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Investigation of the influence of biofertilizer synthesized using microbial inoculums on the growth performance of two agricultural crops

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ABSTRACT

In this work, biofertilizer was synthesized by mixing sawdust and other nitrogenous agricultural wastes into composites in the following ways: S1 (sawdust + chicken litter + vegetable waste), S2 (sawdust + sewage sludge + vegetable waste) and S3 (sawdust only as the control) respectively using actinomycetes as the microbial inoculums. In-vessel method of composting was employed with 120 L capacity polyethylene (PET) container as the bioreactor for the pilot scale study. Microorganisms were isolated from landfill extracts. Aeration was accomplished through turning of the compost twice weekly. Nitrogen, Carbon, Organic Matter and pH were determined at 5days intervals till the end of composting exercise. Flame Atomic Absorption Spectrometer (FAAS), was used for determination of the mineral composition of the raw materials and end products. American Society for Testing and Materials was used in the Analysis of Nitrogen, Carbon, and Organic Matter contents. Biofertilizer was analyzed more for activeness as organic fertilizer in the field studies using two crops (*Maize* and *Okra*). Biochemical test revealed that six genera of Actinomycetes were isolated. Inorganic salt starch agar medium was noticed to be effective isolation media for Actinomycetes. Actinomycetes were found to be good agents for biofertilizer synthesis due to their ability to mineralize nitrogen during composting. Preliminary investigation revealed that *Rothia spp* gave the highest percentage degradation of cellulose (21.6 %) as well as highest percentage mineralization of nitrogen (6.87 %) after 21days of incubation. Dosage ratio of 2:1:1 w: w, moisture content of 50–60 % and 25 days was found to be the optimum condition for nitrogen mineralization. Organic matter content of composts S1 and S2 decreased significantly with time while total kjeldahl nitrogen (TkN) content of the composts increased except compost S3 which on the contrary, reduced. The maximum temperature achieved at the end of 25 days compositing of substrate S2 was 64.6 *ENTITYNOTDEFINED!!!*. Analysis of the leaf area index (LAI) revealed 10th week after transplant to be the period of optimum growth for both crops. In addition to, all the analysis conducted pointed to the fact that influence of biofertilizer on *Okra* and *Maize* growth performance is equivalent to the growth performance of chemical fertilizer on the same crops, affirming that organic fertilizer can comfortably replace chemical fertilizer in future.

1. Introduction

Solid waste management has been a major issue of concern to most urban communities in Africa especially Nigeria. The number of people in Nigeria (population) was estimated at 200 million as quoted by federal department of statistics and demography in 2019. Nigeria is the biggest producer of solid wastes in Africa [1–3]. Host of measures and policies has been put in place over the years to control waste disposal in Nigeria, but with each passing day, solid waste generation and accumulation within the urban cities still assumes alarming rate. As of now, Nigeria produces a roughly 7.2 million tons of solid wastes yearly, yet shockingly, only 20–30%

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is taken off the street and in most cases are not even managed properly [1,4], Christian et al. [36,5]. Just couple of States in Nigeria has demonstrated an extensive level of take steps to make proactive strides in battling this scourge out of the 36 States and the Federal capital in the nation, while the rest have simply paid lip service to issues of waste management. According to Sabiiti et al. [6] and [7], 60 % of solid waste generated worldwide are made up of agricultural waste. Oltien and Beecket, [61], in their research report quoted that agricultural wastes can be processed to alternative fertilizer (biofertilizer) through proper composting.

Nigeria is a very poor country despite her large population due to over-dependency on oil revenue which had made the source of revenue in the country a mono economy. In recent time, attention has been shifted to Agriculture due to dwindling oil price in international market but less than 20 % of the populace are interested in agriculture and these few numbers depends so much on chemical fertilizer (mineral fertilizer) to improve the fertility of the soil. This chemical fertilizer has peculiar problems associated with it which include; degradation of soil structure, pollution of the soil and underground water causing eutrophication, high cost etc [8–10]. In recent time, the demand for safe food and safe environment has forced agriculturist to think and devise a viable alternative to chemical fertilizer and at the same time shifted the attention of the researchers to the best method of converting agricultural wastes into biofertilizer which has been proven to be more eco-friendly than chemical fertilizer according to Agnew and Leonard [11] and [12].

Several authors have bared their mined on the meaning of biofertilizer; Haug, 1993 defined it as substance that contains essential nutrients such as nitrogen, phosphorus, potassium, as well as several other important elements in small amounts, [13] defined it as substances accompanying the oxidative enzymatic polymers in the soil via free radical mechanism process and release of important soil nutrients. Oltien and Beecket [61] defined it as a substance that contains microorganisms and promotes growth of plant by increasing the supply of important soil nutrient. On the other hand Toumela et al. [14], and Thompson et al. [15], quoted composting as the decomposition of degradable organic matter in waste in the presence of microorganisms (*Actinomyces*) under control conditions such as temperature, pH, size of compost, moisture content etc. In other words, composting is a microbiological process. Furthermore, aerobic composting method of wastes transformation into useful materials is the essence of this work. In particular, this investigation is intended to transform agricultural wastes like sawdust, vegetable wastes, chicken litter, and dewatered sewage sludge into Biofertilizer which is plausible alternative to chemical fertilizer. According Asadu et al. [10], they further defined composting as the aerobic microbial decomposition of organic matter of vegetable and animal origin, under conditions that allow the development of thermophilic temperatures as a result of the heat produced by biological reactions. It Involves the mineralization and partial humification of the organic matter, this process will lead to a stabilized and hygienized final product (i.e. free of pathogens and seeds) commonly known as compost

Actinomycetes are living organisms with attributes normal to the two microscopic organisms and growths, yet they have unmistakable highlights that keep them to a particular class [16,17]. They are unicellular like microscopic organisms which create slim non-septate mycelium. Similar to bacteria, they do not have different cell-wall but their cell wall does not have chitin and cellulose (usually present in fungi cell wall). Saprophytic *Actinomycetes* are critical essential colonizers of soil natural material which are generally as insoluble polymers [18]. *Actinomycetes* are heterotrophic in nature, the mesophilic types grow at a temperature range of 25–30 °C while the thermophilic types grow at a temperature range of 55–60 °C in the soil. Therefore, the aim of this study among other things is to produce biofertilizer from agricultural waste using an isolated organisms (*Actinomycetes*) to hasten the rate of nutrient release during composting. This study is very important and there is no better time than now considering that Nigeria is gradually shifting attention from petroleum to agriculture to diversify her economy. Furthermore, the study will ascertain the influence of biofertilizer produced on the growth performance of two agricultural crops

2. Materials and method

2.1. Raw materials and their sourcing

The major raw materials employed in this study was gathered from Nsukka metropolis, Enugu State Nigeria. Fresh sawdust was assembled from Nsukka Timer shed, fresh vegetable waste were collected from Ekpo ref eatery, University of Nigeria Nsukka, Chicken litter was sourced from Monica's poultry farm limited Nsukka, Sewage sludge cake was collected from the waste water treatment plant of Bestie Industrial limited Nsukka. Analytical grade HNO_3, HCl, Agar media, $CaCO_3$, $(NH_4)_2SO_4$, K_2HPO_4, $MgSO_4.7H_2O$, NaCl, $FeSO_4.7H_2O$, $MnCl_2.7H_2O$, and $ZnSO_4.7H_2O$ were purchased from Jecks chemical limited Nsukka main market. The reagents were 99 % pure and was used directly without purifying further. Sawdust was employed in this research to provide the required free air space during composting (Bulking agent) while sewage sludge cake, vegetable waste, and chicken litter were used as the nitrogen rich agro wastes (nitrogen sources). The agricultural wastes were blended before composting to set the initial carbon to nitrogen ratio between 20 and 35. The rate of decomposition of the vegetable waste (made up of Pumpkin stalk, cabbage, fresh plantain peel, green leaves, lettuce etc) were increased by reducing the sizes with a cutter. The properties of the agricultural wastes were presented as evident in Table 1

2.2. Design of composting drums

The composting activities were carried out in polyethylene (PET) drums of diameter 45 cm, volume 120 L and height 115 cm as the reactor. The reactors were effectively transformed (modified) in order to ensure proper air distribution within the waste materials during composting. The changes on the surface of the reactor was characterized by providing 15 mm midway between holes in five layers round the reactors with the help of a mechanical driller to ensure proper circulation of air inside the reactor as shown in Plate 1 . Portions for periodic collection of samples from the reactor for analysis were also provided at the middle and bottom part of the drums. The provisions for the leachate gathering from the bottom of the drums were also provided.

