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RESEARCH PAPER

Effect of Biochar Amended Vermicomposting of Food and Beverage Industry Sludge along with Cow dung and Seed Germination Bioassay

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ABSTRACT

Transformation of food and beverage industrial sludge into vermicompost into value-added product simultaneously can control gaseous emission. Addition of biochar in the vermicomposting as a bulking agent increases fertilizer value. This research aimed to investigate the effect of biochar amendment on vermicomposting of the food and beverage industry sludge (FBIS) and cow dung (CD) in a different ratio using earthworm *Eisenia fetida*. We had further investigated the survival rate of *E. fetida* and the cocoon productions after 35 days of the vermicomposting. Besides, we have also evaluated the seed germination bioassay using Malabar spinach (*Basella alba*) to determine the toxicity and maturity of produced compost. The survival and cocoon production of *E. fetida* were higher in vermicompost amended with 10% biochar. Vermicomposting with biochar resulted in a slight pH shift. Reduction in organic carbon (OC) percentage not so significant in biochar added FBIS and CD. An increase in phosphorus and potassium content and a decrease in nitrogen percentage observed; vermicomposting with biochar resulted in higher seed germination, root elongation, and germination index than vermicomposting without biochar.

Keywords: Vermiculture, biochar, sludge utilization, organic fertilizer, soil conditioner, germination.

INTRODUCTION

In recent years, food processing has emerged as the most important industrial activity in Bangladesh. As a developing country, a significant portion of urban waste is composed of food waste (68 to 81%), which produce CH_4 as they decompose anaerobically. The trends in waste generation indicate a growth rate of 0.1343 million tonnes per year (Shams et al., 2017).

Food processing operations produce significant amounts of waste and sludge rich in nutrients, and in Bangladesh, the data is unavailable. Food industrial sludge is typically disposed of in open dumps or poorly designed landfills that cause surface and groundwater contamination to public health and environmental hazards (Yadav and Garg, 2009). Vermicomposting is a sustainable option for recycling biodegradable organic waste (Lv et al., 2020). The uses of earthworm-mediated vermicomposting technology boost sustainable development by increasing the underlying economic, social, and ecological framework (Singh et al., 2020). Vermicomposting represents a widely used and effective method for converting

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organic waste and sludge into a relatively stable humus-like product used as a soil amendment or organic fertilizer (Bhat et al., 2018). Numerous pollutants present in the sludge materials may restrict earthworms' behavior and the vermicomposting capacity of food and beverage industrial sludge. Hence, sludge vermicomposting involves the introduction of selected materials to provide optimal earthworm conditions.

Biochar is a carbonaceous organic matter applied in agriculture and suggested as a beneficial additive and bulking agent in composting. It has gained significant attention in environmental and agricultural management due to its unique properties such as chemical recalcitrance, high porosity, large surface area, high cation exchange capacity (CEC), and sorption capacity (Wu et al., 2017). Numerous studies have evaluated the impact of biochar on organic waste composting such as improving aeration conditions (Steiner et al., 2010), reducing odours, GHG emissions (Chowdhury et al., 2014; Malińska et al., 2014; Sánchez-García et al., 2015; Steiner et al., 2010), nutrient leaching (Wu et al., 2017),), accelerating the decomposition and humification of organic matter (Dias et al., 2010; Sánchez-García et al., 2015; Steiner et al., 2010), reducing N losses (Chen et al., 2010; Dias et al., 2010) by transforming ammonium into nitrite, improving the quality of end compost (Jindo et al., 2012; Zhang et al., 2014) and reducing the bioavailability of heavy metals (Awasthi et al., 2016). The combined application of biochar and vermicompost mixture has synergistic effects on soil fertility, microorganisms, soil organic matter, water holding capacity, agricultural productivity, and further plant growth (Doan et al. 2015; Huang et al. 2020).

Concerns about environmental quality, biochar vermicomposting can be a fair process of waste and sludge bioconversion into valuable biofertilizers. The objectives of the research were to investigate the effect of biochar addition in the vermicomposting of cow dung and sludge from the Food and Beverage industry, particularly of the dairy industry and compare the physicochemical characteristics of the composts, and to assess the maturity and toxicity by seed germination bioassay of the produced composts.

