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Research Paper

Comparative Study on the Effects of Bio-fertilizers, Organic and Inorganic Fertilizers on Growth and Yield Components of Cowpea (*Vigna unguiculata*)

Itelima, J. U.,[1]* Ogbonna, A. I[2]., Onyimba, I. A[3]., Nwadiaro, P. O[4]., Nyam, M. A[5]., and Agbala, C. C.[6]

[1, 2, 4, 5]Department of Plant Science and Technology, University of Jos, Nigeria.
[3, 6]Department of Laboratory Science and Technology, University of Jos, Nigeria.
*Corresponding author E-mail: janetitelima@yahoo.com

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For optimum plant growth, nutrients must be available in sufficient and balanced quantity. Fertilizers are designed to supplement the nutrients already present in the soil. Increased crop production largely relies on the type of fertilizers used to supplement essential nutrients for plants. The study was carried out to determine the effects of bio-fertilizers produced from plant and animal waste materials such as saw dust, cow dung and poultry droppings inoculated with a fungus (*Aspergillus niger*) and organic and inorganic fertilizers on the growth and yield of cowpea (*Vigna unguiculata*). The experiment was conducted at the Department of Plant Science and Technology Nursery, University of Jos. The saw dust and the cow dung and or poultry droppings were mixed in a ratio of 30:1 and then autoclaved to remove other microbes from the substrates. The substrates were mixed with *A. niger* and water in line with standard method after which they were placed separately into a locally constructed bio-digester. The substrates (cow dung + saw dust mixture and or poultry droppings + saw dust mixture) were stirred daily, while they were allowed to be degraded by the fungus (*A. niger*). The process of degradation of the different waste materials lasted for a period of two weeks after which the nitrogen, phosphorus and potassium (NPK) contents of the bio-fertilizers were analyzed alongside that of organic fertilizers. After biodegradation process, the various substrates were used to amend the soil used for growing cowpea. Six treatments plus control with 5 replicates per treatment were used. The fertilizer types included; bio-fertilizer produced from cow dung-sawdust mixture (BCD), bio-fertilizer produced from poultry droppings-sawdust mixture (BPD), combination of BCD and BPD and organic manures produced from cow dung (CD) and poultry droppings (PD) and inorganic fertilizer (IF). Pots of soils that were not amended with any of the fertilizer served as the control. Plant growth and yield parameters of cowpea plant grown on soil amended with bio-fertilizers were measured and compared with those grown on soil amended with organic and inorganic fertilizers. Both bio-fertilizers and the inorganic manures affected growth parameters of the cowpea variety used at varying degrees. There was a remarkable increase in all the growth parameters for all the treatments over the control (Cowpea grown on soil that was not amended with fertilizer) with the exception of the cow dung manure (CD). The yield of the crop which was obtained from the combination of the two bio-fertilizers (BCD and BPD) gave the best yield with respect to the total weight of the seed per pot 169 g followed by BPD 129 g and BCD 127 g, while IF, PD, control and CD gave the percentage yield of 94, 85, 68 and 61 g respectively. Similar results were also obtained from other crop yield parameters considered in the experiment, where the combination of BPD+BCD gave the highest yield except in the case of the mean length of one pod, with IF giving the highest yield of 0.19 cm. The results of the present study reveals that to obtain optimum yield of cowpea, the combination of the bio-fertilizers produced from cow dung-saw dust mixture and poultry droppings-saw dust mixture should be employed in amending the soil used for cultivating the crop especially in soil lacking essential nutrients.