2.3. Pre-treatment of soil sample for microbial isolation

About 100 g of soil blended with agricultural waste compost was taken from beneath University of Nigeria Veterinary Teaching hospital Compost Nsukka. A sieve of 1.5 mm mesh size was used to remove stones and other large particles. Thereafter, the sample was sun dried for 2 days and incubated at temperature of 42 °C for 7 days in a conical flask using an autoclave (systec-V-75, Shimadzu cooperation, Japan) in order to eliminate the mesophilic microorganisms to pave way for the isolation of thermotolerant *Actinomycetes*.

2.4. Culture media and condition

The method as described by Antoinetta et al. [18] was employed for microbial isolation in this work. The isolation media was

Table 1
Physiochemical Features of the agricultural wastes before composting operation.

Wastes Parameters	Sewage Sludge	Vegetable waste	Chicken Litter	Sawdust
Organic matter (%)	86.4	54.4	76.8	55
Total Carbon (%)	30.2	23	39.25	55.1
pH	7.7	7.6	6.4	5.10
Lignin (%)	4.15	0.46	1.98	22.8
Cellulose (%)	10.07	8.3	7.45	54.7
Nitrogen (%)	5.16	5.9	4.96	0.63
Phosphorus (%)	4.7	6.3	4.87	4.7
Potassium (%)	4.3	2.3	3.12	1.9
Volatile Matter %	19.6	12.4	22.3	25
Ash content (%)	22.8	31.4	18.7	11.3
Moisture content (%)	27.4	33.2	9.8	8.6
Carbon to nitrogen ratio (C/N)	5.85	3.89	8.01	87.5

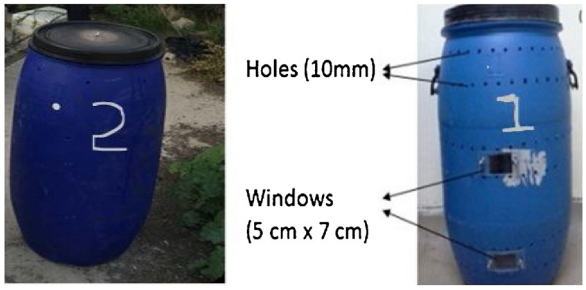

Plate 1. Typical image of the (1) modified and (2) unmodified composting drums used as the reactor.

prepared with Inorganic salt starch agar medium using the following amounts of reagents; Agar- 20 g, Soluble starch- 10 g, Calcium Carbonate ($CaCO_3$) - 2.0 g, Ammonium Sulphate $(NH_4)_2SO_4$- 2.0 g, Di-potassium phosphate (K_2HPO_4) - 1.0 g, Magnesium Sulphate ($MgSO_4.7H_2O$) - 1.0 g, Sodium Chloride (NaCl) - 1.0 g, Iron Sulphate ($FeSO_4.7H_2O$) - 1.0 g, Manganese Chloride ($MnCl_2.7H_2O$) - 1.0 g and Iron sulphate ($ZnSO_4.7H_2O$) - 1.0 g. Preparation of the reagents was done by first, liquifying them with distilled water followed by agitation in a water bath (PolyScience WBO2A11B, Taiwam) at 50 °C and thereafter, sterilized at a temperature of 121 °C for fifteen minutes in autoclave. Some quantity of the mixture was dispensed into 250 ml conical flask and sterilized followed by addition of antifungal reagents, nystatin (20 μg/l) and rifampcin (40 μg/l) to deter the emergence of fungi and other *non-actinomycetes* respectively. Meanwhile, the pretreated soil sample were serially diluted with distilled water in test tubes (Tenfold serial dilution). 0.1 ml of the solution was withdrawn from the test tubes 10^{-3} and 10^{-7} and then transferred aspergically into the inorganic *agar* medium in duplicates using spread plates to form the culture plates. Rifampcin and nystatin were incorporated into the media at concentrations 40 μg/l and 20 μg/l. Thereafter, the culture plates were incubated at 45 °C for five days to ensure the presence of the thermotolerant *Actinomycetes* and to allow growth and sporulation of the colonies. Distinct colonies were picked after 5 days from each plate and inoculated aseptically onto the agar slants in test tubes to obtain pure culture after which stock culture were prepared from the pure culture and stored in refrigerator at 4 °C for mass production and use. The number of colonies formed were monitored using viable plate count (quadrant method). The isolated microbes were characterized via gram staining and biochemical test according to standard microbiological steps and Bergey's Manual of Determinative micro- bacteriology.

2.5. Screening test for degradation of sawdust using the microbial isolates

The method of Kulie and Radojicic [19], was employed for this investigation. 2 g of freshly prepared sawdust was added into seven boiling tubes. In each of the tubes, 25 ml of distilled water was added and the contents were sterilized in autoclave at 121 *ENTITYNOTDEFINED!!!* for twenty mins. The tubes were numbered 1–6 and seventh as the control. Each isolate was inoculated along with 0.5 g of ammonium nitrate into each tube containing sawdust except the controls. The tubes and the contents were left at room temperature for 25days. The fluid contents in the tubes were systematically decanted after 25 days. The sawdust in the tubes were investigated for cellulose compositions. Cellulose content analysis was carried as described by [19].

2.6. Substrate arrangements for biofertilizer synthesis

The agricultural wastes used in this operation were first sterilized using vertical floor stand systec V-150 autoclave and thereafter, combined and inoculated as follow:

Compost S1= Sawdust + Chicken litter + Vegetable Waste + inoculums

Compost S2 = Sawdust + sewage sludge cake + Vegetable waste + inoculums

Compost S3 = Sawdust only + inoculums

There were three substrates S1, S2 and S3. S3 served as the control. *Streptomyces spp* (isolate 4) and *Rothia spp* (isolate 6*)* were selected from the isolates as the types of inoculum as follow: type 1 (*Streptomyces spp*), type 2 (*Rothia spp*) and type 3 *Consortium spp* (*Streptomyces spp* + *Rothia spp*). The organisms were combined at the ratio of 1:1 with bacteria load of 10^6 to form the consortium.

2.7. Composting operation

The pilot scale composting was carried out as follow; 80 kg each of S_1, S_2 and S_3 for every round of composting were formulated by combining the agricultural wastes at the dosage ratio (1:1:1, 1:2:1, 2:1:1, 2:2:1 and 3:1:1) and submitted to separate reactors (drums) as described in Section 2.2. The operation took place in an open environment to give room for natural air. The reactors was supported on the bricks and plastic tray placed below the reactors for leachate collection. Each compost was inoculated with 200 ml of inoculums with microbial load of 10^6 CFU/g at the beginning of

the composting. The oxygen concentration within the compost was monitored using a gas analyzer (Geotechnical instrument, model GA2000) and air sampling probe in stainless steel (1 m long). Compost moisture content was monitored and maintained using electronic instrument (Reotemp 648(800) moisture meter, San Diego CA). The maturity of the composts was determined by monitoring the temperature rise and fall in compost during composting. Stainless steel temperature probe was employed in temperature measurement (semiconductor LM35 monolithic temperature sensor). Oxygen concentration (aeration) within the compost matrix was achieved through forced aeration (manual mixing) once daily. Periodic sprinkling of water and turning of the compost was maintained to ensure proper moisture and proper colonization of the waste by the inoculums. Product quality monitoring and operating parameters such as organic matter, temperature, total carbon and potential hydrogen (pH) were monitored constantly by taking samples from the reactors every five days for analysis in the laboratory. Effect of inoculums type, moisture content, substrate type and dosage ration were studied.

2.8. Characterization of the samples

Standard test method for organic matter, moisture content, volatile matter and ash content as described by American Society for Testing and Materials, ASTMD2974 [20] was adopted. Following this method, organic matter was estimated by placing 10 g of fertilizer sample in muffle furnace for 5 h: 20 min. at an elevated temperature of 450 °C and thereafter allow the sample to cool to a constant weight. Determination of moisture content was done by placing 10 g of sample in oven at 108 °C to a constant weight. 10 g of sun-dried sample was inserted into the furnace at 180 °C for 6 min. and thereafter cooled to room temperature for volatile matter determination. Ash content was estimated from the dry sample by placing 10 g in the furnace at 550 °C for 7 min. Total carbon was estimated using the step by step method as described by [21]. pH was estimated with pH-meter (Model 136E/SET, Precision/sensitivity 0.01/-53.18 mV/pH@25 °C, Hanna Instrument, USA) in three-fold amount of distilled water blended and homogenized with the sample and allowed standing for 1hrs:30 min. Phosphorus and potassium content was analyzed using Flame Atomic Absorption Spectrometer (AAS) (model 230ATS, BUCH scientist). Total kjeldahl nitrogen was determined according to the procedure given by ASTMD3590 [22].

