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ORIGINAL PAPER

Investigation of effective microorganisms bioaugmentation in an on-site aerobic food waste composter

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Abstract

On-site handling of food wastes is an environmental alternative that will divert a significant portion of the wastes from landfills and reduce the collection costs subsequently. Regarding conventional composting systems such as windrows and aerated static piles that need large space and a long time to reach maturity; in-vessel bioreactors besides microbial inoculants can accelerate the process by occupying less space. This bioaugmentation was investigated during 40 days by using effective microorganisms, including fungi and bacteria such as *Yeast*, *Lactic acid bacteria*, and *Actinomycetes*, inside a 40-L composting vessel that was able to rotate and stimulate the temperature of the mass (30, 50, and 70 °C). The moisture content was maintained at over 40% with effective microorganism solutions. Food waste was prepared from the university restaurant kitchen then mixed with yard trimmings and wood chips as feedstock. In experiments with effective microorganisms, the thermophilic phase lasted longer (4 days), decomposition rate increased (16.1%), the compost reached maximum temperature faster (3 days), and the maturation period decreased (4 weeks). The most bioaugmentation occurred in the experiment with 50 °C thermal stimulation and 1.25-L effective microorganism solution which the carbon to nitrogen ratio of feedstock decreases 40.74% compared to 9.5% without effective microorganisms after two weeks. Proper carbon to nitrogen ratio (16 ± 0.5), negligible *E.coli*, *Salmonella*, and heavy metals of the humus, besides good germination index ($> 100\%$), was accepted as a good quality compost. However, the maturation period would be more than 4 weeks without using effective microorganisms and thermal stimulation.

Keywords Accelerated composting · Bioaugmentation · In-vessel bioreactor · On-site treatment · Microbial inoculants · Effective microorganisms

Introduction

Food waste (FW) is characterized by high organic matter and moisture content (Meng et al. 2015; Guo et al. 2018). FW includes uneaten food and food preparation from residences, institutional and industrial sources such as restaurants, school cafeterias, and factory lunchrooms. Most of them are kitchen waste produced from household and restaurant kitchens (Yang et al. 2013). Large amounts of FW generated by homes, restaurants, and food industries cause environmental problems, so the appropriate and efficient disposal of FW is a global environmental concern (Nourbakhshsamani et al. 2021; Charkhestani and Yousefi Kebria 2022).

Regarding FW characteristics, composting has proven to be a proper choice to deal with FW by less environmental pollutants and a more useful final product (Benito et al. 2006; Wei et al. 2017). According to the United States Environmental Protection Agency (EPA) and Alberta Agricultural, Food, and Rural Development (AAFRD) composting of organic municipal solid waste (MSW) can be done using three common methods including aerated windrow, aerated static pile, and in-vessel composting. Aerated or turned windrow composting is suited for large volumes such as that generated by entire communities and collected by local governments. This method includes the placement of the organic waste into long and narrow piles called "windrows" and aerating them periodically by either manually or mechanically turning the piles (EPA 2021). If the windrow

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is ventilated passively, there is a high risk of oxygen depletion, unpleasant odors, and greenhouse gases. The ideal pile height is between four and eight feet with a width of 14 to 16 feet. Windrow composting is a large-scale operation. Some advantages are quick drying with high temperatures, a drier product, the ability to handle high volumes of materials, good product stability, and low capital investment. Disadvantages are high operational costs, not space-efficient, vulnerability to climate changes, and emission of odors.

Alternatively, manure can be piled statically, and airflow can be introduced through the pile by installing ventilation pipes inside the system (Liu and Wang 2020). This method produces compost relatively quickly (within three to six months). The dimensions of the static pile are limited by the amount of aeration that can be supplied by the blowers and the characteristics of the waste (Dentel and Qi 2014). Since there is no physical turning, this method requires careful monitoring to ensure that the outside of the pile heats up as much as the core. Also, it may require significant cost and technical assistance to purchase, install, and maintain equipment such as blowers, pipes, sensors, and fans (EPA 2021). Compared to the windrow method shows that it requires less land and it is more vulnerable to climate impacts. Some other benefits of aerated static piles are high levels of pathogens destruction, good odor control, and good product stabilization (Abdoli et al. 2019).

The third common method which produces compost in just a few weeks is in-vessel composting. It can process large amounts of waste without taking up as much space as the windrow method. This method involves feeding organic materials into a drum, silo, or similar equipment which allows good control of temperature, moisture, and airflow. The material is mechanically turned or mixed to make sure the material is aerated. The size of the vessel can vary in size and capacity. This method is expensive and may require technical expertise to operate it properly. However, it uses much less land and labor than windrow composting besides producing very little odor or leachate (EPA 2021).

So, different factors such as capital and operating costs, land availability, operational complexity, odor emission, maturation, stabilization period, aeration system, and better processing control can be considered in choosing the best method (AAFRD 2005). By considering these tips, in-vessel composting is more controlled, requires less space, has high process efficiency, good odor control, and results in fewer operational problems than other methods (Cekmecelioglu et al. 2005; Kim et al. 2008). However, high capital cost and dependency on specialized mechanical and electrical equipment are some disadvantages of this method. The in-vessel composting technique allows the highest degree of temperature control (Thomas et al. 2020). Furthermore, it can be used to decrease the processing time of composting significantly (An et al. 2012).

In addition, if composting occurs on-site (e.g. at home), it will be diverting a significant portion of the waste stream from landfills and greenhouse gas emission will be reduced subsequently (Adhikari et al. 2013). Home-scale composting has not been studied broadly as compared to industrial-scale composting. Home composting, if conducted appropriately, can produce similar or even better levels of compost stability compared to industrial composting because of better control of the material treated, fewer impurities, higher moisture content, lower heavy metals concentration (Barrena et al. 2014). In addition, it reduces the produced leachate during waste transfer to landfill or composting plants (Nourbakhshsamani et al. 2021). Due to the undersized scale of home composters, the temperature is not sufficient to heat up to eliminate the pathogen effectively (Orthodoxou et al. 2015). Time is also an important factor in on-site composting; the less it takes the better.

The decomposition process is relatively lengthy. Human intervention is attempted to improve the efficiency of this natural process by utilization of microbial inoculants (MI) (Fan et al. 2017). Inoculants are products that have living microorganisms in their composition (Santos et al. 2019). It has been widely studied that the activity of microorganisms causes the decomposition of organic matter into simpler forms. For example, the widely used microbial *Bacillus* sp. is a cellulose and lignin degrader and *Trichoderma* sp. is known to degrade hemicelluloses (Wu et al. 2019). MI is sometimes referred to as activators and accelerators. Co-composting with MI seems to be a sustainable treatment option currently practiced, however, it is not possible to comment on its cost-effectiveness at industrial or small scale applications. A practice in India showed that the overall cost for co-composting of septage with mixed organic wastes was lower compared to other treatments because of higher available microorganisms in the organic waste (Thomas et al. 2020). However, a study in Thailand revealed that it might not be necessary to add commercial inoculants to facilitate composting of household organic waste due to the slight improvement of the finished compost (Karnchanawong and Nissaikla 2014). So, the efficiency of inoculants varies according to the composting conditions (Fan et al. 2018).