MATERIALS AND METHODS

We have collected semi-dried food and beverage industry sludge (FBIS) from the effluent treatment plant of Pran Dairy LTD. Several days old urine-free cow dung was collected from a local dairy farm. Biochar was collected from Bangladesh Biochar Initiative (BBI), made from different organic residues of wood chips, sawdust, bamboo, household solid wastes, agricultural residues, and biogas plant slurry. The biochar is made locally by the women in a cooking stove that is a customized format of "Top-lit updraft (TLUD) gasifiers". The stove automatically makes char in the process of cooking. The carbonization temperature varies between 450– 750 °C in the absence of oxygen (pyrolysis) or with restricted oxygen (gasification) (Bangladesh Biochar Initiative, 2020). According to the International Biochar Initiative (IBI) Biochar Standards Version 2.0 (2014) minimum requirement of organic carbon (OC) should be 10% with the Class 1 biochar containing ≥60%, Class 2 biochar ≥30% and <60% and Class 3 biochar ≥10% and <30%, respectively (International Biochar Initiative, 2014). *E. fetida* earthworm was collected from T.R. Agrocompany LTD.

A 32 cm diameter and a volume of 10 liters buckets were used as media vessels for vermicomposting and kept in the experimental shade of the Department of Environmental Sciences, Jahangirnagar University. In the trial experiments we had used 100% FBIS and 50 earthworms in which the earthworms did not survive. Therefore, to fix such issue on the basis of an increasing percentage of FBIS, three feed mixtures of FBIS and CD in the ratios of 1:3 (25% FBIS and 75% CD), 1:2 (50% FBIS and 50% CD), 3:1 (75% FBIS and 25% CD) were taken to establish 1 kg weight in the first set of buckets (T1, T2, T3). The same mixture ratio was used in the 2nd set of buckets (T1B, T2B, T3B) with 10% biochar (total weight of 1.1 kg). Fifty *E. fetida* was introduced in each mixture, and weekly, we have sprayed water to maintain 60-80% moisture and turned over the media. The environmental temperature was at 30±2 °C.

We have applied a commercial vermicomposting period of 35 days in this experiment, which started on 24 April 2019 and ended on 30 May 2019. After 35 days, we separated the cocoons, earthworms, and vermicomposts manually and recorded the total earthworms and cocoons. The vermicompost samples and all sludge, cow dung, and biochar were air-dried at room temperature, ground, and stored in a plastic zip-lock bag for analysis.

According to the American Public Health Association (APHA) method (APHA 2012) pH was measured by a Griffin pH meter (model-40), organic carbon (OC) by wet oxidation method, and total Kjeldahl nitrogen (TKN) by Kjeldahl method. Total phosphorus (TP) was determined using UV-spectrophotometer (model no: UV-1650PC, SHIMADZU) at 400 nm using a vanadate-molybdate reagent. Total potassium (TK) was determined by Flame photometer (AnA-135, O.S.K OGAWA SEIKI Co. LTD, JAPAN). Heavy metal Cr, Cd were determined by the atomic absorption spectrophotometer (AA240FS Agilent). All solid samples were digested with nitric acid (HNO_3) before TP, TK, Cr, Cd analysis.

Seed germination index (GI) bioassay, a factor of relative seed germination and relative root elongation, is a simple and commonly used ecotoxicological test for evaluating the compost's maturity. In this study, Malabar spinach (*Basella alba*) was used for the determination of germination index bioassay which grown as an annual during the heat of summer.

The seed germination bioassay was evaluated according to Tam and Tiquia (1994), using Malabar spinach (*B. alba*) using the following equation (1-3).

$$Relative\ seed\ germination(\%) = \frac{No.\ of\ germinated\ seeds\ in\ the\ extract}{No.\ of\ seeds\ germinated\ in\ control} \times 100 \quad (1)$$

$$Relative\ root\ elongation(\%) = \frac{Mean\ root\ elongated\ in\ the\ extract}{Mean\ root\ elongated\ in\ the\ control} \times 100 \quad (2)$$

$$Germination\ index(GI) = \frac{(\%\ seed\ germination) \times (\%\ \text{Root elongation})}{100\%} \quad (3)$$

There were three replicates for each feed mixture to carry out statistical analysis. Standard deviation and one-way ANOVA evaluated the significant differences among different vermicomposting treatment parameters for studied parameters. The probability levels used for statistical significance were $p<0.05$ for the tests.

