min with a glass rod. The soil suspension was allowed to stand for about 30 min to allow most of the suspended clay to settle out of the suspension. The electrodes of the pH meter were inserted into the partly settled suspension and the pH was measured (Soil Survey Staff, 2014).

Determination of soil organic carbon (Walkley-Black method): A representative sample was taken and ground to pass through a 0.5 mm sieve. One gram of the sample was weighed and placed in the 250 ml flask. Then 10 ml of 1N K₂Cr₂O₇ solution was pipette accurately into each flask and the flasks were swirled gently to disperse the soil. Then 20 ml conc. H₂SO₄ was rapidly added from a measuring cylinder. The flask was immediately swirled gently until the soil and reagents were mixed and then swirled more vigorously for one minute. The flask was rotated again and allowed to stand on a sheet of asbestos for about 30 min. After that 100 ml of distilled water was added and was allowed to cool again. Then 5 ml of phosphoric acid was added to sharpen the colour change at the end point. Before titration, 3 drops of indicator were added and the solution was titrated with 0.5 N ferrous sulphate solution on a white background. As the end point was approached, ferrous sulphate was added in drops until the colour changed sharply from blue to red in reflected light against a white background. The blank solution was prepared in the same way but without soil to standardize the dichromate (Soil Survey Staff, 2014).

$$\% \text{ Organic Carbon in soil} = \frac{(\text{Blank titre} - \text{actual titre}) \times 0.003 \times m \times f}{\text{g of air dry soil}}$$

where: f = Correction factor = 1.33

m = Concentration of FeSO₄

Determination of soil available phosphorus (Extractable Phosphate - Bray and Kurtz – Method): 1 g of soil was weighed into a 25 ml bottle with screw cap. Then 7 ml of the extraction solution was added (with pipette). The container was shaken for exactly 1 min and the suspension was filtered into a dry bottle or beaker. After which 2 ml of the filtrate (pipette) was transferred to a dry 25 ml beaker. Then 5 ml of ammonium molybdate reagent and 1 ml stannous chloride dilute solute were added to the filtrate in the beaker and mixed. It was then made up to the 10 cm³ mark. The colour development was measured after 10 min at 890 nm on a Spectronic 20. The absorption was read. The standards were measured in the same way (Soil Survey Staff, 2014).

Determination of soil total nitrogen (Kjeldahl method): 1 g of fine mesh soil was weighed into 500 ml Kjeldahl flask or a digestion tube. Few drops of water were added and allowed to stand for about 30 min. 5 g of Kjeldahl catalyst and 20 ml of concentrated sulphuric acid were added. The flask was heated cautiously on a sand bath until frothing ceased. The temperature was then increased until digest cleared and for another 2 h. The flask was cooled and little water was added with care before the content was washed into a 100 ml volumetric flask. The content was allowed to cool completely and was then made up to mark. Then 10 ml of the digest was pipetted into a distillation flask. After that 20 ml 2% boric acid was measured into a 100 ml conical flask and the flask was introduced to the bottom of the condenser (3 drops of mixed indicator were added). Then 10 ml of 10 N NaOH was added to the 10 ml digest in the distillation flask. The distillation unit was immediately introduced and the digest distilled off. About 50 ml of the distillate was then collected. The distillate was titrated with 0.01N H₂SO₄ from green to purple end point (Soil Survey Staff, 2014).

$$\% \text{ Nitrogen} = \frac{0.014 \times (T - 1) \times NA \times \text{Vol. of digest} \times 100}{\text{Weight of soil} \times \text{Aliquot distilled}}$$

Determination cation exchange capacity (CEC) (Ammonium saturation method): 10 g of air-dried (2 mm) soil was weighed into a 500 ml Erlenmeyer flask and 40 ml NH₄OAc was added. The flask was shaken thoroughly and allowed to stand overnight. The soil was filtered with light suction using a 55 mm Buchner funnel (Corning size No. 40). The soil was not allowed to become dry and cracked. The soil was leached with the neutral 1N NH₄OAc reagent until no tests for calcium could be obtained in the effluent solution. (For the Ca test, a few drops of 1N NH₄Cl, 10% ammonium oxalate, and dilute NH₄OH of the leachate were added to a test tube, and the solution was heated close to its boiling point. The presence of Ca was indicated by a white precipitate). The leachate for the determination of exchangeable cations was preserved. The soil was leached about four times with neutral 1N NH₄Cl and once with 0.25N NH₄Cl (25 ml of each was used). The electrolyte was washed out with 200 ml of 99% isopropyl alcohol. The soil was transferred and filtered to 500 ml Kjeldahl flask. After that 200 ml of distilled water, few drops of liquid paraffin, pumice beads to prevent fuming and bumping and 10 ml of 1N NaOH were added. About 60 ml of the solution was distilled into 50 ml of 2 % boric acid solution measured into a 250 ml Erlenmeyer flask. Ten drops of bromocresol green-methyl red mixed indicator was added and the NH₄-borate produced with standard 0.02N H₂SO₄, filtered. Blanks were run on the reagents. The titre was corrected with the blank results. The colour change was from bluish green through bluish purple to pink at the end point (Soil Survey Staff, 2014).