2.9. Experiments in the field

2018 farming season precisely between the month of March and June was the period this field investigation was carried out in the analytical farm of Department of Soil Science along Sullivan Chime drive, University of Nigeria Nsukka. The topographic nature of Nsukka is such that the rainy season (summer) start by March/April till September/October yearly thereby paving way for the winter period which last between November and February annually. Nigerian Metrological service puts the mean yearly rainfall in Nsukka in their 2017 report as between 2340 mm–2610 mm. The average yearly temperature was given as 21.5 °C as minimum and 32.8 °C as maximum. In this part of the country/world, the rain distribution is such that June/July is always characterized by heavy rain. The type of soil that characterizes the area is sandy-loam.

2.9.1. Land preparation for planting operation

280 kg of biofertilizer produced in this research and 25 kg of Chemical fertilizer purchased from Indorama fertilizer company Eleme River State Nigeria was used in this exercise. Genetically improved variety of Maize and Okra seeds was sampled from Nigerian Institute of Root and Crop Research, Umudike, Abia State Nigeria. Soil test was first conducted with samples of soil from the target area to ascertain the amount of various soil nutrients in the untreated soil. The land preparation was in two stages. First, a measured quantity of soil in sample bags into three places were labelled A = soil only (untreated) as the control, B = soil + bio-fertilizer and C = soil + chemical fertilizer. 6 seeds were planted on the dept of 1.5 cm on each soil sample. Germination index were calculated and recorded considering the time of planting to time of transplanting using equation 1. The seeds after growing into seedlings were transplanted to the field for further investigation. Portions of the field measuring 2 m by 5 m were properly arranged by clearing and tilling before application of fertilizers. However, the portions were prepared as follow: Portion T_1 = soil only, Portion T_2 = soil + Biofertilizer, and then portion T_3 = soil plus chemical fertilizer and were fitted into Randomized Block Design without replication as appeared in Table 2. Application of fertilizer was carried in the field on each portion one week before transplant to prevent fertilizer burn inherent with chemical fertilizer. Weeds and other unwanted growth in the farm were manually removed every two weeks. Data on leaf width, plant height, and number of leaves were collected from the portions starting 2 weeks after transplant to 12 weeks after transplant. The data were subjected to statistical analysis using F-test analysis at 95% confidence level.

$$Germination\ \ Index = \frac{Number\ \ of\ \ Seed\ \ Sprouted}{Number\ \ of\ \ Seed\ \ Planted} \times 100 \qquad (1)$$

2.9.2. Leaf width and leaf Area Index

Five (5) randomly selected leaves of tagged plants were measured with a measuring tape and a pair of calipers at 2, 4, 6, 8, 10 and 12 weeks, respectively after transplant. The number of leaves per plant were also determined. Leaf Area Index (LAI) was estimated as stated in Eq. (2), [23].

$$LAI = \text{leaf length (cm)} \times \text{leaf width (cm)} \times \text{No. of leaves/plant} \times 0.72/\text{area/plant} \qquad (2)$$

3. Results and discussion

3.1. Physio-chemical characterization of the raw materials

Table 1 showed that sawdust had high cellulose content (54.7 %), lignin content of (22.8 %) and poor nitrogen content (0.63 %). Lenox et al. [24] reported a cellulose concentration of 53 % and suggested that high resistant to biodegradation by sawdust was due to its high cellulose and lignin content. The initial contents of nitrogen in the sewage sludge (5.16 %), Chicken litter (4.96 %) and vegetable waste (5.9 %) were in conformity with what was presented by Harir et al. [25] and Asadu et al. [26] with the same organic wastes and suggested that the percentages were sufficiently high to maintain the microbial growth for biodegradation during composting. High concentration of organic matter in all the wastes sludge confirms that they are biodegradable and good

Table 2
Randomized Block design of 3 × 3 matrix without replication.

Treatments Block	1	2	3
1	T_1	T_2	T_3
2	T_3	T_1	T_2
3	T_2	T_3	T_1

Soil only → T_1.
Soil + Biofertilizer → T_2.
Soil + Chemical fertilizer → T_3.

substrates for production of biofertilizer. Vegetable waste had the highest percentage nitrogen (5.9 %) and phosphorus (6.3 %). Other wastes as shown in Table 1 had appreciable percentage phosphorus and potassium which make them amenable to be used in biofertilizer synthesis. Carbon to nitrogen ratio of sawdust (87.46 %) is an indication that it is not compactible as composites except when blended with other nitrogen rich agricultural wastes as suggested by Hargreaves et al. [5] and IFA [27]. The carbon to nitrogen ratio of sewage sludge (5.85 %), chicken litter (8.01 %) and vegetable waste (3.89 %) as shown in Table 1, showed that they all have carbon to nitrogen ratio below 20 % and hence are not advisable to be composted alone to avoid excess release of ammonia which causes odor during composting. This agrees with the report by Gajalakshmi and Abbasi [13], that carbon to nitrogen ratio below 20 % is an indication that excess nitrogen per degradable carbon results in ammonia production which can be lost through ammonia voliatilization, leaching from the composites mass and dentrification with associated pungent odors. High carbon to nitrogen ratio makes the process very slow due to insufficient nitrogen for microbial growths resulting in extended time for composting process [28,29]. As such, the sawdust used in this research served as the bulking agent which is the major carbon source for the organisms while other agro wastes which contained relatively high proportion of nitrogen served as the nitrogen source. Therefore, raw materials were blended to provide an initial carbon to nitrogen ratio that is good enough for active composting.

3.2. Identification of the microorganisms isolated from soil extracts blended with agricultural wastes

The microorganisms isolated were designated as 1–6 as evident in Table 3. Using the standard procedure for microbial identification and Bergey's Manual of Determinative bacteriology, the morphological characterization of the isolates were carried out and the results presented in Table 3.

Six organisms were identified (see Table 3). Three were presumed as *Streptomyces spp*, two were established as Rothia spp while one was established as *Actinomadura spp*. *Streptomyces spp* are known to be the largest genera of *Actinomycetes* according to Antoinetta et al. [18]. Identification of *Streptomyces* in this research gave credence on the choice of isolation material and isolation media. Therefore, inorganic starch *agar* medium is a suitable isolation media for *Actinomyces* which further buttress the result by [30]. The organisms were further characterized using biochemical analysis as presented in Table 4

As observed in Table 4, Isolate 2–6 tested positive to nitrate reduction test and therefore an indication that isolate 2–6 has possess the capacity to mineralize and fix nitrogen during composting. This corresponds with the reports by Ayitso and Onyango [31] and Azimi et al. [32]. According to Adeline and Ka [33] and Bakulin et al. [34], nitrate reduction test determines the capacity of an organism to reduce nitrate (NO_3^-) to nitrite (NO_2^-), ammonia, nitrous oxide and nitrogen via enzyme nitrate reductase. Nitrate reduction test also examines the capacity of the isolates to exhibit nitrification ability on nitrite and nitrate as well as the ability of the isolates to transform molecular nitrogen into nitrite. Therefore, isolate 2–6 are good agents for nitrogen mineralization and fixation.

It was observed from Table 4 that most of the bacteria isolates tested positive to sugar fermentation test. As quoted by Lenox et al. [24], sugar fermentation test usually reveal the ability of an organisms to reduce certain sugar/carbohydrate such as sucrose, dextrose, mannitol, maltose and lactose. *Actinomycetes* usually have a dual function which include; mineralization and nitrogen fixation, degradation and breaking down of carbohydrates (lignin and cellulose) and solubilization of phosphorus according to Godliving and Yoshitoshi [35],Asadu et al. [36,37]. Therefore, most of the organisms as evident in Table 4 are good agents for sugar fermentation (degradation of carbohydrates). Furthermore, Table 4 also revealed that isolate 2, 3 4 and 5 tested positive to catalase test. Catalase is an enzyme formed by microorganisms dwelling in oxygenated surroundings to neutralize toxic kinds of oxygen metabolites e.g. H_2O_2 [18,33]. The catalase enzyme neutralizes the bactericidal influences of H_2O_2 and safeguards the organisms [36]. The major metabolite that are usually produced by *Actinomycetes* which inhibits their function is hydrogen peroxide, therefore isolate 2, 3, 4 and 5 revealed in this research are good agents for biofertilizer synthesis since they can breakdown carbohydrate and at the same time produce catalase enzymes which neutralizes hydrogen peroxide.

3.3. Analysis of the potential of the microbial Isolates in decomposing sawdust

Sawdust is somewhat recalcitrant to biodegradation due to high lignin and cellulose content as revealed in Table 1. The ability of the microbial isolates to breakdown this cellulose in sawdust has been demonstrated and the outcome of the investigation presented as shown in Table 5.

