

## Study of commercial effective microorganism on composting and dynamics of plant essential metal micronutrients

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### Abstract

The present study addresses the problem of organic farmers' that needs local organic resources with their enhanced quality to effectively fertilize their agriculture crops. In accordance with the objective of the experiment that is about enhancing quality of compost, a blend of organic resources, comprising cow manure (CM), poultry manure (PM) and kitchen waste (KW) (2:1:1 ratio by volume) was composted with effective microorganisms (EM.1) (Compost<sub>EM.1</sub>) and without (Compost<sub>plain</sub>). During composting, temperature, pH, carbon, nitrogen, C/N ratio, total and diethylene triamine pentaacetic acid (DTPA)-extractable essential metal micronutrient (Fe<sup>3+</sup>, Cu<sup>2+</sup>, Zn<sup>2+</sup>, and Mn<sup>2+</sup>) contents of both the composts were recorded following the standard procedures. Low temperature range (24–24), low pH (6.7–7.2) and higher N-content (1.15–1.40) were recorded for Compost<sub>EM.1</sub> as compared to Compost<sub>plain</sub>. Carbon degradation was also faster in Compost<sub>EM.1</sub> than in Compost<sub>plain</sub>. Consequently, C/N ratio stabilization took 6 weeks in Compost<sub>EM.1</sub> as compared to 18 weeks in Compost<sub>plain</sub>, leading to rapid completion of composting. Total concentration of micronutrients increased while their DTPA-extractable content decreased during the composting. Total micronutrient concentration was augmented more in Compost<sub>plain</sub> samples than in Compost<sub>EM.1</sub>. However, decrease in DTPA-extractable content was similar in both the composts. Increase in micronutrient content was attributed to decrease in organic matter weight, whereas decrease in metal micronutrients was attributed to the formation of organic matter-metal complexes during decomposition. Findings of the study indicated that effective micro-organisms enhanced composting process, however, further studies are required to evaluate its quality, especially effect on plant and soil.

### Key words

Composting, Kitchen waste, Manure, Plant nutrients

### Introduction

Organic composting offers both agriculture and environmental benefits. Composting is decomposition of raw organic materials, such as animal waste, crop residues, food garbage, municipal and industrial wastes into organic fertilizer for extensive and intensive agriculture (Jelin *et al.*, 2013; Mahanta *et al.*, 2014). Different commercial additives are available in market that claims to accelerate the composting process. However, scientifically, there is scarce literature confirming their effectiveness. Himanen and

Himanen (2009) evaluated five commercial additives and found no significant differences between composts created with commercial additives and control. Gabhane *et al.* (2012) indicated that some commercial additives display no noticeable effect, while others facilitate the composting process by aiding in the growth of microbes. Similarly Zhou *et al.* (2014) used three different commercial inoculants, Xu (2015) used DN-1, a commercial inoculants, and both reported improvement in their composting process. Tran *et al.* (2015) explored TM14, a commercially available inoculant and reported for accelerating the degradation

process. Because of the discovery of new products and approaches toward composting, continuous research must be conducted to confirm their specificity, validity, benefits and to update the scientific literature.

This study investigated effective microorganisms1, an additive available worldwide that has been discovered by a Japanese professor, Dr. Teruo Higa which comprised a mixture of microorganisms (*Lactobacillus caseii*, *Rhodopseudomonas palustris*, and *Saccharomyces cerevisiae*) and is used in the treatment of bio-wastes. Previously, Shalaby (2011), Kale and Anthappan (2012), and Boga *et al.* (2014) have reported the effective use of effective microorganisms1 in the cleaning of indoor pollutants, and for waste degradation and management. The focus of this study is to explore the effectiveness of effective microorganisms1 in the composting process, and the dynamics of metal micronutrients during the composting process.

## Materials and Methods

**Experimental setup:** A mixture of three organic materials: cow manure, poultry manure and kitchen waste (2:1:1 ratio by volume) was composted with effective microorganisms1 (Compost<sub>effective microorganisms1</sub>), while another such mixture was composted without effective microorganisms1 (Compost<sub>plain</sub>) served as control. The composting materials were placed in 250 l (900 mm × 1350 mm) black heavy-duty bags. Compost<sub>effective microorganisms1</sub> was stored under anaerobic condition, which was pre requisite for effectiveness of effective microorganisms1, while Compost<sub>plain</sub>, which represented typical aerobic composting (control treatment), bags were drilled with holes to facilitate aeration. There were four replicates of each treatment. Effective microorganisms1 was obtained from the Effective Microorganisms Research Organization (EMRO; <http://www.emrojapan.com>) through Rashed Establishment for Trading and Agriculture (<http://www.rashedagri.com.sa>). The effective microorganisms1 inoculum was prepared by mixing a commercial pack of the effective microorganisms1<sup>®</sup> (EMRO, Okinawa, Japan), molasses and water. (1:1:20 ratio by volume). This mixture was incubated for 5 days for activation of the microbes. Microbial activation was confirmed (as per the manufacturer protocol) by decrease in pH to 3.8 or below, which occurred due to the production of various acids by the microbes. Although the optimized quantity of effective microorganisms has not been established through research by independent study, thus according to manufacturer recommended quantity for decomposing solid wastes, 1L of activated EM.1 per 1 m of the composting blend was used.

