



Effective management and composting of organic wastes using new developed consortia

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Abstract

The process of composting is a feasible biological treatment for the recycling of organic wastes as an amendment or fertilizer. The objective of this study was to qualify an optimum consortium during 120 days composting for different beds (manure, sawdust, leaves and straw) in order to understand the process of composting. Different parameters such as total organic carbon, total nitrogen and carbon-to-nitrogen ratio were measured at the first, middle and end of composting period. The pH widely fluctuated (6.59–9.38) in rotten cow manure (RCM) consortia; but, the same trend has been observed in other consortia during the experiment. There were significant differences among the treatments with increase in organic matter loss (OM loss) and decrease of total organic carbon and dissolved organic carbon (DOC) over time. During composting process, total nitrogen in RCM and *Pantoea Agglomerans* bacteria (PAB) consortia increased to 2.95% and 3.02%, respectively, which led to significant decrease in carbon-to-nitrogen ratio for some treatments at the end of experiment. Manure treated-consortia recorded numerically higher solution forms of potassium, nitrogen and phosphorus and carbon-to-nitrogen ratio normally ranged between standard limit of other studied beds. Manure-composted RCM that reached the standard values to efficiency improves soil and plant properties than other combinations. The new consortia will be able to reduce the required time for decomposition by supplying the nutrient enriched compost products.

Keywords Total carbon balance · Composting process · Consortium · Substrate

1 Introduction

Human population growth and increase of municipal solid, agricultural and food wastes contributed to the emission of greenhouse gases (Elbasiouny et al., 2020). Organic wastes management practices are currently the biggest challenge for improving environmental quality (Adeniran et al., 2017). Most agricultural lands in Iran are located in arid and

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semiarid regions with poorly organic matter (OM) content due to the low rainfall, high temperature and lack of vegetation (Eskandarie, 2012). About 120 million tons of agricultural yields are annually produced in Iran and lead to concerns about poor disposed waste in middle-income countries that are entailed significant health problems. Iran has high potential to produce and consume food, but the proper distribution of agricultural production has not been considered; hence, large amounts of waste are produced (NEXUS, 2019). So, it is necessary to prepare an appropriate condition for organic wastes disposal and improve the carbon sequestration in semiarid soils of Iran.

Organic wastes can be converted to valuable compost in a short period of time by microorganisms under controlled conditions (Wang et al., 2011). Composting is a recycling method, and the final product can be used in agriculture as fertilizer. Because of the increase in fertilizer costs, commodity price fluctuations, decrease in crop productivity and soil fertility, the farmers are persuaded to the organic waste application as a nutrient source. High variation of waste composition, long retention/residence time, temperature sensitive and hygiene concern and odor control are the most challenges in organic waste composting process (Abdullah et al., 2013; Wilson, 2015). The quality and stability of compost are entirely dependent on the physical and chemical characteristics of the raw organic wastes (Benito et al., 2003; Ranalli et al., 2001; Wang et al., 2004). One of the potential sources of organic materials returned to the soils is the agricultural residue wastes during the cropping season and post-harvest (Singh et al., 2019). The active component to facilitate decomposition and involve in biodegradation and conversion processes is a complex component of resident microbial community which plays an important role during composting (Neher et al., 2013). Therefore, optimization of compost quality is directly associated with the microbial community composition and bacterial community succession in the composting process (Taiwo & Oso, 2004).

Many microorganisms are able to drop the carbon-to-nitrogen ratio for improving soil productivity by convert organic wastes to valuable resources for plant nutrients (Novinscak et al., 2008; Umsakul et al., 2010). Although the microbial community that naturally occurs in the wastes usually carries out the process satisfactorily, the inoculation of microorganisms in organic wastes is a process that each microorganism produces by one of polymer degrading extra-cellular enzymes at high level. This is an appropriate way to potentially promote the process and reach the final product (Game et al., 2017).

2 Literature reviews

The presence of microorganisms with capability of lignocelluloses decomposition, thermophiles and nutrient suppliers is important in process of organic wastes microbial decomposition (Chander et al., 2018). Several studies have been conducted on composting process of different organic wastes (Awasthi et al., 2016; Chander et al., 2018; Game et al., 2017; Leow et al., 2018; Zhao et al., 2017). For example, Zhao et al. (2017) studied the effect of a microbial agent on composting of kitchen wastes. In another study, the application of microbial consortium and vermin-composting are also demonstrated to be an effective technology for efficiency disposal of farm wastes and food production (Chander et al., 2018). Awasthi et al. (2016) investigated the effect of various wastes such as wood shaving, agricultural and yard trimming waste combined with an organic fraction of municipal solid waste composting by assessing their influence on microbial enzymatic activities and quality of final compost.

Compared to chemical and thermal methods, the use of an enzyme such as cellulase to treat the organic waste is desirable, but the industrial enzyme is costly if applied at large scale. But the application of cost-effective carbonic amendment that contains microorganisms such as microbial consortia to expedite the composting process is preferable. Lack of information about standardized practices, processes and products with regard to organic composts is one major drawback in precise use of organic wastes (Brinton, 2000; Sayara et al., 2020; Wu et al., 2017). However, due to the diversity of consortia and bacteria, it is particularly important to tailor formulations of microbial consortia and agricultural wastes to its particular use (Sarkar et al., 2011; Odoh et al., 2020; Zhou et al., 2021). Therefore, there is a need to investigate the best combinations of bacteria on the efficacy composting in recycling organic wastes while understanding microbial dynamics of decomposition process, compost quality and evaluation in field application. The objectives of this research were to: (1) the study of organic wastes decomposition with new microbial consortia to identify a suitable composting method; (2) the compare the effects of different consortia on optimum characteristics of compost; and (3) the selection of proper microorganisms to produce qualified compost.

3 Material and methods

The field study was done in part of an organic farm that had a composting unit located in the Zanjan Province, central of Iran. The climate of the region is arid and semiarid. Experimental processes are presented in a graphic in Fig. 1.