It was observed from Table 5, that the microbial isolates possess various capabilities of reduction of the cellulose content of sawdust. This shows that the isolates are proficient in using sawdust as source of carbon and energy for growth. This agrees with the results presented by Lenox et al. [24], Tuomela et al. [14] and Deeble and Lee [38], which demonstrated reduction of carbon content in organic substance by enzymatic hydrolysis under microbial action. Table 5 indicates that isolate 6 (*Rothia spp*) has the maximum percentage cellulose reduction (21.60 %) among the bacteria density isolated followed by isolate 5,(*Rothia spp)* (18.74 %) and then isolate 4 *Streptomyces spp* (15.31 %), isolate 1 *Actinomadura spp* (11.30 %), isolate 2 (9.80 %) and isolate 3 *Streptomycetes spp* (8.58 %). This indicates that isolate 6 *Rothia spp* has greater capacity to secrete "cellulase" enzyme which degrade carbohydrate in organic compounds. Sustainability of cellulose reduction by the bacteria isolates at 42 *ENTITYNOTDEFINED!!!* equally showed that some of the bacteria isolates are thermotolerant *Actinomycetes*, [39,40,36]. These reports have shown that indigenous microbes have the potential to breakdown agricultural wastes. Table 5 further indicates that, there was a massive improvement in the breakdown of cellulose in sawdust when

Table 3
Morphological characteristics of the isolates.

Isolates	Organism genera	Characteristics of hyphae and mycelium
1	*Actinomadura spp*	Detailed branched vegetative hyphae with dense non- fragmenting substrate mycelium.
2	*Streptomyces spp*	Detailed branched mycelium with small fragmentation.
3	*Streptomyces spp*	Detailed branched mycelium with small fragmentation.
4	*Streptomyces spp*	Detailed branched mycelium with small fragmentation.
5	*Rothia spp*	Detailed branched vegetative hyphae which grow and penetrate the agar medium. Aerial mycelium is available.
6	*Rothia spp*	Detailed branched vegetative hyphae which grow and penetrate the agar medium. Aerial mycelium is available.

Table 4
Biochemical identification of the microbes.

Identification Parameter	Organism 1	Organism 2	Organism 3	Organism 4	Organism 5	Organism 6
Gram reaction	positive	positive	positive	positive	positive	positive
Spore staining	positive	positive	positive	positive	positive	positive
Catalase test	negative	positive	positive	positive	positive	negative
Nitrate reduction	negative	positive	positive	positive	positive	positive
Indole test	negative	negative	negative	negative	negative	negative
Urease test	negative	negative	positive	positive	negative	negative
Mannitol test	negative	negative	negative	negative	positive	positive
Glucose test	positive	negative	negative	positive	positive	positive
Sucrose test	negative	positive	negative	negative	positive	positive
Implicated organism	*A.spp*	*S. spp*	*S. spp*	*S. spp*	*R. spp*	*R. spp*

A. spp = Actinomadura spp.
S. spp = Streptomyces spp.
R. spp = Rothia spp.

Table 5
Proportional degradation of cellulose in sawdust by bacteria.

Isolated organisms	Genera	Initial cellulose conc (%)	Cellulose conc (%) after incubation	Percentage difference
1	*A.* spp	54.7	43.40	11.30
2	*S.* spp	54.7	44.90	9.80
3	*S.* spp	54.7	46.12	8.58
4	*S.* spp	54.7	39.39	15.31
5	*R.* spp	54.7	35.96	18.74
6	*R.* spp	54.7	33.1	21.6
Control		54.7	54.54	0.16

compare with prior research by Lenox et al. [24]. Therefore, it can be concluded that isolate 4 (*Streptomycetes spp*) and isolate 6 (*Rothia spp*) has the greater potential to breakdown cellulose in organic wastes.

3.4. Screening of the microbial potential in mineralizing nitrogen during composting

The ability of the microbial isolates in mineralizing nitrogen during composting was investigated and the results presented as shown in Table 6.

From Table 6, it was observed that the nitrogen content of the mixtures increased significantly after composting with individual organisms except the control. This could be as a result of high protein content of chicken litter that might have been changed to nitrate ion and made available as nitrogen through nitrogen fixing bacteria (*Actinomycetes*). The results of the experiments show that the microbes have the capacity to reduce biodegradable organic wastes and release nitrogen and other important soil nutrients needed by plants. This corroborate the findings of Bakulin et al. [34]; Shoji et al. [41] and Godliving and Yoshitoshi [35], that indigenous *Actinomycetes* has the capacity to increase the concentration of nitrogen during degradation of nitrogen rich agricultural wastes. Isolate 5 (*Rothia spp*) recorded the highest increase in percentage nitrogen (6.87 %) followed by isolate 6 (*Rothia spp*) (6.33 %), isolate 4 (*Streptomyces*) (5.76 %), isolate 3 (*Streptomyces spp*) (4.18 %), isolate 2 (*Streptomyces spp*) (3.71 %) and then isolate 1(*Actinomadura spp*) (1.85 %). The least percentage nitrogen recorded by isolate 1 (*Actinomadura spp*) as shown in Table 6 buttressed the results of the biochemical test in Table 4 which showed that *Actinomadura spp* tested negative to nitrate reduction test and hence does not have potential to mineralize nitrogen during composting. These microbes transform atmospheric nitrogen to nitrite which are accessible to plants on dissolution. Nitrogen content of the mixture without inoculums (control) did not show any appreciable increase after composting, and this buttressed the earlier claim by Edriss et al [42] and Dumitrescu et al. [43], that the principal factor which aids the increase in percentage nitrogen during composting are the autochthonous microbes. The results of the experiments also revealed that the isolated microbes have the tendency to reduce biodegradable organic wastes and release nitrogen and other important soil nutrients needed by plants, hence the stock of the organisms are good agents for conversion of organic wastes into biofertilizer. However, it can be concluded that isolate 4,5 and 6 has the greater tendency to mineralize nitrogen during composting.

Table 6
Mineralization of nitrogen by the bacteria isolates.

Isolates	Genera	Initial amount of nitrogen in the mixture (%)	Amount of nitrogen in the mixture after composting (%)	Percentage increase
1	*A. spp*	4.96	6.81	1.85
2	*S. spp*	4.96	8.67	3.71
3	*S. spp*	4.96	9.14	4.18
4	*S. spp*	4.96	10.72	5.76
5	*R. spp*	4.96	11.77	6.87
6	*R. spp*	4.96	11.29	6.33
Control	–	4.96	6.02	1.06

Isolate 4 and 6 were used for further composting operation in this work since they have greater potential to breakdown sawdust and proven ability to mineralize nitrogen.

3.5. Analysis for one factor at a time (OFAT) for the mineralization/solubilization of nutrients during composting

3.5.1. Effect of inoculums type

The effect of using the isolates individually (*Streptomycetes spp* and *Rothia spp*) and then combined effect of the organisms (*Consortium spp*) on the yield of nitrogen from substrates S1, S2, and S3 were investigated and demonstrated as shown in Figs. 1–3. It was observed that Figs. 1 and 2 showed the same trend for the yield of nitrogen but never the case with Fig. 3. The highest percentage yield of nitrogen after 30 days from S_1 during composting as shown in Fig. 1 was (5.2 % with *Streptomyces spp*, 7.9 % with *Rothia spp*, and 9.1 % with *Consortium spp*), from S2 as shown in Fig. 2, it was (10.2 % with *Streptomyces*, 10.9 % with *Rothia spp*, and 12.5 % with *Consortium spp*), while it was (0.23 % with Streptomyces, 0.13 % with *Rothia spp* and 0.12 % with *Consortium spp*) for substrate S3 as shown in Fig. 3. It could be seen that there was a greater yield of nitrogen with combined effect of the organisms (*Consortium spp*) when compared with individual organisms as shown in Figs. 1–3. This is in agreement with the results reported by Christianson et al, [44] and Edriss et al, [42], where they stated that composting with combined organisms with different capacity to mineralize nitrogen has a greater influence on the yield of nitrogen. It was also observed that *Rothia spp* has a greater potential to mineralize nitrogen than *Streptomyces spp* as shown in Figs. 1 and 2. This could be attributed to the ability of *Rothia spp* to secrete more of the enzyme "nitrogen reductase" which is the principal enzyme that converts residual nitrogen in wastes and atmospheric nitrogen into nitrite which further buttressed the results in Tables 4 and 6.

