Soil amendment with poultry manure, single super phosphate and *Bacillus subtilis* inoculant improved performance of cowpea (*Vigna unguiculata* (L.) Walp.) on sandy loam

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Submission: 23 November 2023 Revised: 9 February 2024 Accepted: 12 February 2024

ABSTRACT

Background and Objectives: Phosphorus (P) deficiency often limits the yield of cowpea (*Vigna unguiculata* (L.) Walp.) on highly weathered soils. Applying biofertilizers and organic fertilizers is a promising alternative to fossil-based fertilizers for improving P availability. The effect of organic and inorganic fertilizers and biofertilizers in improving growth and P uptake in cowpeas is poorly understood. Hence, the response of cowpeas to poultry manure, *Bacillus subtilis* inoculant, and single super phosphate (SSP) fertilizer was investigated during the 2020 and 2021 cropping seasons.

Methodology: Treatments comprised two cowpea cultivars (FUAMPEA-2 and Ife Brown) and three soil amendments: poultry manure, *B. subtilis*, SSP fertilizer, and unamended soil, which served as the control. The factorial experiment had eight treatments arranged in a completely randomized design and was replicated six times. Data on collected on leaves, leaf area, stem diameter, yield components, and proximate composition were analyzed with ANOVA and means separated with LSD at $P < 0.05$.

Main Results: The number of leaves (26.58 ± 18.08), leaf area (5,058 ± 3,472 cm^2), and stem diameter (5.61 ± 2.30 mm) of Ife Brown improved significantly on soil augmented with poultry manure. On the contrary, FUAMPEA-2 had a higher number of seeds (33.33 ± 18.97), grain yield/plant (48.23 ± 44.21 g), and harvest index (0.59 ± 0.58) on soil amended with poultry manure. Crude protein (34.2%), crude fat (6.0%), and seed P content of 10.75 g/kg were highest in FUAMPEA-2 sown on soil fertilized with SSP. There was an improvement in essential mineral nutrients in soil supplied with organic fertilizer relative to nutrients recorded on unfertilized soil.

Conclusions: The augmentation of marginal soil with poultry manure enhanced the growth and grain yield of FUAMPEA-2 remarkably while improving biomass accumulation in Ife Brown.

Keywords: Phosphorous augmentation, cowpea cultivars, biofertilizer, cowpea growth and biomass accumulation

Thai J. Agric. Sci. (2023) Vol. 56(4): 248−274

INTRODUCTION

Cowpea (*Vigna unguiculata* (L.) Walp.) has proven over time to be indispensable in the quest for ensuring food and nutritional security (Moreno *et al*., 2022). Cowpea is a good source of plant protein and a better source of income for numerous smallholders in many emerging economies in the tropics and sub-Saharan regions (Rawal and Navarro, 2019). Cultivation of the crop cut across semi-arid tropical regions of Africa, Asia, Europe, the United States, and Central and South America (Fatokun *et al.*, 2002). It is a major legume grown in the West and Central African countries (Adeoye *et al*., 2011).

Ogaraku (2007) reported that the young succulent leaves of cowpea plants are eaten as vegetables, which forms an important source of cheap vegetables containing about 25% protein. Sha and Xiong (2020) and Kołodziejczak *et al*. (2022) have reported the substitution of animal protein with vegetable protein as a meat analog, and this has increased the demand for legumes globally (Maphosa and Jideani, 2017). Cowpea adapts very well to different soil types, has great tolerance to drought, and its cultivation improves soil fertility (Simion, 2018).

Nigeria is the largest cowpeas consumer, accounting for 61% of the production in Africa and 58% worldwide (Nkomo *et al*., 2021). Despite this production level, yields remain low compared to average yields of about 450 kg/ha in other cowpea-growing regions (Kebede and Bekeko, 2020). The low yield has been linked to inherently low soil nutrients, particularly phosphorus (P; Haruna, 2011). The rate of P loss is estimated at an average of 4–19 kg/ha/year (Alewell *et al*., 2020). Phosphorus is a vital macronutrient, highly required by cowpeas for optimum growth and development (Wortman *et al*., 2017). It is essential for cowpeas' foliage production, pod formation, and N-fixation (Abbadi and Gerendás, 2015). Despite the irreplaceability of P in cowpea physiology and crops generally, the available P levels in most Nigerian soil are abysmally low (Tsado *et al*., 2012). The range of available P in most Nigerian soil is between 3.0–35 mg/kg (Shehu *et al*., 2015), while the critical P requirement for optimal cowpea growth is taken as 7.0 mg/kg according to Aune and Lal (1995) and validated by Karikari *et al*. (2015). One thing is for nutrients to be available; another is for such nutrients to be released for crop uptake. Phosphorus is known for its high fixation, especially in soil like Alfisols, Oxisols, and Ultisols with low clay activity. This soil is highly weathered and predominant in the tropics (Pierzynski and Hettiarachchi, 2018; Rawal *et al*., 2022). Soils in cowpea growing regions of Nigeria, such as Southern Guinea Savannah and Derived Savannah, are characteristically low in P content as most of this soil belongs to Alfisol. This made P deficiency one of the most limiting factors in cowpea production. Knowledge of P management in the cowpea field is essential in ensuring that P needs are supplied at specific phenology to realize the potential yield.

Mineral fertilizer is important in increasing soil fertility and crop yield due to its ease of mineralization and availability (Sánchez-Navarro *et al*., 2021). However, applying mineral fertilizer causes undesirable environmental challenges, reducing crop yield (Verzeaux *et al*., 2016). In addition, inadequate supply, adulteration, and high cost are prominent challenges faced by cowpea farmers in Nigeria (Adekiya and Agbede, 2009). Therefore, the need for alternative and sustainable methods of improving soil fertility without compromising environmental purity cannot be underscored.

Organic manures are known for improving soil physical, chemical, and biological properties, thus, stimulating better crop growth and higher yield (Nwaiwu *et al*., 2010). According to Adeoye *et al*. (2011), the beneficial effects of animal manure on soil physical properties and the ease with which they decompose in the soil are major advantages over inorganic fertilizers. Organic fertilizers significantly impact crops as they supply essential nutrients required for optimizing the performance of poor soil (Kutu *et al*., 2019). However, the nutrients embedded in organic manure are not readily available to crops but require a series of microbial activities for mineralization (Sánchez-Navarro *et al*., 2021). Chicken manure constitutes a large chunk of waste generated in the poultry industry to the tune of 93.25 metric tonnes per annum in Nigeria (Waziri and Kaltungo, 2017). The menace that accompanies waste generated from poultry could be appropriated through recycling as fertilizer. Poultry manure has been reported to contain essential plant nutrients like N, P, K, and many micronutrients (Bolan *et al*., 2010). The general effect of chicken manure on crop growth has been widely reported. However, the specific effect of poultry manure on P uptake and proximate composition of cowpea cultivars has not been adequately documented. The need to elucidate this influence is germane for optimizing P uptake and utilization in improving grain yield in cowpeas.

The utilization of numerous soil-inhabiting microorganisms to boost plant mineral nutrient uptake has been well-reported (Dada *et al*., 2020). These microbes belong to different families and species, playing different roles in improving soil fertility status (Bashan *et al*., 2014). Their use as biofertilizers in improving quality of soil and crop yield has been well amplified by several studies (Bhattacharjee and Dey, 2014; Egamberdieva and Adesemoye, 2016). Microbes belonging to the rhizobacteria group, including *Bacillus* species, are beneficial to cowpeas as fertilizers (biofertilizers). Reports have shown that *Bacillus* spp. converts the complex form of essential nutrients, such as P and N, to a simple available form for uptake by plant roots (Kang *et al.,* 2014; Kuan *et al*., 2016). Phosphatases and organic acids secretion from *Bacillus* spp. are known to acidify the surrounding environment, thus facilitating the conversion of inorganic phosphate into free phosphate (Hayat *et al.,* 2010; Kang *et al.,* 2014). However, it remains unknown if the use of *Bacillus* spp. can improve phosphate availability on sandy-loam soil thereby enhancing cowpea yield in comparison to yield produced on soils amended with inorganic fertilizer.

There is a dearth of information on the effect of organic or inorganic fertilizers applied based on their P constituents and biofertilizers in improving P_2O_5 availability on sandy loam and its uptake by cowpeas. Therefore, the performance of two cowpea cultivars in response to poultry manure, inorganic and *Bacillus subtilis,* applied to sandy loam soil in Ibadan was investigated. The need to establish the comparative effectiveness of these soil amendments in improving growth, yield attributes, and agronomic efficiency is critical to increasing cowpea production in the region.

MATERIALS AND METHODS

Experimental Site

The pot experiment was conducted at the rooftop garden of the Crop Protection and Environmental Biology (CPEB) Department (7°27’02”N 3°53’49”E, 234 m above sea level), University of Ibadan, Nigeria, during the 2021 and 2022 early cropping seasons. The average precipitation, temperature, and relative humidity of the study site were 999.9 mm, 33°C, and 71%, respectively. Pots of 18.6 cm diameter and 19 cm depth were filled with 5 kg of soil each.