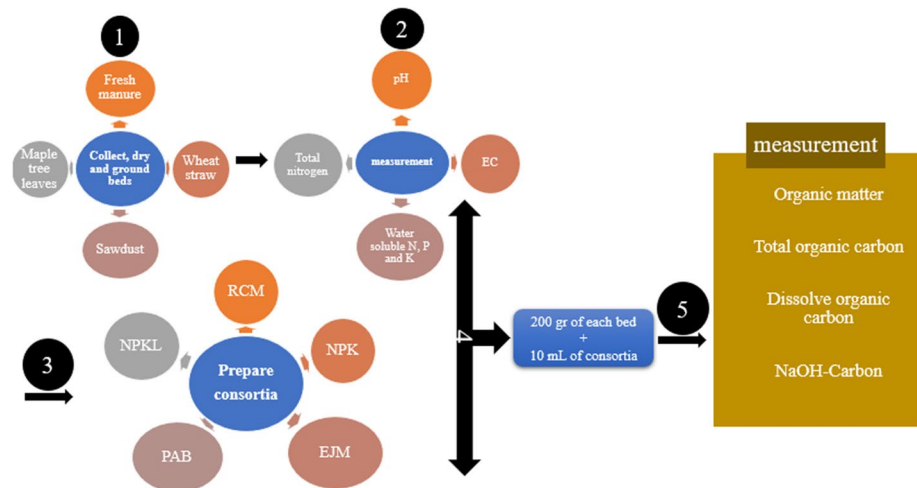


Fig. 1 A schematic graph to describe the experimental framework of this study. Rotten cow manure (RCM); effective Japanese microorganisms (EJM); nitrogen–phosphorus–potassium (NPK); nitrogen–phosphorus–potassium–lignin (NPKL); *Pantoea Agglomerans* bacteria (PAB)

3.1 Organic wastes and chemical properties

Four organic wastes were collected from considered area of Zanjan, Iran. Fresh manure was prepared from a research field of University of Zanjan. Wheat straw was obtained from an agricultural area near university of Zanjan. Sawdust was prepared from a carpentry workshop in Zanjan. Maple tree leaves were collected from campus of University of Zanjan. After acid-washing the organic matter, samples were dried at room temperature, ground to pass through a 0.5-mm sieve and stored until analysis. Meanwhile, all materials and equipment were sterilized before using in each step.

The pH and electrical conductivity (EC) were determined using pH meter (Jenway 3510) and EC meter (Jenway 4520) in a 1:10 dry sample to distilled water. Total nitrogen was measured after digestion of 0.3 g dry samples in 2.5 mL of a mixture of sulfuric acid (H_2SO_4) and salicylic acid ($\text{C}_7\text{H}_6\text{O}_3$) at room temperature overnight. Then, samples were heated in two steps at 120 and 300 °C, and 0.5 mL concentrated H_2O_2 was added several times; this process was continued until the sample became colorless (Sahrawat et al., 2002). Water-soluble potassium, nitrogen and phosphorus were determined after filtering a 1:10 dry sample to distilled water suspension, and then samples were filtered by Whatman No. 42 filter paper. The water-soluble nitrogen was measured by the mentioned method for the total nitrogen (Sahrawat et al., 2002). The water-soluble phosphorus in the extracts was measured by ascorbic acid/ammonium molybdenite method, and the water-soluble potassium was obtained using atomic absorption spectroscopy (AAS) (Varian PG instrument AA500).

3.2 Preparation of microbial consortia

Rotten cow manure (RCM) was obtained from a cow manure that was exposed to the air for one year and used as a microbial source (FAO/TECA 2010; Suhartini et al., 2020). Then, it was mixed into the Hungate culture medium (Hungate, 1947). When decomposition signs were seen, 10 mL of the initial culture medium was added into a new medium to process microbial enrichment. The residual culture medium was filtered, and the solids that remained on the filter were put in distilled water at 100 °C for 0.5 h to remove all microorganisms. Then, the solids were washed several times with distilled water and dried at 80 °C; then, weighed until the weight loss; and then used in the culture medium with constant of 10 times enrichment. The final enriched medium was used as a microbial consortium to accelerate the degradation process of organic wastes. This consortium was used in the amount of 10 mL bed^{-1} .

According to the effective microorganism's research organization (EMRO) company, effective Japanese microorganism (EJM) contains lactic acid bacteria (*Lactobacillus plantrum* and *Streptococcus lactis*), yeast (*Saccharomyces spp*), actinomycete (*Streptomyces spp.*), and photosynthetic bacteria (*Rhodospseudomonas plastris* and *Rhodobacter sphaerode*). This consortium was used in the amount of 10 mL bed^{-1} .

Nitrogen–phosphorus–potassium (NPK) is a combination of *Pseudomonas vancouverensis*, *Pseudomonas koreensis*, and *Pantoea Agglomerans* bacteria. The *Pantoea Agglomerans* bacteria are able to fix N and increase in P and K solubility. The bacteria were prepared from the Soil and Water Institute (SWRI), Iran. These bacteria were mixed in a ratio of 1–10 mL⁻¹ distilled water and used in the beds.

Nitrogen–phosphorus–potassium–lignin (NPKL) is the same as consortium NPK that a *Pseudomonas putida* bacterium has been added to it. Application of *Pseudomonas putida* was due to their ability in lignin decomposition (Bugg et al., 2011). The bacteria were also obtained from SWRI, Iran. The 0.37 g of each inoculum was weighted, mixed with distilled water and added to the beds.

The total population of microorganisms in four studies consortia was 10^8 cells mL^{-1} .

3.3 Experiment performance

The 200 g of each bed was transferred to polyethylene containers. Then, 10 mL of the considered inoculum with distilled water was added to the samples and incubated at 27–30 °C. The whole samples were completely wetted and mixed with the inoculum. The containers were covered with valve while air exchanged with the air. The samples were stirred to ensure uniform ventilation in all parts every 10 days. When the moisture of beds decreased, the required amount of distilled water was added to the samples. Sampling of each treatment was performed at 0, 60, and 120 days after the beginning of experiment and then dried at 60 °C and stored until analysis. Overall, 6 treatments in three replicates and 4 organic wastes (72 containers) were used: (1) control; (2) application of consortium RCM; (3) application of consortium EJM; (4) application of consortium NPK; (5) application of consortium NPKL; and (6) application of *Pantoea Agglomerans* bacteria (PAB).

3.4 Measurement of organic matter different forms

The 5 g of samples was ground to pass through a 2-mm sieve and was dried at 80 °C for 4 h. Subsequently, they were weighted (W_1) and were heated in a muffle furnace at 500 °C for 5 h. Then, they were weighed again (W_2) (Nelson and Sommers 1983). The following equation was used to calculate OM percentage:

$$OM(\%) = \frac{W_1 - W_2}{W_1} \times 100$$

The percentage of OM loss according to the amount of ash in samples was calculated of following equation (Viel et al., 1987):

$$OM_{loss}(\%) = 100 - 100 \times \frac{X_1(100 - X_n)}{X_n(100 - X_1)}$$

where the X_1 and X_n are the ash amounts of 0 and n days, respectively.