The great majority of the early manufactured inoculants contained only one species of microorganism. However, over the past decade, the use of inoculants containing microorganisms of "different types" has increased (Santos et al. 2019). One of these MIs containing many kinds of naturally occurring beneficial microbes is Effective Microorganisms (EM) that have been used widely in nature and farming (Raja Namasivayam and Bharani 2012). The major microbiological components of compost are bacteria and fungi. Moreover, *Actinomycetes*, while a particular type of bacteria is considered as the third main component for their ability to degrade the more recalcitrant compounds (Karnchanawong and Nissaikla 2014). EM is a people-friendly and environmentally safe product of the EM Research Organization (EMRO) that achieves synergistic effects (bioaugmentation) by combining these beneficial microorganisms such as *Lactic acid bacteria*, *Actinomycetes*, *Yeast*, *Fungi*, and *Phototrophic bacteria*. It was developed by Professor Teruo Higa in 1982 (EMRO 2021). In some cases, native microorganisms cannot degrade a particular contaminant (Speight 2017). Therefore, bioaugmentation offers a way to supply enough specific microorganisms in sufficient numbers to improve the existing populations and complete the biodegradation (Maier and Gentry 2015).

Some studies found that the addition of certain EM microbial agents could increase the number of effective microorganisms, which could promote the decomposition of organic matter and accelerate the composting process to achieve rapid biological composting (Qu et al. 2019). Wang et al. (2014) researched cattle manure enhanced composting. Sharma et al. (2014) proved that the application of EM in compost resulted in hastening the composting of paddy straw. They showed high activity of hydrolytic enzymes and microbial activity (Sharma et al. 2014). Jusoh et al. (2013) stated that EM can reduce odor and increase the final product's decomposition rate and nutrient contents. The results showed that the composting period was shortened with the addition of the EM inoculum (Wang et al. 2014). However, Qian et al. (2014) showed that the cost of composting was also increased. They believed that the addition of the EM microbial agent was not worth considering (Qian et al. 2014).

There are different points of view about the bioaugmentation effect of EM in FW composting. Some researchers believe that the existing microbial community in the waste is enough for degradation (Abdullah et al. 2013), and EM does not have a remarkable effect in small-scale FW composting (Nair and Okamitsu 2010). Some others reported that it can enhance enzymatic activities and promote and accelerate biodegradation of organic matter and composting process (Sarkar et al. 2011; Patidar et al. 2013). So, investigation of the current status in on-site composting of FW with EM shows that the differences are in the type of reactors, the amount of feedstock used, and control of influencing factors which finally leads to a reduction in the maturation and stability time.

This study aims to investigate the bioaugmentation effects of EM on the on-site FW composting in an in-vessel bioreactor by different thermal stimulations and EM concentrations. To reach this goal, important physicochemical properties of the feedstock including temperature, pH, electric conductivity (EC), moisture content (MC), total organic carbon (TOC), total organic matter (TOM), total Kjeldahl nitrogen (TKN), and carbon to nitrogen ratio (C/N) were monitored along the composting process. In addition, germination index (GI), heavy metals, pathogens, nitrogen, potassium, and phosphate contents of the optimum experiment were determined to obtain the composting duration and quality. This study was conducted at Noshirvani University of Technology, Babol, Iran. The experiments were carried out from February 2020 to the end of January 2021.

Materials and methods

In-vessel bioreactor

Figure 1 shows the schematic and general view of an aerobic in-vessel bioreactor used in this study. The average rate of Iran's MSW generation in the last decade (2009–2019) was 0.745 kg/capita/day (Esmaeilizadeh et al. 2020). According to the literature, FW constitutes about 59% of MSW with about 0.5 kg/L wet bulk density (Adhikari et al. 2013; Charkhestani and Yousefi Kebria 2022). So, the vessel has been designed with 40-L usable volume which was proper for collecting FW during a week for a family with 5 persons. It has been designed suitably for on-site practice with an interior composting vessel of 35 cm * 30 cm * 45 cm (length*width*height) from stainless steel and an exterior size of 40 cm * 40 cm * 77.5 cm with a top cover from an Iron sheet with a thickness of 1 mm. Aeration occurred by gas outlet fan, which suctions the air through the air inlet window. Three attached bars to a shaft with 15 cm length were used for turning the mass. The feedstock was heated with a curve-shaped thermal element and a thermostat to set different temperatures from 0 to 100 °C as shown in Fig. 1. (II). The rate of turning was set at 2 rpm according to a study that works on the accelerated composting process of garbage for prolonged periods (Nishino et al. 2003). The bars turn the mass for 10 min, then rest for 20 min. During the turning heating element works simultaneously. It seems the mass has been turned and heated for 8 h during the day. It was assumed that if heating was not done simultaneously with the turning protocol, it would lead to a decrease in temperature of the mass and inhibit the activity of microorganisms consequently. On the other hand, heating for more than 8 h leads to a decrease in MC and also higher energy consumption.

Feedstock

For the experiment, the FW was collected from the University restaurant kitchen (Babol Noshirvani University of Technology, Iran), consisting of fruits scraps, vegetable scraps, tea wastes, steamed rice, and other organic discarded parts during a day. For example, it contained scraps of carrots, potatoes, onions, cucumbers, tomatoes, eggshells, etc. For more realistic results, simulated FW did not use. So,

