***Original Research Article***

# Changes in Microbial Population Numbers During Composting of Some Organic Wastes in Greenhouse




## ABSTRACT

**AIM:** The study identified and enumerated microorganisms associated with the composting of some organic wastes using the plate count method

**Study Design:** The different wastes were allowed to decompose for 70 days in a greenhouse using the modified windrow method of composting. Standard microbiological methods were used to monitor temperature changes in compost piles as well as changes in bacterial and fungal populations.

**Place and duration of study:** This study was carried out at an agricultural research farmland in the Federal University of Technology, Owerri, Nigeria.

**Methodology:** Seeds of *Capiscum chinense* were planted in the soil samples in a greenhouse. Rhizosphere soil was collected for analysis to identify the bacterial composition of the rhizosphere soil

.

**Results:** Microbial populations increased concurrently with temperature during the first 3 – 4 weeks of composting except however for faecal coliforms and *Salmonella.* The highest temperature recorded was 60$^{o}$C for cow dung (CD) compost pile while at maturity the temperature in all the compost piles ranged between 27°C to 30°C. The bacterial colony forming units were higher than fungal colony forming units throughout the composting process for both mesopholic and thermophilic microorganisms. The population of mesophilic organisms increased in the first 14 – 15 days; for cow dung, the initial total heterotrophic bacteria count (THBC) and total coliform count (TCC) were 2.4 x$10^7$ cfu/g and 5.0 x$10^5$ cfu/g respectively and increased to 2.5x$10^8$ cfu/g and 1.7x$10^7$ cfu/g for THBC and TCC, respectively, after the 14$^{th}$ day. Thermophilic bacteria dominated all the composting systems after the 21$^{st}$ day and lasted till the 35$^{th}$ day except for cow dung compost where thermophilic temperatures were still observed on the 45$^{th}$ day with a THBC of 6.3x$10^6$ cfu/g on the 49$^{th}$ day. Faecal coliforms and

*Salmonella* were completely eliminated in all the compost systems after the 28th day at temperatures between 47°C – 60°C..

**Conclusion:** Organic wastes when managed properly through the application of knowledge of composting can be transformed into beneficial materials for human and agricultural use.

**Keywords:** composting, mesophilic organisms, thermophilic microorganisms, total heterotrophic bacteria count, total coliform count

## 1. INTRODUCTION

Composting is the process whereby organic wastes are reduced to organic fertilizers and soil conditioners through biological processes [1,2].Organic wastes are potential sources of macronutrients and large quantities of micronutrient required by plants for growth and improvement of soil health [3]. These nutrients are available in huge amounts in farmyard wastes (e.g. cow dung, pig waste and poultry waste), domestic wastes, agricultural wastes, municipal wastes and industrial wastes. Most rural, semi-urban and urban areas of Nigeria lack proper waste collection and disposal system, hence the continuous accumulation of these wastes which presents many unpleasant environmental consequences including land, water and air pollution [4, 5, 6]

The use of organic waste materials as soil amendment is one important approach to sustainable agriculture. To an extent, organic wastes are utilized as nutrient sources in agriculture, however, some of them are not suitable to be applied directly to the soil to improve plant growth [7, 8, 9]. In some countries, like Pakistan, where sewage sludge is directly used as manure without any treatment, the heavy metals and other toxic substances contained in it usually gain entry into the food chain producing serious human health issues [10,6]. Moreover, the availability of organic materials could be limited if it is used in huge bulk volumes, as in the conventional practice where organic wastes are used at several tons per hectare of land for the improvement of crop productivity [11, 12]

Composting offers a remedy and the most sensible way to avoid wasting of useful natural resources, and creating environmental problems. It is a recycling process in which organic materials are biologically converted into stable humus–like substances under controlled conditions of temperature, moisture and aeration [13] The composting process involves mixed populations of microorganisms e.g. bacteria, fungi and actinomycetes that are indigenous to the waste being converted and transforms the waste into a nutrient–rich amendment capable of improving the nutrient level of depleted farmland soils. During composting, the kinds and numbers of microorganisms that develop are usually affected by temperature and nutrient availability.

Initially, mesophiles predominate and proceed to decompose the readily degradable sugars, proteins, starches, and fats typically found in undigested feed stocks and the availability of easily usable organic substances enables the proliferation of the fast-growing microorganisms [14]. At higher temperatures, thermophilic microrganisms dominate the microbial community and continues generating more heat as a result of the decomposition of more organic matter. The higher temperatures will ensure rapid organic matter processing while simultaneously providing optimal conditions for the destruction of human and plant pathogens [15].