Physical and Chemical Analysis of Soil and Poultry Manure

The soil used for the study was collected at a depth of 0–30 cm from the Crop Garden of the Department of CPEB using a spade. The soil was put into plastic bags. The soil sample was sterilized in a dry heat soil sterilizer (COX-115V-N, COX-220V-N) for three hours. Soil samples were air-dried and sieved with a 2 mm mesh sieve. The total organic carbon was evaluated by the wet oxidation method of Walkley (1935), while the total N was determined using the Kjeldahl method (Jackson, 2005). The available P was evaluated using the Olsen method (Schoenau and O’Halloran, 2007). The exchangeable K was estimated by the ammonium acetate extraction method (Jackson, 2005). After equilibration in distilled water, the pH of the soil was determined using a pH meter (H_2O, 1:5). The routine analysis of the soil was performed following standard soil analytical methods described by IITA (1990). Cations were determined by flame photometer and atomic absorption spectrophotometer (AAS) following the methods described by IITA (1990). Five grams of soil were made to pass through a 2 mm sieve, weighed, and 30 mL of 1 N NH_4OAC was added and shaken mechanically for 2 hours. The solution was centrifuged at 2,000 rpm for 5 minutes. The clear supernatant was carefully decanted into a 100 mL volumetric flask. This last step was repeated and transferred into the same volumetric flask. The solution was made up of NH_4OAC solution. The K and Na were determined by flame photometer, while Mn, Fe, Cu, Zn, Ca, and Mg were determined by the flame AAS.

Poultry manure collected from the Teaching and Research Farm at the University of Ibadan was analyzed for its nutrient content before it was applied. Total organic carbon was determined after oxidization with potassium dichromate following the methods of Nelson and Sommers (1996). The total K was measured with a Jenway Flame Photometer (Bibby Scientific Ltd-Stone-Staffs-St15 0SA–UK). The P was estimated by a spectrophotometer as described by Kuo (1996), and total N by the micro Kjeldahl method (Fisher, 1965). The results of both soil and poultry manure analysis are presented in Table 1.

Table 1 Results of pre-trial soil and poultry manure analysis

Parameters	Soil	Poultry manure
Soil particle analysis		
Sand (g/kg)	80.5	na
Silt (g/kg)	12.0	na
Clay (g/kg)	7.5	na
Textural class	Sandy loam	na
pH (H_20)	6.6	8.0
Organic carbon (g/kg)	8.1	59.9
Total nitrogen (g/kg)	1.0	9.8
Exchangeable acidity (cmol/kg)	0.4	nd
Available phosphorous (mg/kg)	30.8	8.9
Exchangeable cation (cmol/kg)		
Ca	3.8	30.0
Mg	1.2	9.1
K	0.2	6.8
Na	0.4	1.9
Extractable micronutrients (mg/kg)		
Mn	83.7	0.3
Fe	13.7	2.8
Cu	3.1	0.2
Zn	12.8	0.3

Note: na = not applicable, nd = not determined.

Plant Materials

Two cowpea cultivars sown were FUAMPEA-2 and Ife Brown, selected based on their growth type and maturity time. FUAMPEA-2 is an erect and early maturing cultivar, while Ife Brown has a semi-erect/creeping growth pattern and medium maturing. FUAMPEA-2 seeds were obtained from an accredited seed outlet, while Ife

Brown seeds were obtained from the Institute of Agricultural Research and Training (IAR&T), both in Ibadan, Nigeria.

Seed Inoculation with *B. subtilis*

B. subtilis LK14 was cultured in the Phytopathology Laboratory of the CPEB Department using a soil sample from the Crop Garden and locust bean seeds. A reference culture of the bacterium was obtained from the Department of Microbiology, University of Ibadan, to validate the identity of the inoculum cultured. The bacterium was sub-cultured in Petri dishes using nutrient agar as the culture medium. One gram (1×10^{10} CFU/mL) of the *B. subtilis* LK14 inoculum was used to inoculate 30 seeds (approximately 16 g) from each variety following the method described by Burton (1984).

Application of Poultry Manure and Single Super Phosphate Fertilizer

In each pot containing 5 kg of soil, poultry manure was applied based on P concentration (16.9 g/5 kg soil ≡ 60 kg P_2O_5/ha). The soil and poultry manure mixture was left for a week to aid mineralization before seeds were sown. Single super phosphate (SSP) was applied as a side-banding immediately after the seeds were sown at the recommended rate of 60 kg P_2O_5/ha. The recommended P rate for growing cowpea on many soil types found in the cowpea growing regions in Nigeria is 60 kg P_2O_5/ha. The quantity applied was based on the expression:

$$\frac{\text{Weight of soil}}{2 \times 10^6} \times \frac{\text{Conc. of P}}{\text{Rate of P}} \times 100 \times 1{,}000 \text{ g}$$

Sowing, Management, and Pest Control

Two seeds were sown into the prepared pots at a depth of 1–2 cm. Seven days after emergence, the seedlings were thinned to one stand per pot. A soil moisture meter (LMS-714) was used to determine the moisture level to ascertain irrigation needs whenever the moisture level was low (≤ -0.5 MPa). Weeding was done manually using hands to remove any weeds sighted in the pots. Insect pests were controlled by spraying Lamda-cyhalothrin at the rate of 2 mL/liter of water. The mixture was sprayed regularly with a portable hand sprayer to control insect pests. Insect pests were managed every other week.

Treatments and Experimental Design

The 2 × 4 factorial experiment comprised 8 treatments, including 2 cowpea varieties and 3 soil amendments with a control (unamended): poultry manure, SSP, and *B. subtilis* inoculum, while unamended soil served as control. The eight treatments were arranged in a completely randomized design (CRD) in six replicates. A total of 48 pots were used for this study. The trial was conducted twice during the growing seasons of 2020 and 2021.

Data Collection

Data were collected on growth parameters, yield, and its components, as well as biomass accumulation, following standard procedures.

Measurement of Growth, Yield, and Yield Components

The period of data collection covered vegetative, flowering, pod formation, and physiological maturity stages. Growth parameters such as plant height were measured with the meter rule from the soil level to the tip of the fully formed leaf at the plant apex. Leaf area was determined using a hand-held leaf area meter (LI-3000C), while stem diameter was measured with an vernier caliper, and the number of leaves and branches was assessed by counting. At the final harvest, the number of nodules formed was detached, counted, and weighed on a sensitive balance.

The days to flowering were taken as the number of days from sowing to the emergence of the first flower. The number of flowers formed per plant was counted. Pod parameters like the number of pods, length of pods, weight of dried pods, and the number of unfilled pods were determined following standard procedures. The pods were shelled to determine the number of seeds per plant. The yield was determined by weighing the cumulative grains yield from shelled pods per plant.

At the final harvest, shoot, root, and total biomass accumulation were determined. Plants were carefully uprooted and washed under running water. Afterward, distilled water was used to decontaminate the plants and then dried with a paper towel. The samples were separated into shoot and root parts and were oven-dried at 80°C for 48 hours to attain constant weight. The dried weight of the shoot, root, and total dry samples were determined using a balance. The harvest index was evaluated as a ratio of economic yield to the total biological yield (Amanullah and Inamullah, 2016).

Determination of Seed Phosphorus Content

Pods were harvested and sun-dried under the open shed for seven days. The dried pods were kept in a paper bag and shelled by breaking the pods with a club. Seeds were collected and taken for P determination. The P content in cowpea seeds was determined using the vanado-molybdophosphoric colorimetric method after nitric-perchloric acid digestion, as described by Poitevin (2016).

Proximate Composition and Seed Phosphorous Uptake

At harvest, shelled seeds were processed for proximate analysis using standard procedures described by Poitevin (2016). The sample of the seeds was dried at 70°C for 72 hours using a Gallenkamp oven (300 plus series) to obtain a constant weight. The percentage moisture content of the seed was evaluated from the dried seeds as follows:

$$\% \text{ Moisture} = 1 - \frac{\text{Weight of dry sample}}{\text{Weight of wet sample}} \times 100$$

The Soxtec method was adopted to determine the crude fat concentration in the seed extracted using petroleum ether, and the SoxtecTM 2050 automated analyzer (FOSS Analytical, Hillerød Denmark) was used following the method of Noureddini and Byun (2010). The crude fat (%) was estimated as follows:

$$\% \text{ Crude fat} = \frac{\text{Weight (extraction cup + residue)} - \text{Weight (extraction cup)}}{\text{Weight of sample}} \times 100$$

The protein concentration was calculated by determining the amount of total N in the seeds using the KjeltecTM 2200 Auto Distillation Unit. The amount of total N was used to multiply N-to- the protein factor of 4.4 to estimate the protein quantity of the seeds, according to Ng *et al.* (2008). Also, the ash content was determined by dry ashing the samples in a Muffle furnace (Muffle furnaces series 642, JBP003 model 642/8, RS-France) at 600°C for 6 hours. The sample was then cooled and weighted. The P concentration in the seed was determined using a NOVA 400 atomic absorption spectrometer (Analytik Jena AG, Jena, Germany) with an air/acetylene flame, and respective hollow-cathode lamps were used for absorbance measurements. The wavelength was calibrated at 766.5 nm, slit at 0.8 nm, and lamp at 4.0 mA to determine the P elements in the seed. The results for mineral contents were expressed as g/kg dry weight.