Keywords: Bio-fertilizers, Organic and inorganic fertilizers, growth and yield components, *Vigna unguiculata*

INTRODUCTION

Plant nutrients are essential for production of crops and healthy food for the world's expanding population. For optimum plant growth, nutrient must be available in sufficient and balanced quantities. Soil contains natural reserves of plant nutrients, but these reserves are largely in forms unavailable to plants, and only a minor portion is released each year through biological activity or chemical processes. This release is too slow to compensate for the removal of nutrients by agricultural production and to meet crop requirement. Therefore fertilizers are designed to supplement the nutrients already present in the soil (Chen, 2006). In recent years agrochemicals such as chemical pesticides and fertilizer are extensively applied to obtain high yield. Intensive application of agrochemicals leads to several agricultural problems and poor cropping systems. Some farmers use more chemical fertilizers than the recommended levels for many crops. This practice accelerates soil acidification but also risks of contaminating ground water and the atmosphere and also weakens the roots of plants and making them easy prey to unwanted diseases (Chun-Li, 2014). Establishing an environmental friendly co-existing mechanism on earth is of vital importance.

Bio-fertilizers are important components of integrated nutrient management. These potential biological fertilizers would play key role in productivity and sustainability of soil and also protect the environment as eco-friendly and cost effective inputs for the farmers (Khosro and Yousef, 2012). Bio-fertilizers are renewable sources of plant nutrient which supplement chemical fertilizers (Hari *et al*., 2010). Utilization of bio-fertilizer is one of the ways to increase crop production by naturally optimizing the nitrogen and phosphorous level of the soil and by enriching the compost waste used as a natural fertilizer (Khosro and Yousef, 2012). The main sources of bio-fertilizer are bacteria, fungi and cyanobacteria (blue-green algae). Vessey, (2003) defined bio-fertilizer as a substance which contains living microorganisms which, when applied to seeds, plants surfaces, or soil, colonizes the rhizosphere or the interior of the plant and promotes growth by increasing the supply or availability of primary nutrients to the host plant. According to Hari *et al*. (2010) bio-fertilizer is most commonly referred to as selected strains of beneficial soil microorganisms cultured in the laboratory and packed in suitable carriers.

Bio-fertilizers differ from chemical and organic fertilizers in the sense that they do not directly supply any nutrients to crops and are cultures of special bacteria, fungi and algae. The production technology for bio-fertilizers relatively simple and installation cost is very low compared to chemical fertilizer plants. Cowpea (*Vigna unguiculate*) is a legume grown in savannah region, the tropics and sub-tropics. It is largely grown in the West and Central African countries. Its value lies with its high protein content. Its ability to tolerate drought and poor soil makes it an important crop in the savannah region where these constraints restrict other crops. Cowpea seed is nutritious and is a cheap source of protein for both rural and urban consumers. The seed contains about 25% protein and 64% carbohydrate (Chinma *et al*., 2008). The study therefore, was carried out to determine the effects of bio-fertilizers, organic and inorganic fertilizers on the growth and yield components of cowpea (*Vigna unguiculata*) .

MATERIALS AND METHODS

Sample collection

With the aid of a shovel about 50 kg of fresh cow dung was collected from abattoir (slaughter house) in Jos, Nigeria. Also about 50 kg each of poultry droppings and saw dust were collected from the section of a local poultry farm and timber market respectively in the above mentioned city. About 30 kg of potato peels were collected from food sellers, sundried and milled into powder using a grinding machine to serve as one of the substrates for the growth of the fugus (*Aspergillus niger*). Soil samples were collected from the garden in the study area and screened for the presence of *A. niger*. Ten gram of soil sample was dissolved in 100 ml of sterile distilled water and mixed thoroughly to obtain the dilution of 10^{-1}. The soil sample was serially diluted in sterile distilled water up to 10^{-7} (each tube containing 9 ml of sterile water). After then, 10^{-5}, 10^{-6}, 10^{-7} dilutions were taken for spread plate technique culture. Sterilized Potato Dextrose Agar (PDA) was prepared and poured into Petri dishes. After solidification of the medium, 0.1 ml of each dilution was poured into agar medium plate in duplicates. By using L-rod, the sample was spread evenly over the agar surface and then incubated at room temperature (25°C) for 3 days.

Identification of fungal culture

To evaluate the colony characteristics of *A. niger*, it was necessary to subculture the suspected colony of the fungus to PDA to obtain a pure culture. The Morphology and cultural characteristics were determined and used for identification of the fungi. The fungal isolates were identified by making references to Domsch *et al*. (1980).