RESULTS AND DISCUSSIONS

After 35 days of the vermicomposting cycle, we have observed a consistent reduction in the sludge granule size. Visually, the vermicompost amended with biochar was darker in color homogeneously granular, odorless humus-like material. Vermicomposting without biochar was brown and less homogeneous and contained loosely slacked material. However, both treatments of T3 contained some pieces of the softly loosen, slacked part of indigested sludge by the earthworms. Characteristics of the collected sludge from the food and beverage industry, cow dung, and biochar are shown in Table 1.

Table 1. Characteristics of the collected sludge from the food and beverage industry, cow dung and biochar

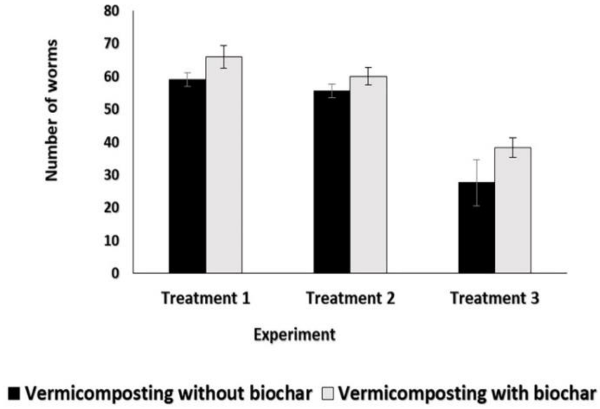
Parameters	Food and beverage industry sludge	Cow dung	Biochar	Compost standard (Waste concern of Bangladesh, 2016)
pH	7.03±0.058	8.2±0.17	7.03±0.058	6.0-8.5
OC (%)	2.7±0.2	12.87±0.15	45±1	10-25%
TKN (%)	3.5±0.2	1.07±0.12	2.043±0.029	0.5-4.0%
TP (ppm)	4815±5	5067±2	6042.33±2.52	5000-15000 ppm
TK (ppm)	137.13±2.21	443.63±1.52	970.13±0.153	1-3% (10000-30000) ppm
Cr (ppm)	2.13±0.153	0.005±0.004	0.0413±0.002	Max. 50 ppm
Cd (ppm)	0.02±0.01	0.044±0.0006	0.003±0.002	Max. 5 ppm

* All values are the mean and standard deviation of three replicates.

Vermicomposting with biochar had 5% to 12.33% higher cocoons compared to the vermicomposting without biochar. Gong et al. (2018) and Malińska et al. (2016) indicated that biochar to green waste and sewage sludge increased *E. fetida* growth, cocoons, juvenile earthworms, and reproduction. However, in this experiment, the total number of *E. fetida* and cocoons after 35 days was not significantly different ($p<0.05$). Treatment T1(B), T2(B) showed the highest number of total earthworms and cocoons production (Figure 1 (a) and (b)).

The Malabar spinach seed germination (%) and root elongation (%) in vermicompost without biochar ranged from (28.33±5.77-66.67±5.77) % and (20±5.0-73.33±2.89) %, whereas seed germination (%) and root elongation (%) ranged from (40±5.0-78.33±5.77) % and (38.33±5.77-81.67±5.77) % when vermicomposting of food and beverage industrial sludge mixed with 10% of biochar (Figure 1(c) and (d)). Treatment T1(B) and T2(B) had the highest GI 65.33±6.28 and 54.48±2.85, whereas T1 and T2 had 48.83±3.75 and 33.08±2.88, respectively (Figure 1(e)). The seed germination, root elongation, and germination index results have not shown significant differences ($p>0.05$). Germination index (GI) higher than 60 indicated the maturity of vermicompost (Zucconi and de Bertoldi, 1987), while GI values below 50 indicate the presence of phytotoxic compounds that remained un-metabolized while inhibiting germination (Easha *et al.*, 2015).

