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Vermiconversion of biowastes with low-to-high C/N ratio into value added vermicompost

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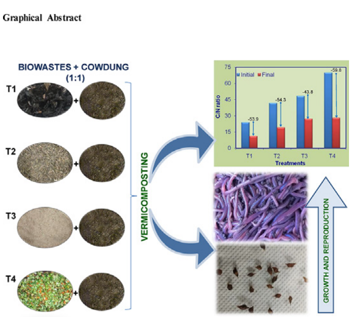
GRAPHICAL ABSTRACT



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ABSTRACT

Seaweed (T1), sugarcane trash (T2), coir pith (T3) and vegetable waste (T4) with cowdung (1:1, w/w) were vermicomposted using *Eudrilus eugeniae* (50 days). The pH in vermicomposts showed a decrease while electrical conductivity showed increment. The organic matter content, organic carbon, lignin, cellulose, C/N and C/P ratios in vermicompost was significantly lower than compost. Total NPK contents of vermicompost were significantly elevated ($P < 0.05$) with 12.04–63.75%, 19.05–31.58% and 22.47–42.55%, respectively. The significantly higher growth rate of 1.41 and 7.74 mg/worm/day was observed in T1 on 10th and 50th day respectively, with 23.91 initial C/N ratio; while it was 0.85 and 4.81 mg/worm/day in T4 with 69.81 initial C/N ratio. A similar pattern was reflected in cocoon production, hatchling success and hatchling number/cocoon. Results revealed that vermicompost quality, worm growth, and reproduction depend on C/N ratio. The study suggests that amendment materials like cowdung are necessary to reduce C/N ratio for effective vermicomposting.

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1. Introduction

The sustainable option for utilizing organic biomass residues is vermicomposting technology which employs earthworms for the conversion and recovery of nutrients from biowastes (Das et al., 2019a; Soobhany, 2019). Vermicomposting has been found to be an environment-friendly effective technology for recycling waste biomass generated during the course of agricultural and industrial activities, and the generation of organic solid wastes by rural and urban population (Bhat et al., 2018; Boruah et al., 2019; Paul et al., 2018). Of course, the phytomasses such as seaweeds and weeds can also be vermistabilized for the production of nutrient-rich vermicompost (Ananthavalli et al., 2019a,b; Devi and Khwairakpam, 2020). However, the organic feedstock materials for the vermicomposting need to be pre-decomposed and/or amended with suitable amendment materials to make them acceptable for earthworm and microbial activity which are the key factors of vermiconversion process (Esmaeili et al., 2020; Gong et al., 2019; Karmegam et al., 2019).

The carbon-to-nitrogen (C/N) ratio plays a major role during the vermicomposting process where the correct ratio provides nutrition for the earthworms (Ndegwa and Thompson, 2000) and it is also essential for microbial activity and multiplication (Jiang et al., 2011; Lv et al., 2018). With the initial C/N ratios of 15, 22 and 27, Wang et al. (2019) observed that the organic matter is the critical factor for the redistribution of copper, zinc, and phosphorus fractions in composting. Lv et al. (2018) reported while vermicomposting sewage sludge with *Eisenia fetida* that the increase in C/N ratio (12, 16 and 20) results in the reduction of greenhouse gas emissions. Moreover, the compost maturity is mainly contributed by C/N ratio among the various other factors (Guo et al., 2015). Normally C/N ratio tends to decrease due to the decomposition of organic materials and reduction of organic carbon. The reduction of organic carbon during vermicomposting is chiefly attributed to the respiratory activity of microorganisms and earthworms with a synchronized increase in nitrogen added through the mucus and nitrogenous excretion by the worms (Boruah et al., 2019; Lv et al., 2018). Even, the C/N ratio in soils applied with vermicompost essentially improves soil physico-chemical and biological activities apart from crop growth and yield (Das et al., 2019b). Though the studies on vermicomposting of different sources of organic substrates focus on the changes in C/N ratio and associated parameters, the studies on the effect of low-to-high C/N containing biowastes on the nutrient quality, and worm growth and reproduction is least studied. Hence in the present study, four different biowastes with a wide range of C/N ratio (23–70) have been subjected to vermicomposting using the earthworm, *Eudrilus eugeniae* addressing the relationship of C/N ratio to that of vermicompost quality, and worm growth and reproduction.