Composting has resolved problems associated with the use of raw organic wastes as soil amendments, which include maladors, human pathogens, toxic heavy metals, toxic organic compounds and other undesirable physical and chemical properties [16, 9, 17]. It also provides a way to manage big volumes of organic wastes in environmentally sound manners [13,18].

The present investigation studied the changes on the microbial population numbers during the composting of some organic wastes using the modified windrow metho

## 2. MATERIALS AND METHODS

### 2.1 Location of the Study Area

This study was carried out at the farmland of Centre for Agricultural Research, Federal University of Technology, Owerri (FUTO), Imo State – Nigeria.

### 2.2 Duration of the Study

The study was done between between September 2017 and January 2018.

### 2.3 Composting of Organic Wastes

The organic wastes used in this study included Poultry Litter (PL), Pig waste (PW), Cow dung

(CD) and Source-Separated Municipal Solid Waste (MSW). MSW was obtained from a dumpsite located at Ikenegbu, Owerri while PL, PW and CD were obtained from the research farm of the School of Agriculture, FUTO.

The organic wastes were composted/cocomposted as following:

a) Pig waste (PW) only
b) Poultry litter (PL) only
c) Cow dung (CD) only
d) Municipal solid waste (MSW) only
e) Pig waste + MSW
e) Poultry litter + MSW
f) Cow dung + MSW

The windrow method of composting as modified by Malone [19] was employed.Sixty kilograms (60) each of PW, PL, CD and MSW were introduced respectively into 100-litre(L) buckets that had previously been perforated at several points. For the co-composted piles,30kg of both samples were introduced into the same 100L bucket that had previously been perforated and then mixed thoroughly.. The compost bins were left open and contents turned every seven days. The organic wastes were allowed to decompose at room temperature in a corner of the greenhouse. The contents of the composting bins were watered with 200 mls of sterile distilled water at intervals of three weeks until the compost samples matured. Composting was done for a period of 70 days (10 weeks).

### 2.4 Determination of Temperature of Composting Piles

The temperature of the composting piles and that of the environment were monitored daily during the entire period of the composting i.e. for 70 days. Process temperatures were determined by taking the average readings from the two thermometers that were inserted 5 cm deep into each pile at different spots. The ambient temperature was continuously monitored by taking average reading of the two different thermometers (Salmoiraghi Co. thermometer model, 1750) fixed permanently at two different spots in the green house.

### 2.5 Isolation and Enumeration of Isolated Bacteria

The media employed included Nutrient Agar, Mackonkey Agar, Eosine Methylene Blue Agar and Salmonella- Shigella Agar. They were all prepared according to manufacturer’s guideline

(Oxoid, England). The initial microbial populations as well as subsequent populations in the compost bins were studied using standard microbiological methods as described by Harley-Prescott [20]. The Total Heterotrophic Bacteria Count (THBC), Fecal Coliform count (FCC), Salmonella Count(SC), Total Coliform Count (TCC) and Total Fungal Count(TFC) of composting organic wastes were determined on day 0, day 4, day 7, day 10, day 14, day 21, day 28, day 35, day 42, day 49, day 56, day 63, and day 70 on the appropriate growth medium. Compost suspensions were prepared by the addition of 10 g compost samples to 90 ml of normal saline (0.85% w/v). Serial dilutions of these initial suspensions were made in normal saline. Aliquot (0.1 ml) of each appropriate dilution was inoculated in duplicate and spread with sterile rod spreader in the Petri plates containing the required medium. Fecal coliforms were counted on Eosin Methylene Blue Agar plates incubated at 44.5°C while *Salmonellae* were counted on Salmonella-Shigella agar plates incubated the at 37°C according to the method described by APHA [21]. The colonies that developed on the plates were counted and recorded as colony forming units using standard methods [19, 22].

## 3. RESULTS AND DISCUSSION

Table1 represents changes in the temperature of the composting piles during composting. Initial temperature of the compost piles ranged from 28 – $30^{o}$C. The temperature of the piles increased at different rates. For CD the temperature increased from $30^{o}$C to $46^{o}$C after two weeks while it took the PL, MSW and PW+MSW 21 days to attain a temperature of $45^{O}$C. The highest temperature of $60^{O}$C was recorded for CD compost on the $28^{th}$ day. However, by the $7^{th}$ week (day 49) the temperature of the compost piles dropped to between $34^{o}$C – $40^{o}$C and stabilized at between $27^{o}$C – $30^{o}$C by the $9^{th}$ week (day 63). During the cooling stage that lasted for about 21 days (i.e. day 50 – day 70), the pile temperatures remained in the range of $27^{o}$C – $37^{O}$C in all the compost piles.

