PHYSICOCHEMICAL CHARACTERIZATION AND MICROBIAL IDENTIFICATION OF COMPOST PRODUCED FROM MUNICIPAL SOLID WASTE IN SHEWA ROBIT TOWN, ETHIOPIA

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ABSTRACT: Compost from solid decomposable organic waste collected in Shewa Robit town was produced and its physicochemical properties and microbial composition studied. Results of pH showed increment from initial value of 5.20 to 9.27 at the end of the third month of the composting time whereas, during the same period electrical conductivity decreased from initial value of 1.35 to 0.29 ds/m. Contents of total organic carbon, total nitrogen, available potassium and available phosphorus analysis conducted at the initial, intermediate and final stages showed that all values decreased consistently from the beginning to the end. A total of 8 microbes, 5 fungi, and 3 bacteria genera were isolated. While results of the carbon to nitrogen ratio, temperature, nitrogen, phosphorus, and potassium macronutrients, and its composition of bacteria and fungi genera of the end product of the compost indicated its maturity and stability and hence its potential application in agriculture as soil conditioner and fertilizer, its highly alkaline nature observed indicates care to be taken before its direct application for agricultural purpose.

Key words: Compost, solid organic waste, C/N, electrical conductivity, microbes

INTRODUCTION

Waste generation is a natural process and generally increases with one's per capita income [1]. The most commonly used method of eradicating municipal solid waste generated in developing countries like Ethiopia is disposing of in waste disposal sites, rivers, roadsides or land filling mainly in the outskirts of a town [2]. In the absence of a proper waste management system, the waste could be not only a source of human and animal diseases but could negatively affect the environment and deface the aesthetics beauty of a town [3]. Several methods are adopted to deal with waste nuisances including using it for biogas production, composting, recycling, incinerating, landfilling, etc. [4, 29]. From these solid waste control methods, composting is one of the well-known viable options with double advantages; replenishing soil fertility and at the same time efficiently and economically avoiding undesired waste [26]. Composting is an oxidative decomposition process of organic matter (OM) assisted by microbial consortia under controlled conditions producing a stabilized and sanitized end product called compost which is ready to be used as a fertilizer and soil conditioner [5].
During the process, several factors affect the rate of degradation and composting time. These include the type as well as homogeneity and particle size of compost source, temperature, rate of aeration, moisture content, C/N, presence of animal manures [6, 7]. Mixing the solid waste with cow dung at the initial stage of the composting process has proven to be effective in enhancing the degradation process by optimizing the optimal C/N ratio and introducing microorganisms to the compost [8]. A group of microorganisms mainly bacteria and fungi break down the organic matter under optimal conditions and during the process temperature of the composting pile goes up from mesophilic to thermophilic stage, in which most of the pathogenic microorganisms and weed seeds are killed and then later goes down to the mesophilic stage again in which there is less easily decomposable organic matter left giving rise to an accompanied decline in microbial community and hence results in a matured, stabilized humus-like end product [9, 10]. In this work, compost has been produced from a mix of solid organic waste collected from households, restaurants, vegetable and fruit shops, Khat selling shops, juice houses and street droppings in Shewa Robit town and characterized for its physicochemical properties and microbial composition. Shewa Robit is a small town located at 225Km Northeast of Addis Ababa at a latitude of 9°59'60.00"N and Longitude of 39°54'0.00"E with an elevation of 1280m above sea level. According to the 2007 census survey report of the Ethiopian Central Statistical Agency, the town has a total population of 17,575.

MATERIALS AND METHODS
Site Description
The compost preparation was carried out at the Agricultural Demonstration Farm of Debre Berhan University in Shewa Robit town during the period from April, 2014 – July, 2014.

Waste Collection, Processing and Pile Design
Waste was collected from households and restaurants, street droppings, fruits and vegetables selling shops refuse disposal sites and Khat selling shops. Cow dung was obtained from surrounding farmers in the town. Indecomposable materials including iron, glass, plastic bags, bottles, etc. were sorted out and segregated from the waste and then larger materials mechanically chopped down into smaller pieces with the help of an ax and gadget. The waste and cow dung in an approximately 1:4 (wet mass basis) was then mixed thoroughly with pitchfork and formed into 2.5m width by 1.5m height windrow pile. Initially, the moisture content was 58.4% and remained in the range of 52±2% for the rest of the composting period by turning and watering action. Dry grass clipping was used at the end of every turning and watering action time to minimize the extent of water loose from the surface of the pile due to evaporation.