Post-trial Soil Analysis

After the plants had been uprooted, soil samples were collected per treatment and analyzed for residual mineral nutrients. The routine post-trial analysis and available P content in the soil were performed following standard soil analytical methods of IITA (1990).

Data Analysis

Analysis of variance (ANOVA) of the pooled data sets collected during the two seasons was performed using the general linear model of the Statistical Analysis System (SAS, version 9.1). The differences in means were separated with the least significant difference (LSD) at $P < 0.05$.

RESULTS AND DISCUSSION

Effect of Poultry Manure, Single Super Phosphate Fertilizer and *B. subtilis* Inoculant on Growth Parameters of Two Cowpea Cultivars

The responses of the two cowpea cultivars to different soil amendments applied based on P content are indicated in Figures 1–2. The results showed that Ife Brown formed more leaves than FUAMPEA-2 (Figure 1A). Also, the response of the two cowpea cultivars to different amendments showed that the two cultivars were statistically ($P > 0.05$) similar with respect to leaf area at the early growth stage and flowering. However, at pod formation and maturity phases, the leaf area of FUAMPEA-2 was significantly higher than that of Ife Brown (Figure 1B). The height of the two cultivars was not significantly different at the active vegetative stage. Nonetheless, FUMPEA-2 was significantly ($P < 0.05$) taller than Ife Brown at reproductive and maturity stages (Figure 1C). There was no significant difference in the stem diameter of the two cultivars as shown in Figure 1D.

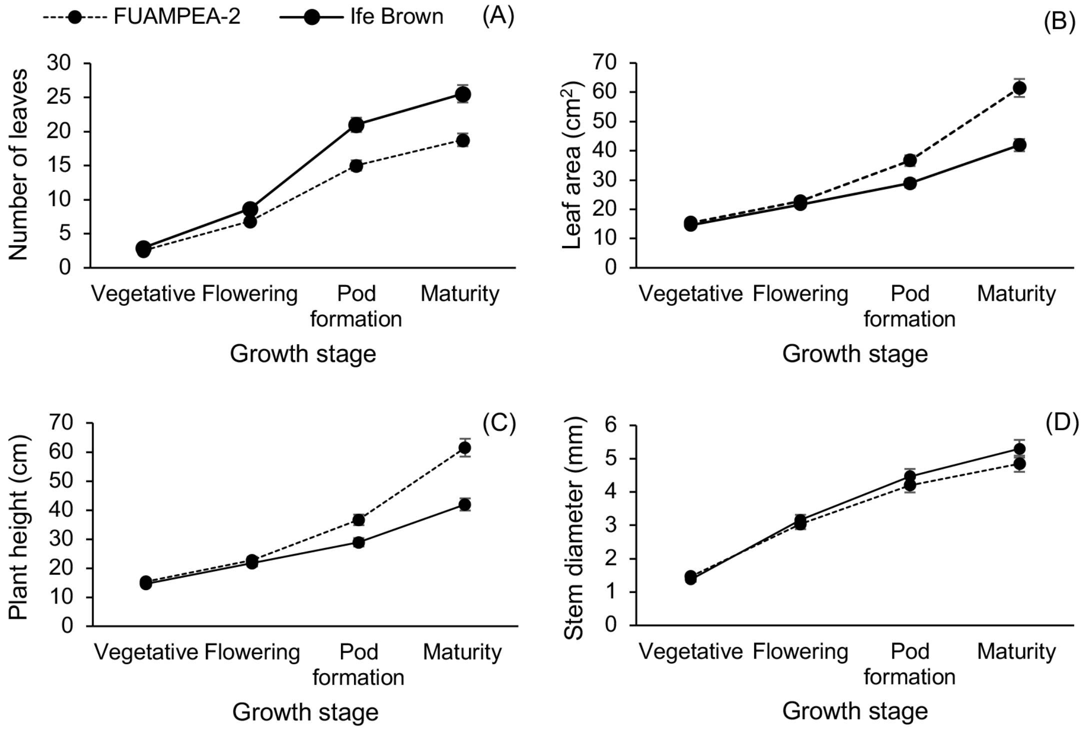

Figure 1 Growth responses of two cowpea cultivars to soil amendments: number of leaves (A), leaf area (B), plant height (C), and stem diameter (D).

The effect of different soil amendments on leaf formation at the vegetative stage showed that cowpeas produced more leaves under *B. subtilis* treatment than under other treatments. However, at the flowering and pod formation stages, leaf formation increased tremendously in soil amended with poultry manure relative to other amendments. The highest leaves formed were recorded in plants grown on soil fertilized with poultry manure, which was 15%, 7.8%, and 5.0% higher than leaves formed in *B. subtilis* treatment, SSP, and unamended soil, respectively, at the maturity stage (Figure 2A).

The effect of different amendments on the leaf area showed that *B. subtilis* treatment enhanced the leaf area more than other treatments at the early growth stage. As the crop grew further, leaf area was higher in soil amended with poultry manure at flowering (1,966.30 cm^2) and pod forming (5,205.30 cm^2) stages. At the final harvest, leaf area was not affected by the amendments but was highest (4,636.30 cm^2) in soil fertilized with poultry manure (Figure 2B). Cowpea plants were taller under *B. subtilis* treatment at vegetative and reproductive phases. Nevertheless, at the pod-forming stage, the tallest plant (34.69 cm) was observed in soil augmented with poultry manure, but this was not statistically taller (30.92 cm) than the height of the plant treated with *B. subtilis*. At the final harvest, different soil amendments had no significant effect on the height of the cowpea but was taller (55.73 cm) on soil augmented with poultry manure than on soil treated with other amendments (Figure 2C).

Wider stem diameter was recorded at the early growth stage in *B. subtilis* treatment compared to the other soil amendment treatments. Beyond the vegetative stage, the amendments did not affect the diameter of cowpea stem differently. Nonetheless, at the final harvest, cowpeas had wider stem diameter (5.42 mm) on soil amended with poultry manure, while the thinnest stem diameter (4.83 mm) was recorded in cowpeas sown on unamended soil (Figure 2D).

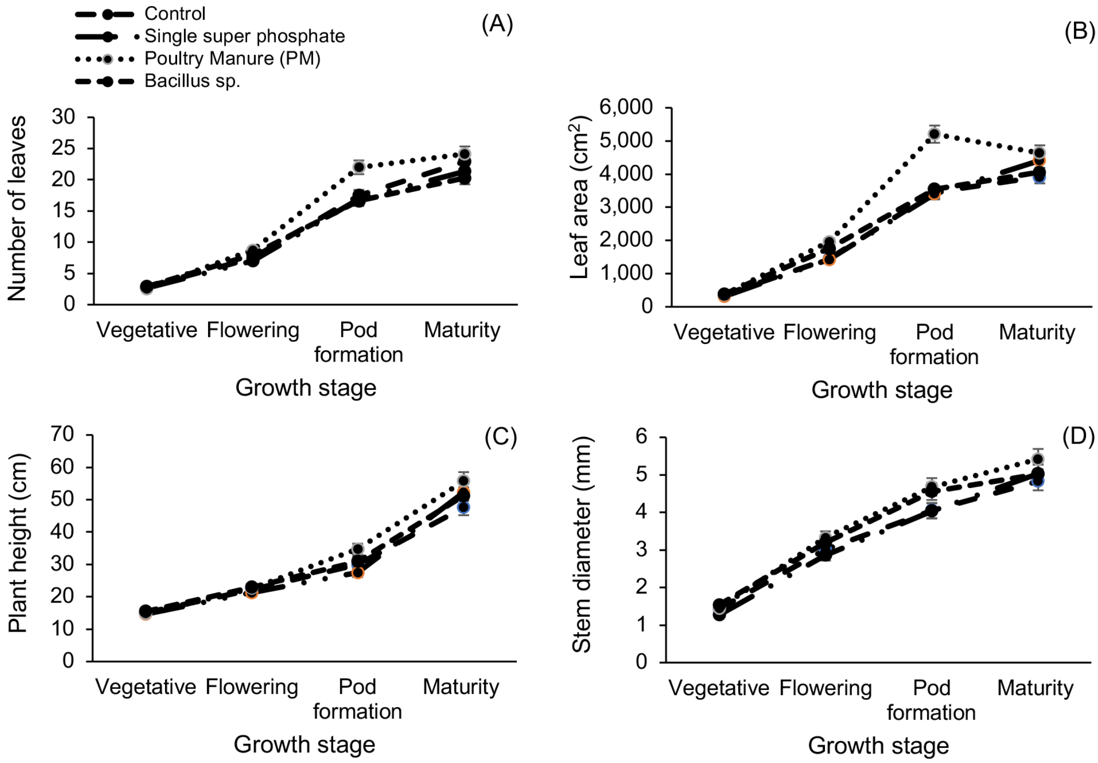

Figure 2 Effect of different soil amendments on the growth of cowpeas: number of leaves (A), leaf area (B), plant height (C), and stem diameter (D).