Figures 1 to 5 show the changes in the microbial populations of the different organic wastes. The same pattern was observed for Total Fungal Count (TFC), Total Coliform Count (TCC) and Total Heterotrophic Bacteria Count (THBC). As temperature increased, the microbial populations increased until a peak was attained as determined by the type of organic waste. Faecal coliforms and Salmonellae were not detected in some of the compost bins when temperatures as high as between 47°C – 60°C were recorded.

THBC as high as $7.2\times10^9$ cfu/g was recorded for CDC on day 28 when pile temperature was $60^0$C and PL had the lowest THBC throughout the composting period, from day zero to maturity, when compared to the others. Meanwhile, fungal counts were lower than THBC when compared and the lowest fungal count of $1.0\times10^3$ cfu/g was recorded for PW.

**Table 1**: **Changes in Temperature (°C) during composting of organic wastes**

| Day | PLC<br>T | PWC<br>T | CDC<br>T | MSWC<br>T | PLC +MSWC<br>T | PWC+MSWC<br>T | CDC+MSWC<br>T |
|---|---|---|---|---|---|---|---|
| 0 | 28 | 29 | 30 | 28 | 28 | 29 | 28 |
| 4 | 31 | 30 | 31 | 31 | 30 | 30 | 32 |
| 7 | 31 | 32 | 34 | 30 | 31 | 31 | 33 |
| 10 | 35 | 33 | 37 | 33 | 36 | 33 | 33 |
| 14 | 37 | 39 | 44 | 34 | 36 | 35 | 39 |
| 21 | 45 | 50 | 53 | 45 | 47 | 45 | 48 |
| 28 | 54 | 55 | 60 | 47 | 53 | 52 | 52 |
| 35 | 50 | 49 | 52 | 45 | 45 | 45 | 46 |
| 42 | 45 | 42 | 50 | 42 | 44 | 43 | 44 |
| 49 | 37 | 36 | 40 | 35 | 36 | 44 | 37 |
| 56 | 31 | 31 | 32 | 29 | 29 | 29 | 31 |
| 63 | 28 | 27 | 30 | 27 | 27 | 27 | 29 |
| 70 | 28 | 28 | 30 | 28 | 28 | 27 | 28 |

**Key**

PLC = Poultry Litter Compost

PWC = Pig Waste Compost

CDC = Cow dung Compost

MSW = Municipal Solid Waste Compost

T = Temperature (°C)