2. Materials and methods

2.1. Collection and preparation of organic substrates for vermicomposting

Based on the C/N ratio and availability, the organic substrates such as sugar cane trash (ST), coir pith (CP), vegetable market waste (VW) and seaweed (*Spatoglossum asperum* J. Agardh, Phaeophyceae) (SW) were collected and processed for vermicomposting experiments. The organic waste substrates were chopped into small pieces and allowed for shade drying and pre-decomposed for 15 days by sprinkling water and regular turning in rectangular cement tanks. The substrates were then mixed with urine free cowdung in 1:1 ratio on dry weight basis as shown in Table 1. The treatments, T1-T4 were maintained in triplicates in rectangular plastic containers of 45 × 30 × 15 cm size. Adequate water was added to each substrate to hold 60–70% moisture content. After 24 h stabilization of the substrates, few worms were placed in the dampened medium and observed for any mortality for 48 h. If no mortality occurred, the medium was deemed fit for the experimental animals. The epigeic earthworm, *E. eugeniae* cultured in mass culture tanks with cowdung at Vermiculture Unit, Alagappa University, Karaikudi was used for the study. Fifty earthworms of uniform size (765.6 ± 21.1 mg) were sorted out and transferred to vermibeds of each treatment. The substrates without earthworms were also maintained to compare the effect of earthworms in vermiconversion of different substrates. Before introducing the earthworms, the initial physico-chemical parameters were analyzed adopting the standard methods as described below. The vermicomposting trials were conducted for 50 days and the final end product obtained from the treatments with earthworms—vermicompost and the treatments without earthworms—compost was subjected to physico-chemical analysis.

Table 1
Vermicomposting treatments.

Treatments	Earthworm species used (no./vermibed)	Organic substrate mix (1:1, w/w)
T1	*E. eugeniae* (50)	SW + CD
T2	*E. eugeniae* (50)	ST + CD
T3	*E. eugeniae* (50)	CP + CD
T4	*E. eugeniae* (50)	VW + CD

ST – Sugarcane Trash; CP – Coir Pith; VW-Vegetable Market Waste; SW-Seaweed; CD – cowdung.

Table 2
Initial physico-chemical composition of the various substrates used for vermicomposting.

Parameters	Substrates			
	T1	T2	T3	T4
pH	7.49 ± 0.63	7.41 ± 0.62	7.48 ± 0.59	7.57 ± 0.54
EC (dS m^{-1})	1.49 ± 0.06	2.09 ± 0.18	1.65 ± 0.09	1.76 ± 0.08
TOC (%)	30.85 ± 1.76	33.04 ± 1.98	35.20 ± 2.04	48.17 ± 2.86
OMC (%)	53.37 ± 3.19	57.16 ± 3.30	60.90 ± 4.27	87.33 ± 5.64
TKN (%)	1.29 ± 0.07	0.79 ± 0.04	0.73 ± 0.03	0.69 ± 0.04
TP (%)	0.67 ± 0.05	0.46 ± 0.03	0.41 ± 0.03	0.40 ± 0.02
TK (%)	0.84 ± 0.06	0.65 ± 0.03	0.40 ± 0.02	0.48 ± 0.03
Cellulose (%)	14.37 ± 0.92	26.15 ± 1.58	22.03 ± 1.18	23.97 ± 1.64
Lignin (%)	17.49 ± 1.23	23.47 ± 1.42	26.34 ± 1.58	22.53 ± 1.41
C/N ratio	23.91 ± 1.29	41.82 ± 2.76	48.23 ± 4.01	69.81 ± 5.22
C/P ratio	46.04 ± 2.91	71.83 ± 4.57	85.85 ± 5.39	120.43 ± 8.98
Cellulose/lignin ratio	0.82 ± 0.06	1.11 ± 0.04	0.84 ± 0.06	1.06 ± 0.08
Lignin/nitrogen ratio	13.56 ± 0.97	29.71 ± 1.51	36.08 ± 2.47	32.65 ± 2.20

The data represent the mean ± standard deviation of three replicates.