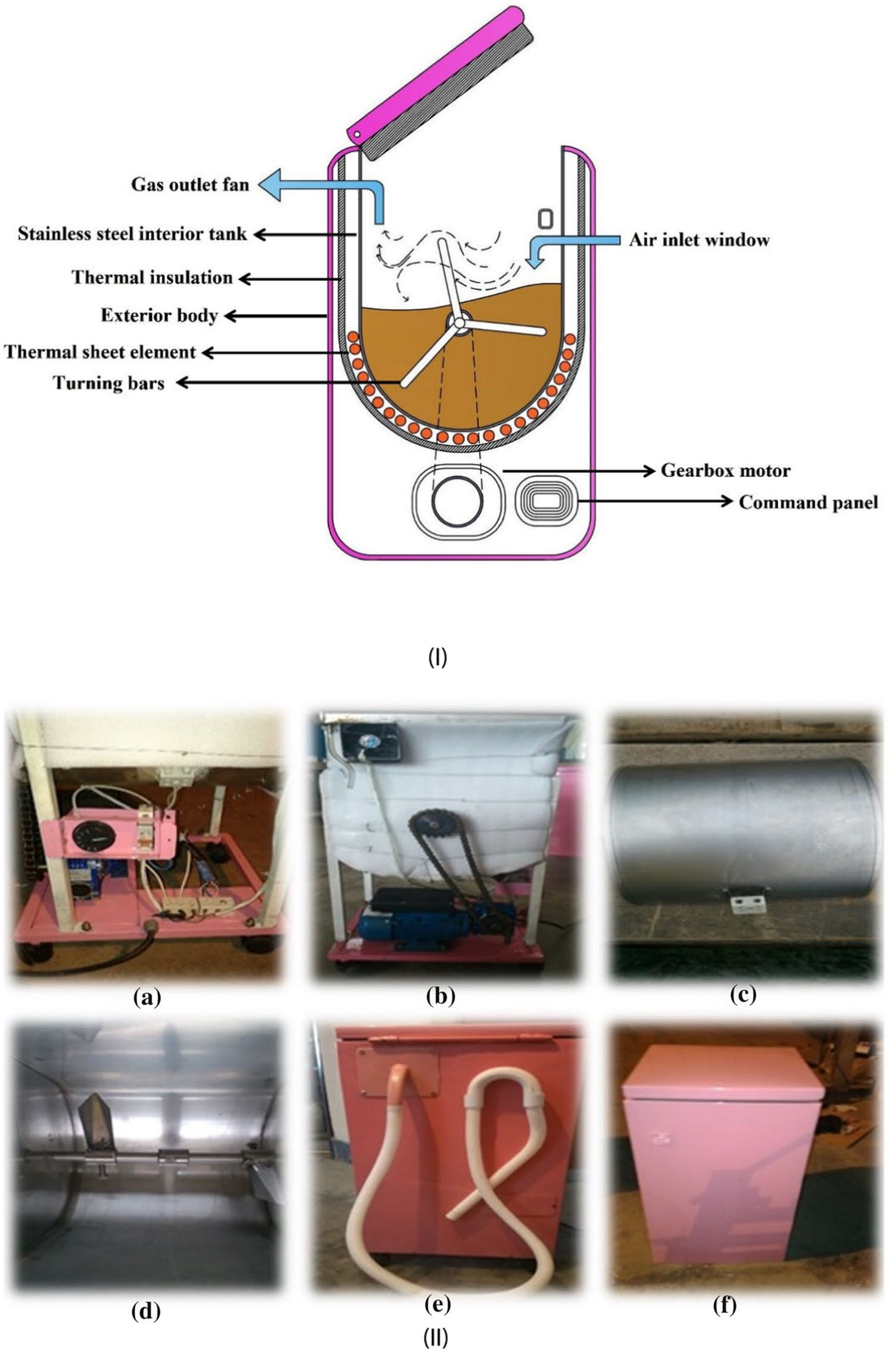


Fig. 1 **I** Schematic parts of in-vessel bioreactor. **II** Photo of **a** command panel, **b** Gearbox motor and exhaust fan, **c** Curve-shaped element, **d** The bars and curve-shaped empty vessel, **e** Back of the bioreactor, and **f** Front of the bioreactor

one plastic bin was placed in the kitchen on a specific day of the week and the shredded wastes were dumped into it. Then, the required volume of sample was taken from the bin then mixed with bulking agents (BA) and EM according to "Experimental design section". BA like yard trimmings and wood chips can reduce odors and leachate generation besides C/N and MC regulation (Adhikari et al. 2009). Also, the proportion of starting materials used in the composting mixture influences the degradation of organic matter, nitrogen dynamics of the process, and its toxicity on germinating plants. The proportions with greater amounts of FW to a BA (70:30 v:v) had higher concentrations of mineral matter, higher peak temperature, and a better initial carbon-to-nitrogen ratio compared to 50:50 or 30:70 ratio (Guidoni et al. 2018). According to Adhikari et al. (2013), FW combined with yard trimmings gives higher MC and a lower C/N ratio than wood chips. So, in this study, both of them have been used equally as BA which was collected from the landscape in the university's campus. By considering 50% of the bioreactor's volume (20-L from 40-L), the 8 kg feedstock, including 6 kg restaurant kitchen waste, 1 kg yard trimmings, and 1 kg wood chips, were fed to the batch composting bioreactor (3:1 FW/BA ratio on a wet mass basis). The main characteristics of feedstock are presented in Table 1.

Effective microorganisms (EM)

The beneficial microorganisms such as *Lactic acid bacteria*, *Actinomycetes*, and *Fungi* are the major microbiological components of compost which have abilities to degrade the more recalcitrant compounds (Karnchanawong and Nissaikla 2014). These microorganisms are physiologically compatible with one another and can coexist in liquid culture. *Lactic acid bacteria* contributed to modify of environmental conditions allowing for the activity of important microorganisms in composting (Tran et al. 2015). They accelerated composting by solving the low pH problem, inhibiting the production of acetic acids, and increasing the proliferation of fungi having the ability to degrade organic matter (Tran et al. 2015; Nakasaki et al. 2019). *Actinomycetes* improved cellulase activities, increased the content of humic substances, and accelerated production of the key enzymes, including *CMCase*, *Xylanase*, *lignin peroxidase*, etc. (Zhao et al. 2017; Wei et al. 2019). They also alleviated CO_2 emissions during composting. *Fungi* are the most abundant and fastest emerging microorganisms during composting which can produce extracellular enzymes accountable for cellulose and lignin degradation during composting (Awasthi et al. 2014). *Yeast* as a kind of fungi can degrade organic acids and thus increase the pH value beyond the neutral pH of composting material (Nakasaki et al. 2013). So, bioaugmentation with these EM offers a way to provide specific microorganisms in sufficient numbers to enhance the existing populations and complete the biodegradation (Maier and Gentry 2015). The commercial EM used in this study contains fungi like *Yeast,* and bacteria including *Actinomycetes*, *Lactic acid*, and *Photosynthetic bacteria* (Daly and Stewart 1999). EM includes both aerobic and anaerobic microorganisms which co-exist in an environment of around 3.5 pH (Mandalaywala et al. 2017). EM solution is a brownish liquid with a pleasant odor. The activated commercial EM (EM-1®) used in this study was supplied by Emkanpazir Pars Co., LTD., Iran. According to Fan et al. (2018), the activated EM contains ~ 107 Colony Forming Unit (CFU)/mL of fungi and ~ 108 CFU/mL of bacteria. The specified species are *Streptomyces albus*, *Propionibacterium freudenreichii*, *Streptococcus lactis*, *Aspergillus oryzae*, *Mucor hiemalis*, *Saccharomyces cerevisiae*, *Candida utilis* by 105 minimum viable organisms per mL and an unspecified number of *Lactobacillus* sp., *Rhodopseudomonas* sp., and *Streptomyces griseus* (Fan et al. 2018).

Experimental design

Nishino et al. (2003) worked on thermoacidophilic conditions for composting of garbage in prolonged periods with 85 °C constant temperature. It was ultrahigh temperature and consequently consumed high energy. The development of thermophilic composting with ultrahigh temperature (over 75 °C) would inactivate the potential in viral pathogens, but it can affect microbial activity if it lasts longer than 24 h (Chang et al. 2019). In addition, mesophilic microorganisms thrive in 20–45 °C, normal thermophilic microorganisms thrive in 45–65 °C, extreme thermophiles thrive in 65–80 °C and hyperthermophiles that grow optimally at 80 °C and above (Noll 2001; Zeldes et al. 2015; Schiraldi and De Rosa 2016). Therefore, to reduce energy consumption and highlight the bioaugmentation effect of EM, three intermittent heating regimes below 80 °C were applied in Run 4 (70 °C), Run 3 (50 °C), and Run 2 (30 °C). Also, Run 1 was conducted without heating stimulation to compare the results.