Analysis of Physicochemical Parameters
Temperature (T) of the pile was measured with a digital thermometer. Fresh samples collected from the periphery, 50cm distance apart and the middle, depth of 0.75m from the pile at every two weeks interval were taken rapidly to laboratory and dried in an oven whose temperature maintained at 105°C for 24h, cooled in desiccators, reweighed and moisture content (MC) determined. Electrical conductivity (EC) and pH values were measured using digital pH (Model No. CP 505) and EC (Model No. CC 411) meters in 1:50 extracts of compost powder passed through 1mm sieve and double distilled water as solvent. Total Organic carbon (%OC) content was determined by ignition at 550°C, and that of Total Nitrogen (%TN) was determined by Kjeldahl method. Available potassium (Av. K) was determined using flame photometry in ammonium acetate extract and available phosphorus (Av. P) was estimated using Olsen et.al. method [11].

Microbial Identification
The isolation of microorganisms was carried out once at the end of cooling phase, the third month of the composting time. One (1) g of compost sample added to 9 ml of distilled water was shaken vigorously. Then 1 ml of the sample was taken and added into 9ml of distilled water. Ten-fold serial dilutions up to 10⁻⁵ were made and the odd numbered ones were used. An amount of 0.1 ml was placed on Standard Plate Count Agar (SPCA) for cultivation of bacteria and Potato Dextrose Agar (PDA) for fungi; for cultivation of anaerobic bacteria, plates were wrapped with plastic plasters. All culture media were prepared according to the instruction of manufacturing company. Petri plates were incubated at ambient temperature (35°C) for 24 h for bacteria, 5 days for fungi at 28°C. Isolates were maintained on respective media slants at 4°C. Prevalence of different groups of microorganisms was calculated in terms of percentage. These isolates were identified on the basis of conventional cultural and morphological characteristics and diagnostic biochemical test for Gram-positive and Gram-negative bacteria was also carried out.
RESULTS AND DISCUSSION
Temperature, electrical conductivity, and pH was measured at every two weeks interval over the 3 months of composting whereas %OC, %TN, available nitrogen and phosphorus was measured in the beginning, intermediate and final stages of the composting period only.

Physicochemical Parameters (pH, T, and EC)
The temperature measured at the middle point of the pile was initially 31°C. This increased slowly and reached a maximum of 68°C when measured at the 45th day and then decreased dramatically to 36°C in the 60th day and then declined slightly to the surrounding temperature of 35°C when measured at the 75th day and remained same when measured at the end of the third month, table 1. During Composting process, microorganisms degrade the organic matter in the waste with the production of heat, NH₃, CO₂, organic acids and water vapor [13, 27]. The heat released is trapped within the pile leading to the phenomenon of self-heating which is responsible for the rise in the temperature of the pile at the initial phase of composting [14]. In well controlled composting, temperature increases rapidly during the initial phase, reaches maximum at the active phase and then comes back to the ambient temperatures at the cooling phase [15]. The measured values obtained here are in agreement with this trend and indicate that the compost has come to a stable stage. If the temperature exceeds 70°C, most microorganisms begin to die and decomposition process slows down. Frequent turning is, therefore, crucial to ensure proper composting by removing excess heat developed in the pile and creating conducive microbial environment [16]. The pH was acidic measured in the beginning, 5.20 and became alkaline, 8.80 after a period of two weeks. Then after, it remained at slightly greater than 9 with minor fluctuations with the value at the 90th day being 9.27, table 1. This dramatic increase in pH observed after 30 days may be due to the release of ammonia under acidic environment by the mineralization of organic nitrogen sources supported by microbial action [12]. The EC value indicates the concentration of ions available in the compost. It was 1.35 ds/m in the beginning and came down decreasing to 0.29 ds/m at the end of the third month. The cause for the decrease could be due to ammonia volatilization and precipitation of mineral salts and similar continuous decline of EC values during the 17th day of composting carried out in a rotary drum composter has been reported by Sudharsan Varma V, and Ajay S. Kalamdhad [17].