Fig. 3 shows that percentage nitrogen was in downward trend for the three organisms during composting of substrate S_3. The decrease in percentage nitrogen in substrate S3 buttressed the earlier report by Edriss et al, [42] and Dumitrescu et al., [43], that sawdust traps down nitrogen while trying to breakdown carbohydrate thus depriving the microbes of nitrogen. Edriss et al., [42], reported 2.75 % of nitrogen yield after 46 days of composting of a mixture of sawdust plus sewage sludge plus vegetable wastes without innoculums; Tseng et al. [45], reported 6.45 % of nitrogen after 35 days of composting of mixtures of sawdust plus sewage sludge plus vegetable wastes with *Streptomyces spp*; reported 5.6 Tumuhairwe et al, [7] reported 5.6% yield of nitrogen after 30 days of composting of sawdust plus cow dung plus vegetable waste with *Chrysteobacterium spp*. Tiquia et al., [40], reported 7.4 % yield of nitrogen after composting of sawdust plus vegetable waste plus sewage sludge with *Gluconacetobacter spp*. After comparing the results of this investigation with prior research, it can be concluded that the use of microbial inoculums

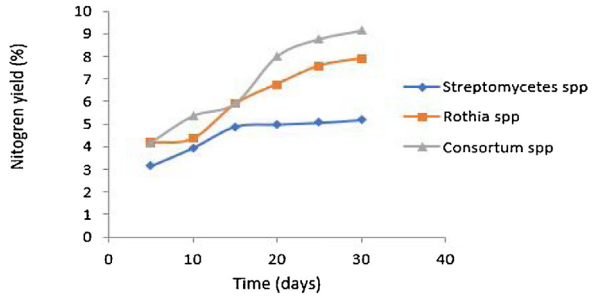

Fig. 1. Effect of inoculums type on substrate S_1 during composting.

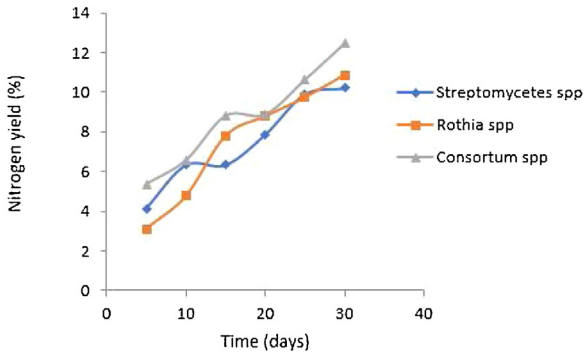

Fig. 2. Effect of inoculums type on substrate S_2 during composting.

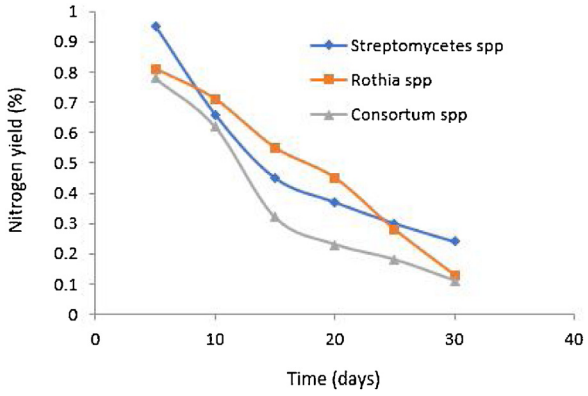

Fig. 3. Effect of inoculums type on substrate S_3 during composting.

improved nutrient yield but greater improvement was achieved with combined effect of the organisms (consortium) of the organisms.

3.5.2. Effect of substrate type

The effect of varying the substrate combination on the mineralization of important soil nutrients such as nitrogen was investigated with three different substrates S_1, S_2 and S_3 and each was composted with three different inoculums as shown in Figs. 4–6. Substrate S_3 served as the control. From Fig. 4, the highest yield of nitrogen from the substrates after 30 days of composting with *Streptomyces spp* are 5.24 % from S_1, 10.30 % from S_2 and 0.25 % from S_3. From Fig. 5, the percentage yield after 30 days of composting with *Rothia spp* was 7.9 % from S_1, 10.99 % from S_2 and 0.14 % from S_3 while it was 9.33 % from S_1, 12.58% from S2 and 0.38% from S3 after

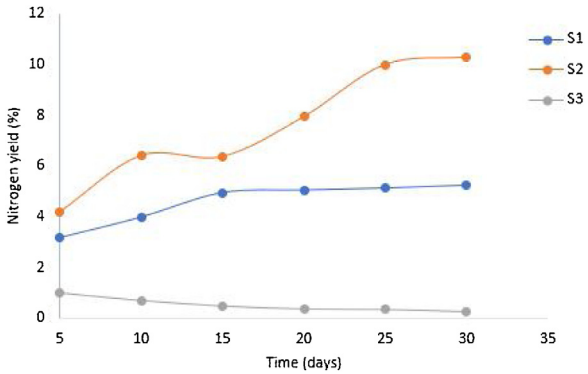

Fig. 4. Mineralization of nitrogen during composting of substrates with *streptomycetes spp*.

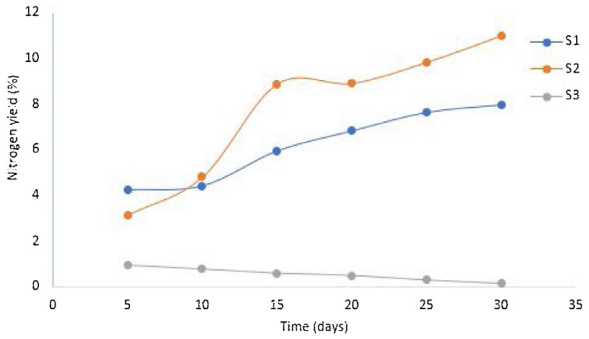

Fig. 5. Mineralization of nitrogen during composting of substrates with *Rothia spp*.

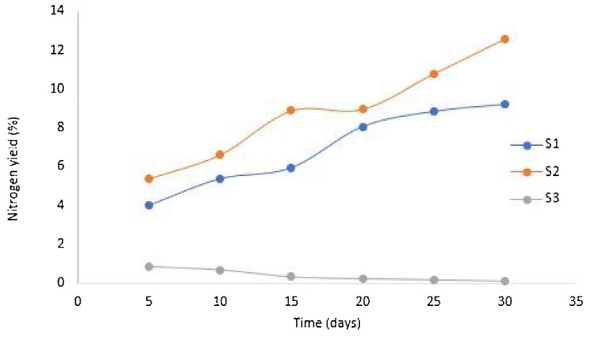

Fig. 6. Mineralization of nitrogen during composting of substrates with *consortium spp*.

30 days of composting with *Consortium spp* as shown in Fig. 6. It was observed that substrate S_2 gave a greater yield of nitrogen after composting with the three organisms followed by substrate S_1 as shown in Figs. 4–6. This could be as a result of high concentration of protein in sewage and high percentage composition of nitrogen in vegetable waste. Similar results have been reported by Samaras et al. [46] and Maboeta and Rensburg [47]

Substrate S_1 and S_2, showed a good tendency to release nitrogen during composting as shown in Figs. 4 and 5 which indicates that they are good biofertilizers. Greater percentage of nitrogen was recorded after composting the substrates with consortium spp as shown in Fig. 6, which further indicates that consortium spp is more active in mineralization of nitrogen during composting. Nitrogen in substrate S3 (control) decreased significantly with time after composting with the three inoculums as shown in Figs. 4–6. This is in concurrence with the report by Christian et al. [36]; Edriss et al. [42]; Dumitrescu et al. [43], Olayinka and Adebayo [48] and Adeoye et al. [8] that sawdust withdraws nitrogen from the compost as it decomposes. From the results of this investigation, it can be concluded that, substrate S_2 has more potential to release nitrogen during composting.

3.5.3. Effect of composting time on potential of hydrogen (pH)

The main factor that decides the rate at which nutrient is released during composting is pH even though it is a dependent variable during composting. The variation in pH of the composts as noticed during composting was stated as depicted in Figs. 7–9. It was discovered that the pH of the composts S_1 and S_2 reduced slightly between the 5th day and 10th day from the commencement of the composting exercise and then increased progressively from 15th day till the end of composting. This could be due to the accumulation of proteins in sewage sludge and chicken litter in S_2 and S_1 which was acted upon by the microbial isolates and converted to peptides then to amino acids, and thereafter

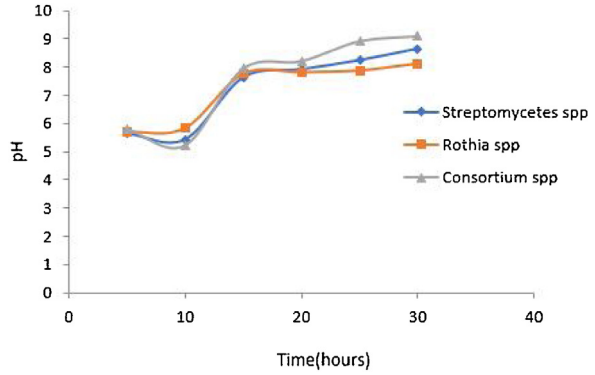

Fig. 7. Effect of time on pH of substrate S_1.