Experimental conditions are presented in Table 2. Four experiments (Run 1–4) were considered to investigate the bioaugmentation effect of EM during the composting

Table 1 Physical and chemical properties of feedstock used for composting

Parameters	FW	Yard trimmings	Wood chips	Mixture (6:1:1 w/w)
pH	5.5 ± 0.2	6.5 ± 0.2	5.6 ± 0.1	5.75 ± 0.09
EC (mS/cm)	6 ± 0.1	6.4 ± 0.1	2.3 ± 0.2	6.3 ± 0.17
MC (%)	80 ± 3	35 ± 1.5	10 ± 1	67 ± 1.8
OM (% dry weight basis)	81.9 ± 2.7	63.1 ± 2.5	90 ± 1	82.3 ± 1.5
TOC (% dry weight basis)	43.7 ± 1.5	34.5 ± 1.9	47 ± 2	43.2 ± 1
N (% dry weight basis)	2 ± 0.3	1.8 ± 0.1	1 ± 0.1	1.6 ± 0.08
C/N (dry weight basis)	21.8 ± 2.2	19.2 ± 1.8	47 ± 2.3	27 ± 1.2
Bulk density (kg/L)	0.55 ± 0.1	0.1 ± 0.03	0.2 ± 0.02	0.45 ± 0.07

Values represent means and standard deviations ($N = 3$)

Table 2 Different experiment conditions during the study

Runs	Element Temperature (°C)	MC (%)	Moisture modifying (EM)*	Turning and heating plan
Run 1	Off	> 40	0.75 L in the first day + 0.5 L when MC reaches 40%	10 min turning and heating then 20 min rest
Run 2	30			
Run 3	50			
Run 4	70			

*Distilled water used as moisture modifier in control runs instead of EM

process compared to four control groups that used distilled water instead of EM. In all Runs, 0.75-L EM was sprayed on the feedstock initially to regulate the MC around 75%. Many researchers reported that effective MC during composting is between 40 and 60% (Hubbe et al. 2010). So, the MC during the process was maintained above 40% using 0.5-L activated EM in main experiments or distilled water in control groups whenever MC reached 40 ± 2%. To reduce the number of study variables, EM was used as an MC regulator. Regarding different thermal regimes of the experiments, different concentrations of EM were used during the processes subsequently.

Monitoring parameters

Temperature measurements were done daily throughout the process by Testo 925 thermometric probe. MC was controlled by a portable REOTEMP long stem compost moisture meter during a day. This method is not very accurate like the hand-squeeze test but it gives good knowledge about the MC of the feedstock without sampling. So, some days, for more accuracy, the MC was also determined as weight loss through drying samples at 105 °C in an electronic oven (Memmert, UNE 400, Germany) for 24 h and reported as the exact MC of feedstock.

To determine pH and EC, each sample was mixed with distilled water (1:10 w/v ratio) to make a solution. The prepared solutions were stirred at 300 rpm for 30 min. Then, after 30 min rest, the electrodes (827 pH-lab Metrohm and WTW 9310) were dipped in each sample prepared solution and recorded when it was stabilized (Sánchez-Monedero et al. 2001; Fan et al. 2018). TOC and TKN were measured by Walkley–Black wet digesting and Kjeldahl (Vaposest 30 s, Gerhardt, Germany) methods (Walkley and Black 1934; Leege 1998). Subsequently, the C/N ratio was monitored during the process, too.

Statistical analysis

One-way analysis of variance (ANOVA) was applied using SPSS ver. 26.0 Software to find out whether there was a significant difference ($p < 0.05$) among the Runs and their controls. In Post Hoc Multiple Comparisons, LSD for normally distributed data sets and Games-Howell for not normally distributed data sets was used. Statistical analysis provides proper data to determine the optimum Run.

Maturation and quality of the final compost

The C/N is one of the essential parameters to determine the maturity of compost. When waste is composted, generally C/N ratio is stabilized in the range of 15–20 (Goyal et al. 2005). Maturity is not described by a single property, and therefore, it is best assessed by measuring two or more compost characteristics. So, to find the compost quality and minimum time for maturation, other parameters like germination index, heavy metals, nutrients, and pathogen contents were determined for the Run which its C/N ratio was placed faster in the range of 15–20.

According to Bernal et al. (2009), germination tests present an index for phytotoxicity of the final compost (Bernal et al. 2009). Zucconi et al. (1981) stated that a germination index below 50% is considered an immature compost. To determine germination index, the mixture for measurement of pH and EC was filtered through a 0.45 µm filter membrane, then 10 cress seeds (Lepidium sativum) were distributed on paper tissue in Petri dishes and moistened with 10 mL of the filtrate extraction (Yang et al. 2013). Three replicates for each sample were incubated at 25 °C for 3 days in Binder FD 23 drying and heating chambers, Germany, while distilled water was used as control. The germination index (GI %) was calculated using the following equation:

$$\mathrm{GI}(\%) = 100 \times \frac{G_{\mathrm{sample}}}{G_{\mathrm{water}}} \tag{1}$$

where G_{sample} and G_{water} are the numbers of cress seeds germinated in the assay and water control.

According to Mao et al. (2021), pathogens are airborne and foodborne. Researchers are usually more concerned with foodborne pathogens like Escherichia coli (*E. coli*) and Salmonella because they use the compost for vegetable cultivation. (Mao et al. 2021). So, for the pathogen test, *E. coli*i and Salmonella were determined according to the methods stated by Fan et al. (2018) based on the Food and Drug Administration's (FDA) Bacteriological Analytical Manual. The nutrient contents (N, K_2O, and P_2O_5) were determined according to the standard methods of compost maturity analysis (Leege 1998). To determine heavy metals (Zn, Cu, Cr, Ni, Co, As, Hg, Mo, Cd, and Pb) concentrations in the final compost, one gram of the sample was dried and ground then digested by 36 mL HNO_3 and $HClO_4$ (5:1 ratio, v/v) followed by atomic absorption spectrometry (Bazrafshan et al. 2016).

Results and discussion

Temperature

Temperature is one of the important parameters that reveal microorganisms' activity (Sarkar et al. 2010; Kopčić et al. 2014). Figure 2 shows the temperature profile of interior mass during the experiments. As shown in Fig. 2, Runs with EM have a higher mass temperature, reach faster to maximum and the thermophilic phase (> 45 °C) lasts longer than the controls. However, statistical analysis shows that the temperature profile in each Run with EM is not significantly different ($p < 0.05$) from its control Run. Furthermore, ANOVA analysis between four main experiments with EM as well as four control groups indicates a significant difference in both of the groups that were acceptable regarding different thermal regimes.

Figure 2c reveals that mass temperature in Run 3 with 0.75 L initial EM reached thermophilic phase (> 45 °C) on the first day and reached its maximum temperature (76 ± 1 °C) after three days that is faster than other EM runs with the same concentration of EM. It indicates easy decomposing of organic matter and pathogens elimination (Strauch and Ballarini 1994; Awasthi et al. 2018). Fan et al. (2018) reached 45 °C after 5 days by using 1.2-L EM for 4 kg feedstock with the highest temperature (50 °C) on the 7 day. However, Awasthi et al. (2018) reached the thermophilic phase after 1 day by using a 2-L bacterial consortium for 25 kg feedstock with the highest temperature (68 °C) on the 5 day. One of the main differences between this study and the literature is thermal stimulation.