Nutrient Analysis
Nitrogen, potassium, and phosphorus are macronutrients essential for proper plant growth. The measurements, table 2, of available phosphorus, available potassium, total organic carbon (%) and total nitrogen (%) showed similar consistently decreasing tendency from the initial to the final stage. The amount of available potassium was 67.59 initially, 43.86 at the 45th day, and 36.01 cmol/kg at the 90th day whereas that of available phosphorus was 1965.58, 687.03, and 549.65 mg/kg, all on a dry weight basis, at the beginning, 45th and 90th day, respectively. Potassium salts are highly soluble in water and the observed decreasing tendency of available potassium could be due to its leakage and run-off as a result of the frequent turning and watering action [28] while the cause for the decrease in available phosphorus could be due to precipitate formation as a results of its reaction with positively charged ions of Fe, Mg, Ca metals in the compost [17]. The OC and TN content also showed consistently decreasing tendency over the composting time. The %OC was 55.12 at the beginning and 15.69 at the end and that of %TN was 1.32 initially and 1.03 in the end. This decrease of carbon and nitrogen contents in the compost is due likely to their consumption by microorganisms for their proper functioning. Carbon is used as energy source and nitrogen for protein synthesis by bacteria which leads to their rapid proliferation during the initial stage of composting [8, 17].

<table>
<thead>
<tr>
<th>Table-1. Temperature, pH, EC and MC values of compost samples</th>
</tr>
</thead>
<tbody>
<tr>
<td>Days</td>
</tr>
<tr>
<td>------</td>
</tr>
<tr>
<td>1</td>
</tr>
<tr>
<td>15</td>
</tr>
<tr>
<td>30</td>
</tr>
<tr>
<td>45</td>
</tr>
<tr>
<td>60</td>
</tr>
<tr>
<td>75</td>
</tr>
<tr>
<td>90</td>
</tr>
</tbody>
</table>

If compost is to be applied to land, its maturity and stability has to be assessed first for otherwise it may adversely affect the growth of plants due to the possible presence of pathogenic microorganisms and nutrient composition which disturbs the nutrient balance of the soil where it is applied [18, 19, 25].
Though there is no one universally accepted method of evaluating compost maturity and stability, one of the most commonly used method of checking is the carbon to nitrogen ratio [18]. In stabilized and matured compost, C/N ratio of less than or equal to 15 has been accepted to be appropriate for land application as a fertilizer and conditioner [20] and our result for the final compost sample of 15.2 reasonably lies within the acceptable range for use in agriculture.

<table>
<thead>
<tr>
<th>Days</th>
<th>Av. K (cmol(+)/kg)</th>
<th>Av. P (mg/kg)</th>
<th>%OC</th>
<th>%TN</th>
<th>C:N</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>67.59</td>
<td>1965.58</td>
<td>55.12</td>
<td>1.32</td>
<td>41.8</td>
</tr>
<tr>
<td>45</td>
<td>43.86</td>
<td>687.03</td>
<td>21.30</td>
<td>1.31</td>
<td>16.3</td>
</tr>
<tr>
<td>90</td>
<td>36.01</td>
<td>549.65</td>
<td>15.69</td>
<td>1.03</td>
<td>15.2</td>
</tr>
</tbody>
</table>

### Microbial Analysis

Composting is aerobic decomposition of OM facilitated by a group of microorganisms under controlled conditions. Temperature of the compost affects and is being affected by the group of microorganisms available in it. MC, pH, C/N also affect the composition of microorganisms in a compost. In the initial mesophilic stage where the temperature is < 50°C, microbial activity becomes high, decreases in the intermediate thermophilic stage where the temperature is between 55–70°C, and then increases in the second mesophilic phase when they re-colonize the compost pile as the temperature becomes convenient for them to live [21]. Identifying microbes in compost is essential in that it not only helps in determining its quality and soil application but also helps to detect hazardous pathogens for plants and animals and optimize compost quality and standards [22].