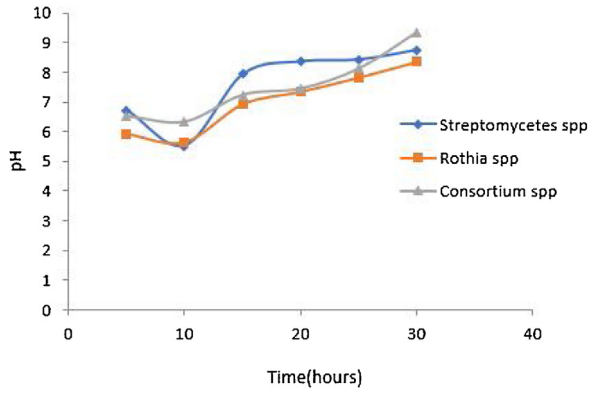

Fig. 8. Effect of time on pH of substrate S_2.

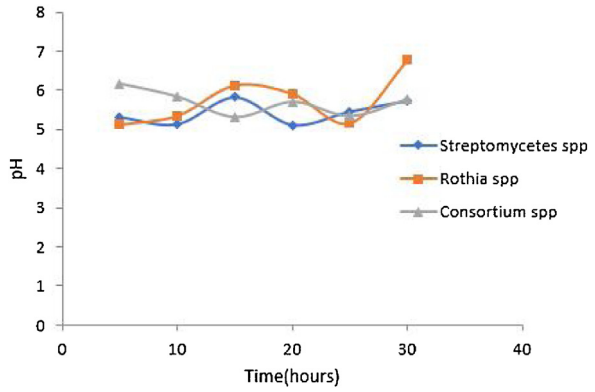

Fig. 9. Effect of time on pH of substrate S_3.

ammonia gas that dissolved in water to form ammonium ion (NH_4^+) which made the compost alkaline and moved the pH up progressively. Also, the conversion of carbohydrates in sawdust to simple sugar which was further converted to inorganic acid (H_2CO_3) lowered the pH by increasing the acidity level of the compost. Similar results were reported by Fang and Wong [49] and Gajalakshmi and Abbasi [13], were they reported the range of pH values suitable for bacteria development as between 6.0–7.5 while fungi survive an environment in the range of pH 5.5–9.0. pH of substrates S_1 and S_2 initially decreased due to the breaking down of organic matter in agricultural wastes containing carbohydrate which led to organic and inorganic acids formations which lowers the pH (increases acidity). However, further increasing in pH as observed in Figs. 7 and 8 was due to decomposition of organic

matter with nitrogenous compounds which led to NH_3 formation which react with water **to** form ammonium compound.

The pH of Substrate S_3 remained acidic and irregular althrough the composting time as depicted in Fig. 9. This could be attributed to the breakdown of cellulose in sawdust which produced organic acid and inorganic acid during composting without any nitrogenous compound to produce ammonia to counter balance the acid. This buttresses the fact that composting sawdust alone would be counterproductive in bio-fertilizer production and need to be blended with other nitrogen rich agricultural wastes for self-buffering of the compost; this is in agreement with the view of [50] and [43].

3.5.4. Effect of solid water ratio (moisture content)

Effect of water solid ratio on the mineralization of nutrients during composting of substrates S_1 and S_2 was investigated using three different microbial inoculums (*Streptomycetes spp*, *Rothia spp* and *Consortium spp*) and the results obtained were presented as shown in Figs. 10 and 11. Effect of moisture content was not conducted with substrate S_3 because in this research, there has been a substantial evidence that composting sawdust alone cannot give a good biofertilizer.

From Figs. 10 and 11, it was observed that the range of moisture between 50–60% gave the highest amount of nitrogen with consortium spp. This agrees with the report by [13], Trautmann and Krasny [50,51], Christian et al. [36,2,9] and [10] where they reported the optimum moisture for the mineralization of nutrients in organic waste during composting with microbial inoculums to be between 50–60 %. Edriss et al. [42] reported optimum moisture between 55–65 %. Substrate S_2, gave the highest percentage nitrogen at the moisture range 50–60 % during composting with *Consortium spp*. This might be due to high protein concentration in sewage sludge which is the main component of S_2.

Constant aeration of a compost system improves the quality of biofertilizer (products). Excess moisture content restricts the diffusion of oxygen to the compost system and as such change the system to anaerobic fermentation rather than desired aerobic fermentation; Also, low moisture content reduces microbial growth and multiplication rate thereby delaying the system [52,53] and [54]. Therefore, one can conclude that the optimum moisture for the mineralization of nutrients in substrates S_1 and S_2 falls between 50–60 %.

3.5.5. Effect of substrate dosage

Effect of substrate dosage ratio on the mineralization of nutrients from substrate S_1 and S_2 was investigated by composting each substrate with three different inoculums (Streptomycetes, Rothia and Consortium) at varying ratios. The results were presented as shown in Figs. 12 and 13.

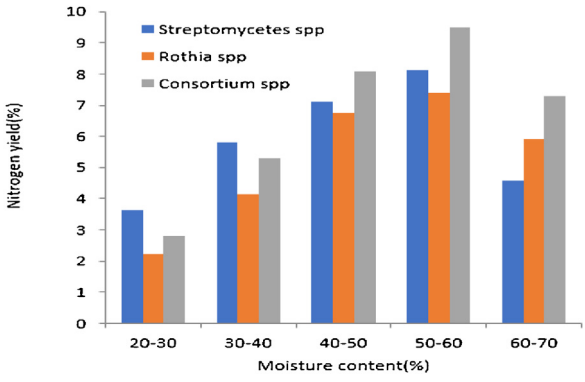

Fig. 10. Effect of moisture content on mineralization of nitrogen in substrate S_1.

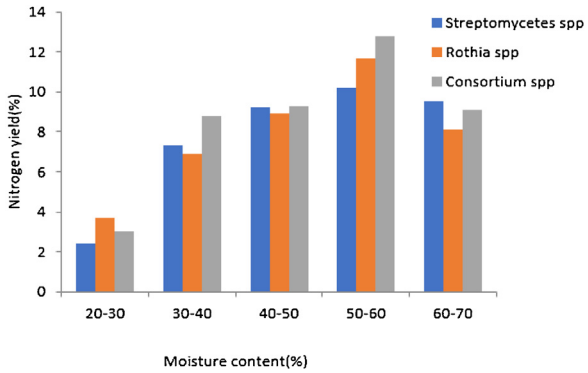

Fig. 11. Effect of moisture content on mineralization of nitrogen in substrate S_2.

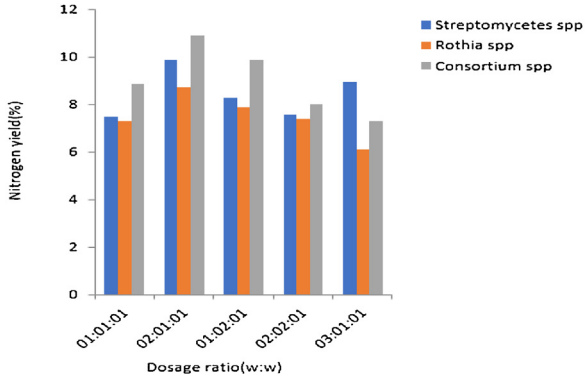

Fig. 12. Effect of dosage ratio on mineralization of nitrogen in substrate S_1.

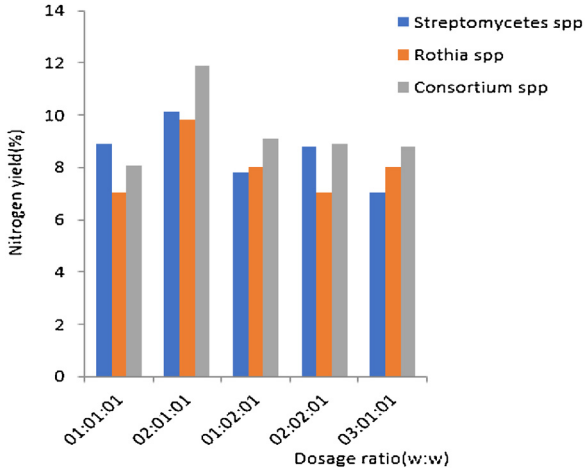

Fig. 13. Effect of dosage ratio on mineralization of nitrogen in substrate S2.