The interaction between cowpea cultivars and different soil amendments, including *B. subtilis* treatment, indicated that Ife Brown produced more leaves in *B. subtilis* treated soil than FUAMPEA-2 on soil treated with SPP or poultry manure at the vegetative stage (Table 2). However, leaf formation in Ife Brown at flowering and pod-forming stages was highest on soil amended with poultry manure relative to leaves produced by FUAMPEA-2 sown on unamended soil. At maturity, the number of leaves produced by either Ife Brown or FUMPEA-2 was not significantly affected by the applied soil amendments as shown in Table 2.

The interaction between cultivars and different soil amendments had a significant effect on leaf area. FUAMPEA-2 had the highest leaf area under *B. subtilis* treatment at the vegetative phase. Subsequently, Ife Brown grown on soil fertilized with poultry manure had a leaf area, which was significantly higher than the leaf area of FUAMPEA-2 under *B. subtilis* treatment, as indicated in Table 2.

The interaction between the two cultivars and different soil amendments did not influence the diameter of the cowpea stem differently. Similarly, the interaction between the two cultivars and different soil amendments did not influence the height of the cowpea cultivars differently at vegetative and flowering stages. As the growth progressed, specifically at pod forming stage, FUAMPEA-2 became taller (35.33 cm) than Ife Brown in soil amended with poultry manure, but the difference was not significant (Table 3).

Ife Brown was superior to FUAMPEA-2 regarding foliage development, and this could be attributed to the genetic variations in the two cowpea cultivars in response to the amendment applied to the marginal soil. Environmental modification in terms of variation in plant mineral nutrient availability and abundance in soil may also be responsible for differences recorded in the two cultivars. This agrees with the submission of Ndor *et al*. (2012) and Animasaun *et al*. (2015), who ascribed significant variations in growth and yield characteristics of some improved cowpea varieties to their genetic makeup and environmental factors like soil available nutrients especially P. In the same vein, the improved performance of Ife Brown with respect to growth indices at the active vegetative and reproductive stages over FUAMPEA-2 in soil amended with poultry manure suggests that poultry manure is likely to have supplied adequate nutrients, which were better utilized by this cultivar for biomass accumulation than experienced in the other cultivar and soil amendments. Efficient utilization of any applied amendment is often linked to the potential of the crop to convert the applied materials to usable forms for the metabolic process. Hence, the improvement recorded in growth traits of Ife Brown over FUAMPEA-2 suggests that Ife Brown converts the minerals supplied by the amendment for morphological or structural development better than FUAMPEA-2. This observed improvement in structural development at these growth stages could be connected to the availability of essential minerals in sufficient quantities necessary to enhance cowpea growth and development, as reported by Reyhan and Amiraslani (2006) and Mohammed *et al*. (2020).

Ife Brown formed more branches (2.38) than FUAMPEA, while FUAMPEA-2 had a higher number of flowers (12.06) and flowered earlier (55.10 days) than Ife Brown. The highest number of branches and flowers were observed in plants grown on soil augmented with poultry manure relative to those on unamended soil with the lowest number of branches and flowers. Different soil amendments did not influence the days to flowering by the crop differently (Table 4).

The number of branches and flowers and days of flowering were significantly affected by the interaction between cowpea cultivars and the soil amendments (Table 4). The Ife Brown had the highest number of branches on soil fertilized with poultry manure, but this was statistically similar to the number of branches formed by Ife Brown under *B. subtilis* treatment. FUAMPEA-2 formed the highest number of flowers (16.67) on soil fertilized with poultry manure and flowered earlier (52.50 days) under *B. subtilis* treatment, as shown in Table 4.

The Ife Brown sown on soil fertilized with poultry manure branched profusely contrary to the extent of branching observed in FUAMPEA-2. This suggests that Ife Brown is more efficient in absorbing the released nutrients, particularly N, known to enhance branching and phosphorous, which typically promotes reproductive development in crops. The observation is similar to the findings of Agbede *et al*. (2008), who found that the number of branches in plants appreciably increased under an adequate quantity of nutrients supplied by poultry manure. Poultry manure improved branching and flower production, while *B. subtilis* promoted early flowering makes both; a suitable soil fertility resources in low-income and organic farming systems. Cowpea is a cheap source of plant protein among the poor resource community. Hence, its production using available, cheap, and easily accessible inputs will go a long way to increasing its production, thus boosting food availability and food security.

The basis for the increased height experienced in FUAMPEA-2 over Ife Brown on soil fertilized with poultry manure could be linked to the growth type of the crop. FUAMPEA-2 is an indeterminate cultivar, while Ife Brown exhibited a determinate growth pattern. Hence, it is unlikely that FUAMPEA-2 had a longer assimilatory capacity than Ife Brown. This view is similar to that of Chaturvedi *et al*. (1980), who linked longer growth habits to better assimilatory potential. Ikram *et al*. (2012) pointed out that the application of poultry manure caused an improvement in the height of arable crops.

It appeared that the effect of *B. subtilis* on the height of cowpea cultivars was comparable to that of poultry manure. This implies that both amendments probably have some components that induce stem elongation in cowpeas. Stem elongation is a measure of growth in plants as influenced by some mineral elements like N as well as P that contributes to the synthesis of certain growth hormones like auxin or gibberellins. These hormones are known to influence cell division and multiplication, thus stimulating stem elongation, as reported by Adedeji *et al*. (2020). Examining in detail the main component in these materials that causes induction of stem extension is very necessary.

Early flowering observed in FUAMPEA-2 is genetically related, as this cultivar is an early maturing genotype contrary to Ife Brown, a medium maturing genotype. This clearly explained why FUAMPEA-2 flowered earlier than Ife Brown (Ubah *et al*., 2012). The early flowering may have played a significant function in ensuring that photo-assimilates were partitioned over a longer period thus, compensating for indeterminate growth habit. The assimilatory capacity of the crop could have been enhanced such that early flowering gets synchronized with a longer growth period, causing the sink to benefit from the prolonged biomass synthesis from the source (Chaturvedi *et al*., 1980; Owusu *et al*., 2018). However, the relationship between indeterminate growth patterns and early flowering in cowpea genotypes across different agroecologies needs to be clearly elucidated.

Table 2 Effect of interaction between cultivar and soil amendment on number of leaves and leaf area of cowpea

Cultivars	Soil amendment	Number of leaves				Leaf area (cm^2)			
		VEG	FLR	PDF	MAT	VEG	FLR	PDF	MAT
FUAMPEA-2	Control	2.58 ± 0.67^{b}	6.83 ± 1.47^{c}	16.00 ± 6.13^{bc}	19.25 ± 9.04^{ab}	385 ± 234	$1,352 \pm 519^{c}$	$3,305 \pm 1,480^{b}$	3,980 ± 2,184
	SSP	2.50 ± 0.52^{b}	6.67 ± 1.56^{c}	13.33 ± 3.17^{c}	17.33 ± 5.41^{ab}	345 ± 180	$1,425 \pm 527^{c}$	$3,018 \pm 965^{b}$	3,767 ± 1,350
	PM	2.50 ± 1.00^{b}	7.25 ± 2.42^{c}	18.08 ± 7.82^{bc}	23.00 ± 9.08^{ab}	389 ± 278	$1,578 \pm 787^{c}$	$4,495 \pm 2,873^{ab}$	4,766 ± 2,507
	B. subtilis	2.58 ± 0.90^{ab}	6.75 ± 1.71^{c}	12.67 ± 4.29^{c}	15.58 ± 5.12^{b}	399 ± 308	$1,395 \pm 538^{c}$	$2,740 \pm 1,116^{b}$	3,159 ± 1,399
Ife Brown	Control	3.17 ± 0.72^{a}	8.00 ± 1.86^{bc}	19.00 ± 6.06^{bc}	25.25 ± 11.34^{a}	342 ± 156	$1,554 \pm 544^{bc}$	$3,598 \pm 1,397^{b}$	5,058 ± 2,873
	SSP	2.83 ± 0.58^{ab}	7.33 ± 1.15^{bc}	18.42 ± 6.41^{bc}	25.33 ± 12.09^{a}	280 ± 101	$1,439 \pm 531^{c}$	$3,802 \pm 1,890^{b}$	5,056 ± 2,912
	PM	2.67 ± 1.07^{ab}	10.17 ± 5.11^{a}	25.92 ± 15.34^{a}	26.58 ± 18.08^{a}	353 ± 254	$2,354 \pm 1,692^{a}$	$5,916 \pm 4,122^{a}$	5,058 ± 3,472
	B. subtilis	3.17 ± 0.83^{a}	9.33 ± 2.57^{ab}	20.58 ± 7.45^{ab}	25.00 ± 15.05^{a}	364 ± 183	$2,129 \pm 792^{ab}$	$4,365 \pm 1,978^{ab}$	4,967 ± 3,095
LSD		0.66	2.05	6.39	9.29	179.62^{ns}	674.09	1,792.60	$2,092.20^{ns}$

Note: ^{a,b,c} Means in the same column with different superscripts differ significantly ($P < 0.05$). LSD = least significant difference at $P < 0.05$, ns = not significant. SSP = single super phosphate fertilizer, PM = poultry manure, VEG = vegetative phase, FLR = flowering phase, PDF = pod formation phase, Mat = maturity phase.