Organic carbon in the samples was determined by the Walkley–Black method (Walkley & Black, 1934). The 0.2 g samples was ground to pass through a 0.5-mm sieve. Then, it was transferred into 250 Erlenmeyer flask and 20 mL potassium dichromate and 40 mL concentrated H_2SO_4 to Erlenmeyer flask. Finally, the samples were titrated with ferroammonium sulfate until the solution color turned to green and then changed to red. Percentage of organic carbon was calculated using the following equation:

$$OC(\%) = \frac{T_B - T_S}{S} \times M \times 0.39$$

where the T_B and T_S are the ferro-ammonium sulfate volumes for control and sample, respectively. Weight of sample is S and M is the normality of ferro-ammonium sulfate.

A 1:10 ratio suspension of dry sample to distilled water was prepared to measure of dissolved organic carbon (DOC). Then, sample was shaken on a rotary shaker for 30 min at 120 rpm. The suspensions were filtered using a filter paper (Whatman, 42). Then, DOC in the extract was measured by wet oxidation method with potassium dichromate and sulfuric acid (Nelson and Sommers, 1983). A 1:10 ratio suspension of dry sample to NaOH (0.5 M) was used to measurement of dissolved carbon. The 5 g sample was added to 50 mL NaOH 0.5 M and mixed on a rotary shaker 30 min at 120 rpm. Then, suspension was filtered and carbon content was determined with the method mentioned above.

3.5 Statistical analysis

Experimental design included the five consortia effect on the decomposition process of four organic waste beds in three times (0, 60 and 120 days). An experiment is completely randomized design in the form of a three-factor experiments with three replications. Statistical analysis was conducted using SAS 9.4 for Windows. The ANOVA model and Duncan's Studentized range test was used to distinguish significant differences among treatments.

4 Results

Chemical and biological characteristics of new consortia are presented in different figures and tables which are indicators for optimizing them in soil fertility and productivity.

4.1 Initial properties of organic wastes

Some chemical properties of organic wastes are summarized in Table 1. Initial pH ranged from 6.42 for straw to 8.05 for manure, but the differences were not statistically significant. There were much significant differences in EC and K of manure compared to the other organic wastes. The organic carbon of sawdust and straw was significantly higher than that of manure and leaves (Table 1).

Table 1 Some properties of organic wastes in this study

Parameter	Manure	Sawdust	Leaves	Straw
pH	8.05 ^a ± 0.51	7.15 ^a ± 0.31	7.72 ^a ± 0.13	6.42 ^a ± 1.33
EC	14.50 ^a ± 3.13	0.98 ^b ± 0.21	3.97 ^b ± 0.68	4.31 ^b ± 0.75
OC (%)	45.80 ^b ± 2.12	55.79 ^a ± 2.14	45.24 ^b ± 2.79	52.66 ^a ± 1.78
TN (%)	1.25 ^a ± 0.20	0.65 ^b ± 0.07	0.72 ^b ± 0.23	0.88 ^{ab} ± 0.20
K (mg kg ⁻¹)	144.60 ^a ± 3.82	3.78 ^b ± 0.41	4.23 ^b ± 0.59	1.42 ^b ± 0.16
C/N	36.60 ^c ± 2.12	85.80 ^a ± 2.12	62.70 ^b ± 3.25	59.80 ^b ± 3.54

EC electrical conductivity; OC organic carbon; C/N carbon/ nitrogen ratio; TN total nitrogen

Values followed by same letter in raw do not differ at the level of 5%

Table 2 The EC and pH of four organic wastes treated by different consortia at three times. Rotten cow manure (RCM); effective Japanese microorganisms (EJM); nitrogen–phosphorus–potassium (NPK); nitrogen–phosphorus–potassium–lignin (NPKL); *Pantoea Agglomerans* bacteria (PAB) and unamended control (no consortium added)