It was observed that ratio 2:1:1 w:w produced the best results for the mineralization of nitrogen from S_1 and S_2. Also, greater amount of nitrogen was produced at the same ratio (2:1:1w:w) with consortium spp for each substrates. At the same ratio (2:1:1), substrate S_2 released greater amount of nitrogen. This would likely be as a result of high protein concentration in the sewage sludge. It could be concluded that ratio 2:1:1(w: w) is the best combination for the substrates for the mineralization of nutrients during composting. Similar ratio was reported by [2]; and [51]

Table 7
Analysis of treated and untreated Soil.

Parameters	Untreated soil	Soil + biofertilizer	Soil + chemical fertilizer
Sodium	0.0245 mg/100 g	0.034 mg/100 g	0.0245 mg/100 g
Calcium	0.871 mg/100 g	0.630 mg/100 g	0.871 mg/100 g
Phosphorous	1.9 %	5.81 %	7.32 %
Potassium	1.23 %	3.74 %	5.1 %
Aluminum	0.231 mg/100 g	0.043 mg/100 g	0.231 mg/100 g
Magnesium	0.325 mg/100 g	0.6134 mg/100 g	0.325 mg/100 g
pH	5.9	8.12	6.5
Free air space/Porosity	39.6 %	58.3 %	40.8 %
Bulk density	2.386/1000 g	3.152 g/1000 g	2.435 g/1000 g
Texture class	Sandy-loam	Sandy-loam	Sandy-loam
Carbon	11.24 %	56.7 %	25.7 %
Organic Matter	3.88 %	79.8 %	28.1 %
Nitrogen	0.214 %	6.86 %	7.88 %

3.6. Analysis of results from field exercise

Viability and effectiveness of biofertilizer produced are usually ascertained through field study. After preparing the soil with biofertilizer produced using substrate S_2, Soil investigation was carried out and the results are shown in Table 7. Fertilizer produced from substrate S_2 was chosen due to higher nutrient composition. As can be seen in Table 7, the concentration of nitrogen in soil only (untreated) is as low as 0.214 %, in soil blended with biofertilizer it is 6.86 %, however, it is 7.88 % in the soil blended with mineral fertilizer (chemical fertilizer).

Furthermore, Table 7 demonstrated that there was in like manner higher enhancement in the concentrations of organic matter and organic carbon of the soil blended with biofertilizer compared to the untreated soil and soil blended with chemical fertilizer. These observations agreed strongly with the report by Daramola et al. [55] and Asadu et al. [9,10], where they claimed that biofertilizer improves the organic matter content of soil which is a precursor for soil structure improvement due to high concentration of polymer compounds called polyhydroxyalkanoate and poly-acrylamide. Additionally, there was greater enhancement in free air space (58.3 %) as against (39.6 %) in the untreated soil which guaranteed a proficient gaseous exchange among the soil microorganisms. It can also be seen from Table 7 that there was improvement in the amount other soil mineral elements such sodium, calcium, aluminum and magnesium as against the untreated soil and soil blended with chemical fertilizer. These few observations demonstrated the advantages biofertilizer (organic fertilizer) have over chemical fertilizer.

Impact of soil treatments on germination time and germination rate were calculated using (Eq. 1) and the results presented as depicted on Fig. 14. 100 % germination index was recorded within five days when both biofertilizer and chemical fertilizer were applied after planting, compared with 80 % germination index recorded for the untreated soil (control) after same period of time. Germination of seeds in the soil media treated with both fertilizers was progressively quick. It could be concluded that application of fertilizer to the soil improved the breakdown of the seed coats, thereby increasing the pace at which the seeds absorbs water. These observations further demonstrated the efficiency of the biofertilizer produced in this work to enhance seed germination and ability to compete with chemical fertilizer.

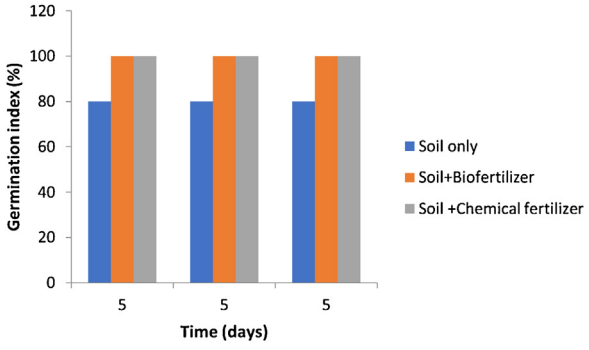

Fig. 14. Effect of different soil media on germination of seed.

Between week 8 and 10 as evident in Table 8, the percentage nitrogen accumulated in the leaves of okra and maize planted in the soil blended with biofertilizer had increased significantly beyond the original percentage nitrogen in the soil. This could be credited to the nitrogen fixing microorganisms (bacteria isolates) in biofertilizer which dependably changes over environmental nitrogen (Nitrogen in the air) into available nitrites for plants immobilization. This submission concurred with the view of Bakulin et al. [34] Chang and Yang [56] and Adeoye et al. [8].

Increase in percentage nitrogen in leaf of okra and maize as shown in Table 8 could be as a result of the immobilization of the nitrogen released by the fertilizers in the soil media. According to Nottidge et al. [57] and Bakulin et al. [34], percentage increase in nitrogen in the leaf of crops planted with biofertilizer is mainly a direct result of microbial nitrogen fixation. However, Table 8 further demonstrated that the percentage nitrogen in leaf of crops widened as the number of weeks goes by. Crops grown in soil media with chemical fertilizer gave higher percentage accumulation of nitrogen. This trend gave credence to the earlier results of Daramola et al. [55] and Asadu et al. [9,10] that chemical fertilizer releases its nutrients faster compared with the biofertilizer. Subsequently, immobilization is higher in crops with little time and exposes the crops to nutrient deficiency due to downward movement of the nutrients in the soil as a result of rain and soil erosion.

However, as observed also in Table 8, there was a slow growth in percentage concentration of nitrogen in crops grown with biofertilizer which demonstrated the famous nature of gradual and consistent release of nutrients by organic fertilizer as quoted by Allen [58]. 6th, 7th and 8th week were the period more accumulation was recorded as evident in Table 8 for both crops. This period was explained by Jean-Claude et al. [23] as the optimum growth time. Also, there was a sharp decrease in percentage nitrogen in the leaves of crops planted with chemical fertilizer while it was a slow pace for crops planted in the soil media with biofertilizer between 10th week and 12th week. This may not be unconnected to the crops growing to their pick and movement of nutrients from old leaves to younger ones (re-imobilization). Limited percentage of nitrogen in the leaves of crops grown in the untreated soil (control) as demonstrated also in

Table 8
Effect of soil media on crops performance.

Weeks after transplant Treatments	2	4	6	8	10	12	Average
	Amount of nitrogen in okra leaf (%)						
Soil only	0.65	0.79	0.86	0.83	0.64	0.45	0.703
Soil + Bio-fertilizer	1.67	2.34	4.92	7.27	8.37	8.13	5.42
Soil + Chemical Fertilizer	2.31	3.65	5.74	8.12	8.06	6.48	5.96
	Amount of nitrogen in maize leaf (%)						
Soil only	0.71	0.81	0.77	0.89	0.41	0.36	0.66
Soil + Bio-fertilizer	1.23	2.76	3.82	6.47	7.12	8.01	4.74
Soil + Chemical Fertilizer	1.87	4.32	5.33	7.94	8.77	7.83	6.01
	Plant height for okra (cm)						
Soil only	5.1	6.4	7.7	15.6	19.3	23.1	12.87
Soil + Bio-fertilizer	10.8	12.3	23.2	34.6	36.8	38.9	26.1
Soil + Chemical Fertilizer	10.7	12.7	26.8	35.8	39.1	42.3	27.9
	Plant height for maize (cm)						
Soil only	5.7	15.6	19.7	25.3	37.8	44.3	24.73
Soil + Bio-fertilizer	10.6	38.4	57.6	88.4	123.1	153.1	78.53
Soil + Chemical Fertilizer	10.8	44.1	61.8	90.6	128.4	155.2	81.82
	Leaf number (okra)						
Soil only	4	5	7	8	9	9	7.0
Soil + Bio-fertilizer	6	7	11	13	15	15	11.2
Soil + Chemical Fertilizer	6	7	12	14	17	17	12.2
Treatment	Leaf number (maize)						
Soil only	3	3	4	5	7	7	4.83
Soil + Bio-fertilizer	5	7	9	10	15	15	10.2
Soil + Chemical Fertilizer	5	8	9	11	16	16	10.8
Treatment	Leaf width for okra (cm)						
Soil only	6.2	8.8	11.6	13.7	14.2	16.6	11.85
Soil + Bio-fertilizer	11.4	15.9	18.1	25.3	27.6	27.7	21.0
Soil + Chemical Fertilizer	11.5	16.3	17.9	26.8	28.4	28.9	21.63
Treatment	Leaf width for maize (cm)						
Soil only	1.8	2.3	2.8	3.3	3.6	3.6	2.36
Soil + Bio-fertilizer	2.4	4.9	6.1	8.7	8.9	10.4	6.90
Soil + Chemical Fertilizer	2.5	5.3	6.8	9.1	9.7	11.2	7.43

Table 8 could be credited to insufficient amount of important soil nutrients (nitrogen) required by crops which gave credence to the case of Lesueur et al. [59], which opined that fertilizer usage stimulates the nutrients availability in the soil.