**Parameters and data collection procedure:** Temperature, pH, carbon, nitrogen, C/N ratio, total and DTPA extractable contents of micronutrients (Fe<sup>3+</sup>, Cu<sup>2+</sup>, Zn<sup>2+</sup>, and Mn<sup>2+</sup>) were

analyzed. All the data were recorded at the beginning of the composting process. Following this, temperature and pH were measured once every two weeks. Temperature was measured by inserting a composting thermometer at the center of the compost; while for pH, representative sample was collected from each experimental unit using a sampling probe. A suspension of the obtained sample was prepared (compost and deionized water at a ratio of 1:2), and its pH was measured with pH meter. The contents of each bag were mixed and turned over thoroughly every 6<sup>th</sup> week. After mixing process, samples were collected for analysis of C, N and micronutrient content. Moisture was also checked during these intervals using a moisture meter probe, and maintained at 50%. Nitrogen and carbon contents were quantified using a Perkin-Elmer CHNS/O Analyzer (Model 2400) following manufacturer's protocol (Perkin Elmer, Inc., Waltham, MA, USA).

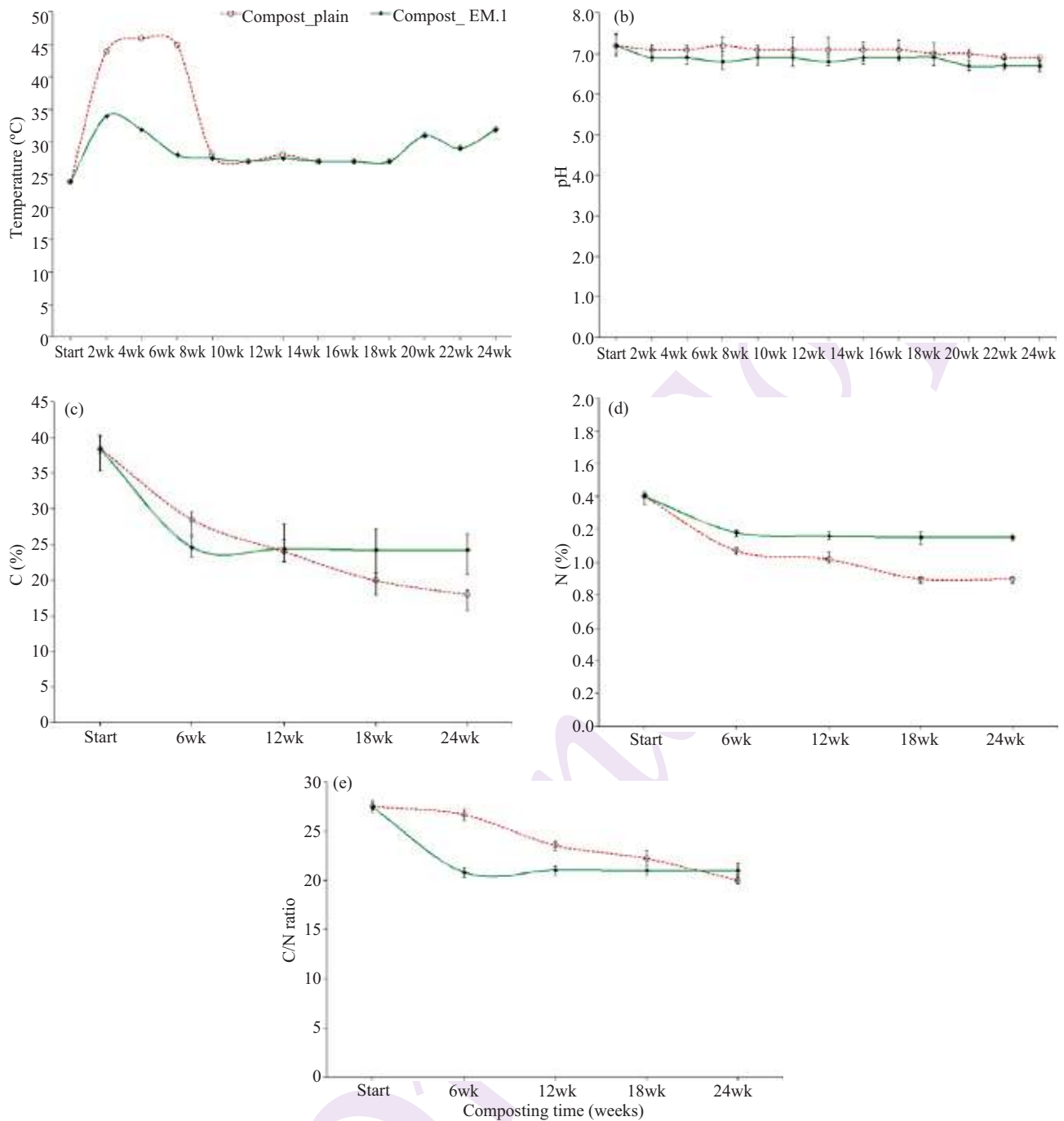
Micronutrient (Fe<sup>3+</sup>, Cu<sup>2+</sup>, Zn<sup>2+</sup> and Mn<sup>2+</sup>) contents were determined following the method of Ryan *et al.* (2001). Briefly, 0.5 g of compost was dissolved in 10 ml HNO<sub>3</sub>/HClO<sub>4</sub> (6:4), and digested. The final volume of each sample was increased to 50 ml by adding of distilled water, and solutions were filtered with Whatman filter paper grade 1 (Sigma Aldrich, St. Louis, MO, USA). Filtrate from each sample was analyzed for Fe<sup>3+</sup>, Cu<sup>2+</sup>, Zn<sup>2+</sup> and Mn<sup>2+</sup> content, by atomic absorption spectrophotometer.