Table 3 Effect of interaction between cultivar and soil amendments on stem diameter and height of cowpea

Cultivar	Soil amendment	Stem diameter (mm)				Plant height (cm)			
		VEG	FLR	PDF	MAT	VEG	FLR	PDF	MAT
FUAMPEA-2	Control	1.40 ± 0.56	2.82 ± 0.90	3.86 ± 0.59	4.60 ± 0.80	15.49 ± 2.60	22.75 ± 3.14	34.10 ± 8.59	56.66 ± 30.41
	SSP	1.30 ± 0.41	2.88 ± 0.97	3.99 ± 0.89	4.70 ± 0.93	15.38 ± 2.48	21.41 ± 3.60	28.58 ± 2.79	64.18 ± 36.05
	PM	1.55 ± 0.38	3.26 ± 1.19	4.48 ± 0.85	5.22 ± 0.78	15.28 ± 3.36	23.78 ± 4.35	35.33 ± 8.52	65.33 ± 27.39
	B. subtilis	1.61 ± 0.78	3.20 ± 1.19	4.46 ± 1.43	4.88 ± 1.16	15.31 ± 3.96	23.03 ± 3.49	32.50 ± 6.82	59.75 ± 30.54
Ife Brown	Control	1.45 ± 0.59	3.20 ± 1.09	4.25 ± 1.20	5.07 ± 1.44	14.63 ± 1.33	21.48 ± 2.10	25.83 ± 3.03	38.50 ± 20.21
	SSP	1.24 ± 0.30	2.85 ± 0.84	4.10 ± 0.92	5.36 ± 1.54	13.93 ± 1.94	21.02 ± 2.37	26.38 ± 1.92	40.33 ± 30.39
	PM	1.36 ± 0.40	3.40 ± 1.62	4.88 ± 2.34	5.61 ± 2.30	14.23 ± 3.68	21.36 ± 5.00	34.05 ± 18.26	46.13 ± 31.50
	B. subtilis	1.47 ± 0.49	3.19 ± 0.98	4.67 ± 1.11	5.17 ± 0.78	15.60 ± 2.36	22.81 ± 2.53	29.33 ± 6.57	42.75 ± 24.67
LSD		0.41^{ns}	0.91^{ns}	1.03^{ns}	1.07^{ns}	2.31^{ns}	2.80^{ns}	6.97^{ns}	23.72^{ns}

Note: LSD = least significant difference at $P < 0.05$, ns = not significant, SSP = single super phosphate fertilizer, PM = poultry manure, VEG = vegetative phase, FLR = flowering phase, PDF = pod formation phase, Mat = maturity phase.

Table 4 Effect of different soil amendments on number of branches, number of flowers, and days to flowering of two cowpea cultivars

Factors		Number of branches	Number of flowers	Days to flowering
Cultivar				
FUAMPEA-2		1.23 ± 1.31^{b}	12.06 ± 7.14^{a}	55.10 ± 12.17^{b}
Ife Brown		2.38 ± 1.58^{a}	8.52 ± 11.48^{b}	60.94 ± 11.48^{a}
LSD		0.57	2.44	4.87
Soil amendment				
Control		1.42 ± 1.49^{b}	8.46 ± 3.76	57.54 ± 13.43
SSP		1.54 ± 1.28^{b}	10.04 ± 6.12	59.04 ± 10.18
PM		2.54 ± 1.89^{a}	11.83 ± 9.25	58.00 ± 11.64
B. subtilis		1.71 ± 1.42^{b}	10.83 ± 4.32	57.42 ± 13.63
LSD		0.80	3.44^{ns}	6.88^{ns}
Cultivar × Soil amendment				
FUAMPEA-2	Control	1.50 ± 1.50^{bcd}	9.58 ± 3.34^{b}	57.83 ± 16.40^{ab}
	SSP	0.92 ± 0.90^{cd}	10.92 ± 5.38^{b}	55.67 ± 7.60^{ab}
	PM	1.83 ± 1.59^{bc}	16.67 ± 10.88^{a}	54.42 ± 13.56^{ab}
	B. subtilis	0.67 ± 0.89^{d}	11.08 ± 5.21^{b}	52.50 ± 10.25^{b}
Ife Brown	Control	1.92 ± 1.50^{bc}	7.33 ± 3.96^{b}	57.25 ± 10.38^{ab}
	SSP	2.17 ± 1.34^{ab}	9.17 ± 6.90^{b}	62.42 ± 11.58^{a}
	PM	3.25 ± 1.96^{a}	7.00 ± 3.07^{b}	61.75 ± 8.39^{ab}
	B. subtilis	2.17 ± 1.27^{a}	10.58 ± 3.42^{b}	62.33 ± 15.19^{a}
LSD		1.14	4.72	9.77

Note: ^{a,b,c,d} Means in the same column with different superscripts differ significantly ($P < 0.05$). LSD = least significant difference at $P < 0.05$, ns = not significant, SSP = single super phosphate fertilizer, PM = poultry manure.

B. subtilis inoculation improved cowpea growth at the earlier growth stage, but the influence declined as the crop progressed. Li and Hu (2020) have shown that *B. subtilis* promotes growth at the early stage of plant life. The improved growth performance of cowpeas on *B. subtilis* treatment may be linked to some soil factors that encouraged survival and colonization of *B. subtilis.* This observation is similar to that of Adhya *et al*. (2015), who listed soil environment as one of the factors that influence the efficacy of rhizospheric microbes in solubilizing unavailable P. This comparable performance recorded in *B. subtilis* inoculated treatment agrees with the earlier reports that inoculation of these microbes improved solubilization, availability, and uptake of fixed P, which ultimately improved cowpea performance. At the early stage, *B. subtilis* was more efficient in improving the growth indices of the cowpea cultivars than other treatments, especially soil fertilized with SSP. Therefore, to stimulate growth at the early stage, it would not be out of place if *B. subtilis* is added to complement some soil amendments that have low P mineralization potential. The need to evaluate the P mineralization potentials of diverse organic fertilizers on the soil of cowpea growing regions is very pertinent.

FUAMPEA-2 grown on poultry manure-amended soil performed better than Ife Brown in terms of flowering, pod formation, and other yield components. One of the attributes of FUAMPEA-2 over Ife Brown was the superior yield performance, which could be attributed to its ability to utilize minimal nutrients for greater yield (Omoigui *et al*., 2018). This is most probably because this cowpea line was bred for high grain yield and yield components potential under minimum fertilizer input. This agrees with the submission of Owusu *et al*. (2018), who ascribed the significant variation in yield characteristics of some improved cowpea varieties to growth habits, maturity indices, and yield components. Also, Owusu *et al*. (2018) reported that flowering, pod, and seed characteristics contribute significantly to discriminating genetic diversity among cowpea cultivars. Perhaps this sufficed to buttress the basis for the increased yield observed in FUAMPEA-2.

Effect of Poultry Manure, Single Super Phosphate and *B. subtilis* Inoculant on Yield Components of Two Cowpea Cultivars

The response of the two cultivars to different soil amendments showed that there was no significant difference in the cowpea yield attributes such as number of pods, unfilled pods, weight of pods, and length of pods (Table 5). Nevertheless, different soil amendments exerted significant effects on the number of pods, unfilled pods, and weight of pods produced by cowpea cultivars. Cowpea grown on soil fertilized with poultry manure had the highest number of pods which was statistically similar to the number of pods recorded under *B. subtilis* treatment.

Table 5 Effect of different soil amendments on components of yield of two cowpea cultivars

Factors		Number of pods	Number of unfilled pods	Weight of pods (g)	Length of pods (cm)
Cultivar					
FUAMPEA-2		6.25 ± 3.73	1.19 ± 2.15	4.84 ± 3.13	10.24 ± 2.49
Ife Brown		5.52 ± 2.87	0.81 ± 1.35	4.30 ± 3.02	10.05 ± 2.88
LSD		1.32^{ns}	0.72^{ns}	1.20^{ns}	1.09^{ns}
Soil amendment					
Control		4.96 ± 2.79^{b}	1.25 ± 1.75^{ab}	3.68 ± 2.73^{b}	9.82 ± 2.67
SSP		5.17 ± 3.10^{b}	0.71 ± 1.27^{ab}	3.69 ± 2.50^{b}	10.28 ± 2.53
PM		7.25 ± 4.07^{a}	1.54 ± 2.71^{a}	5.70 ± 3.48^{a}	9.61 ± 2.75
B. subtilis		6.17 ± 2.90^{ab}	0.50 ± 0.72^{b}	5.23 ± 3.07^{ab}	10.87 ± 2.74
LSD		1.86	1.02	1.71	1.54^{ns}
Cultivar × Soil amendment					
FUAMPEA-2	Control	4.58 ± 2.68^{b}	1.00 ± 1.65^{b}	3.91 ± 3.03^{b}	9.45 ± 3.34
	SSP	5.42 ± 3.20^{b}	0.67 ± 1.07^{b}	3.88 ± 2.45^{b}	10.11 ± 1.94
	PM	9.33 ± 4.54^{a}	2.50 ± 3.55^{a}	6.53 ± 3.76^{a}	9.98 ± 2.49
	B. subtilis	5.67 ± 2.64^{b}	0.58 ± 0.79^{b}	5.06 ± 2.71^{ab}	11.42 ± 1.74
Ife Brown	Control	5.33 ± 2.96^{b}	1.50 ± 1.88^{ab}	3.44 ± 2.63^{b}	10.19 ± 1.86
	SSP	4.92 ± 3.12^{b}	0.75 ± 1.48^{b}	3.50 ± 2.64^{b}	10.46 ± 3.09
	PM	5.17 ± 2.12^{b}	0.58 ± 0.90^{b}	4.87 ± 3.12^{ab}	9.23 ± 3.05
	B. subtilis	6.67 ± 3.17^{b}	0.42 ± 0.67^{b}	5.40 ± 3.51^{ab}	10.33 ± 3.46
LSD		2.54	1.41	2.44	2.19^{ns}