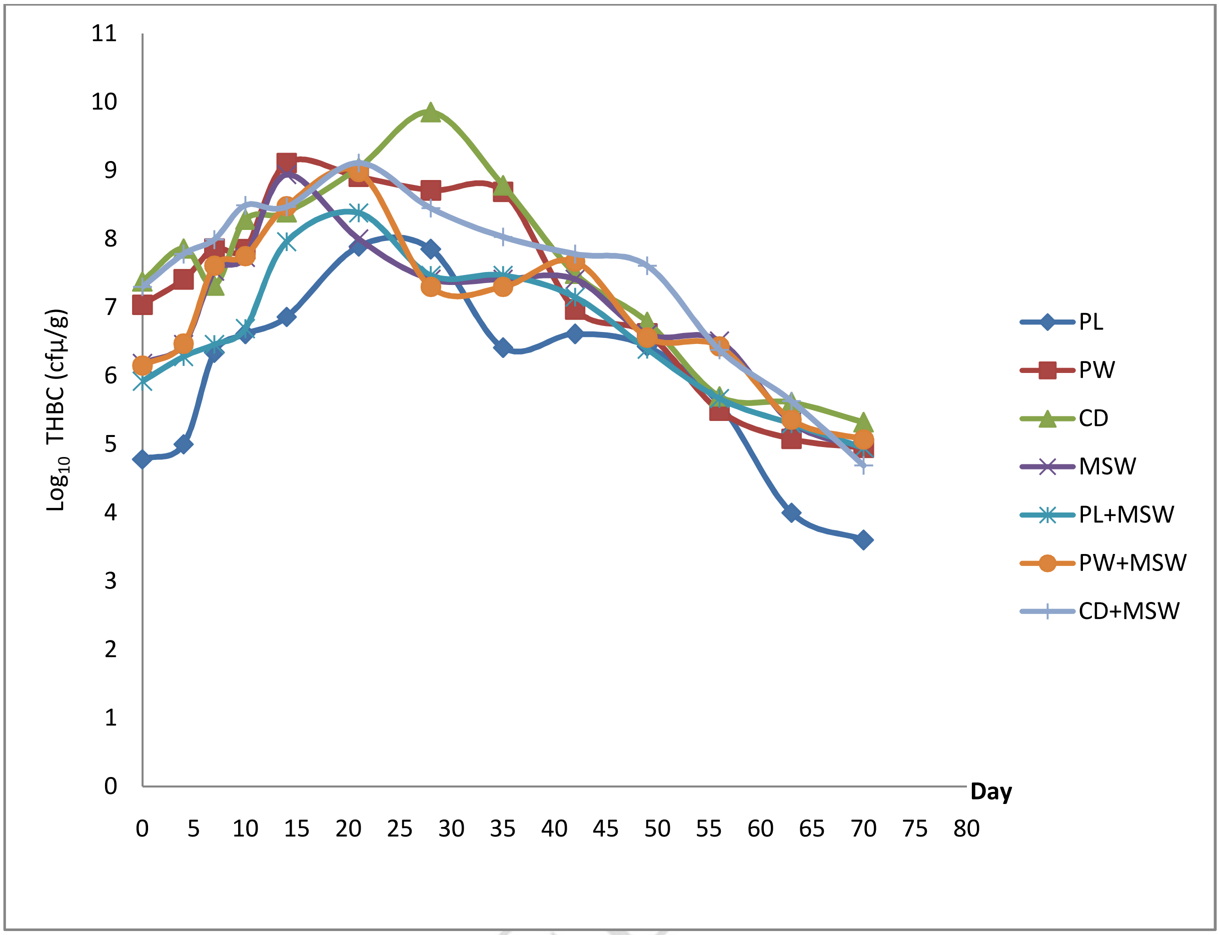


**Fig. 1: Changes in the Total Heterotrophic Bacteria Count (THBC) of the organic wastes during the composting period.**

Key

CDC = Cow dung Compost

PLC = Poultry Litter Compost

PWC = Pig Waste Compost

MSWC = Municipal Solid Waste Compost

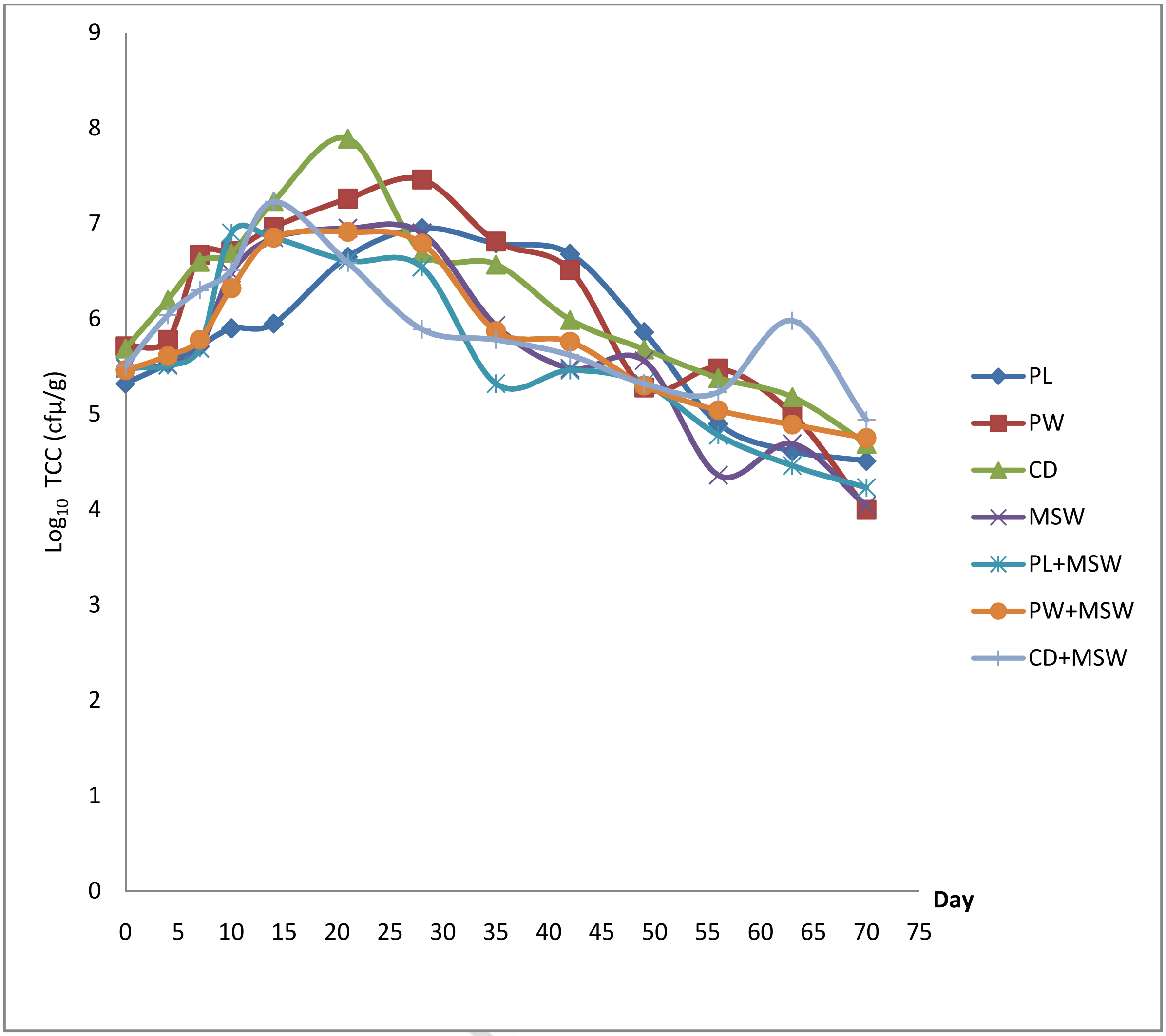


**Fig. 2: Changes in the Total Coliform Count (TCC) of the organic wastes during the composting period.**

Key

CDC = Cow dung Compost

PLC = Poultry Litter Compost

PWC = Pig Waste Compost

MSW C = Municipal Solid Waste Compost

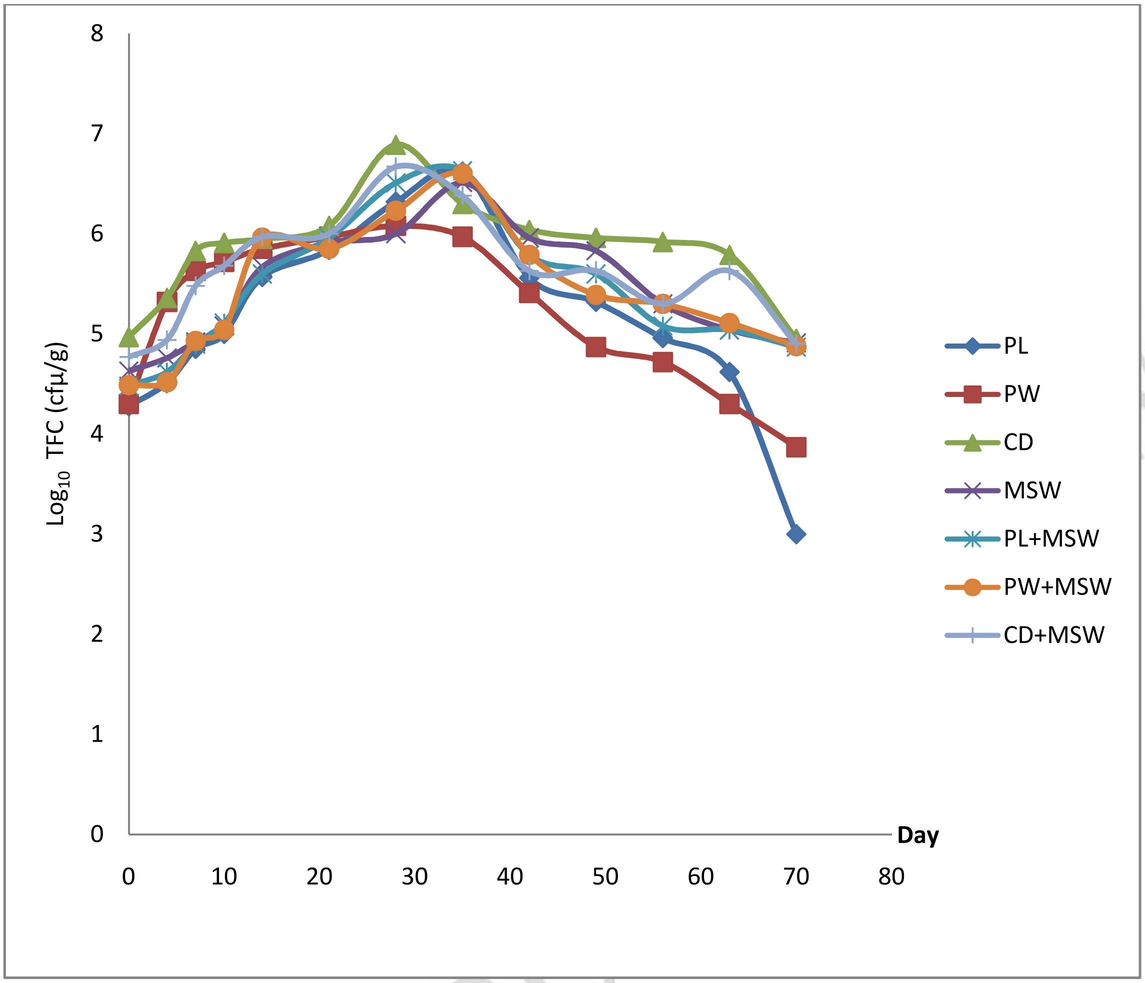


**Fig.3: Changes in the Total Fungal Count (TFC) of the organic wastes during the composting period**.

Key

CDC = Cow dung Compost

PLC = Poultry Litter Compost

PWC = Pig Waste Compost

MSWC = Municipal Solid Waste Compost

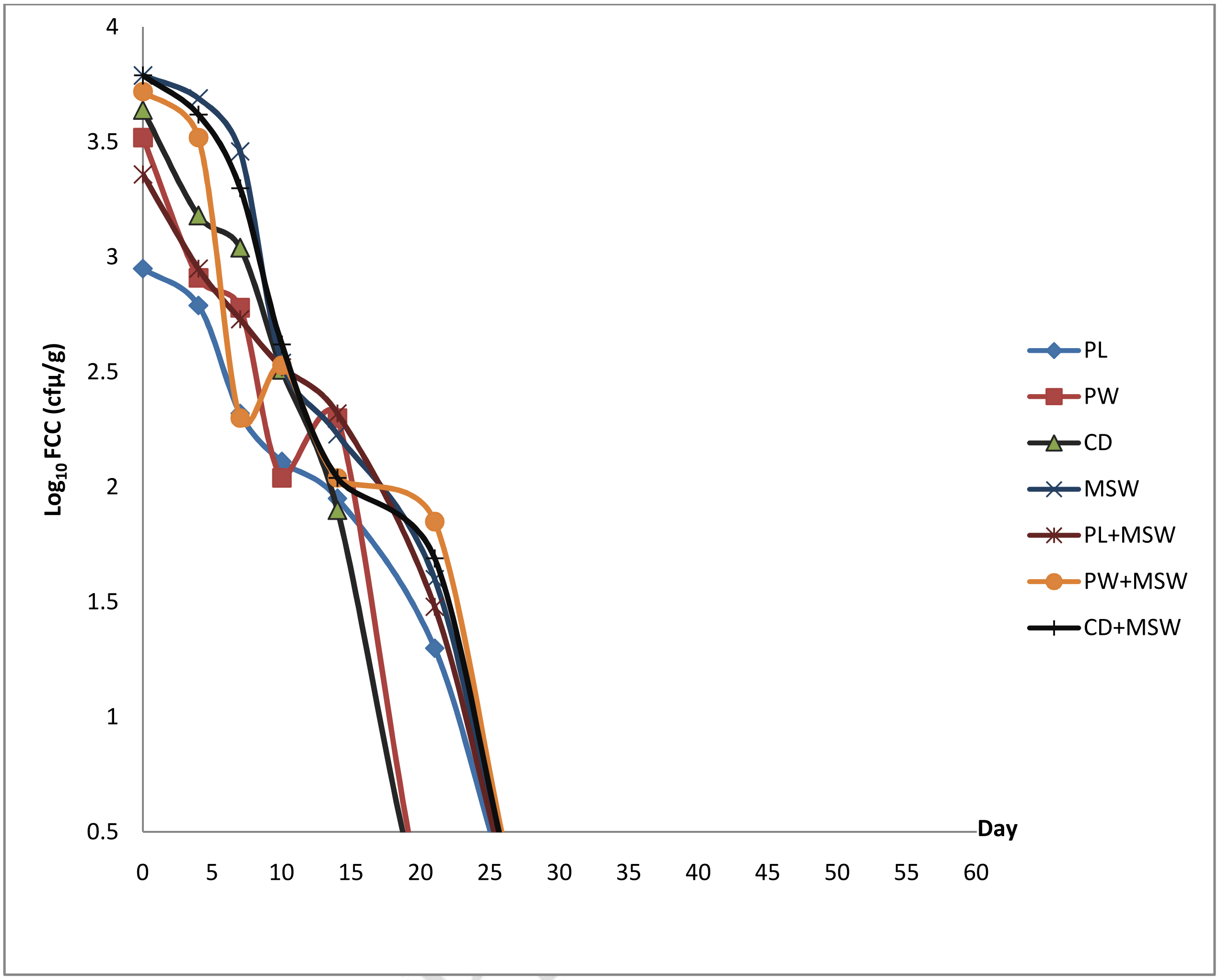


**Fig. 4: Changes in the Fecal Coliform Count (FCC) of the organic wastes during the composting period.**

Key

CDC = Cow dung Compost

PLC = Poultry Litter Compost

PWC = Pig Waste Compost

MSW C = Municipal Solid Waste Compost

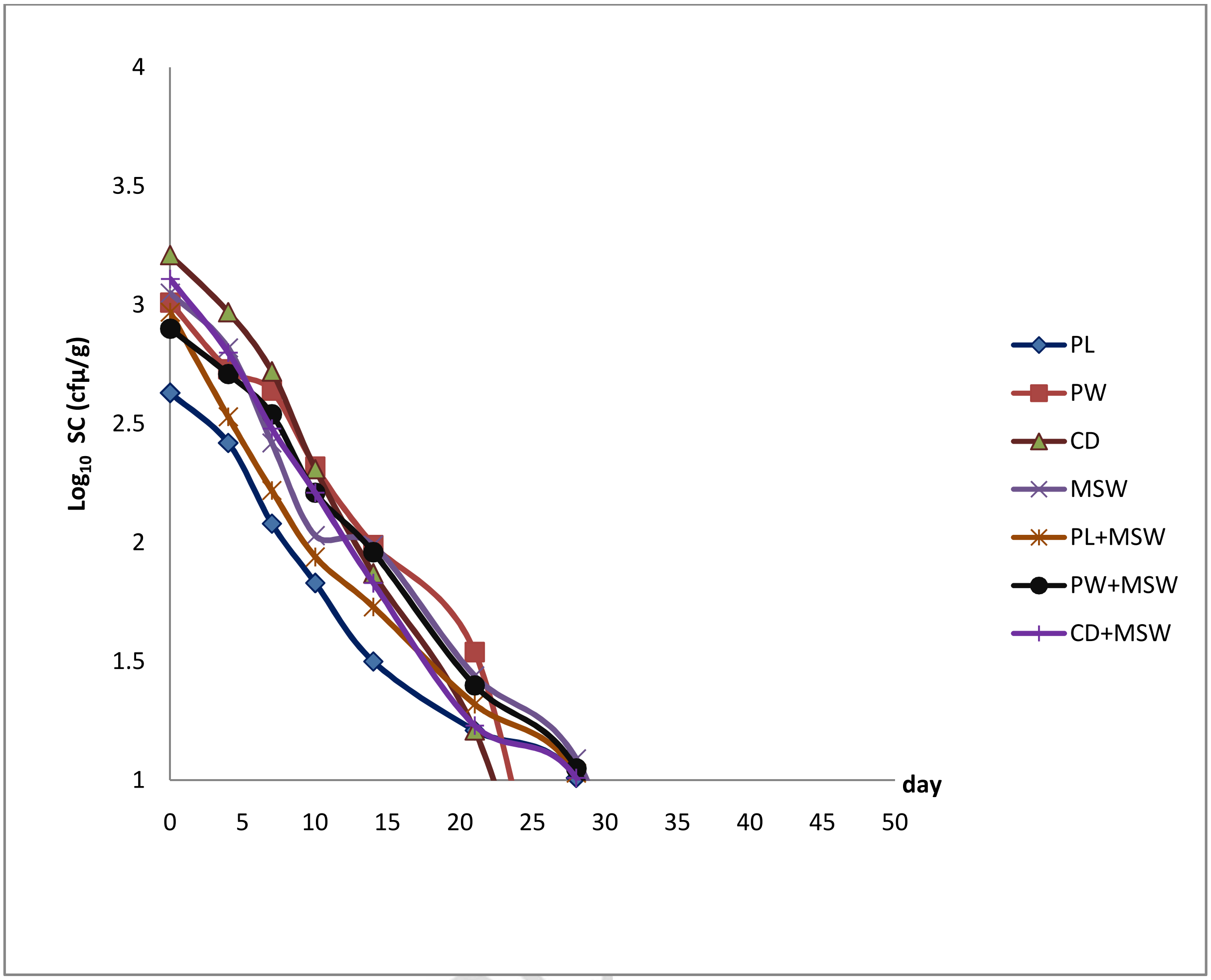


**Fig. 5: Changes in the Salmonella Count (SC) of the organic wastes during the composting period.**

Key

CDC = Cow dung Compost

PLC = Poultry Litter Compost

PWC = Pig Waste Compost

MSWC = Municipal Solid Waste Compost