Preparation of growth medium and inoculum

The growth medium used for preparing the *A. niger* inoculum consisted of 30 g of milled potato peels, peptone, 0.1%; malt extract, 0.1% (w/v), calcium carbonate 0.2% (w/v); ammonium phosphate, 0.2% (w/v), and ferrous sulphate. $7H_2O$, 0.001% (w/v) (Abouzeid and Randy, 1986). *A. niger* inoculum was prepared in 250 cm^3 cotton–plugged conical flasks containing 100 cm^3 of the growth media. The flasks containing the growth medium were sterilized and inoculated with *A. niger* spores. A sterile glass rod was inserted in each of the flasks and the flasks were kept at room temperature for 7 days to allow for sufficient mycelial growth and sporulation of the fungus

Production of bio-fertilizer

With the aid of a shovel, the saw dust and the cow dung/poultry droppings were mixed in a ratio of 30:1 respectively. The C:N of manure is a very important factor that affects the whole degradation process because microbes need 30 times more carbon than nitrogen to grow and carry out their biochemical activities. The ratio used in this experiment was obtained by weighing the materials with the aid of a weighing balance. 20 kg of the cow dung + saw dust mixture and the poultry droppings + saw dust mixture were placed separately in locally constructed plastic container bio-digester of 20 liters having a cover and a stirrer. The manures which served as carries of fungal inoculum were autoclaved to remove other microorganisms. After this, the liquid medium containing the fungus and one litre of water was added to every 2 kg of the manure to form slurry. Water is needed for active degradation of the substrates in the bio-digester. The bio-digesters were covered tightly and left for a period of two weeks during which the substrates were turned regularly to ensure faster and complete degradation. The untreated cow dung and poultry droppings without saw dust were dried and kept at room temperature so that their effects on plant parameters can be compared with those of the biodegraded manures.

Aging of the substrates

After the process of bio-digestion, the substrates were kept in a cool dry place to dry. Sun drying was not employed because elements such as nitrogen can escape when exposed to high temperatures (Swathi, 2010). Aging was done in order to dry the degraded substrates and end the activity of microorganisms that might have survived degradation temperatures. Aging lasted for two weeks after which the substrates were sieved to remove lumps.

Packing and storage

The bio-fertilizer was packaged in a black polythene bag, sealed and labeled according to the type of organic manure, microorganisms and the quantity of NKP. They were stored in a cool dry place away from direct sunlight.

Analysis of nutrient content of bio-fertilizers

The two bio-fertilizers produced from cow dung (BCD), poultry droppings (BPD), and the two untreated manures; cow dung (CD) and poultry droppings (PD) were analyzed for the presence of nitrogen, phosphorous and potassium according to methods described by Reginald, (2012). Nitrogen and phosphorous content were determined using calorimeter method, while potassium was measured with an ion- selective electrode.

Experimental design and measurement of plant parameters

The experiment was conducted at the Department of Plant Science and Technology Nursery, University of Jos. The experimental design employed in this study was the complete randomized design (Rangaswamy, 2007). Six treatments plus control with 5 replicates per treatment were used. The manure types were weighed and 1.6 kg of each manure type mixed with soil at ratio of 1:4 respectively (Adeoye *et al*., 2011) and then filled pots having diameter of 30 cm and length of 35 cm . The manure types included; bio-fertilizer produced from cow dung + sawdust mixture (BCD), bio-fertilizer produced from poultry droppings + sawdust mixture (BPD), and organic manures produced from cow dung (CD) and poultry droppings (PD), combination of BCD and BPD, inorganic fertilizer (IF). Pots containing mixture of sandy and loamy soils that were not amended with any of the manure served as the control. All the pots were kept in a green house to avoid wilting which may result from harsh weather of the experimental site. The applied manure was allowed to mix well with the soil by light irrigation with a portable sprinkler system, ones daily for 3 days. The cowpea variety (TVx3236), were then planted at a rate of 3 seeds per pot. The irrigation of the plants continued all through growth of the plants and up to the harvesting of the pods. Weeds were removed when found growing in the pots. The plant parameters measured during the six weeks of the experiment included plant height (measured with a meter rule), leaf area index (calculated as a product of total length and breadth at the broadest point of the longest leaf on the plant), and stem girth (measured with a venier caliper). After fruiting and maturity the harvested pods were taken and assessed for parameters needed to calculate the yield such as number of pods, length of pods, and number of seeds per pod and weight of seeds (Adeoye *et al*. 2011).