$$CEC \left(\frac{meq}{100g} \right) = (T - B) \times N \times \frac{100}{W}$$

where: T = Vol of acid used in titration
 B = Titre value for blank
 N = Normality of acid used
 W = Weight of soil used

Determination of exchangeable bases (Ca, Mg, K and Na): The leachates preserved from the CEC determination (after dilution) were used for the determination of Ca, Mg, K and Na with AAS Perkin Elmer AAnalyst 400. The standard readings for the different concentrations of each cation were used to plot a graph of Absorbance vs Concentration (ppm) (Soil Survey Staff, 2014). The operational conditions for the metals are:

- i. Wavelengths at 422.7 nm, 285.2 nm, 766.5 nm and 589.0 nm for Ca, Mg, K and Na respectively.
- ii. Slit of 0.7 nm, 0.7 nm, 0.7 nm and 0.2 nm for Ca, Mg, K and Na respectively.
- iii. Relative noise at 1.0, 1.0, 1.0 and 1.0 for Ca, Mg, K and Na respectively.
- iv. Linear range of 5.0 mg/L, 0.5 mg/L, 2.0 mg/L and 1.0 mg/L for Ca, Mg, K and Na respectively.

$$\text{Exchangeables bases (meq/100g)} = \frac{\text{ppm graph} \times \text{V.E.} \times \text{DF} \times 100}{1000 \times \text{W} \times \text{Eq Wt. of element}}$$

where: V.E. = Volume of extractant
 DF = Dilution factor
 W = Wt of soil used

Determination of heavy metals: Lead (Pb) and Cadmium (Cd) were determined using atomic absorption spectroscopy as described for the method of determination of mineral content. A Perkin Elmer Analyst 400 was used. The operational conditions for the metals are:

- i. Wavelengths at 228.8 nm and 283.3 nm for Cd and Pb respectively.
- ii. Slit of 0.7 nm and 0.7 nm for Cd and Pb respectively.
- iii. Relative noise at 1.0 and 0.43 for Cd and Pb respectively.
- iv. Linear range of 2.0 mg/L and 20.0 mg/L for Cd and Pb respectively.

Statistical analysis: Data was analyzed using the Statistical Package for Social Sciences (SPSS) version 23.0 computer package. Descriptive statistics and One Way ANOVA were used to analyze data. Post hoc analytical tool used was Duncan Multiple Range test.

Results

Tables 1 and 2 show the chemical composition of soil and poultry manure used in this study. It was observed the pH of the soil is slightly acidic with a value of 5.72, while that of the poultry manure is alkaline with a value of 8.48. The most acceptable pH range for the growth of *Amaranthus caudatus* is 5.5-7.5. The pH of poultry manure could retard the growth of *A. caudatus*. However, when added to the soil, the interaction will cause it to be less alkaline. In this study, the growth parameters investigated showed the highest values were recorded for the 10% treated plants (PM1). The tables show increase in organic carbon with the value of 12.17±0.25 % compared the control (0.49±0.04 %). Phosphorus had a value of 3.82±0.01 g/kg compared to control with 0.31±0.01 g/kg. Other exchange bases were higher in poultry manure than in the soil.