Generally, the microbial population of the different composting systems increased during the first 3 – 4 weeks of composting except however, faecal coliforms and salmonella. This could be attributed probably to the utilization of the various nutrients available to the microorganisms in the compost due to vigorous microbial activity during this period. The mesophilic population starts the process, oxidizing readily available substrates such as proteins, sugars, starch. As temperature increased, thermophilic microbes developed. This is the period of fastest decomposition, and more resistant compounds such as lignin are degraded to form humus [23].The microorganisms make use of the organic matter in the compost as food source and this process generates heat, water vapor and humus as a result of the growth and activities of microorganism [24]. Hargerty *et al*.[25] reported that there is usually maximum increase in the microbial population of composts during the first 4 weeks of composting provided all other environmental conditions are favourable.

During composting, the population of mesophilic bacteria increased rapidly for the first 14 – 15 days for cow dung. The initial THBC and TCC for cow dung compost were $2.4x10^7$ cfu/g and $5.0x10^5$ cfu/g respectively. Meanwhile, after the 14^th^ day the THBC and TCC increased to $2.5x10^8$ and $1.7x10^7$ respectively. For the other wastes namely; poultry litter, pig waste and the co-composted wastes, mesophilic temperature still manifested between days 14 and 21 of composting. Thermophilic bacteria became dominant in all the composting systems after the 21$^{st}$ day and lasted till the 35$^{th}$ day except for cow dung compost where thermophilic temperature were still observed on the 45$^{th}$ day with a THBC of $6.3x10^6$ cfu/g on the 49$^{th}$ day. Mesophilic populations were again noticed after the thermophilic phase and this lasted for between 21 to 30 days.

The Faecal Coliform Count (FCC) and Salmonella Count (SC) decreased as the composting process progressed. After 21 days, faecal coliforms were completely eliminated in the pig waste and cow dung composts but it took 28 days of composting to completely eliminate *Salmonella* sp in the same compost systems i.e PW and CD. There was complete elimination of faecal coliforms and *Salmonella* in all the compost systems after the 28$^{th}$ day with temperature range between $47^OC$ – $60^OC$. This was probably due to the high temperatures generated in the different compost bins. Many pathogenic bacteria carried via animal are found in high concentration in their waste and the numbers and types depend on the source of the waste and the physico-chemical composition of the wastes [26].