Note: ^{a,b} Means in the same column with different superscripts differ significantly ($P < 0.05$). LSD = least significant difference at $P < 0.05$, ns = not significant, SSP = single super phosphate fertilizer, PM = poultry manure.

Also, a higher number of unfilled pods were recorded on soil augmented with poultry manure, while the lowest number of unfilled pods was observed on soil treated with *B. subtilis* (Table 5). The weight of pods (5.70 g) was highest in cowpeas grown on soil augmented with poultry manure, but this was not significantly higher than the mass of pods recorded in *B. subtilis* treatment. The length of the pods of cowpea was not significantly influenced by different soil amendments, but the pods were longer in *B. subtilis* treatment than others in other soil amendments.

The interaction effect between the two cowpea cultivars and different soil amendments on the number of pods, unfilled pods, total weight of pods, and length of pods is shown in Table 5. FUAMPEA-2 had a higher number of pods (9.33), number of unfilled pods (2.50), and the total weight of pods (6.53 g) on soil amended with poultry manure and these were significantly higher than Ife Brown on soil treated with diverse soil amendments. Ife Brown had the lowest unfilled pods (0.42) under *B. subtilis* treatment. Length of pods was not significantly affected by the interaction between cowpea cultivars and soil amendments (Table 5).

The partitioning of assimilation into economic components was greatly influenced by different types of soil amendments. The obvious improvement observed in components of yield of cowpeas grown on soil amended with poultry manure showed that the amendment could be a good source of essential minerals for enhancing photosynthate accumulation in cowpeas. The amendment was a rich source of essential nutrients like P, K, and micronutrients necessary for building effective sources for synthesizing partitioning and assimilating into the sink. The impressive performance recorded under *B. subtilis* augmentation suggests that the organism promoted cowpea growth and development. Hence, it could be a suitable biofertilizer for promoting crop growth on poor soil. A similar view was shared by Kour *et al*. (2020), where bioinoculants like *B. subtilis* were identified as good rhizospheric plant growth-promoting organisms (Korir *et al*., 2017; Kour *et al*., 2020). The poor performance recorded in cowpeas grown on unfertilized soil implies that under native nutrients, cowpeas will suffer tremendous yield reduction.

Effect of Poultry Manure, Single Super Phosphate and *B. subtili*s Inoculant on Nodulation, Number of Seeds and Grain Yield of Two Cowpea Cultivars

The two cowpea cultivars differed significantly concerning the weight of fresh nodules and the number of nodules formed. Cultivar FUAMPEA-2 had a higher weight and number of nodules than Ife Brown (Table 6). The different amendments had no significant effect on cowpea nodulation, but higher (59.83 nodules/plant) and heavier nodules (2.67 g/plant) were recorded in cowpea grown on soil fertilized with poultry manure. Similarly, the number of nodules (95.50) and the weight of fresh nodules (4.72 g/plant) produced by FUAMPEA-2 in soil fertilized with poultry manure were higher than values recorded in Ife Brown treated with other amendments.

Cowpea typically forms symbiotic relationships with soil-inhabiting bacteria, but the success of the symbiotic relationship could be varietal dependent. This probably explains why FUAMPEA-2 modulated better than Ife Brown. Although nodulation improved in FUAMPEA-2 grown on soil augmented with poultry manure, this was not the case in Ife Brown. This observation regarding higher nodulation in FUAMPEA-2 is in agreement with the findings of Gerald (2004), that the addition of organic manure to soil enhanced the symbiotic relationship between microorganisms in the soil. In contrast, the lowest number of nodules and light-weighted nodules observed in Ife Brown grown on soil amended with poultry may be inherent in this genotype. This finding is similar to that of Ndor *et al*. (2012), who observed a significant variation in yield characteristics of some improved varieties of cowpea, and these were attributed to the genetic makeup of the assessed cultivars. It, therefore, means that *B. subtilis* LK14 may not be compatible with the colonizing roots of Ife Brown compared to FUAMPEA-2. Hence, the compatibility of biorganisms is germane in deploying biofertilizers for improving crop growth.

The effect of different amendments on the number of seeds showed that the highest number of seeds (30.92) was recorded in plants treated with *B. subtilis* inoculant, which was significantly higher than the number of seeds observed in plants grown on unamended soil. The different amendments exerted significant influence on the yield of the crop. Grain yield of cowpeas increased tremendously in soil amended with poultry manure over unamended soil, SSP, and *B. subtilis* by 55.7%, 28.1%, and 19%, respectively (Table 6).

The two cultivars were not significantly different with respect to yield and its components. The interaction between the two cultivars and different amendments significantly influenced the number of seeds, weight of seeds, and nodulation by the two cowpea cultivars. FUAMPEA-2 produced a higher number of seeds (33.33) and grain yield (48.23 g/plant) on soil fertilized with poultry manure than those produced by Ife Brown on other treatments (Table 6).

Effect of Poultry Manure, Single Super Phosphate and *B. subtili*s Inoculant on Weight of 100 Seeds and Harvest Index of Two Cowpea Cultivars

The weight of 100 seeds and the harvest index of the two cowpea cultivars were statistically similar. Furthermore, the type of amendments did not influence the weight of 100 seeds of cowpea. However, the harvest index was significantly influenced by soil amendments (Table 6). The weight of 100 seeds (13.08 g/plant) and harvest index (0.69) were highest in cowpeas grown on soil amended with poultry manure and lowest in unamended soil (Table 6).

The interaction showed that FUAMPEA-2 had a higher weight of 100 seeds (13.32 g/plant) in the soil fertilized with poultry manure comparable to Ife Brown under *B. subtilis* inoculant (13.28 g/plant). The lowest weight of 100 seeds (10.62 g/plant) was recorded in Ife Brown grown under native soil nutrients. The FUAMPEA-2 had the highest harvest index (0.59) on soil sown with seed inoculated with *B. subtilis*, while Ife Brown had the lowest harvest index (0.24) on unfertilized soil (Table 6).

The weight of 100 seeds and harvest index are important yield components in cowpeas. These components were higher in FUAMPEA-2 than Ife Brown. This infers that FUAMPEA-2 is more productive than Ife Brown as it appeared that the cultivar efficiently partitioned photoassimilate into economic yield better than biological components. Assimilate mobilization from source to sink could be linked to adequate internal transportability and remobilization potentials (Prieto *et al*., 2017). This perhaps explains the high grain yield recorded in FUAMPEA-2 over and above the other cultivar. Similar findings were also reported by Abebe and Feyisa (2017).

Table 6 Effect of different soil amendments on nodulation, number of seeds, and grain yield of two cowpea cultivars