All vermicompost sample amended with biochar (6.86±0.15-7.13±0.15) were higher in pH value than those of without biochar (6.23±0.23-6.50±0.4). The pH showed significant differences, $p<0.05$ (Figure 2(a)). Literature also showed that biochar in composting leads to an increase in pH value than composting without biochar (Wu et al., 2017). This may due to biochar's cation exchange capacity. The use of biochar in composting accelerates organic matter degradation and may absorbs large amounts of NH_3 derived from the mineralization of organic matter (Dias et al. 2010; Sánchez-García et al. 2015; Zhang and Sun, 2016) and adsorption of humic substances (Hua et al., 2009). Another study showed that mature vermicompost might increase pH mainly due to the utilization of organic acids and an increase in nutrient constituents of the organic wastes (Jadia and Fulekar, 2008).



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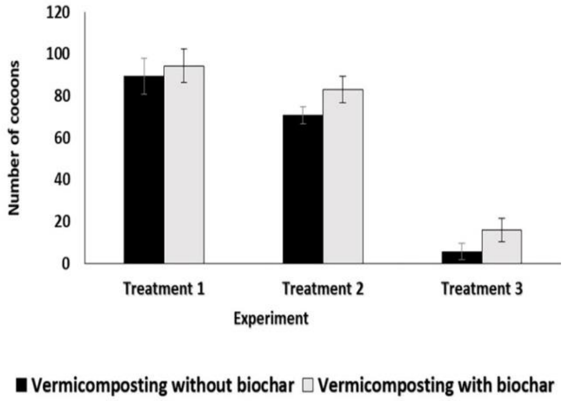

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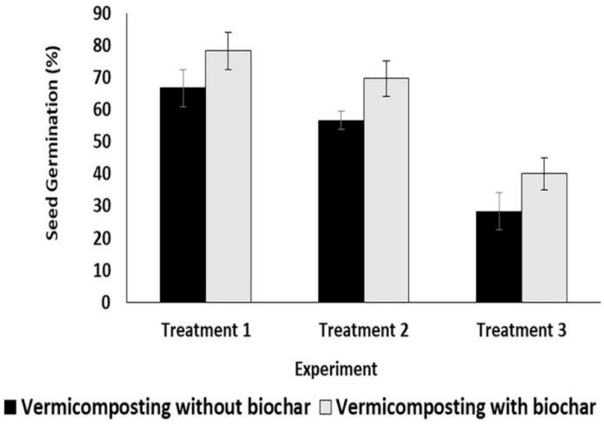

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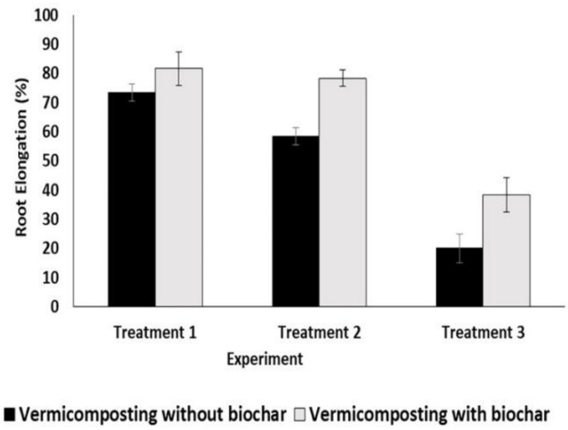

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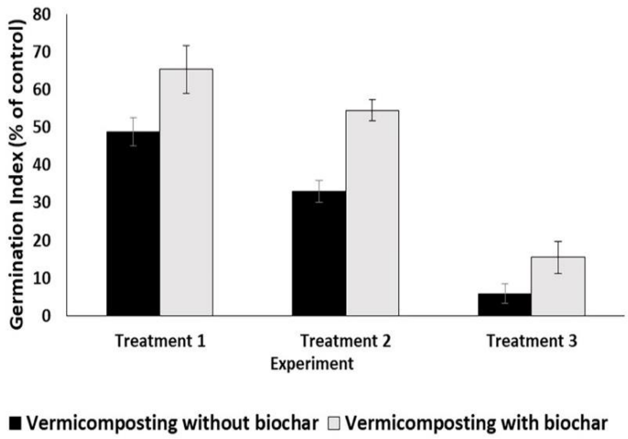