During the mesophilic stage, lots of pathogenic organisms proliferated, and so, the thermophilic stage is considered important for destroying thermo-sensitive pathogens [27,28]. In the cause of this study, it was observed that most common human pathogens, like fecal coliforms and *Salmonella* spp. etc. that dominated the mesophilic phase were eradicated from the composts when temperature reached $45^{O}C$. Previous studies had indicated that temperatures between 45 – $55^{O}C$ for 3 consecutive days is sufficient to destroy pathogenic bacteria [29, 30, and 31]. Liao *et al*.[32] had also reported that reduction in the number of fecal coliforms and *Salmonella* was due probably to high temperatures and unfavourable conditions.

The Fungal Counts(FC) showed slight increases in the first 28 days of composting from a range of $1.9x10^6$ – $5.9x10^6$ cfu/g to a range of $1.2x10^6$ – $7.8x10^6$ cfu/g. After the $35^{th}$ day, the fungal counts began to decrease until cooling and maturation phase (table 4.1c). Insam *et al* [33] had earlier reported that mesophilic bacteria and fungi were the dominant active degraders of the organic wastes, and the interaction between the various groups of microorganisms depended on the nutrient resources and the biochemical mechanisms of organic and inorganic matter transformation changes. Microorganisms play key roles in the composting process and the presence of some microorganisms reflects the quality of the maturing compost. Ryckeboer *et al*. [34] further reported that bacterial and fungal populations were fundamentally influenced by temperature, pH and the nutritional composition of the organic wastes.

During the first 4 weeks of composting diverse populations of mesophilic fungi proliferated and degraded the readily available nutrients and raised composting system temperatures to above $45^{O}C$ The microbial counts showed a decline during the later weeks of composting and at maturity. Reasonable numbers of microorganisms were still present in all the composts at maturity and these depended on the nutrients available and other environmental factors such as temperature, pH, aeration and moisture content [25,35].

## 4. CONCLUSION

Microorganisms play key roles in the composting process and the presence of microorganisms were fundamentally influenced by the temperature of the compost piles. The microorganisms made use of the organic matter in the compost as food source and this process generated beneficial materials for agricultural usage.

## COMPETING INTERESTS

Authors have declared that there are no competing interests .

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