Parameter	Bed	Time	Treatment						
			Control	RCM	EJM	NPK	NPKL	PAB	
pH	Manure	0	8.05 ^{d-i}	7.80 ^{f-o}	7.93 ^{d-k}	8.20 ^{c-h}	7.88 ^{e-m}	8.69 ^{bc}	
		60	7.61 ^{j-r}	7.83 ^{f-n}	7.39 ^{k-u}	7.39 ^{k-u}	7.45 ^{j-t}	7.57 ^{h-r}	
		120	7.56 ^{j-r}	7.67 ^{j-r}	7.37 ^{k-u}	7.24 ^{o-v}	7.40 ^{k-t}	7.51 ^{i-s}	
	Sawdust	0	7.15 ^{p-x}	7.64 ^{i-r}	7.39 ^{k-u}	7.46 ^{i-t}	7.28 ^{l-w}	7.21 ^{n-x}	
		60	7.36 ^{k-u}	7.88 ^{d-k}	6.89 ^{s-y}	7.10 ^{p-x}	7.03 ^{r-x}	7.32 ^{k-v}	
		120	7.43 ^{j-t}	8.34 ^{c-f}	7.52 ^{i-r}	7.26 ^{l-w}	7.21 ^{n-x}	7.24 ^{m-w}	
	Leaves	0	7.72 ^{j-p}	7.43 ^{j-t}	7.89 ^{f-l}	8.10 ^{e-i}	8.23 ^{c-g}	7.69 ^{j-q}	
		60	7.46 ^{j-t}	9.21 ^{ab}	7.74 ^{f-p}	7.66 ^{j-q}	7.80 ^{f-o}	7.69 ^{j-q}	
		120	7.05 ^{r-x}	9.38 ^a	6.88 ^{s-y}	6.69 ^{v-y}	7.61 ^{h-r}	6.88 ^{s-y}	
	Straw	0	6.42 ^y	6.59 ^{xy}	7.12 ^{p-x}	6.84 ^{t-y}	6.66 ^{wxy}	7.28 ^{l-w}	
		60	8.34 ^{c-f}	9.20 ^{ab}	8.52 ^{cd}	8.49 ^{cde}	8.07 ^{d-j}	8.49 ^{cde}	
		120	6.78 ^{u-y}	9.38 ^a	7.16 ^{p-x}	7.14 ^{p-x}	6.85 ^{t-y}	6.89 ^{s-y}	
	EC (dS m ⁻¹)	Manure	0	14.5 ^{b-e}	12.1 ^{gh}	11.5 ^h	13.2 ^{d-g}	12.6 ^{fgh}	11.5 ^h
			60	16.0 ^b	16.5 ^a	15.8 ^{abc}	14.1 ^{def}	14.9 ^{a-d}	14.3 ^{c-f}
			120	13.4 ^{d-g}	14.5 ^{b-e}	13.7 ^{d-g}	12.9 ^{e-h}	13.6 ^{d-g}	13.1 ^{e-h}
Sawdust		0	0.98 ^o	1.1 ^o	0.95 ^o	0.86 ^o	0.93 ^o	0.88 ^o	
		60	0.88 ^o	1.08 ^o	0.83 ^o	0.77 ^o	0.79 ^o	0.88 ^o	
		120	0.91 ^o	0.83 ^o	0.78 ^o	0.96 ^o	0.76 ^o	0.89 ^o	
Leaves		0	3.97 ^{j-n}	3.20 ^{lmn}	4.10 ^{j-n}	4.6 ^{i-l}	3.55 ^{j-n}	3.41 ^{k-n}	
		60	3.48 ^{j-n}	3.91 ^{j-n}	4.11 ^{j-n}	3.72 ^{j-n}	3.62 ^{j-n}	3.71 ^{j-n}	
		120	2.45 ^{no}	3.48 ^{j-n}	3.74 ^{j-n}	2.89 ^{lmn}	2.73 ^{mn}	3.05 ^{lmn}	
Straw		0	4.31 ^{i-m}	3.66 ^{j-n}	3.81 ^{j-n}	3.69 ^{j-n}	3.73 ^{j-n}	3.98 ^{j-n}	
		60	5.3 ^{ij}	5.9 ^{li}	5.8 ⁱ	5.16 ^{ijk}	4.31 ^{i-m}	3.63 ^{j-n}	
		120	3.54 ^{j-n}	4.64 ^{i-l}	4.5 ^{i-m}	3.59 ^{j-n}	3.79 ^{j-n}	3.52 ^{j-n}	

Values followed by same letter in column for each parameter do not differ at the level of 5%

The pH of four treated beds generally decreased over time except in sawdust and straw beds (Table 2). It ranged from 6.42 in control treatment of straw at the beginning of study to 9.38 for RCM treatment of straw and leaves at the end of experiment. The highest pH of cow manure belonged to the PAB treatment (8.69) at the beginning, and there were significant differences between control treatments of all consortia compared to the 120 days. In some treatments, the pH of beds had significant differences at the beginning and the end of experiment. An increase was observed in pH value of sawdust and straw at the 60 days; then, it has decreased at the end of experiment. The effects of different consortia on the EC changes in the treatment of four beds over time are presented in Table 2. In the cow manure, EC was significantly 1 to 10 times higher than other beds. The highest initial EC belonged to the cow manure for RCM (16.5 dS m⁻¹) and the lowest EC observed in sawdust for NPKL at the end of experiment (0.76 dS m⁻¹).

4.2 Total organic carbon and OM losses

The effect of different consortia on the total organic carbon for four studied beds during composting is shown in Fig. 2. The highest total organic carbon was observed in sawdust in the beginning of experiment (56.8%) and the lowest total organic carbon belonged to dried leaves (29.8%). The total organic carbon (%) decreased over time, and the most changes with time were significantly observed in the leaves, while the lowest changes of total organic carbon were observed in sawdust for all consortia and no significant differences were found except for NPKL treatment after 120 days. In the most treatments, the total organic carbon for straw at the end of study was significantly higher than the beginning and middle times of experiment. The RCM treatment in four beds has significantly affected more successful organic carbon decomposing than other consortia, and then, EJM consortium has been relatively succeed for this purpose.

The OM losses (%) in different treatments were increased at the first, middle and end of experiment (Fig. 3). This could be because the organic wastes contained easily degradable compounds and increasing humidity with time which resulted in the high OM loss during the composting process (Kadir et al., 2016). The most OM loss (%) was observed in straw bed (72.0% for RCM), and then dried leaves bed had the highest OM loss (%) in RCM treatment (58.7%) at 120 days. Also, it has increased over time in the manure bed. The minimum OM loss (%) was observed in sawdust bed at the beginning of experiment for NPKL treatment (9.3%) and did not significantly change during the experiment. The highest OM loss (%) was generally observed in RCM treatment for all organic wastes as bed on studied times.

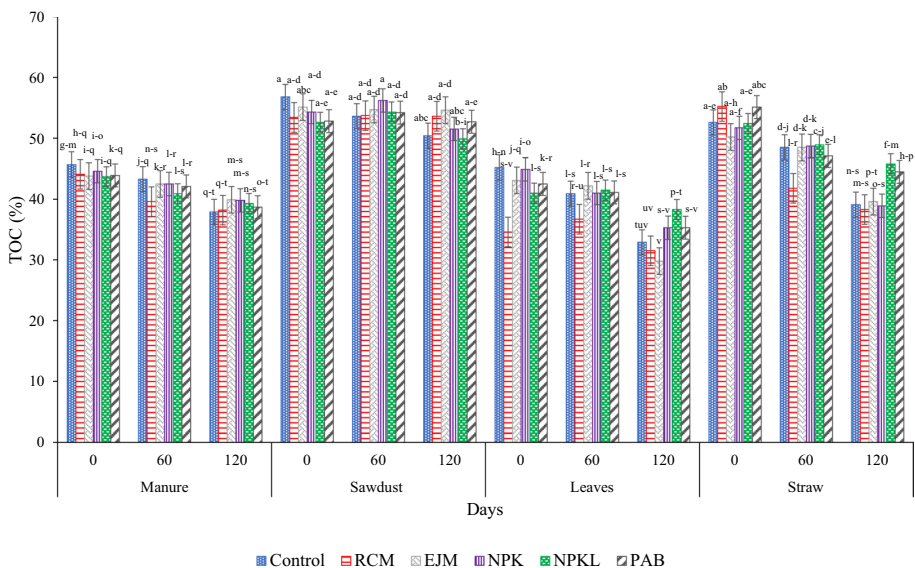


Fig. 2 TOC (%) of four organic wastes treated by different consortia at three times. TOC: total organic carbon; rotten cow manure (RCM); effective Japanese microorganisms (EJM); nitrogen–phosphorus–potassium (NPK); nitrogen–phosphorus–potassium–lignin (NPKL); *Pantoea Agglomerans* bacteria (PAB) and unamended control (no consortium added). Values followed by same letter in treatment do not differ at the level of 5%. Error bars represent standard errors ($n=3$)