(e)

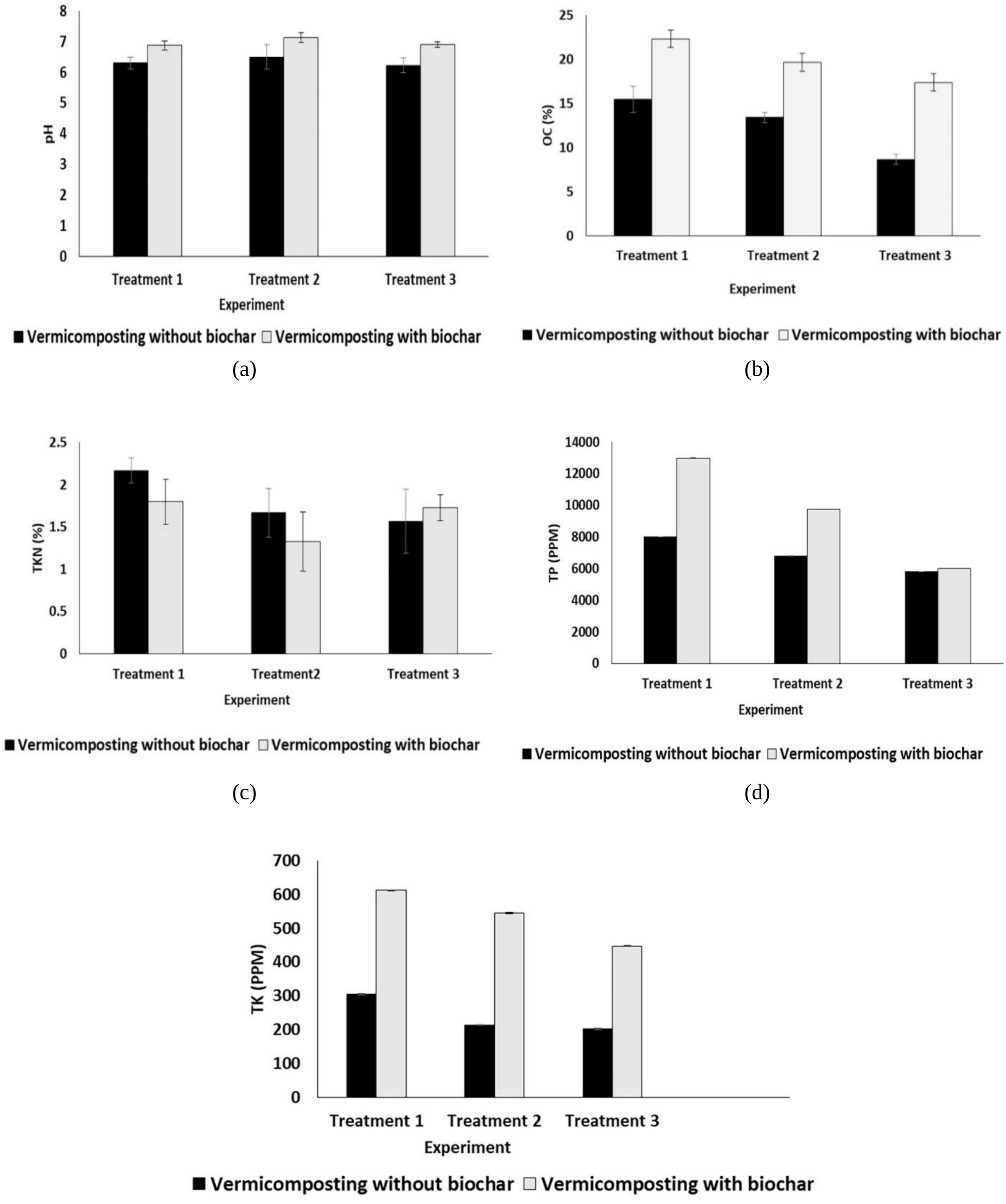
Figure 1. (a) Number of *E. fetida* population and (b) the total number of cocoons after 35 days of vermicomposting (all values are the mean of three replicates; ANOVA, $p>0.05$). (c) Seed germination (%), (d) Root elongation (%), and (e) Germination index in different treatments. (All values are the mean and standard deviation of three replicates; ANOVA, $p>0.05$). Capped bars indicate the standard deviation of the replicates.