Thermal stimulation may help the mass temperature to be in the range of the thermophilic phase during the process. So, it is not clear the difference between real thermophilic phase duration with EM and control. However, Run 1 (with 0.75 L initial EM and without heating stimulation) highlighted this difference which showed a longer thermophilic phase for EM (7 days) compared to the control (3 days) that was due to the higher microbial activity. In addition, the thermophilic phase reached a faster rate rather than the control without EM (3 days faster).

Regarding Fig. 2, intermittent heating, while feedstock is turning, prevents the loss of mass temperature. In addition, at the early stages of the process when the activity of microorganisms is high, the temperature of the mass sometimes rises above the temperature of the element, and gradually, with the decrease of the activity of microorganisms, the temperature of the mass is almost constant and approaches the temperature of the element.

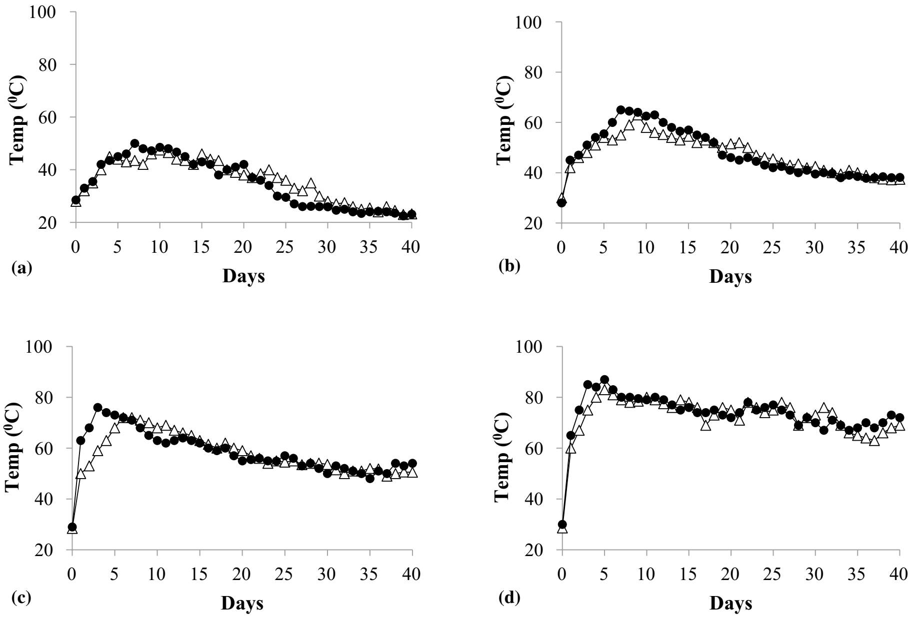


Fig. 2 Temperature profile during different experiments; **a** Run 1, **b** Run 2, **c** Run 3, and **d** Run 4. —●— EM; —△— Control

Moisture content

For all Runs, 0.75 L EM was sprayed on feedstock initially then obtained MC as presented in Fig. 3. In control Runs, distilled water was used. Regarding conducted pretests, except initial augmentation of EM, we decided to modify MC by spraying 0.5 L EM on feedstock in the bioreactor when MC reaches around 40 ± 2% which is clear in the Figures. As demonstrated, by increasing the bioreactor temperature, MC drastically decreases and control of the MC needs more EM. Apparently, in terms of additive consumption, 4 times addition of EM (2.75 L) in Run 4 is not cost-effective compared with the literature (Awasthi et al. 2018; Fan et al. 2018); however, discussing the proper augmentation of EM to obtain the best results needs the investigation of other maturation indices as follows.

pH and electrical conductivity

Microbial activity can affect pH during the initial stages of composting (Awasthi et al. 2018). Several studies suggested neutral pH for optimal composting (Zucconi et al. 1981; Kuok et al. 2012; Kopčić et al. 2014). Although in this study, pH was around 5.8 ± 0.15 and 5.08 ± 0.14 in the controls and Runs with EM, respectively, after initial moisture modifying. As shown in Fig. 4a, b, c the drop in pH occurred in the early stages of composting that is due to the formation of organic acids (Bazrafshan et al.

Fig. 3 MC changes during the experiments and EM addition according to experimental design; **a** Run 1, **b** Run 2, **c** Run 3, and **d** Run 4. Results are expressed as the mean value and standard deviation of triplicate samples

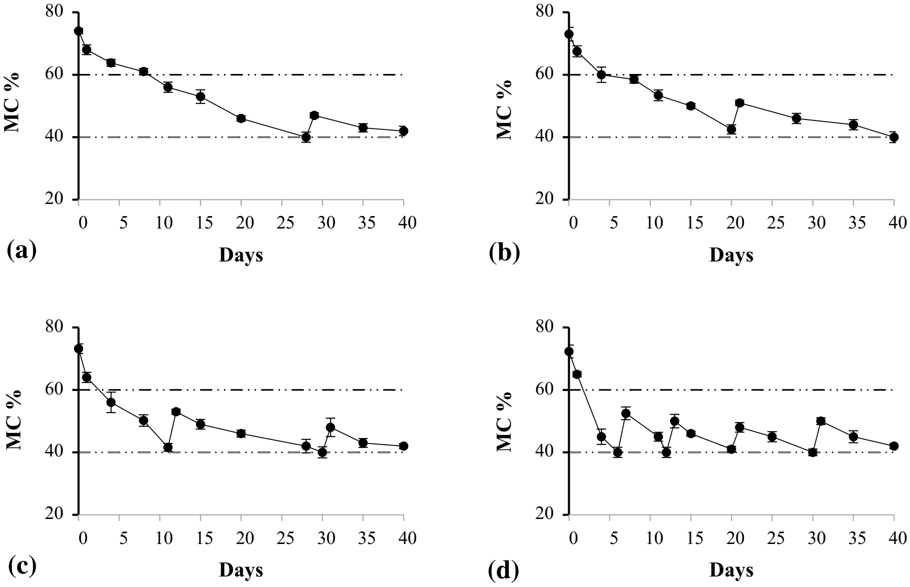
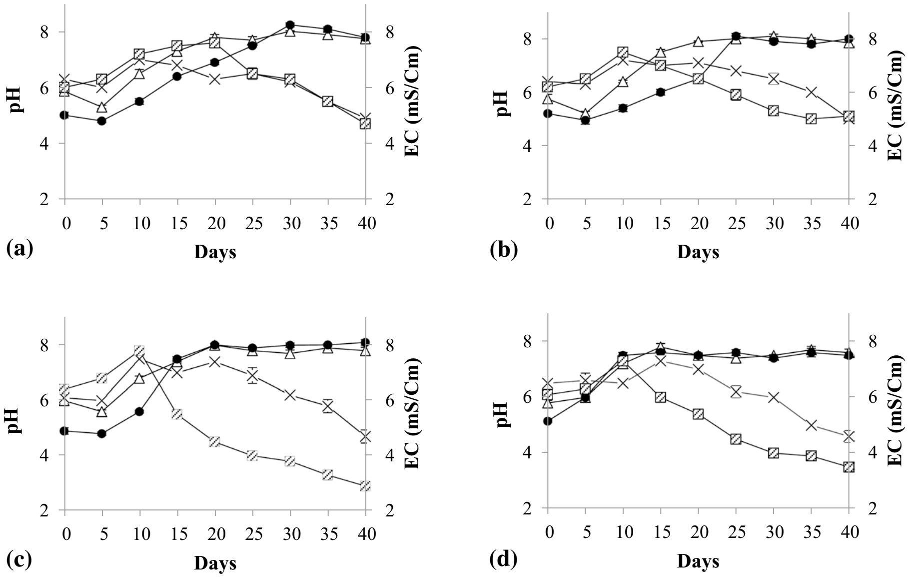