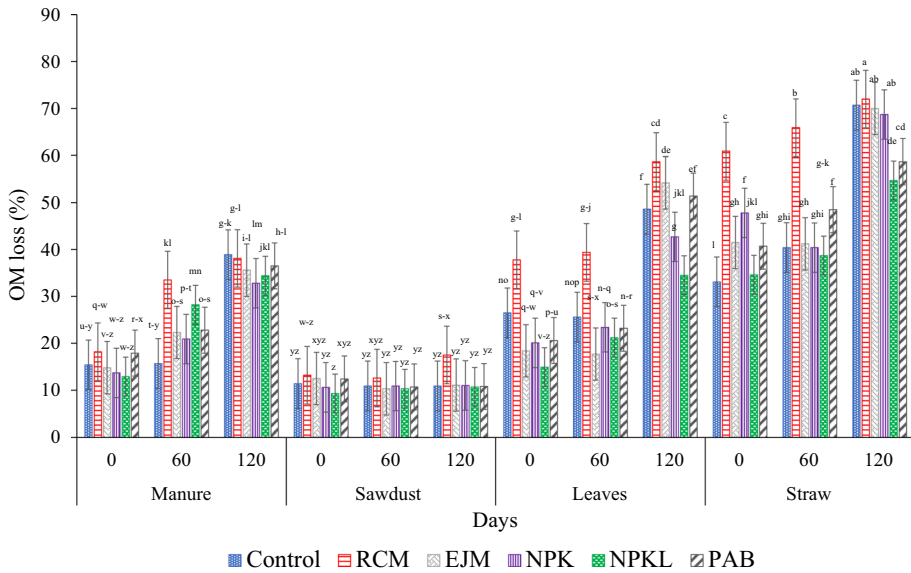


Fig. 3 OM loss (%) of four organic wastes treated by different consortia at three times. OM: organic matter. Rotten cow manure (RCM); effective Japanese microorganisms (EJM); nitrogen–phosphorus–potassium (NPK); nitrogen–phosphorus–potassium–lignin (NPKL); *Pantoea Agglomerans* bacteria (PAB) and unamended control (no consortium added). Values followed by same letter in treatment do not differ at the level of 5%. Error bars represent standard errors ($n=3$)

Significant differences were obtained between the OM loss (%) of straw, leaves and manure at the end of experiment than first and middle times. In other treatments, there were significant differences in the OM loss (%) for EJM, NPK and PAB consortia than control and other treatments for manure, leaves and straw at three times. Overall, the effect of these consortia on the decomposition of OM depended on the bed material type and time.

4.3 Soluble carbon

The content of DOC (g kg^{-1}) in four beds during the experiment is shown in Fig. 4. The difference between DOC in the present study may be due to the nature of the organic wastes, e.g., sawdust and straw which have low initial DOC compared to the other beds. The DOC decreased in all beds with time, but this reaction was slower for sawdust. The reason for reduced DOC is related to the mesophilic and thermophilic reactions and the quickly consumption of small and soluble molecules during the composting process that make up the dissolved fraction (Zmora-Nahum et al., 2005). The initial concentration of DOC depended on the bed types, and the highest initial DOC was observed in cow manure for NPKL treatment (7.60 g kg^{-1}). The lowest initial DOC (1.02 g kg^{-1}) was obtained in sawdust for RCM treatment. The highest DOC was observed in the manure (2.52 g kg^{-1} for RCM), and the lowest DOC was measured in the sawdust (0.75 g kg^{-1} for control) at the end of experiment. There were significant decreases in DOC over time in all treatments of manure and the most treatment of leaves and straw. Also, decreased DOC indicates effective biological activity in the four studied beds.