Organic carbon (OC) percentage of the study showed that composting amended with 10% biochar (0.1 kg) had increased organic carbon from the treatments resulting in a 20.33% to 17.4% decrease in three treatments. In comparison, OC% decreased from ~15.5% to 8.67% in the treatment of vermicomposting without biochar. The OC% showed significant differences p<0.05 (Figure 2(b)). A large fraction of OC lost through microbial respiration and earthworm's activity in the form of CO_2 in vermicomposting without biochar compared to vermicomposting with biochar. The higher OC might have added from the 10% biochar. According to the International Biochar Initiative (IBI) Biochar Standards Version 2.0 (2014) minimum requirement of organic carbon (OC) should be 10% with the Class 1 biochar containing ≥60%, Class 2 biochar ≥30% and <60% and Class 3 biochar ≥10% and <30%, respectively.

TKN in vermicompost amended with biochar was nearly the same percentage as vermicompost without biochar (Figure 2(c)). The increase in vermicompost's nitrogen content depends on the collected sample's initial nitrogen content and mineralization level. However, the literature showed biochar amended vermicompost attributed to the bio-oxidation and reduces N loss and NH_3 volatilization due to high absorption properties and increasing nitrogen content (Malińska et al., 2014; Vandecasteele et al., 2016 Sánchez-García et al. 2015; Zhang and Sun, 2016). Hua et al. (2009) reported that incorporating 9% biochar into sludge composting significantly lessened 64.1% total N loss. This effect is attributed to the bio-oxidation of biochar surfaces itself and the adsorption of humic substances.

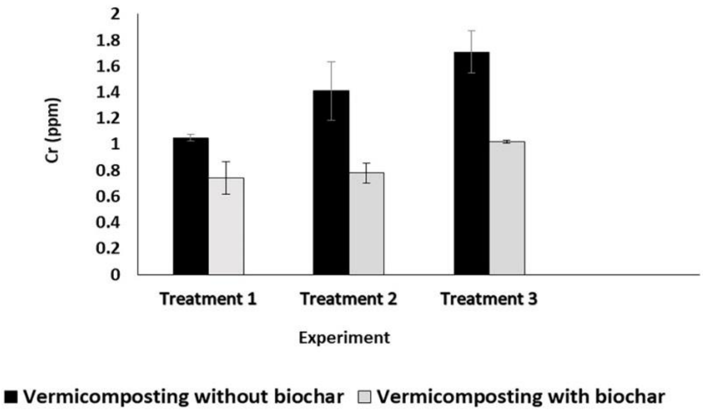
Composting amended with biochar had increased the value of total phosphorus (TP) than composting without biochar but without significant differences (p<0.05). TP in vermicomposting with biochar had a higher concentration ranged from (6010±3.0-12978.67±9.61) ppm, whereas TP in vermicomposting without biochar had (5820±2.65-8015.67±1.15) ppm (Figure 2(d)). The observed increase of TP in vermicompost is probably due to mineralization and mobilization of phosphorus resulting from the enhanced phosphatase activity by microorganisms in the gut epithelium earthworms (Kiyasudeen et al., 2016, Malińska *et al.*, 2016).