Fig. 4 Change in pH and EC during the experiments; **a** Run 1, **b** Run 2, **c** Run 3, and **d** Run 4. Results are expressed as the mean value and standard deviation of triplicate samples. ——●—— EM (pH); ——△—— Control (pH); -------▨------- EM (EC); --------×-------Control (EC)



2016). According to Mandalaywala et al. (2017) acidic environment is favorable for the growth of EM. By decomposition of organic acids, releasing volatile ammonia, and mineralization, increasing pH was observed. Comparing the trends in Fig. 4 shows that pH increases 3.4, 2.7, 3.2, and 2.4 from the least to highest in 25, 20, 15, and 10 days in Runs 1 to 4, respectively. However, in control Runs, it increases 2.7, 2.9, 2.4, and 2. As demonstrated, in thermal stimulation above 30 °C (Run 3 and Run 4), pH changes have been stabled 2 weeks earlier due to the EM bioaugmentation effect that causes an increase in decomposition rate which is more significant when EM concentration and mass temperature have been increased. Statistical analysis showed that there is no significant difference ($p > 0.05$) between Run with EM and without EM in all 4 Runs.

Electrical conductivity is a factor of salinity and affects the quality of final compost. Figure 4 shows the variation of EC. As seen in Fig. 4, EC has been increased then decreased in Run 1 to 4. The increase occurs during the early thermophilic phase due to the release of mineral salts through the decomposition of organic substances (Gao et al. 2010). However, the reduction of water-soluble substances such as organic acids attributed to the reduction of EC in control groups (Tang et al. 2004). By thermal stimulation in different Runs, EC reduction has been accelerated and the largest reduction happened in Run 3 (6.4 to 2.9 ± 0.14 ds/m). Run 4 shows that more concentration of EM does not have any special bioaugmentation effect than Run 3 (6.1 to 3.5 ± 0.1 ds/m). In addition, statistical analysis revealed that a significant difference ($p < 0.05$) with its control has been observed only in Run 3. Although, ANOVA analysis between four main experiments with EM as well as four control groups showed no significant difference ($p > 0.05$).

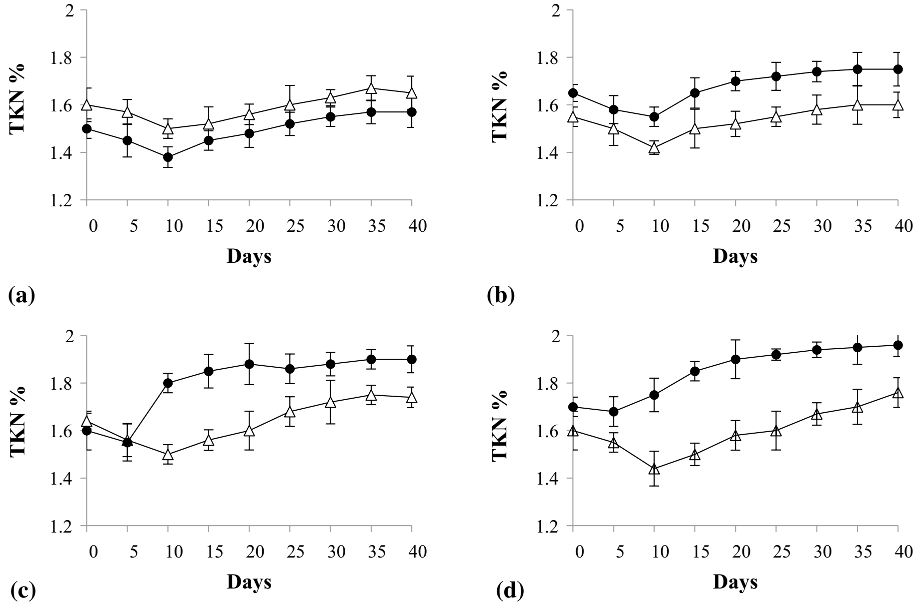
Total Kjeldahl's nitrogen

TKN depends on the rate of decomposition and initial nitrogen content of the waste (Kaviraj and Sharma 2003). TKN has a declining trend in the initial stage of composting due to the loss of nitrogen in the form of ammonia (Mengistu et al. 2017). Figures 5c, d show that the increasing rate of TKN begins about 5 days faster for EM Runs than the controls. It may be attributed to the higher loss of organic matter rather than the loss of NH3 during the active phase (Bernai et al. 1998). As demonstrated in Fig. 5c, d, the most TKN augmentation was 18.75 (Run 3) and 15.3% (Run 4) in the EM Runs verses 6.09 and 10% in the controls, respectively. So, compost with EM has more N due to the higher nitrogen-fixing bacteria activity (Seal et al. 2012; Jusoh et al. 2013). However, some studies state that EM did not have any special effect on composting of kitchen waste (Nair and Okamitsu 2010). Statistical analysis between the main Run and its control shows a significant difference between them in all Runs. Unlike Runs with EM, no significant difference was observed between the 4 control groups. LSD analysis showed that Run 1 (EM) has far from other EM Runs which highlights the effects of thermal stimulation and the role of EM.

Total organic carbon and C/N ratio

TOC contents in both EM and control Runs gradually decreased during composting. As shown in Fig. 6a–d, TOC has been reduced 25.7, 37.3, 42.1, and 42% in Runs 1 to 4, respectively, by the addition of EM and thermal stimulation. In all Runs, TOC reduction was higher in the EM inoculated mass rather than the controls (21.6, 29.9, 26, and 28.3%)

Fig. 5 Changes in TKN during the experiments; **a** Run 1, **b** Run 2, **c** Run 3, and **d** Run 4. Results are expressed as the mean value and standard deviation of triplicate samples. ——●—— EM; ——△——Control