Factors		Number of nodules/plant	Weight of nodules (g/plant)	Number of seeds/plant	Grain yield (g/plant)	Weight of 100 seeds (g/plant)	Harvest index
Cultivar							
FUAMPEA-2		66.65 ± 38.96^{a}	3.41 ± 2.26^{a}	27.00 ± 15.67	26.20 ± 29.36	12.17 ± 3.47	0.43 ± 0.48
Ife Brown		38.92 ± 37.89^{b}	0.96 ± 0.93^{b}	24.96 ± 17.33	21.54 ± 24.65	12.36 ± 3.41	0.35 ± 0.54
LSD		15.69	0.70	6.51^{ns}	10.74^{ns}	1.39^{ns}	0.21^{ns}
Soil amendment							
Control		46.83 ± 28.44	2.08 ± 1.78	19.25 ± 13.52^{b}	15.40 ± 16.67^{b}	11.40 ± 4.31	0.22 ± 0.20^{b}
SSP		55.04 ± 28.89	1.80 ± 1.31	23.71 ± 13.31^{ab}	18.42 ± 21.51^{b}	11.89 ± 2.45	0.25 ± 0.34^{b}
PM		59.83 ± 54.48	2.67 ± 3.00	30.04 ± 16.32^{a}	34.04 ± 35.42^{a}	13.08 ± 2.50	0.69 ± 0.81^{a}
B. subtilis		49.42 ± 46.01	2.11 ± 2.06	30.92 ± 19.96^{a}	27.62 ± 28.33^{ab}	12.70 ± 2.67	0.47 ± 0.45^{ab}
LSD		22.19^{ns}	0.99^{ns}	9.21	15.19	1.97^{ns}	0.29
Cultivar × Soil amendment							
FUAMPEA-2	Control	61.25 ± 30.53^{b}	3.33 ± 1.76^{b}	20.42 ± 13.36^{ab}	15.26 ± 15.32^{b}	12.17 ± 3.94	0.27 ± 0.17
	SSP	56.67 ± 24.38^{b}	2.42 ± 1.46^{bc}	23.33 ± 12.94^{ab}	17.58 ± 19.40^{b}	11.08 ± 2.74	0.38 ± 0.47
	PM	95.50 ± 56.69^{a}	4.72 ± 3.07^{a}	33.33 ± 18.97^{a}	48.23 ± 44.21^{a}	13.32 ± 3.09	0.49 ± 0.56
	B. subtilis	53.17 ± 22.33^{bc}	3.17 ± 2.06^{b}	30.92 ± 14.85^{ab}	23.76 ± 19.25^{b}	12.11 ± 4.01	0.59 ± 0.58
Ife Brown	Control	32.42 ± 17.48^{bc}	0.84 ± 0.42^{d}	18.08 ± 14.16^{b}	15.55 ± 18.61^{b}	10.62 ± 4.68	0.24 ± 0.23
	SSP	53.42 ± 33.84^{bc}	1.32 ± 0.88^{cd}	24.08 ± 14.24^{ab}	19.27 ± 24.27^{b}	12.70 ± 1.89	0.24 ± 0.11
	PM	24.17 ± 14.73^{c}	0.62 ± 0.46^{d}	26.75 ± 13.16^{ab}	19.86 ± 15.17^{b}	12.84 ± 1.85	0.56 ± 1.02
	B. subtilis	45.67 ± 62.43^{bc}	1.05 ± 1.48^{d}	30.92 ± 24.74^{ab}	31.48 ± 35.72^{ab}	13.28 ± 4.00	0.34 ± 0.21
LSD		29.81	1.35	13.19	21.03	2.78^{ns}	0.43^{ns}

Note: ^{a,b,c,d} Means in the same column with different superscripts differ significantly ($P < 0.05$). LSD = least significant difference at $P < 0.05$, ns = not significant, SSP = single super phosphate fertilizer, PM = poultry manure.

Effect of Poultry Manure, Single Super Phosphate and *B. subtilis* Inoculant on Dry Matter Accumulation by Two Cowpea Cultivars

The two cultivars are similar concerning biomass accumulation. Also, the type of soil amendments did not influence dry matter accumulation by the two cowpea cultivars differently. The interaction between two cultivars and different soil amendments on biomass accumulation and partitioning showed that Ife Brown had the highest weight of root (1.78 g/plant) and shoot (13.28 g/plant) on soil fertilized with poultry manure, and this was statistically similar to results obtained in other treatments (Table 7).

The improved dry matter yield recorded in Ife Brown over FUAMPEA-2-grown soil amended with poultry manure implies that the cultivar efficiently utilized the nutrients supplied by poultry manure for biological yield. Similar findings were reported by Agbogidi and Egho (2012) and Animasaun *et al*. (2015), who observed a significant variation in growth and yield characteristics of some improved varieties of cowpea and attributed it to the genetic makeup of the varieties examined. The same findings were reported by López-Arredondo *et al*. (2017). Interestingly, the cultivar had superior vegetative growth than FUAMPEA-2, implying that the Ife Brown accumulated dry matter for structural components rather than economic yield. This suggests that the cultivar may be a better fodder crop than FUAMPEA-2. The classification of this cultivar as fodder rather than grain cowpea needs to be more elucidated.

Table 7 Effect of different soil amendments on dry matter accumulated by two cowpea cultivars

Factors		Dry matter (g/plant)		
		Root	Shoot	Total biomass
Cultivar				
FUAMPEA-2		1.45 ± 0.80	10.19 ± 7.13	11.64 ± 7.77
Ife Brown		1.61 ± 0.74	11.69 ± 8.17	13.30 ± 8.51
LSD		0.32^{ns}	3.13^{ns}	3.33^{ns}
Soil amendment				
Control		1.47 ± 0.74	10.99 ± 7.93	12.43 ± 8.60
SSP		1.49 ± 0.63	10.31 ± 6.55	11.80 ± 7.09
PM		1.69 ± 0.92	12.57 ± 9.40	14.26 ± 9.67
B. subtilis		1.47 ± 0.75	9.89 ± 6.67	11.40 ± 7.18
LSD		0.45^{ns}	4.43^{ns}	4.71^{ns}
Cultivar × Soil amendment				
FUAMPEA-2	Control	1.52 ± 0.86	12.38 ± 9.46	13.90 ± 10.27
	SSP	1.22 ± 0.54	8.47 ± 5.24	9.69 ± 5.60
	PM	1.59 ± 0.88	11.87 ± 6.97	13.46 ± 7.51
	B. subtilis	1.48 ± 0.92	8.04 ± 5.95	9.52 ± 6.82
Ife Brown	Control	1.42 ± 0.63	9.60 ± 6.14	10.95 ± 6.67
	SSP	1.76 ± 0.73	12.16 ± 7.40	13.92 ± 8.01
	PM	1.78 ± 0.98	13.28 ± 11.63	15.06 ± 11.73
	B. subtilis	1.47 ± 0.58	11.73 ± 7.08	13.29 ± 7.33
LSD		0.63^{ns}	6.28^{ns}	6.66^{ns}

Note: LSD = least significant difference at $P < 0.05$, ns = not significant, SSP = single super phosphate fertilizer, PM = poultry manure.

The lowest biomass accumulated by Ife Brown grown in soil inoculated with *B. subtilis* could probably be due to the reduction in biomass accumulation as the growth proceeded beyond the vegetative stage. One cannot rule out the possibility of incompatibility of the bacterium with the root of the Ife Brown cultivar compared to FUAMPEA-2. Berg and Smalla (2009) have reported that plant morphological and physiological factors influence rhizospheric colonization and diversity of microbes. Sharma *et al*. (2013) also showed that mineral nutrients, especially N, contribute significantly to microbial functionality and root colonization in rhizospheric. It may be wise to conclude that the nutrient in this treatment was not adequate to support cowpea growth and biomass yield potentials. This infers that under inadequate nutrient availability, there was remobilization and redistribution of accumulated biomass to sustain the crop through its lifecycle. Hence, the complementary interaction of this organism with other soil amendments needs to be further studied to optimize its beneficial roles in improving crop performance on marginal soil.

Proximate Composition and Phosphorus Uptake Concentration of Two Cowpea Cultivars as Influenced by Different Soil Amendments

The proximate compositions of the two cowpea cultivars were not statistically different. Similarly, different soil amendments did not influence proximate components and seed P-concentration of the crop differently (Table 8). However, the concentration of crude protein in seeds of FUAMPEA-2 (32.0 g/kg) was significantly higher than the concentration recorded in Ife Brown. The P concentration in seeds was higher in Ife Brown than FUAMPEA-2, while crude protein was higher in FUMPEA-2 but not statistically different from the concentration recorded in Ife Brown. Crude protein, fiber, carbohydrates, and seed P-uptake constituents of cowpea seed improved tremendously on soil amended with poultry manure compared to other soil amendments.

The interaction between cowpea cultivars and different soil amendments showed variation in the proximate and P content of the two cowpea cultivars (Table 8). Seeds of Ife Brown had the highest moisture content (11.6%) on soil fertilized with poultry manure, but this was statistically similar to the moisture content in seeds of FUAMPEA-2 (11.5%) with the same amendment. FUAMPEA-2 had the lowest moisture content (8.9%) on soil fertilized with SSP. The seeds of FUAMPEA-2 had the highest ash (8.5%) on soil fertilized with poultry manure, whereas the highest crude protein (34.2%) was recorded in seeds harvested from soil amended with SSP. There was no significant difference in the crude protein content of FUAMPEA-2 seeds obtained from soil treated with poultry manure or *B. subtilis* inoculant. Seeds of FUAMPEA-2 obtained from soil treated with poultry manure had the highest crude fiber (5.3%), while Ife Brown had the lowest crude fiber (4.5%) on soil amended with SSP. The dry matter was highest (91.2%) in seeds of FUAMPEA-2 obtained from soil amended with SSP relative to dry matter (88.4%) of Ife Brown seeds obtained from soil fertilized with poultry manure. The seeds of Ife Brown had the highest carbohydrate (43.2%) under *B. subtilis* inoculant, while FUAMPEA-2 seeds had the lowest carbohydrate (39.8%) in seeds obtained from unamended soil. The seed P concentration ranged from 3.15 mg/kg in FUAMPEA-2 seeds harvested from *B. subtilis* inoculant to 10.75 mg/kg in seeds of the same cowpea cultivar obtained from soil fertilized with SSP.