Table 1: Physical properties of soil used

Particle size/Parameters	Rate
Clay (%)	5.00±0.012
Silt (%)	29.00±0.14
Sand (%)	65.93±0.12
Texture class	Sandy loam
pH (in water)	5.72±0.003
Organic carbon (%)	0.49±0.04
Available phosphorus (g/kg)	0.46±0.006
Total nitrogen (g/kg)	0.31±0.01
Organic matter (%)	0.92±0.027
Exchangeable bases	
Calcium (cmol/kg)	1.13±0.02
Magnesium (cmol/kg)	0.83±0.035
Potassium (cmol/kg)	0.22±0.014
Sodium (cmol/kg)	0.080±0.00
Cation exchange capacity (CEC) (ml/kg)	36.43±0.49

Values are mean ± standard error of mean of triplicate analysis

Table 2: Chemical Properties of poultry manure used

Parameter	Rate
pH (H ₂ O)	8.48±0.00
Total Nitrogen (g/kg)	3.82±0.01
Available phosphorus (g/kg)	1.21±0.006
Potassium (cmol/kg)	12.48±0.035
Calcium (cmol/kg)	18.61±0.006
Magnesium (cmol/kg)	27.54±0.78
Organic Carbon (%)	12.17±0.25
Organic matter (%)	20.72±0.38

Values are mean ± standard error of mean of triplicate analysis

The result of the effect of treatment with poultry manure on the height of *A. caudatus* is presented in Table 3. The treatment with poultry manure enhanced growth (height) of *A. caudatus* compared to control. Growth enhancement was however against the concentration gradient. At 3WAT, control height was 5.96±0.15 cm, while PM1, PM2 and PM3 had heights of 24.40±0.72, 10.83±1.25 and 9.73±2.65 cm respectively. At the end of

the experiment, the treatment with PM1 recorded the highest of 83.40±16.49 cm which was significantly different from the other treatments and control.

Table 3: Effect of poultry manure on the height (cm) of *A. caudatus*

Treatment	Weeks After Planting (WAT)					
	1	2	3	4	5	6
PM1	5.30±0.44 ^c	12.40±0.45 ^c	24.40±0.72 ^d	38.56±3.27 ^c	55.90±9.28 ^c	83.40±16.49 ^c
PM2	4.56±0.12 ^{bc}	6.13±0.54 ^{ab}	10.83±1.25 ^{bc}	19.33±5.89 ^b	31.16±5.54 ^b	47.73±8.38 ^b
PM3	2.96±0.77 ^a	4.76±1.27 ^a	9.73±2.65 ^{ab}	19.60±2.77 ^b	30.16±7.03 ^b	44.86±4.69 ^b
PM0	4.20±0.25 ^{ab}	5.17±0.17 ^{ab}	5.96±0.15 ^a	6.73±0.15 ^a	7.83±0.17 ^a	8.46±0.14 ^a

** PM1 = 10 % poultry manure, PM2 = 20 % poultry manure, PM3 = 30 % poultry manure, PM0 = Control (no manure)

Values are mean ± standard error of mean of triplicate analysis

Values with different subscript down the column are significant (p<0.05) different

Table 4 shows the effect of treatment with poultry manure on number of leaves of *A. caudatus*. The application of poultry manure to the soil enhanced growth in the number of leaves of *A. caudatus*, in comparison with control. There was however decrease along the concentration gradient. Thus at 3WAT, the values of 46.33±7.35, 20.66±9.17 and 14.33±0.66 were obtained for PM1, PM2 and PM3, while control (PM0) had a value of 11.00±0.57. At the end of the experiment, statistics showed that the PM1 treatment had the highest number of leaves with the value of 138.66±35.37. There was significant difference between control and the treatments.

Table 4: Effect of poultry manure on the number of leaves of *A. caudatus*

Treatment	Weeks After Planting (WAT)					
	1	2	3	4	5	6
PM1	8.00±0.57 ^c	22.00±2.64 ^b	46.33±7.35 ^d	66.33±11.28 ^b	96.66±22.65 ^b	138.66±35.37 ^c
PM2	5.33±0.88 ^{ab}	8.00±0.57 ^a	20.66±9.17 ^{abc}	35.66±18.00 ^a	59.66±18.97 ^{ab}	92.33±9.59 ^c
PM3	4.33±0.66 ^a	7.66±1.20 ^a	14.33±0.66 ^{ab}	28.33±8.41 ^a	47.33±13.96 ^a	73.00±25.03 ^b
PM0	6.33±0.33 ^b	9.33±0.33 ^a	11.00±0.57 ^a	12.00±0.00 ^a	14.66±0.33 ^a	17.33±0.33 ^a

** PM1 = 10 % poultry manure, PM2 = 20 % poultry manure, PM3 = 30 % poultry manure, PM0 = Control (no manure)

Values are mean ± standard error of mean of triplicate analysis

Values with different subscript down the column are significant (p<0.05) different

The result of the effect of poultry manure on the leaf area of *A. caudatus* is illustrated in Table 5. Leaf area was enhanced by treatment with poultry manure and the growth declined along the concentration gradient. At 4 WAT, control had 4.00±0.57 cm² while PM1, PM2 and PM3 had values of 83.66±14.74, 29.66±8.01 and 35.66±10.68 cm² respectively. At the end of the experiment (6WAT), PM1 had the highest leaf area value of 111.66±24.11 cm². The control (5.33±0.33 cm²) was significantly different from all other treatment. PM1 was also different from control and the other treatments.