The available plant fractions of micronutrients were extracted following the method of Lindsay and Norvell (1978). Briefly, 5 g of each sample was extracted using 10 ml of DTPA extractant (0.005M DTPA + 0.01M CaCl<sub>2</sub> + 0.1M triethanolamine, pH 7.3) by mechanical agitation for 2 hours. All the samples were centrifuged at 10000 rpm for 15 min and the supernatants were subsequently quantified for micronutrient content by atomic absorption spectrophotometer.

**Statistical analysis:** Statistical analysis was done by MStatC. Statistical significance between the values was tested by Fisher's least significant difference-test ( $p < 0.05$ ). Graphs were created using Microsoft Excel (Microsoft Corp., Redmond, WA, USA).

## Results and Discussion

Temperature dynamics during the trial are displayed in Fig.1 (a). Temperature was found to be higher in Compost<sub>plain</sub> during 2<sup>nd</sup>-6<sup>th</sup> week, as it fluctuated between 44°C and 46°C. In contrast, during the same period, temperature in Compost<sub>effective microorganisms1</sub> was found between 28°C and 34°C. On 8<sup>th</sup> week, temperature dropped to 28°C in both the compost types, and varied between 27°C and 32°C till end of the experiment. The obtained results revealed that, in the presence of effective microorganisms1, the composting



**Fig. 1 :** Dynamics of (a) temperature, (b) pH, (c) carbon, (d) nitrogen and (e) C/N ratio during composting process. Error bar indicates standard deviation (SD) of mean values of 4 replicates; E.M.1: effective microorganisms 1

process occurred at low temperatures, which indicated that the composting/break down process occurred as a result of microbial enzyme production. During normal composting process, as represented by Compost<sub>plain</sub>, temperature rose due to oxidation of carbon to CO<sub>2</sub> and subsequent generation of heat. Actually, the bacteria in the compost utilize carbon as a

source of energy and nitrogen to build protein in their bodies. They obtain energy by oxidizing organic material, especially the carbon fraction. This oxidation process heats up the compost pile from ambient air temperature. These results are supported those obtained by Wongwilaiwalin *et al.* (2010) and Gondek *et al.* (2014). The pH dynamics for Compost<sub>plain</sub>

**Table 1** : Total and DTPA-extractable micronutrient content tested during the composting of Compost<sub>plain</sub> and Compost<sub>effective microorganisms1</sub>