Total potassium (TK) showed significant differences (P<0.05) and an increase in potassium concentration in vermicomposting with biochar than vermicomposting without biochar (Figure 2(e)). Total potassium in vermicomposting with biochar had a higher concentration ranged from (447.67±0.58-612.33±1.53) ppm, where total potassium in vermicomposting without biochar had (203±1.73-304.67±2.08) ppm. The increase of potassium concentration in vermicompost with biochar could be due to biochar containing a higher potassium concentration than cow dung and sludge and increase these essential bioavailability macronutrients. Biochar could also enhance essential potassium in compost due to its cation exchange capacity (Malińska et al., 2016; Yadav and Garg, 2016; Vandecasteele et al., 2016). FBIS also contained some negligible amount of heavy metals such as Cr and Cd, which decreased after vermicomposting with biochar addition (Figure 3(a) and (b)) may not contain much toxicity.

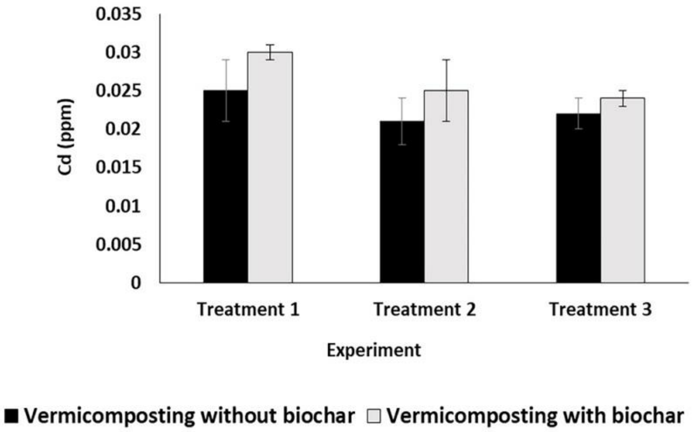


(e)

Figure 2. (a) Changes of pH, (b) organic carbon (OC), (c) total Kjeldahl nitrogen (TKN), (d) total phosphorus (TP), and (e) total potassium (TK) in different treatment (all values are the mean and standard deviation of three replicates; ANOVA, $p<0.05$ for pH, $p<0.05$ for OC but $p>0.05$ for TKN, $p<0.05$ for TK but $p>0.05$ for TP). Capped bars indicate the standard deviation of the replicates.



(a)



(b)

Figure 3. Heavy metals (a) Chromium and (b) Cadmium content in different treatments (All values are the mean and standard deviation of three replicates; and ANOVA, $p>0.05$ both for Cr, Cd). Capped bars indicate the standard deviation of the replicates.

The effect of biochar on vermicomposting of food and beverage industrial sludge and cow dung can be a more effective way to improve conventional vermicomposting efficiency. In addition, Gong et al., 2018 determined 6% biochar addition promoted earthworm growth and the vermicomposting of green waste as well as toxicity to germinating seeds. Wu et al., 2017 reviewed the combined application of biochar and compost that had great potential for waste management, remediating contaminated soils, and increasing plant growth. Malińska et al. (2016) obtained the addition of biochar in vermicomposting facilitated the growth and reproduction of *Eisenia fetida*. Biochar could allow faster and more efficient conversion of sewage sludge into vermicompost.

CONCLUSION

Vermicomposting sludge from the food and beverage industry, particularly the dairy industry, is little explored and reported from Bangladesh. In this study, we had compared the addition of cow dung and biochar as a bulking agent in vermicomposting of dairy industry sludge.

Biochar can be an effective way to improve conventional vermicomposting efficiency. Despite the decrease in nitrogen content, all other results indicated that 10% of the biochar addition enhanced the quality of vermicomposting and high GI and increased cocoons and earthworm numbers within 35 days. The proposed product can improve soil quality, both in terms of crop production and counteracting soil degradation effects. However, one of the significant limitations of this study was adopting a commercial composting time of 35 days. Moreover, due to the unavailability of the standards, we could check only two heavy metals. All heavy metals should be explored in the future. Therefore, further studies are required to determine the quality of the dairy industry's vermicomposting sludge with biochar addition and their role in the circular economy.

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CONFLICT OF INTEREST

The authors declare that there is not any conflict of interests regarding the publication of this manuscript. In addition, the ethical issues, including plagiarism, informed consent, misconduct, data fabrication and/ or falsification, double publication and/or submission, and redundancy has been completely observed by the authors.

LIFE SCIENCE REPORTING

No life science threat was practiced in this research.

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