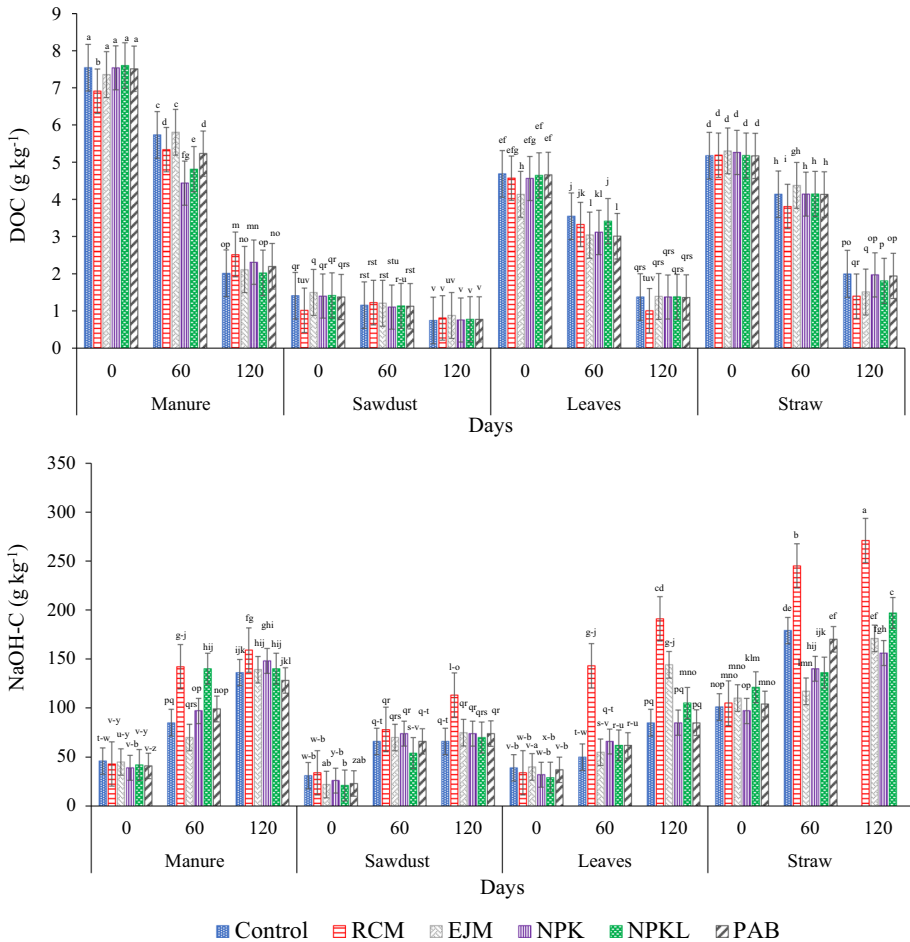


Fig. 4 Soluble carbon of four organic wastes treated by different consortia at three times. DOC: dissolved organic carbon and NaOH-C: NaOH-carbon. Rotten cow manure (RCM); effective Japanese microorganisms (EJM); nitrogen–phosphorus–potassium (NPK); nitrogen–phosphorus–potassium–lignin (NPKL); *Pantoea Agglomerans* bacteria (PAB) and unamended control (no consortium added). Values followed by same letter in treatment do not differ at the level of 5%. Error bars represent standard errors ($n=3$)

The concentration of NaOH-C in the four beds during experiment is shown in Fig. 3, and their amounts were much higher than the DOC in treatments. At the beginning of the test, humic compounds were gradually degraded in manure, leaves straw and sawdust beds and had a large amount of soluble carbon over time. The NaOH-C is actually a collection of DOC, fulvic acid, and humic acid and increased over time. The highest initial concentration of NaOH-C was observed in straw (121 g kg⁻¹), and the lowest NaOH-C was measured in sawdust bed (31 g kg⁻¹) in the NPKL.

The highest NaOH-C (159 g kg⁻¹) belonged to the RCM treatment for manure, which was significantly different from other treatments at the 120 days (except NPK treatment). In sawdust bed at the end of the experiment, there was significant difference between treatments than the beginning of experiment. In dried leaves, the highest NaOH-C was demonstrated in RCM consortium which was significantly different from other treatments during

the experiment. The highest NaOH-C between all treatments was observed in RCM consortium for straw bed that had a greater impact on the changes of OM or humic matter in straw and dried leaves beds than the other consortia.

4.4 Total nitrogen and carbon-to-nitrogen ratio

The percentage of total nitrogen includes ammonium N, organic N and any forms of nitrate in all treatments increased and followed upward trend during the experiment (online Appendix A; Fig A1). The minimum of total nitrogen was observed in sawdust bed at the beginning of experiment (from 1.19% for PAB to 1.65% for control). For manure, leaves and sawdust beds, the same trend was observed in total nitrogen and there were significant differences between some treatments at the 60 and 120 days with the beginning of experiment.

The effects of different consortia on the carbon-to-nitrogen ratio in four beds are shown in Fig. 5. The carbon-to-nitrogen ratios for the manure were low, and the range was narrow due to the lack of C-rich bedding materials and the high nitrogen content of the manure. There were significant decreases of carbon-to-nitrogen ratio in the most treatments at the beginning of experiment compared to the end of study in four studied beds, due to the reduction of total organic carbon and loss of labile carbon as CO₂ during compost process. The highest carbon-to-nitrogen ratio was observed in sawdust (44.4 for PAB treatment at the beginning of experiment) and the lowest carbon-to-nitrogen ratio obtained in dried leaves (10.4 for 120 days in EJM consortia). The carbon-to-nitrogen ratio in OM decomposition is one of the important characteristics to determine the decomposition process. In this study, the carbon-to-nitrogen ratio has decreased over time and the highest reduction

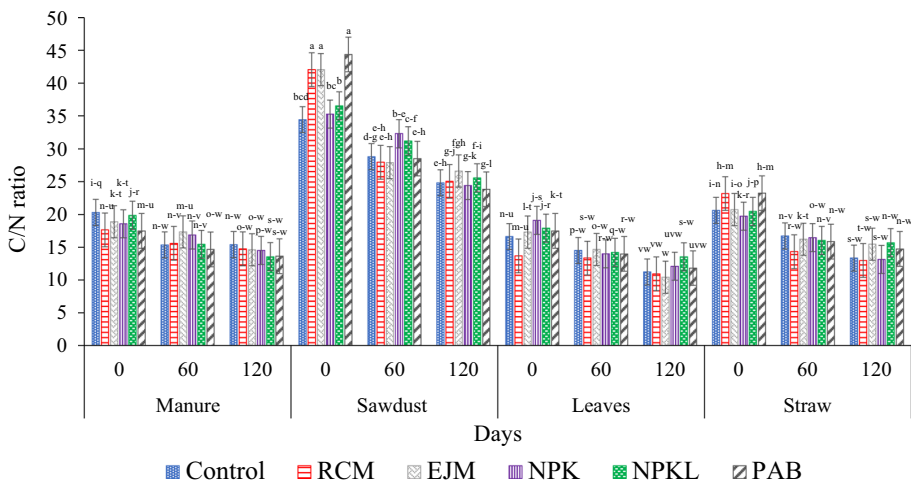


Fig. 5 The C/N ratio of four organic wastes treated by different consortia at three times. C/N ratio: carbon-to-nitrogen ratio. Rotten cow manure (RCM); effective Japanese microorganisms (EJM); nitrogen–phosphorus–potassium (NPK); nitrogen–phosphorus–potassium–lignin (NPKL); *Pantoea Agglomerans* bacteria (PAB) and unamended control (no consortium added). Values followed by same letter in treatment do not differ at the level of 5%. Error bars represent standard errors ($n = 3$)

was observed in sawdust and in PAB treatment with 20.5 units. In this context, the lowest decrease of carbon-to-nitrogen ratio was seen leaves in RCM treatment with 2.7 units.

4.5 Water-soluble potassium, nitrogen and phosphorus

The concentrations of water-soluble potassium in the treated-four beds are shown in Table A1 (online Appendix A). The highest concentration of initial water-soluble potassium was observed in cow manure bed after application of EJM (158.3 mg L^{-1}), and the lowest concentration was observed in straw for control treatment (1.4 mg L^{-1}). The water-soluble potassium has increased over time in four beds and all treatments. There were significant differences between manure and the other beds. In all treatments of cow manure, especially in RCM, significant higher concentration of water-soluble potassium was observed than other consortia at the end of experiment. In other beds, there were no significant differences between treatments.