Calculations:

Leaf Area (LA) = Laminal length x maximum width x 0.75.

Statistical analysis

The data obtained from the experiment were subjected to analysis of variance (ANOVA) and fishes least significant (F-LSD) was employed to separate the means.

RESULTS AND DISCUSSION

The results in (Figure 1) represent the nutrient content of the various manure types which include; bio-fertilizer produced from cow dung-sawdust mixture (BCD), bio-fertilizer produced from poultry droppings + sawdust mixture (BPD) and organic manures produced from cow dung (CD) and poultry droppings (PD). The results showed that nutrient contents of organic manure samples varied widely, while the nutrient contents of bio-fertilizers were comparable. In general, PD had the highest concentrations of nitrogen (482 ppm), followed by CD (465 ppm), while BPD and BCD had similar concentrations of potassium, 379 ppm and 376 ppm respectively which were higher than those of PD and CD (271 ppm) and (250 ppm) respectively. Although, the phosphorous contents were very low in all the manure samples but slight increases were recorded for BCD and BPD. The rise in potassium and phosphorus levels in the bio-fertilizers is as result of the activities of *A. niger* inoculated into the substrates. *A. niger* has the ability to secrete metabolites capable of converting the insoluble forms of phosphorus and potassium to soluble forms. The mean NPK contents of the bio-fertilizers obtained in this study is suitable for plant growth, as the mean NPK requirement of plants is usually in the ratio of 1.5-0.2-1.0. It has also been reported that excess phosphorus in soil causes early maturity of plants and low crop yield in the plants, whereas appreciable amount of nitrogen and potassium are needed for good crop yield (Scalenghe *et al*., 2012).

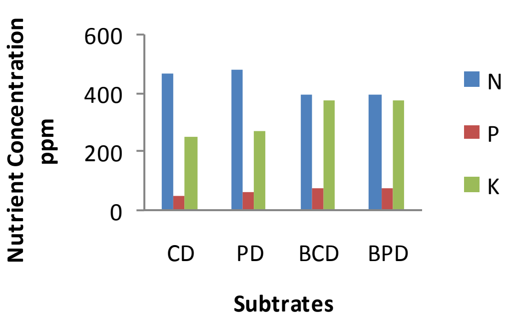

Figure 1. Nutrient contents of bio-fertilizers and organic fertilizer. (N=Nitrogen, P=Phosphorus, K=Potassium) CD =Cow dung (organic manure), PD = Poultry dropping (organic manure), BCD = Bio-fertilizer from cow dung, BPD=Bio-fertilizer produced from poultry droppings.