Table 5: Effect of poultry manure on leaf area (cm²) of *Amaranthus caudatus*

Treatment	Weeks After Planting (WAT)					
	1	2	3	4	5	6
PM1	5.20±0.10 ^b	29.36±1.28 ^b	56.13±6 ^c	83.66±14.74 ^c	96.66±15.01 ^c	111.66±24.11 ^d
PM2	1.13±0.26 ^a	4.63±2.23 ^a	18.73±10.48 ^b	29.66±8.01 ^b	65.00±10.58 ^b	74.33±3.17 ^c
PM3	1.03±0.08 ^a	2.20±0.58 ^a	14.86±1.53 ^{ab}	35.66±10.68 ^b	53.66±17.33 ^{ab}	64.66±22.34 ^{bc}
PM0	2.00±0.00 ^a	3.00±0.57 ^a	4.00±1.00 ^a	4.00±0.57 ^a	5.33±0.33 ^a	5.33±0.33 ^a

** PM1 = 10 % poultry manure, PM2 = 20 % poultry manure, PM3 = 30 % poultry manure, PM0 = Control (no manure)

Values are mean ± standard error of mean of triplicate analysis

Values with different subscript down the column are significant (p<0.05) different

Table 6 shows the effect of poultry manure on the number of branches of *A. caudatus*. The addition of poultry manure to the soil enhanced growth (number of branches) of the plants. At 3WAT, the various treatments PM1, PM2 and PM3 recorded values of 7.33±1.33, 0.66±0.66 and 1.66±1.66 when control was 1.00±0.00a. At the end of the experiment, PM1 had the highest number of branches (25.33±2.18). The control was significantly different from all the treatments. PM1 was also different from other treatments and control.

Table 6: Effect of poultry manure on number of branches of *A. caudatus*

No. of Branches	Weeks After Planting (WAT)					
	1	2	3	4	5	6
PM1	0.00±0.00	2.66±0.66 ^c	7.33±1.33 ^c	12.66±1.33 ^c	16.00±2.51 ^c	25.33±2.18 ^c
PM2	0.00±0.00	0.00±0.00 ^a	0.66±0.66 ^a	4.33±1.76 ^{ab}	8.33±2.72 ^b	13.66±4.37 ^b
PM3	0.00±0.00	0.66±0.67 ^b	1.66±1.66 ^{ab}	4.66±2.72 ^{ab}	9.33±2.33 ^b	14.00±2.00 ^b
PM0	0.00±0.00	0.00±0.00 ^a	1.00±0.00 ^a	1.00±0.00 ^a	1.00±0.00 ^a	1.66±0.33 ^a

** PM1 = 10 % poultry manure, PM2 = 20 % poultry manure, PM3 = 30 % poultry manure, PM0 = Control (no manure)

Values are mean ± standard error of mean of triplicate analysis

Values with different subscript down the column are significant (p<0.05) different

The Table 7 shows the effect of poultry manure on proximate composition of *A. caudatus*. The addition of poultry manure to the soil enhanced yield of *A. caudatus*. The values for protein, lipids, fibre and carbohydrate on soil treated with poultry dungs are 6.54±0.19, 3.52±0.14, 30.13±0.52 and 27.36±0.82 respectively and 4.45±0.11, 2.64±0.15, 28.07±0.23 and 31.44±0.67 for control. The nutritional composition of plants treated with poultry manure had the higher values for protein, lipids and fibre. However, the carbohydrate content in the control was greater than that of the poultry manure treated plants.

Table 7: Effect of poultry manure on proximate composition of *A. caudatus*

Treatment (%)	Percent (%)						
	Moisture	Lipid	Ash	Fibre	Protein	Carbohydrate	Dry matter
PM0	30.04±0.24 ^a	2.64±0.15 ^a	3.05±0.20 ^a	28.07±0.23 ^a	4.45±0.11 ^a	31.44±0.67 ^a	69.96±0.24 ^a
PM1	28.16±0.24 ^b	3.52±0.14 ^b	4.29±0.24 ^b	30.13±0.52 ^b	6.54±0.19 ^b	27.36±0.82 ^b	71.84±0.23 ^b

** PM1 = 10 % poultry manure, PM0 = Control (no manure)

Values are mean ± standard error of mean of triplicate analysis

Values with different subscript down the column are significant (p<0.05) different

Plants derived from PM1 had significantly higher amounts for Ca, Mg, K, Na, P, Fe, Cu, Zn and Mn compared to the untreated plants which did not produce as much nutrients (Table 8).