The concentration of water-soluble nitrogen extracted from four treated beds with different consortia is shown in Table A1 (online Appendix A). In manure, the higher water-soluble nitrogen was significantly observed than other beds and the water-soluble nitrogen maximum was found in RCM (65.1 mg L^{-1}) treatment. This reflected that the manure had stronger capability of nitrification between all beds. After RCM treatment, it seems that the high level of water-soluble nitrogen in the treatment of PAB in dried leaf litter and straw is related to the decomposition of nitrogen compounds and release soluble nitrogen into the solution.

The concentration of water-soluble phosphorus in four beds treated by different consortia also is shown in Table A1 (online Appendix A). The highest water-soluble phosphorus at the end of experiment was observed in the cow manure (53.1 mg L^{-1} for RCM) which was significantly different from other treatments, and the lowest water-soluble phosphorus was measured in the straw bed (3.0 mg L^{-1} for NPK). The water-soluble phosphorus has significantly increased in cow manure than the other beds. But in the other beds, measured water-soluble phosphorus did not differ significantly during the experiment. In dried leaves, the water-soluble phosphorus was increased over time although its slope was not significantly more than sawdust and straw.

5 Discussion

Yang and Zhu (2015) indicated that the percentage of organic carbon (44.4 to 47.8%) and total nitrogen (1.5–2.1%) in some dried leaves was similar to our studied leaves. It has been observed that the cow manure had lower organic carbon (30.8%) and total nitrogen (2.9%) and carbon-to-nitrogen ratio $> 10:7$ compared to the swine and chicken manures (Huang et al., 2017).

The optimum range of pH for organic wastes decomposition was reported between 6.5 and 8.5 (Hoitink and Fahy, 1986). Some studies have indicated that with the reduction of carbon-to-nitrogen ratio, the pH stabilized between 7 and 8 (Gade et al., 2010; Himanen and Hanninen, 2011; Sarker et al., 2013).

This increasing trend has been shown in other similar works (Dastpak et al., 2020; Meng et al., 2018; Vig et al., 2011). Yu et al. (2019) reported that pH decreased until day 4, then increased until day 12 and then decreased until the end of the composting period. Increasing pH has been affected by the formation of ammonia and other soluble alkaline elements

when the protein of organic wastes was decomposed during the composting process while organic acids and nitrification production decreased pH of beds at the end of composting (Pant et al. 2012; Martin 2012).

Huang et al. (2004) observed that EC increased at the beginning of the period and reached its maximum on 14 days and then decreased until the end of the pig manure and sawdust mixture composting. They also concluded that the increased EC at the beginning of decomposition is depending on ammonia and ortho-phosphates release. But over time, the volatilization of ammonia and deposition of ortho-phosphates might have reduced EC at the end of experiment (Huang et al., 2004). Fornes et al. (2012) also reported that the EC showed a sharp decrease at the beginning and then increased in the middle of the experiment and finally decreased during livestock manure composting.

Verma et al. (2014) have observed a reduction in total organic carbon after 175 days composting, because the OM was broken down by microorganisms and part of the C was released as CO₂ following microbial respiration during composting process (Nada, 2015). In another study, decreasing of total organic carbon was also observed during the wheat straw and manure decomposition (Yu et al., 2019). Some authors have reported the reduction of total organic carbon during composting (Gupta & Garg, 2008; Malińska et al., 2016). Nada (2015) observed that the more than 36% of the initial total organic carbon was lost after 60-day composting. They conducted that this reduction could be due to the mineralization of the OM by microorganisms. Also, Hanajima et al. (2010) reported that the volatilization of CH₄, methyl mercaptan, dimethyl sulfide and other organic compounds may cause carbon losses in composting process. Organic matter addition is an important management such as changes in total organic carbon, and it was related to water stability of aggregates and mineralization (Chan et al., 2002). Then, the increase of DOC led to release some compounds that are the source of mineralizable N and P. The availability of these nutrients is closely linked to DOC dynamics (Silveira, 2005).

Among the studied consortia, RCM consortium had a significant effect on the OM decomposition in the most beds and it indicated that this consortium will be able to properly decompose organic wastes by emission of C to CO₂ during composting (Tiquia et al., 1998). The OM loss in this study for leaves and straw at the end of experiment was higher than that losses reported by some authors for other beds (Flynn & Wood, 1996; Tiquia et al., 1998). Michel Jr. et al. (2004) demonstrated that the OM losses were from 45 to 74% for sawdust and from 54 to 79% for the straw amended windrows. Their results indicated that OM losses of straw during 120 days of composting were similar to sawdust and were lower (2–3 times) than sawdust amendments. Nada (2015) conducted that the emission rate of CO₂ during the thermophilic phase (60–95%) was higher than the curing phase. Therefore, the most OM loss occurs when thermophilic microorganisms are active during composting.

Some studies suggested a threshold level of DOC (4 g kg⁻¹) for compost maturity (Hue & Liu, 1995; Zmora-Nahum et al., 2005). The DOC reached below levels of compost maturity (3 g kg⁻¹) toward the end of experiment in the manure, leaves and straw beds. In the sawdust bed, the DOC concentrations in all treatments were lower than threshold limit for compost maturity during the experiment. Zmora-Nahum et al. (2005) reported that the initial DOC in various components of municipal and manure changes from 5.5 to 3.5 g kg⁻¹ and eventually reached less than 4 g kg⁻¹. The DOC of various beds (agricultural and municipal waste) at the composting process reduced and reached less than 10 g kg⁻¹ (Hu et al., 2011). Also, it has demonstrated that DOC of wheat straw and manure decreased during the decomposition (Yu et al., 2019). Simple and small compounds of organic wastes are consumed by microorganisms, and DOC decreased during the various

stages of decomposition (Dias et al., 2010). Microorganisms break down hard carbon compounds such as cellulose, and this process continues until the simple carbon compounds enter to the solution (David, 2013). In the present study, it seems that RCM has been more successful in this case than other treatments.

Humification is actually one of the processes that occur during the process of decomposition and composting of OM (Ribeiro et al., 2017; Vargas-Garcia et al., 2006). The concentration of humic acid increased than the total OM with promoting the decomposition process in organic wastes (Mbarek et al., 2019). So, the NaOH-C could increase significantly during 120 days. Zorpas and Loizidou (2008) reported that NaOH-C increased in the final products of sewage sludge compost. This can be due to the biodegradation and mineralization activities of the soil microflora, which intensify in response to applied OM (Schroder et al., 2008).