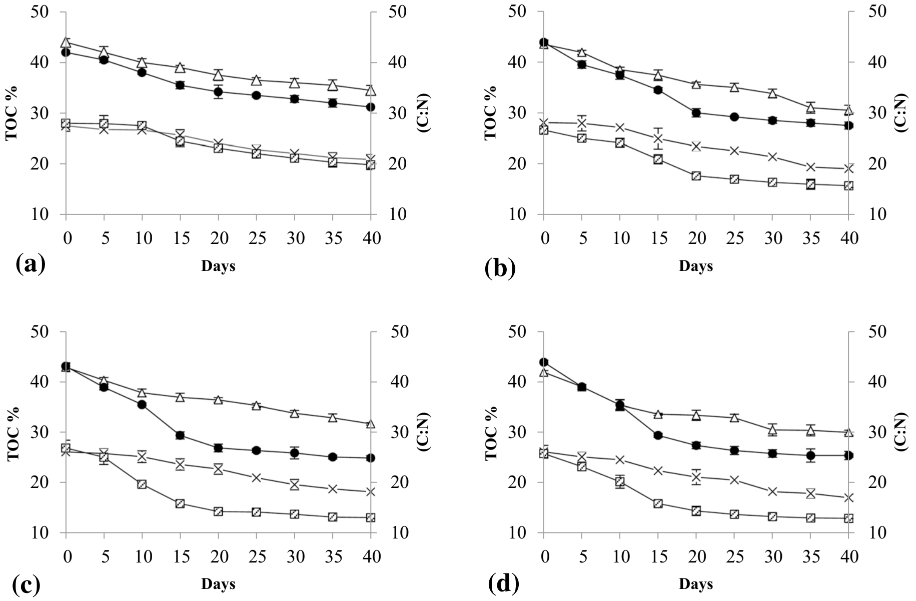


Fig. 6 Changes in TOC and C/N ratio during the experiments; **a** Run 1, **b** Run 2, **c** Run 3, and **d** Run 4. Results are expressed as the mean value and standard deviation of triplicate samples. ——●—— EM (pH); ——△—— Control (pH); -------▨------- EM (EC); -------×------- Control (EC)

which is similar to the results of Awasthi et al. (2018) that worked on in-vessel co-composting of FW with a bacterial consortium. Kim et al. (2008) worked on centralized in-vessel composting of FW. They reported TOC content decreases from 55 to 34% (38% reduction) during 35 days for 14,000 kg/day flow rate of mixing material. It is near to the best TOC reduction efficiency of this study (42.13% reduction in 40 days for Run 3) considering its different scale. By comparing Fig. 6c, d, it is obvious the temperature above 70 °C and subsequently higher EM consumption have no significant role in the TOC reduction trend. In addition, ANOVA analysis between the Runs by EM and without EM shows a significant difference ($p < 0.05$) only in Run 3.

The C/N ratio is an important factor to investigate compost maturity (Goyal et al. 2005). As seen in Fig. 6, by decomposition of organic matter through the microorganisms, the C/N ratio has been decreased. Run 3 and 4 demonstrated that enhancing bioreactor temperature and EM concentration make the C/N declining trend faster. In this study, the initial C/N ratio of the feedstock was 27 ± 1.2. To avoid nitrogen immobilization when matured compost is applied to soil, it should ideally have a value of about 15 (Erhart and Burian 1997). By considering this issue, in control Runs without EM during 40 days, C/N did not drop below 17.1 ± 0.56. In the experiments with EM, it dropped to 19.87 (−29.03%), 15.71 (−40.96%), 13.16 (−51.26%), and 13.01 (−49.73%) during 40 days in Run 1 to 4, respectively. According to Goyal et al. (2005), the C/N ratio stabilizes in the range of 15–20 when compost is matured. In Run 3 and 4, C/N reached 16 from an initial ratio of 27 and 25.9 after two weeks, respectively. Since the C/N ratio is one of the important indices of maturation, it is clear that most EM bioaugmentation effect occurs in Run 3 with a 40.7% reduction in C/N after 2 weeks which can decrease the maturation period to 14 days or less compared to 40 days or more without EM. In fact, in this stage, the compost is matured but not stabilized. Regarding the Temperature, OC, and C/N declining trend, it can be said that mature compost needs 1–2 weeks more to stabilize completely.

In comparing with literature, Awasthi et al. (2018) composted 25 kg FW mixed with sawdust using 2 L enriched bacterial consortium without thermal motivation, and the C/N ratio reached 16.18 (36.8% reduction) after 40 days. However, they could decrease only about 10% of C/N after 2 weeks. Fan et al. (2018) obtained a 61–66% final reduction of C/N after 8 weeks with 1.2 L EM for 4 kg simulated FW without daily turning, moisture-controlling, and thermal stimulation. However, they stated there was no significant difference in the C/N changes when using distilled water instead of EM. In addition, after 2 weeks, they did not obtain significant results in C/N reduction with EM compared to control. Abdullah et al. (2013) used a microbial solution consisting of seven types of bacteria and eight types of fungi isolated from soils as a starter culture for the kitchen-waste composting. They showed that the mixture with added starter culture and control have no significant difference in terms of TOC content, total nitrogen content, and C/N ratio and reached similar values at the end of composting. Nair and Okamitsu (2010) revealed that no significant difference was observed between the control and those inoculated with Trichoderma and EM in terms of the C/N ratio of the final product for on-site small scale household organic waste treatment during 28 days. However, it was observed that EM inoculation enhanced the reproductive rate of earthworms, and so probably created the best environment for vermicomposting. Patidar et al. (2013) observed significant decrement in MC, TOC, and C/N ratio and increment in temperature, and phosphate in thermophilic composting with microbial inoculation followed by vermicomposting. Sarkar et al. (2011) combined 35 different enzyme-producing bacterial strains by permutation combination to make different microbial consortia. They showed more than 50% degradation in 21 days for organic kitchen wastes while control with no inoculation showed only 36% degradation at the same time.

In this study, maximum C/N reduction occurred in Run 3 by 51.27% during 40 days compared to 30.28% in the control without EM. The significant results occurred after 2 weeks which C/N decreases 40.74% in Run 3 (with 1.25 L EM) compared to 9.5% in Run without EM. This trend was 38.17% compared to 14.4% for Run 4 (with 1.75 L EM). Lower energy consumption and less utilization of EM rather than Run 4, make Run 3 more cost-effective in C/N reduction.

In addition, statistical analysis of C/N changes during the experiments showed that there was a significant difference between all four Runs with EM ($p < 0.05$). However, this analysis did not present any differences between control groups ($p > 0.05$). So, Post Hoc Multiple Comparisons (LSD) were conducted between Runs with EM and results showed that Run 1 was far from the other Runs. It highlights the role of thermal stimulation that occurred in the other three Runs (2, 3, and 4). Comparing the C/N changes of each EM Run with its control demonstrated that there was a significant difference only between Run 3 and Run 4 with their control groups. Regarding higher consumption of EM and energy (higher temperature) in Run 4 besides other investigated conditions, Run 3 could be considered as the optimum Run of experiments. So, correlation analysis was conducted among parameters including Temperature, pH, EC, TOC, TKN, and C/N ratio for Run 3 and its control which has been presented in Table 3.

According to Table 3, in the optimum Run with EM, C/N is significantly correlated with parameters like pH and TKN compared to the control which is not. Regarding Fig. 4c pH of feedstock is almost acidic during the first 2 weeks which is favorable for the growth of EM (Mandalaywala et al. 2017). In addition, the greatest increase in pH (4.8–7.5) has occurred during this period that was coincided with the greatest decrease in C/N (40.74%) due to the decomposition of organic acids, releasing volatile ammonia and mineralization.