Composting time	Compost <sub>plain</sub> micronutrient content								Compost <sub>EM,1</sub> micronutrient content							
	Total				DTPA-extractable				Total				DTPA-extractable			
	Fe	Cu	Mn	Zn	Fe	Cu	Mn	Zn	Fe	Cu	Mn	Zn	Fe	Cu	Mn	Zn
Start	7446 <sup>c</sup>	61C	128d	240e	610a	34a	45a	132a	7424	64	132	238	446a	34a	48a	136a
	±178	±3.3	±4.0	±12.2	±6.3	±1.6	±4.1	±2.8	±288	±6.4	±7.0	±10.0	±8.4	±4.2	±3.2	±7.0
Week 6	7580c	74c	136d	322d	324b	30ab	44a	128a	7585	66	140	244	422a	33a	40ab	132a
	±324	±2.2	±3.6	±10.1	±14.1	±2.2	±4.1	±3.6	±256	±5.0	±6.5	±16.1	±11.1	±4.2	±2.8	±6.2
Week 12	8956b	100b	150c	466c	350b	28b	44a	122ab	7644	63	138	236	336b	30a	40ab	134a
	±410	±3.0	±3.2	±4.0	±12.2	±1.3	±2.2	±3.0	±198	±4.8	±6.4	±18.4	±10.1	±1.6	±3.0	±3.8
Week 18	9484ab	114a	164b	540b	240c	22c	34b	114b	7642	70	146	256	316b	24ab	36bc	124ab
	±186	±4.0	±3.9	±5.3	±11.0	±1.0	±2.2	±3.2	±160	±8.1	±14.0	±6.8	±4.2	±3.0	±2.0	±4.3
Week 24	9911a	124a	180a	588a	222c	18c	30b	100c	7780	68	176	262	312b	21b	32c	118b
	±244	±3.1	±2.4	±9.8	±8.0	±1.2	±3.0	±4.0	±254	±3.0	±4.4	±14.1	±5.1	±2.2	±1.4	±6.0

Each value in the table is the mean of four replicates ±SD. Mean values with different superscript letters in the same column differ significantly ( $p < 0.05$ )

and Compost<sub>effective microorganisms1</sub> are shown in Fig.1 (b). pH decreased in both composts, and ranged between 7.2–6.9 and 7.2–6.7 in Compost<sub>plain</sub> and Compost<sub>effective microorganisms1</sub>, respectively. These results show that pH of Compost<sub>effective microorganisms1</sub> remained lower as compared to Compost<sub>plain</sub>. A probable reason for this could be that the microbes effective microorganisms1 presented in released organic acids, for breakdown of organic matter, resulting in lowering of pH. The decrease in pH occur as a result of organic acids produced by microbes as supported by the research conducted by Himanen and Hänninen (2009), Gondek *et al.* (2014) and Kharrazi *et al.* (2014).

Fig. 1(c) shows a faster rate of carbon degradation in Compost<sub>effective microorganisms1</sub> as compared to Compost<sub>plain</sub>. However, it disappeared after week 12. Faster degradation of carbon that the composting process was slower in the compost not supplemented with effective microorganisms1. The results were supported by the research conducted by Hu *et al.* (2013) and Sharma *et al.* (2014). Total nitrogen content (Fig. 1(d), initially decreased up to week 12 in Compost<sub>effective microorganisms1</sub>, and after that it remained stable. In Compost<sub>plain</sub>, the total nitrogen content decreased till week 18, after that became steady. The changes in nitrogen content could be attributed to nitrogen loss in the form of gaseous ammonia and nitrification (Himanen and Hänninen, 2009). The C/N ratio, Fig. 1(e) was noticed to be stable at week 6 in Compost<sub>effective microorganisms1</sub>, while in Compost<sub>plain</sub> the ratio stabilized at week 12. This indicated earlier completion of the composting process in Compost<sub>effective microorganisms1</sub>. These results are in confirmation with the previous reports of Chaturvedi *et al.* (2010) and Nakasaki *et al.* (2013), where microbes were discovered to stimulate and accelerate degradation of organic matter.

The total and DTPA extractable metal micronutrients content observed in the present study are detailed in Table 1. A similar increase in total micronutrient content was found in both compost types, during the composting process. This increase could be attributed to decrease in mass of composting material, caused by mineralization of organic matter. Interestingly, in both types of compost, the micronutrient content extracted by DTPA was observed to have reduced with the composting process. The DTPA-extractable portion is considered important, as it is a fraction of total nutrients that are available to plants (Tsadilas *et al.*, 2013; Zhang *et al.*, 2014). A decrease in DTPA extractable metal micronutrients could be attributed to the formation of organic matter-metal complexes during composting. This supposition is supported by the results of Thiyagarajan *et al.* (2009), who showed that the oxide-bound fractions and organic matter-metal complexes [non-available (residual) forms] of nutrients are produced during composting and maturation.

In conclusion, these results suggest that effective microorganisms1 could be useful for rapid composting and improving compost quality. Therefore, further studies are required to assess the fate of pathogenic microbes in effective microorganisms1-induced composting. Further, to improve the content of micronutrients available to plants in composts, exploitation of various nutrient solubilizing bacteria or neutral-salt addition should be examined.

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