Table 8: Effect of poultry manure on the mineral composition of *A. caudatus*

Treatment (%)	mg/g								
	Ca	Mg	K	Na	P	Fe	Cu	Zn	Mn
PM0	0.47±0.47 ^a	1.42±0.03 ^a	15.90±0.28 ^a	0.36±0.06 ^a	0.087±0.01 ^a	0.17±0.01 ^a	0.03±0.01 ^a	0.04±0.01 ^a	0.005±0.01 ^a
PM1	0.87±0.03 ^b	4.78±0.02 ^b	42.54±0.65 ^b	0.73±0.01 ^b	0.57±0.06 ^b	0.36±0.01 ^b	0.05±0.01 ^b	0.07±0.02 ^b	0.01±0.01 ^b

** PM1 = 10 % poultry manure, PM0 = Control (no manure)

Values are mean ± standard error of mean of triplicate analysis

Values with different subscript down the column are significant (p<0.05) different

Lead was found to be higher in plants treated with poultry manure and was significantly (p<0.05) different from control plants (Table 9). Cadmium was not found.

Table 9: Effect of poultry manure on heavy metals of *A. caudatus*

Treatment	mg/kg	
	Pb	Cd
PM0	0.34±0.09 ^a	0.002±0.01 ^a
PM1	1.66±0.14 ^b	0.003±0.02 ^b

** PM1 = 10 % poultry manure, PM0 = Control (no manure)

Values are mean ± standard error of mean of triplicate analysis

Values with different subscript down the column are significant (p<0.05) different

Discussion

The growing population of many tropical countries raises awareness of the significance of vegetables as a source of essential nutrients and various chemicals that may not be found in other food sources (Ihekoronye and

Ngoddy, 1985; Schreinemachers *et al.*, 2018). Organic fertilizers play an important role in improving soil fertility by adding essential nutrients to soil that aid in adequate plant growth and yield.

The soil used in this experiment is typically sandy loam which indicates that the water holding capacity of the soil is good. The essential nutrients such as organic carbon, nitrogen, phosphorus, sodium, magnesium and potassium in soil used were very low which shows the low fertility status of the soil which is in agreement with work by Akanbi and Togon (2002) and Basak *et al.* (2022). This observation is confirmed by Ogedegbe *et al.* (2013), who reported that most agricultural soils were impoverished due to weathering, leaching, erosion and intensive cultivation. A good soil has an organic matter value above 3% (Alam *et al.*, 2007). This implies that the variable response of amaranth to poultry manure observed in this study was expected and also the application of organic fertilizers to increase soil fertility for amaranth production was justified which was also reported by Law-Ogbomo and Ajayi (2009). The poultry manure used for this experiment had higher nitrogen, potassium, phosphorus, organic carbon, organic matter content as well as a higher pH compared control. This could be attributed to the high nutrient dense feed chickens consume and concentrate over a short period of time. In this study, dosage of poultry manure had incremental effect on the plant height, average number of leaves and leaf area compared to control. However the increase was against the concentration gradient. It has been recognized that the application of poultry manure above agronomic rates may result in the release of salts into the soil either because of a straight forward nutrient excess or imbalance between nutrients (Marschner, 1986; Singh *et al.*, 2017) which may adversely affect crop yield.

The highest plant height of amaranth at maturity was observed in the PM1 plants which was significantly ($p < 0.05$) different from the other treatments. This position was earlier reported by Egharevba and Ogbe (2002) and Okokoh and Bisong (2011). The enhanced height exhibited by PM1 plants might have been due to the presence of the primary nutrients plus other minerals found in organic manure at optimum level and also it may be due to favourable nutrient mineralization of poultry manure as a result of the influence of the mineral component on the organic content of the manure (Abayomi and Adebayo, 2014). The control plants had the lowest plant height as they had to depend mainly on the intrinsic soil fertility as exhibited by the soil chemical analysis (Table 1). A similar effect for control was reported for *Amaranthus caudatus* by Abayomi and Adebayo (2014) and on radish stems amaranth-indian spinach by Islam *et al.* (2011). The height of the plant is an important growth character directly linked with its productive potential of plants. An optimum plant height is claimed to be positively correlated with productivity of plants (Saeed *et al.*, 2001).