The changes in total nitrogen in the compost material were affected by a variety of factors. Decomposition of organic N led to ammonium N volatilization at high temperatures which result in a decrease in total nitrogen contents (Yang et al., 2019). After the thermophilic phase, total nitrogen increased due to the higher dry matter loss rate (as CO₂) than the ammonium N loss rate and water evaporation (Huang et al., 2006). When the organic compounds strongly degraded, the amount of total nitrogen increased in the compost, due to the enrichment effect (Awasthi et al., 2016; Jiang et al., 2015), which led to the reduction of total dry mass and increase in the N content (Li et al., 2012). In other study, the increase of total nitrogen content has been observed during composting process (Dastpak et al., 2020).

The microorganism's energy is obtained from the oxidation of organic carbon and nitrogen. The proper carbon-to-nitrogen ratio of organic bed for decomposing is from 20 to 30, because microorganisms need 1-part nitrogen for 30-parts carbon (Hotta & Funamizu, 2019; Palaniveloo et al., 2020). During decomposition, this ratio usually decreases because of the organic carbon decomposition and the reduction of organic carbon content in the bed. The content of total nitrogen increases with the decomposition process, ultimately leading to a decrease in the carbon-to-nitrogen ratio (Bernal et al., 2009).

During the composting process, the utilization of carbon by microorganisms was higher than nitrogen, which reduced the ratio of carbon to nitrogen. The most part of OM decomposed to the humus and some part was comparatively difficult or very difficult to decompose. So, thermophilic microorganisms are not able to growth and the rate of degradation decreased and reduction of carbon became slower and carbon-to-nitrogen ratio increased remarkably during the last stage of decomposition (Wu et al., 2010; Zeng et al., 2006). On the other hand, bed weight loss, mineralization of nitrogen and ammonia, organization of mineral nitrogen by microorganisms, nitrification and denitrification processes are the most contribution in N dissolution (Elvira et al., 1998; Liang et al., 2018; Paredes et al., 2002).

Singh et al. (2019) reported that the water-soluble phosphorus and potassium concentration increased during decomposition. Increase in water-soluble phosphorus and potassium of manure was also demonstrated over time in another study at the end of composting process which was consistent with the results of this study (Fornes et al., 2012). They concluded that the water-soluble phosphorus and potassium increased with weight decreasing of the beds and the lack of leaching or gradually increasing of phosphorus and potassium mineralization. Manure is a good source of potassium, nitrogen and phosphorus and contains readily available forms of them for plant growth.

Efficient management practices as acid and thermal pretreatments are necessary in order to sufficiently decomposition of organic wastes (Rouches et al., 2016). In the EJM consortia, lactic acids bacteria may lead to more efficient decomposition and reduced

carbon-to-nitrogen ratio; but, EJM was only highly affected in the leaves bed. The presence of phosphate-solubilizing bacteria such as *Pseudomonas putida* in the NPKL did not have significant influence on water-soluble nitrogen concentration, and water-soluble nitrogen was measured higher in the RCM than NPKL. Nitrifying bacteria are combined in NPK and NPKL consortia and PAB treatments and so increased the nitrogen solubility during composting. These results were indicated in the water-soluble nitrogen and total nitrogen results and were higher than other treatments in studied beds. Overall, between all studied consortia, RCM consortium with higher total nitrogen, water-soluble potassium, water-soluble nitrogen and nitrogen and lower carbon-to-nitrogen ratio could more precisely describe the characteristics of a proper consortium for composting process.

Some microorganisms are important to specified decomposition process of organic waste such as cellulolytic microbes, actinomycetes, and mycorrhiza (Ward et al., 2005). Manure contains a combination of these microorganisms and during its anaerobic digestion; C in the simple compound forms is converted to CO₂ and methane. Also, N will be transformed and lost to NH₄ gas and P changed to the bacterial P and some of the insoluble P as a result of microbial decomposition. However, highest responses of the evaluated parameters were obtained from cow manure between studied beds.

The effect of microorganism's type on the OM decomposition has also been studied in different studies. Gong et al. (2017) observed that the *Trametes versicolor* and *Phanerochaete chrysosporium* increased the rate of lignin, cellulose degradation and composting process. It was also reported that the addition of a consortium contained *Trametes versicolor*, *Aspergillus niger*, and *Aspergillus flavus* increased carbon and nitrogen mineralization which result in stabilization of compost (Awasthi et al., 2016). The microorganisms normally involved in compost stability and main producers are bacteria and fungi including bacteria from *Pseudomonas* and *Bacilli* genera which are efficiently able to degrade the organic wastes and eventually turn them into humus and improve several other physico-chemical properties of soils (Cesaro et al., 2019; Pant et al., 2012).

5.1 Recommendation for consortia and beds

In this study, results suggested that the consortium is a better option than the isolated single or mixed strains. The use of the microbial consortium is better for buffering of pH and temperature changes and have tendency to change better resistance for heavy metals presence, toxic organic compounds or contamination by other strains (Zhang et al., 2011). The combination of many microorganisms decomposes effectively organic wastes using cellulolytic and hemicellulolytic enzymes (Leow et al., 2018). Also, the sterilization of beds is an important parameter to the increase of composting quality and it can be used to improve the agronomic value of composts related to their nutrient availability or metal mobility.

6 Conclusion

The tendency to increase in productivity will create the need to improve the management and efficiency recycling of organic wastes. The decomposition of OM depends a lot on the nature of the organic wastes and other properties such as the initial C and N, OM composition type and carbon-to-nitrogen ratio. The pH in RCM treatment increased significantly compared to the other treatments. Highest OM loss (72.0%) and DOC during 120 days were found in straw bed, and the carbon-to-nitrogen ratio was higher than in other beds.

The general trend of total organic carbon changes was downward over time, and most of the carbon changes over time were observed in the dried leaves and EJM treatments. The most changes of total nitrogen during the experiment were observed in the manure and straw and in RCM and PAB with nitrification bacterium. The highest water-soluble potassium, water-soluble nitrogen and nitrogen were observed in manure and RCM treatments. Therefore, the RCM consortium plays an important role in the decomposition of OM in all treatments, especially in manure bed with significant differences than other consortia. Therefore, this study contributes significant new knowledge for the combination of manure bed and RCM consortium to remediate productivity and fertility in applied soils.

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