Table 8 Proximate components (%) and phosphorus content (g/kg) of two cowpea cultivars as influenced different soil amendments

Factors		Moisture content	Ash	Crude protein	Crude fiber	Crude fat	Dry matter	CHO	Seed phosphorus content
Cultivar									
FUAMPEA-2		10.17	7.02	32.0	4.85	5.53	89.93	40.74	5.85
Ife Brown		10.90	7.15	30.15	4.69	5.5	89.10	41.34	8.45
LSD		0.29^{ns}	0.64^{ns}	1.82^{ns}	0.23^{ns}	0.48^{ns}	0.29^{ns}	1.04^{ns}	2.74^{ns}
Soil amendment									
Control		10.60^{ab}	6.65	32.16	4.75	5.25	89.41^{ab}	40.61	6.25
SSP		9.51^{b}	6.80	33.28	4.65	5.75	90.49^{a}	40.02	8.65
PM		11.50^{a}	7.75	28.89	4.95	5.45	58.50^{b}	41.47	7.45
B. subtilis		10.55^{ab}	7.15	329.97	4.73	5.55	89.46^{ab}	42.06	6.25
LSD		1.83	1.92^{ns}	6.39^{ns}	0.69^{ns}	0.81^{ns}	1.83	3.38^{ns}	9.69^{ns}
Cultivar × Soil amendment									
FUAMPEA-2	Control	10.25^{c}	6.50^{e}	33.69^{b}	4.60^{c}	5.20^{ed}	89.75^{c}	39.79^{g}	3.45^{g}
	SSP	8.85^{e}	6.50^{e}	34.13^{a}	4.80^{b}	5.90^{a}	91.15^{a}	39.82^{g}	10.75^{a}
	PM	11.40^{a}	8.40^{a}	27.58^{f}	5.20^{a}	5.10^{e}	88.60^{e}	42.32^{b}	6.05^{e}
	B. subtilis	9.99^{d}	6.50^{e}	32.38^{c}	4.60^{c}	5.70^{b}	90.01^{b}	40.83^{d}	3.15^{h}
Ife Brown	Control	10.94^{b}	6.80^{d}	30.63^{d}	4.90^{c}	5.30^{ed}	89.06^{d}	41.43^{c}	9.05^{c}
	SSP	10.17^{d}	7.10^{c}	32.42^{c}	4.50^{b}	5.60^{c}	89.83^{c}	40.21^{f}	6.55^{e}
	PM	11.60^{a}	7.10^{c}	30.19^{e}	4.70^{d}	5.80^{b}	88.40^{f}	40.61^{e}	8.85^{d}
	B. subtilis	11.05^{b}	7.76^{b}	27.51^{f}	4.76^{c}	5.34^{d}	88.91^{d}	43.19^{a}	9.35^{b}
LSD		0.15	0.14	0.16	0.15	0.14	0.16	0.17	0.16

Note: ^{a,b,c,d,e,f} Means in the same column with different superscripts differ significantly ($P < 0.05$). LSD = least significant difference at $P < 0.05$, ns = not significant, SSP = single super phosphate fertilizer, PM = poultry manure, CHO = carbohydrate.

The superior proximate constituents observed in cowpea seeds harvested from poultry manure fertilized soil suggests that seed quality in terms of proximate content is related to the extent of soil mineral nutrient availability, assimilate mobilization, and partitioning into economic yield (Weih *et al*., 2017). This influences the quality of the seed obtained. The improvement in P uptake by FUAMPEA-2 grown in amended relative to the unamended soil may be connected to efficient translocation of the acquired minerals adequately mobilized and partitioned into the grains. Phosphorus concentration in seed harvested from unfertilized soil was low, suggesting that adequate mobilization of P is necessary for improving grain quality in cowpeas.

Residual Effects of Different Soil Amendments on Soil Physical and Chemical Characteristics

The influence of different soil amendments on the chemical and physical characteristics of soil showed that the application of poultry manure improved the chemical characteristics of the soil greatly (Table 9).

Table 9 Influenced of poultry manure, single super phosphate and *Bacillus subtilis* inoculant on soil chemical and physical characteristics

Soil amendment	pH	OC (%)	Total N (%)	Av. P (mg/kg)	Ca	Mg	K	Na	CEC	Mn	Fe	Cu	Zn	Particle size (%)		
					(cmol/kg)					(mg/kg)				Sand	Silt	Clay
Control	6.56^{b}	1.90	0.21	23.70^{b}	4.52^{b}	1.04^{b}	0.07^{a}	0.21	6.21^{b}	119.50^{bc}	128.00^{b}	1.15	1.29	79.5	12.0	8.5
SSP	6.74^{b}	1.99	0.22	23.89^{b}	4.67^{b}	1.04^{b}	0.06^{a}	0.24	6.33^{b}	126.50^{ab}	147.00^{ab}	1.16	1.23	80.0	12.5	7.5
PM	7.21^{a}	2.02	0.25	72.21^{a}	5.92^{a}	2.01^{a}	0.08^{a}	0.24	8.59^{a}	141.00^{a}	151.00^{a}	1.16	1.26	79.5	12.5	8.0
B. subtilis	6.57^{b}	1.68	0.19	20.82^{b}	4.33^{b}	1.04^{b}	0.04^{b}	0.21	5.96^{b}	110.00^{c}	130.00^{ab}	1.05	1.20	81.5	11.0	7.5
LSD	0.32	0.52^{ns}	0.08^{ns}	8.10	0.93	0.13	0.02	0.04^{ns}	0.92	15.52	21.33	0.24^{ns}	0.21^{ns}	3.26^{ns}	2.41^{ns}	4.28^{ns}

Note: $^{a, b, c}$ Means in the same column with different superscripts differ significantly ($P < 0.05$), LSD = least significant difference at $P < 0.05$, ns = not significant, SSP = single super phosphate fertilizer, PM = poultry manure, OC = organic carbon, Total N = total nitrogen, Av. P = available phosphorus, CEC = cation exchange capacity.

The organic and N contents of the soil were higher in the soil fertilized with poultry manure but not significantly higher than the contents from the other treatments. The available P, Ca, Mg, Mn, and Fe concentrations improved significantly in soil amended with poultry manure, while these nutrients were depleted in the unfertilized soil. The particle size was not influenced by the application of different P-based soil amendments (Table 9).

Applying organic manure increased soil pH after harvest, with a substantive increase in macro and micronutrients observed in soil amended with poultry manure. The applied poultry manure was more effective in alleviating pH, P, Ca, Mg, cation exchange capacity (CEC), Mn, Fe, Cu, Zn, and sandy loam. This agrees with the findings of Abumere *et al*. (2019), who reported that poultry manure was superior to other soil amendments commonly utilized in enhancing the fertility of soil low in essential minerals. Reports have shown that the application of farmyard manure, such as chicken manure, often heightens soil P levels. The high residual P content in soil mixed with poultry manure presupposes a far-reaching P mineralization stemming from enormous dissolved organic matter components compared to unfertilized soil (Camberato, 2001; Singh *et al*., 2014). Dissolved organic matters are known to regulate soil P dynamics and facilitate its availability for crop use (Jindo *et al*., 2023). Poor P mineralization and high fixation are characteristically associated with soil low in organic matter, which is predominant soil in cowpea growing regions in the Tropics. This may probably explain why unamended soil with high initial P content had low residual P compared to soil amended with poultry manure.

CONCLUSIONS

The two cultivars responded differently to organic, inorganic fertilizers as well as *B. subtilis,* inoculant. Generally, the growth of both cultivars improved tremendously at the early phenological stage under biofertilizer (*B. subtilis*) treatment. The agro-botanical traits of Ife Brown improved greatly under organic fertilizer, while the yield and yield components of FUAMPEA2 improved tremendously under organic fertilizer. The proximate components of the cultivars exhibited superior performance under organic fertilizer treatment. Nevertheless, this had a similar effect to the single super phosphate fertilizer. The essential mineral nutrients like N, P, and K increased remarkably in soil treated with organic fertilizer. The effect of the amendments in improving the performance of cowpea was in the order: poultry manure > single super phosphate > *B. subtilis* > unamended. This study demonstrated the practical benefits of utilizing poultry manure and biofertilizers in improving cowpeas' P uptake and grain yield. Phosphorus uptake and proximate composition of a grain of FUAMPEA-2 increased considerably on marginal soil amended with poultry manure. To enhance the performance of cowpeas in marginal fields, the application of poultry manure is recommended. FUAMPEA-2 is superior to Ife Brown in terms of yield and grain quality, while Ife Brown could be explored as a fodder crop because of its high agro-botanical yield. It is concluded that the application of poultry manure, an organic fertilizer, and *B. subtilis* a biofertilizer, improved the yield and quality of cowpea grain.

ACKNOWLEDGEMENTS

The authors acknowledge the technical assistance of Mrs. Olufunmilola K. Odeh of the Plant Pathology Laboratory, Department of Crop Protection and Environmental Biology in culturing the *B. subtilis*. Contributions and expertise of O.O. Popoola of Phytopathology Laboratory, Department of Botany, University of Ibadan for identification and validation of the cultured organism is thankfully appreciated.

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