At maturity, the PM1 showed the highest average number of leaves of Amaranth, which was also reported by Law-Ogbomo and Ajayi, (2009) for *Amaranthus cruentus*. This also agrees with reports by previous workers such as Sanwal *et al.* (2007) in turmeric (*Curcuma longa*) and Premesekhar and Rajashree (2009) in Okra (*Abelmoschus esculentus*), who separately attributed higher leaf yield to released nutrients from organic manure application which improved the chemical, physical and biological properties of soil.

In this study, the best succeeded treatment (PM1) on *A. caudatus* vegetative parameters was used to analyse for proximate and mineral compositions as well as heavy metals content of *A. caudatus*. The moisture content for *A. caudatus* was higher in control (PM0) plants. This is contrary to the report by Mofunanya *et al.* (2014) in which poultry manure treatment elicited the highest moisture content. This study corroborated similar results reported by Oyedeji *et al.* (2014) on the effect of NPK and poultry manure on growth, yield and proximate composition on *Amaranthus hybridus*.

PM1 plants had the highest amount of fat. The findings of this study agree with previous work by Kahu (2017) and Mofunanya *et al.* (2014), who reported similar findings with this study, that plants grown with organic fertilizer had the highest fat value. The reason for having higher fat for plants grown with organic fertilizers is attributed to the high organic content of the poultry manure, thereby having high level of carbon containing compounds which are attacked by bacteria, actinomycetes and fungi under aerobic conditions and the carbon is primarily mineralized into CO₂ and some lost as methane. The CO₂ is absorbed by the plants and used to produce fatty acids.

PM1 plants yielded the highest level of crude protein and differed significantly ($p < 0.05$) from control. This is consistent with previous reports on *Amaranthus hybridus*, *Amaranthus cruentus*, and *Amaranthus deflexus* by Oyedeji *et al.* (2014). This could be due to the fact that poultry manure contains a high concentration of nitrogen. When applied to the soil, the nitrogen is converted into ammonia through the nitrogen fixation process by bacteria such as *Azotobacter*, *Clostridium*, *Rhizobium*, and *Azospirillum*. Also, the ammonia can be further converted to nitrate by nitrifying bacteria. The ammonia in the form of ammonium and nitrate are taken by the plants and used for synthesis of nucleotides and protein.

Crude fibre was found to be significantly higher in PM1 plants. Oyedeji *et al.* (2014) reported similar results for *Amaranthus cruentus*. The high crude fibre may be due to the high levels of carbon containing compounds in the poultry manure which are easily converted to CO₂ and absorbed by the plants which readily synthesize fibre. PM1 plants also showed significantly higher ash content in *A. caudatus*. Oyedeji *et al.* (2014) reported similar results for *Amaranthus hybridus*, *Amaranthus deflexus* and *Amaranthus cruentus*. The ash content in the poultry

organic fertilizers were significantly higher and may be due to the balanced nutrients in the manures which contains a lot of minerals and trace elements. Control plants recorded the higher available carbohydrate. There was significant ($p < 0.05$) difference from PM1 plants. The plants derived from control produced high level of carbohydrate because the plants derived from PM1 had more available nutrients. Plants derived from PM1 produced more nutrients like mineral elements, fat, and protein, resulting in less carbohydrate than the untreated plants, which could not produce as many nutrients. The PM1 had the higher dry matter and was significantly ($p < 0.05$) different from control. This is because poultry manure treatment had higher levels of fat, ash, fibre, and crude protein, all of which are energy sources.

Lead was found to be higher in plants treated with poultry manure and was significantly ($p < 0.05$) different from control plants. The reason for this may be due to the fact that lead contamination may have originated from the chemicals used to treat diseases in commercial chickens (Bolan *et al.*, 2010) and subsequently absorbed by the plants. Cadmium was not found in any of the treatments and may have been due to heavy metals absorption being governed by soil characteristics such as pH and organic matter content. The magnitude of accumulation and the toxic level depend on the plants and heavy metals species. Cadmium could therefore not have been present in the soil and fertilizers or if it is present, might not be readily available for use by amaranth.

Conclusion

The results of this study justify the use of poultry manure for improved yield. An obvious drawback however is the elevated level of lead in the treatments.

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