The results of the means of the plants' growth parameters are presented in (Tables 1 and 2). Both bio-fertilizers and the inorganic manures effected growth parameters of the cowpea variety used at varying degrees. There was a remarkable increase in all the growth parameters for all the treatments over the control (Cowpea grown on soil that was not amended with fertilizer) with the exception of the cow dung manure. All the growth parameters were affected by manure application. The application of inorganic fertilizer and poultry droppings (PD) led to remarkable difference in stem height, number of leaves and leaf area index of the cowpea plant. The yield of the crop which was obtained from the combination of the two bio-fertilizers (BCD and BPD) gave the best yield with respect to the total weight of the seed per pot 169 g followed by BPD 129 g and BCD 127 g, while IF, PD, control and CD gave the percentage yield of 94, 85, 68 and 61 g respectively. BPD and BCD also had significant ($p<0.05$) higher yield in most of the parameters than IF, PD and CD. Similar results were also obtained from the other crop yield parameters considered in the experiment, where the combination of BPD+BCD and BPD and BCD gave better yield than other treatments except in the case of the mean length of one pod, with IF giving the highest yield of 0.19 m. The highest yield obtained by combining BPD and BCD in most of the parameters is in consonance with work of Adeoye *et al*. (2011) who reported that the combination of poultry and cattle manures gave better leaf areas of cowpea compared to the use of either of the manure alone. Large leaf area may likely give better seed yield as a result of higher rate of photosynthetic process taken place in the leaves. The improvement in cowpea yield is associated with increase in the phosphorous and potassium concentrations in the soil amended with bio-fertilizers is affirmed by Reyghan and Amiraslani (2006). *A. niger,* a phosphorus and potassium solubilizing fungus, can enhance plant growth by increasing the uptake of phosphorus and potassium by the crop. In addition, *A. niger* has the ability to enhance the availability of other elements such as iron to the plants (Fe) zinc (Zn) (Ngoc *et al*., 2006). Also, it can synthesize enzymes that modulate plant hormone level and can also kill pathogen with antibiotic (Akhtar and Siddiqui, 2009). The lesser numbers of pods and longer length of pods recorded in cowpea plants grown in soil amended with poultry droppings and the inorganic manure (PD&IF) than those grown in soil amended with bio-fertilizer could be as result of very high concentration of nitrogen in PD&IF. This agrees with the report of Davis *et al*. (1991) which stated that excess nitrogen in cowpea promotes lush vegetable growth, suppresses nitrogen fixation and may reduce pod and seed yield. The excessive application of poultry manure in some cropping systems has also resulted in nitrate contamination of ground water (Chun-Li, 2014). Thus, proper estimation of nitrogen content of poultry manure and proper manure handling are necessary for optimum crop growth and minimal nitrate leaching. One will expect that the soil treated with cow dung (CD) should give a better yield than the control but

Table 1. Means of the plants growth parameters (6 weeks after seed sowing).

Treatment (Manure types)	Height (cm)	Leaf area (cm^2)	Stem girth (cm)
Control	24.3	1.161	0.067
CD	22.7	1.125	0.061
PD	32.6	1.192	0.074
BCD	27.5	1.176	0.080
BPD	28.1	1.179	0.086
BCD+BPD	28.5	1.185	0.098
IF	33.1	1.198	0.079

Table 2. Effects of various manure types on cowpea yield.

Treatment (Manure types)	Mean number of pod per plant	Mean length of pod (m)	Mean number of seed per pod	Mean number of seed per pot	Total weight of bean seed (g) harvested
Control	12.00	0.11	9.00	324	68
CD	11.00	0.10	8.00	264	61
PD	15.00	0.18	10.00	450	85
BCD	18.00	0.15	13.00	702	127
BPD	19.00	0.11	14.00	741	129
BCD+BPD	23.00	0.16	16.00	1104	165
IF	14.00	0.19	11.00	462	94

CD=Cow dung (organic manure), PD=Poultry droppings (organic manure), BCD= Bio-fertilizer produced from cow dung + saw dust mixture, BPD=Bio-fertilizer produced from poultry droppings + saw dust mixture, BCD+BPD= Bio-fertilizer produced from cow dung saw dust mixture combined with Bio-fertilizer produced from poultry droppings-saw dust mixture, IF= Inorganic fertilizer.

the reverse is the case. This present result is in line with the findings of Reyhan and Amiraslani, (2006) which stated that cowpea yield, can be inhibited by cattle manure because of its high salinity level.

Conclusion

In conclusion, the present study has shown that using bio-fertilizers offers a better option for the growth and yield of cowpea and will help in reducing the use of agrochemical, and also help to maintain soil fertility and strength. The yield components of cowpea plants were increased remarkably as a result of the soil being amended with bio-fertilizers. However, the combination of both bio-fertilizers gave the best yield. Thus, for optimum yield, it is recommended that the combination of the bio-fertilizers produced from cow dung + saw dust mixture and poultry droppings + saw dust mixture be employed in amending the soil used for growing cowpea.

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