<table>
<thead>
<tr>
<th>S.No</th>
<th>Fungi isolated</th>
<th>Macroscopic morphology</th>
<th>Microscopic morphology</th>
<th>Frequency</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Aspergillus</td>
<td>The surface growth is downy or powdery, showing various shades of blue-green to a grey-green with a narrow white border.</td>
<td>Hyphae are septate with smooth walled conidiophores. Conidia are (smooth or slightly rough), subspherical and about 2-3.5µm in diameter and develop in chains</td>
<td>9</td>
</tr>
<tr>
<td>2</td>
<td>Penicillium</td>
<td>Colonies are usually blue-green or grey-green.</td>
<td>Microscopically Penicillium species are identified by their dense broom-like spore-bearing structure called phialides. Phialides are flask-shaped and produce chains of spores (conidia) from their tips. The spores are typically green in color and vary in shape.</td>
<td>8</td>
</tr>
<tr>
<td>3</td>
<td>Mucor</td>
<td>Colonies are very fast growing, cottony to fluffy, white to yellow, becoming dark-grey, with the development of sporangia.</td>
<td>Sporangiohophores are erect, simple or branched; They are hyaline, gray or brownish, globules to ellipsoidal, and smooth-walled or finely ornamented.</td>
<td>4</td>
</tr>
<tr>
<td>4</td>
<td>Alternaria</td>
<td>- Rapidly grow downy or cottony colonies maturing within 5days - Grey to olive brown on the surface with short aerial hyphae</td>
<td>Alternaria structures appeared brown to black in color. - Dark septate hyphae</td>
<td>3</td>
</tr>
<tr>
<td>5</td>
<td>Fusarium</td>
<td>A purple color, particularly on Potato-Dextrose Agar</td>
<td>Hyphae are hyaline (clear/non-pigmented) and are septate (show divisions or walls within the hyphae)</td>
<td>6</td>
</tr>
</tbody>
</table>
Table 4. Bacteria genera isolated from compost

<table>
<thead>
<tr>
<th>S.No</th>
<th>Bacteria isolated</th>
<th>Colony Shape</th>
<th>Gram Stain</th>
<th>Microscopy</th>
<th>Motility</th>
<th>E*</th>
<th>Oxygen</th>
<th>Biochemical Test</th>
<th>Frequency</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Bacillus</td>
<td>Round translucent smooth edge</td>
<td>+</td>
<td>Straight rod</td>
<td>Motile, (pf)</td>
<td>+</td>
<td>Aerobic/f*</td>
<td>Catalase (+)</td>
<td>13</td>
</tr>
<tr>
<td>2</td>
<td>Clostridium</td>
<td>Circular, mucoid, translucent colony</td>
<td>+</td>
<td>Short rod</td>
<td>None</td>
<td>+</td>
<td>Anaerobic</td>
<td>Catalase (-)</td>
<td>4</td>
</tr>
<tr>
<td>3</td>
<td>Pseudomonas</td>
<td>Circular with pp</td>
<td>-</td>
<td>Straight rods</td>
<td>Motile polar flagella</td>
<td>-</td>
<td>Aerobic</td>
<td>Oxidase (+) with pp</td>
<td>6</td>
</tr>
</tbody>
</table>

pf = peritrichous flagella, E* = Endospore, f* = facultatively anaerobic, pp = pigment production

A total of 8 microbes, 5 fungi, and 3 bacteria genera were isolated. The fungi genera include *Aspergillus*, *Penicillium*, *mucor*, *Alternaria*, and *fusarium*. Table 3 shows the macroscopic and microscopic morphology observed including their frequency (number) in a culture medium. The bacteria genera were *Bacillus*, *Clostridium*, and *Pseudomonas*. *Bacillus* and *Clostridium* were both gram stain and endospore formers whereas *Pseudomonas* genera were gram-negative. The shape of the colony, microscopic morphology and catalase and oxidase biochemical tests including the frequency observed in a culture of PDA and SPCA media is given in table 4. From the tables, it could be seen that *Bacillus* from bacteria and *Aspergillus* and *Penicillium* from fungi genera were the most dominant ones in the compost sample after 90 days of composting. These same dominant bacterial and fungal genera were also found during the cooling down stage in different composting piles [22, 23] and are beneficial microorganisms in agriculture [24].

CONCLUSION AND RECOMMENDATION

Compost from solid biodegradable organic waste collected from households, streets and refuse disposal sites in Shewa Robit town has been produced and its physicochemical properties and microbial composition determined. The EC value showed decreasing tendency over the three months of composting time which could be due to the low concentration of ions due to ammonia volatilization or precipitation of ions. Similarly, TOC, TN, and C/N also showed consistently decreasing tendency over the same period due to their consumption by microorganisms for their proper functioning in the compost pile. Assessment of Available K and P contents showed decreasing tendency from the initial to the final stage and that the decrease in K may be due to its leakage and run-off as its salts as highly soluble in water during the turning and watering action and that of P may be due to its precipitation by forming insoluble phosphates by reactions with cations such as Fe, Ca, Mg available in the compost. While the temperature, C/N, EC, nutrient, and microbial composition values indicate compost stability and maturity and its potential application for agricultural or horticultural purposes safely, the higher than suitable pH range for the proper growth of most crops observed suggest care to be taken during application to soil.

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