TKN is negatively correlated with TOC in a run with EM that is related to the activity and population of the presented humus. Higher consumption of organic carbon by microorganisms facilitates increasing the nitrogen content compared to the control Run without EM. In a control Run without EM, TOC is correlated with temperature which indicates that temperature stimulation can aid the composting process where the temperature is not high enough.

Quality of compost

During the first two weeks of the 40 days monitoring, Run 3 showed the best efficiency in terms of TOC and C/N reduction, temperature, and pH profile. Since it was assumed that EM shows their bioaugmentation effect in reducing compost production time, other parameters like heavy metals, nutrient contents, and germination index were examined to investigate the maturity and quality of the produced humus, after 2 weeks for Run 3. The results have been presented in Table 4.

The germination test presents the phytotoxicity and maturity of the compost. It directly indicates if the compost has an inhibitory effect on plant growth or not (Awasthi et al. 2018). Initially, the germination indices were very low due to the active decomposition of organic materials that generated a variety of toxic compounds and thus reduced the seed germination (Wang et al. 2018).

Table 3 Correlations of the investigated parameters for the optimum Run

	Temperature	pH	EC	TOC	TKN	C/N
Optimum run (with EM)						
Temperature	1	0.689264	−0.39424	−0.78915	0.710915	−0.79014
pH		1	−0.87726	**−0.97654**	**0.930119**	**−0.97309**
EC			1	0.850379	−0.70616	0.800797
TOC				1	**−0.9279**	**0.990872**
TKN					1	**−0.96861**
C/N						1
Control (without EM)						
Temperature	1	0.837341	−0.15837	**−0.95259**	0.532	−0.87398
pH		1	0.037928	−0.85803	0.530894	−0.818
EC			1	0.329468	−0.69231	0.493862
TOC				1	−0.64725	**0.950168**
TKN					1	−0.8516
C/N						1

Correlation above 90% between the parameters are shown in bold

Table 4 Quality of the compost and comparison with standards after 2 weeks (optimum run)

Parameters	Content	Iran standards[a]	Washington state standard, USA[b]	European commission countries[c]
N (%)	1.85 ± 0.07	1.25–1.66	> 1	–
K (K_2O%)	0.9 ± 0.05	0.5–1.8	–	–
P (P_2O_5%)	0.6 ± 0.08	0.3–3.8	–	–
TOC (%)	29.5 ± 0.7	> 25	–	–
OM (%)	50.8 ± 1.2	> 35	> 50	> 30
C/N	16.0 ± 0.5	15–20	15–20	–
MC (%)	49 ± 1.55	< 35	40–60	–
pH	7.5 ± 0.08	6–8	5.5–8	–
EC (ds/m)	5.5 ± 0.07	< 8	< 3	-
Zn (mg/Kg)	30.1	1300	1400	200–4000 (600)
Cr (mg/Kg)	15.8	150	600	70–200 (100)
Pb (mg/Kg)	5.2	200	150	70–750 (120)
Cd (mg/Kg)	0.06	10	10	0.7–10 (1.5)
Cu (mg/Kg)	17.1	650	750	70–1000 (200)
Ni (mg/Kg)	12.2	120	210	20–300 (50)
As (mg/Kg)	0.3	10	20	10–25 (15)
Germination index (%)	105 ± 5.8	> 70	> 95	-
E.Coli (MPN/g)	< 0.4	< 1000	< 1000	< 1000
Salmonella (MPN/4 g)	absent	< 3	absent	absent

[a] Institute of Standards and Industrial Research of Iran, standard No. 10716 and 13,321 (ISIRI 2008, 2011)

[b] Washington composting rules about composting facilities, permit requirements, and operating 17–350-220 (WAC 2019)

[c] European commission scientific and policy reports (Saveyn and Eder 2014)

The germination index of the humus with EM reached over 100% after two weeks, while in the control Run, it did not reach over 80% after 40 days. It shows proper mineralization due to the bioaugmentation effect of EM. Comparing with literature shows that Fan et al. (2018) reached 306% in the compost with EM after 8 weeks, however, control Run without EM reached 401.6%. So, giving more time to the discharged compost until completely stabilized can improve the GI.

To control microbial parameters of the humus, *E.Coli* and *Sallmonella* were determined. They were negligible and met the standards for pathogen content according to Table 4. It is maybe due to the separated feedstock that is less likely to be contaminated by pathogens or thermophilic temperatures during the process (Fan et al. 2018). Temperature above 55 °C for at least three days inactivates pathogens (Wichuk and McCartney 2007). It was achieved by a curved-shaped element in the bioreactor besides the activity of EM.

In the case of nutrient contents of the composted raw mixture, initial Nitrogen, Potassium and Phosphorus of humus were 1.60 ± 0.07%, 0.79 ± 0.06% and 0.21 ± 0.05% that after composting were 1.85 ± 0.07%, 0.9 ± 0.05% and 0.6 ± 0.08%, respectively in Run 3 with EM compared to 1.56 ± 0.04%, 0.85 ± 0.05% and 0.54 ± 0.03% in control. So, no significant bioaugmentation effect of EM was seen except in the N content compared to the control. Fan et al. (2018) reported the same finding of the EM effect on the nutrient contents. Although Jusoh et al. (2013) revealed that composting of rice straw with EM had a significantly more N, P, and K content ($P < 0.05$) compared to compost without EM.

The compost with hazardous components (i.e., heavy metals) limited its use in horticulture (Sánchez-Monedero et al. 2001). Heavy metals present in kitchen waste are extremely low which could be ignored in the humus that is following our results after two weeks (Manungufala et al. 2008). This low level of heavy metals or toxic compounds is a proper feature for the quality improvement of soil (Hogg et al. 2002).

The heavy metals, nutrient contents, microbial parameters, and germination index of the compost after two weeks were qualified as a first-grade compost according to the presented standards and it was suitable to use as fertilizer.

Conclusion

EM bioaugmentation in an on-site in-vessel FW composter was investigated. Thermal stimulation, turning the mass, and moisture controlling highlighted this bioaugmentation to reduce the time required for the maturation of compost. In experiments with EM, the thermophilic phase lasted longer, the decomposition rate of organic carbon increased, C/N reduced rapidly and the compost reached maximum temperature faster compared to controls without EM. The application of EM alone without an in-vessel bioreactor and suggested stimulations do not significantly affect the bioaugmentation of the process. Temperature stimulation above 50 °C not only had no significant effect but also increased energy consumption and moisture evaporation. The results indicate that compost inoculated with EM has rapid mineralization rates in the first 2 weeks. Proper C/N, negligible *E.coli* and *Salmonella* of the humus besides low-level content of heavy metals and good germination index (> 100%) after two weeks showed that maturation period decreases about four weeks compared to run without EM and thermal stimulation. This is an environmental alternative for on-site treatment of FW that will divert a significant portion of the waste stream from landfills, reduce collection costs and greenhouse gas emissions subsequently.

Of course, accurate discussion about the energy consumption and cost-effectiveness of the studied method compared to other alternatives, bedsides investigation of feedstock entry continuously instead of batch mode can be suggested as topics for future studies.

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Declarations

Conflict of interest The authors declare that they have no conflict of interest.

Ethics approval and consent to participate This article does not contain any studies with human participants or animals performed by any of the authors.

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