Table 8 further demonstrated the effect of fertilizers on plant height. It was observed that crops responded positively to the impact of fertilizer in the soil. Week four and eight demonstrated the period of speedy growth, although it was noticed to be sluggish towards week ten and twelve. The sluggish growth observed from both crops could be a prove that the crops were tending towards maturity. The difference in average height between the crops planted in soil subjected to biofertilizer and that of chemical fertilizer was observed to be very small which demonstrated that biofertilizer can be used in place of chemical fertilizer. The stunted growth observed with crops grown in the untreated soil is an evidence of importance of fertilizer application before farming. The little differences in average performance achieved by the plants in the soil subjected to organic fertilizer could be attributed to the organic fertilizer's rate of nutrient release. According to Allen [58] and Daramola et al. [55], the rate at which organic fertilizer releases its nutrients to plants is gradual and does not support boom and burst usually demonstrated by chemical fertilizers. This causes great harm to the soil structure. Employment of organic fertilizers are of immense advantage because chemical fertilizer causes great harm to the soil structure and are also more expensive compared to organic fertilizers.

However, the leaf number emergence observed by both crops demonstrated that the average number of leaves developed by crops grown with biofertilizer is almost equivalent to the average number of leaves developed by crops grown with chemical fertilizer except the control which developed a limited number within the same weeks as shown in Table 8. Limited number of

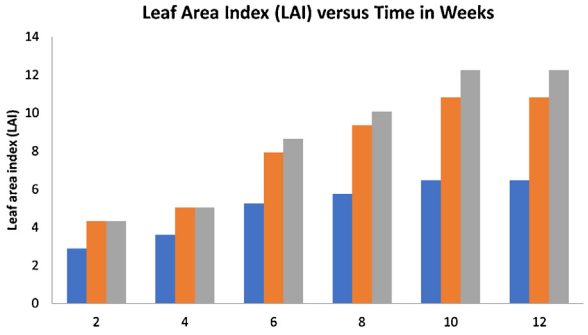

Fig. 15. Effect of soil media on leaf area index (LAI) of Okra.

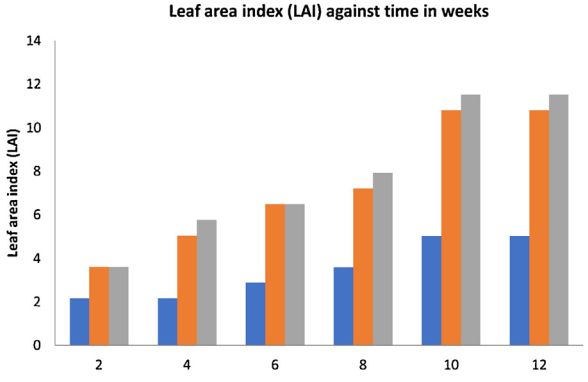

Fig. 16. Effect of soil media on leaf area index (LAI) of maize.

Table 9a
Analysis of variance table for *Okra* performance in soil media.

Source of Variance	Degree of freedom (df)	Sums of Squares (SSQ)	Estimate of Variance	F-value
Blocks	2	30.44	15.22	2.04
Treatments	2	120.87	60.43	8.10
Error	4	29.84	7.46	–
Total	8	152.15	–	–

Table 9b
Analysis of variance table for *Maize* performance in soil media.

Source of Variance	Degree of freedom (df)	Sums of Squares (SSQ)	Estimate of Variance	F-value
Blocks	2	201.23	100.62	7.01
Treatments	2	251.32	125.66	8.75
Error	4	57.41	14.35	–
Total	8	509.96	–	–

The Critical F-value ($F_{0.05,\ 2,4}$) = 6.94.

leaves affects the rate of photosynthesis in plants which further affects the number of fruits produced.

The leaf area was also studied as demonstrated in Table 8. The results showed that the average leaf area of crops grown with chemical fertilizer and biofertilizer were very close which further demonstrated the effective competitiveness between biofertilizer and chemical fertilizer. The improvement in the leaf area as against the control highlights the importance of fertilizer application because wider leaves attracts more energy from the sun and hence increase in photosynthesis and improved fruit yield.

Using Eq. 2, leaf width and number of leaves as shown in Table 8 were applied in the simulation of leave area index (LAI) for each plant as demonstrated in Figs. 15 and 16. It was observed that organic fertilizer and chemical fertilizer influenced positively the LAI of both crops when compared with crops grown with soil only (control). The LAI of crops grown with organic fertilizer are in a very good competition with the LAI of crops grown with chemical fertilizer which further proved the efficacy of organic fertilizer produced in this work. The optimum growth of plants in the soil media as observed from Figs. 15 and 16 occurred at 10th week after planting since the LAI were at maximum at 10^{th} week and thereafter remained the same. The optimum growth of plants which promoted the LAI for both crops could be attributed to role of nitrogen on the synthesis of chlorophyll, enzyme and proteins which in turn increases vegetative growth [23].

Fruit emergence was observed in okra plants from 8th week after transplant. 2 fruits were observed in some okra plants grown with chemical and organic fertilizer in the 8^{th} week after transplant while none was observed in the control. In the 10^{th} week, 6 fruits were observed in some plants grown with biofertilizer, 7 were observed in some plants grown with chemical fertilizer, while 3 were observed in the control. In the 12th week, some plants grown with chemical and biofertilizer produced 11 fruits while 5 were observed in the control.

Tasseling was observed in the maize plants grown with chemical and biofertilizer in the 8th week followed by silking in the 9th week, while tasseling was observed in the control the in the 10th week followed by silking in the 11th week with a reduced height. The delay in tasseling and silking development in the control could be attributed the stunted growth that characterized the plants due to insufficient nitrogen in the soil. Blister started in the maize plants grown with both fertilizers 12th day after silking.

From the observations, it could be seen that chemical fertilizer slightly had a greater yield of okro fruits in the 10th week but in the 12th week, the yield was equal. The reproductive developments in maize plants for both fertilizers are almost the same as observed. This is an indication that biofertilizer can comfortably replace chemical fertilizer with amplified investigation.

Statistically, the results from the field (Randomized block design) were examined using F-test analysis according to the technique proposed by Grolemud and Wickman [60], to choose if there is any accurate contrast among the three treatments as designated by T_1, T_2 and T_3 in the field. From Tables 9a and 9b, it can be deduced that application of biofertilizer and chemical fertilizer significantly and statistically affected the performance of okra and maize. This conclusion was in accordance with the suggestion of the calculated F-value and critical F-value. The calculated F-value of the treatments for okra was found to be 8.10 while that of maize was found to be 8.75 which are both more than critical F-value of 6.94 and hence depict that the observation is against null hypothesis (H_0) i.e ($F_{observed} > F_{critical}$). Therefore, application of organic fertilizer and chemical fertilizer had positive influences on the plant height, number of leaf emergence and leaf width.

4. Conclusion

Biochemical characterization of six genera of organisms isolated revealed that apart from *Acinomadura spp*, the rest are good agent for the breakdown of lignin and cellulose in organic waste during composting while *Streptomycetes spp* and *Rothia spp* have the potential to secrete an enzyme nitrogen reductase, which mineralize nitrogen during composting. Composting of agricultural wastes with microbial inoculums *Streptomycetes spp* and *Rothia spp* produced a good biofertilizer as regards to mineralization of important soil nutrient such as nitrogen but combination of the two organisms (*Consortium spp*) produced better results. Finally, Substrate S2 (Sawdust + Sewage + Vegetable waste) was found to be a better source of nitrogen after comparing the results of the characterization of biofertilizer produced from it with others.

Declaration of Competing Interest

The authors above categorically declare there no competing interest of any form that exist

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Appendix A. Supplementary data

Supplementary material related to this article can be found, in the online version, at doi:https://doi.org/10.1016/j.btre.2020.e00493.

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