2.2. Physico-chemical analysis of initial and final substrates

The physico-chemical composition of the vermibed substrate were analyzed for the selected parameters such as pH, electrical conductivity (EC) (Jackson, 1973), total organic carbon (TOC), organic matter content (OMC) (Walkley and Black, 1934), potassium (TK) (Tandon, 1993), cellulose (Updegraff, 1971) and lignin (Chesson, 1978). The total nitrogen (total Kjeldahl nitrogen, TKN) was estimated after digesting the sample with conc. H_2SO_4 and conc. $HClO_4$ (9: 1, v/v) using Kjeldahl distillation unit (Tandon, 1993). The total phosphorus (TP) content was analyzed with molybdenum in conc. H_2SO_4 using digital colorimeter (Systronics, India) (Tandon, 1993).

The ratio of the percentage of carbon to that of nitrogen (i.e. C/N ratio) was calculated by dividing the percentage of carbon estimated for the sample with the percentage of the nitrogen estimated for the same sample. The ratio of the percentage of carbon to that of phosphorous (i.e. C/P ratio) was calculated by dividing the percentage of carbon estimated for the sample with the percentage of phosphorus estimated

Table 3
pH, EC, OMC and TOC in different treatments after composting (50 days).

Treatments	pH		% Change	EC (dS/m)		% Change
	Compost	Vermicompost		Compost	Vermicompost	
T1	7.68 ± 0.55	7.14 ± 0.37	−7.56	2.04 ± 0.12	2.45 ± 0.19	20.10
T2	7.33 ± 0.42	7.18 ± 0.23	−2.08	2.09 ± 0.14	2.39 ± 0.18	14.35
T3	7.37 ± 0.35	7.22 ± 0.29	−2.07	1.63 ± 0.08	1.92 ± 0.06	17.79
T4	7.50 ± 0.33	7.29 ± 0.45	−2.88	1.91 ± 0.11	2.11 ± 0.15	10.47
ANOVA	*F-value*	*P-value*		*F-value*	*P-value*	
Between treatments	0.653581	0.632387^{NS}		27.04955	0.011305*	
Between end products$	7.869379	0.067559^{NS}		48.64865	0.006048*	
Treatments	**OMC (%)**		**% Change**	**TOC (%)**		**% Change**
	Compost	**Vermicompost**		**Compost**	**Vermicompost**	
T1	44.26 ± 3.68	37.65 ± 2.34	−14.93	25.67 ± 1.45	21.84 ± 1.53	−14.92
T2	54.81 ± 2.62	48.12 ± 2.43	−12.21	31.79 ± 2.54	27.91 ± 1.68	−12.21
T3	54.15 ± 4.77	43.50 ± 3.25	−19.67	31.41 ± 2.12	25.23 ± 1.42	−19.68
T4	75.56 ± 5.80	63.36 ± 5.55	−16.15	43.83 ± 3.65	36.75 ± 2.94	−16.15
ANOVA	*F-value*	*P-value*		*F-value*	*P-value*	
Between treatments	72.5482	0.00268*		72.28554	0.002695*	
Between end products$	40.82957	0.007762*		40.68222	0.007802*	

The data represent the mean ± standard deviation of three replicates. '*' and 'NS' in P-values indicate significant and non-significant difference at P < 0.05 by Two-way ANOVA, respectively. $ - refers to final compost and vermicompost obtained without and with *E. eugeniae*;

Table 4
C/N and C/P ratios in different treatments after vermicomposting (50 days).

Treatments	C/N ratio		% Change	C/P ratio		% Change
	Compost	Vermicompost		Compost	Vermicompost	
T1	17.83 ± 1.02	11.03 ± 0.72	−38.14	36.15 ± 2.04	20.60 ± 1.13	−43.02
T2	34.03 ± 2.45	19.12 ± 1.16	−43.81	61.13 ± 5.20	36.72 ± 1.45	−39.93
T3	37.84 ± 3.06	27.13 ± 1.00	−28.30	69.80 ± 5.83	43.50 ± 2.75	−37.68
T4	54.79 ± 3.85	28.05 ± 1.86	−48.80	104.36 ± 8.25	73.50 ± 6.28	−29.57
ANOVA	*F-value*	*P-value*		*F-value*	*P-value*	
Between treatments	6.883373	0.073709^{NS}		61.41064	0.003427^{S}	
Between end products$	11.75499	0.041582^{S}		57.23174	0.00479^{S}	

The data represent the mean ± standard deviation of three replicates. '*' and 'NS' in P-values indicate significant and non-significant difference at P < 0.05 by Two-way ANOVA, respectively. $ - refers to final compost and vermicompost obtained without and with *E. eugeniae*;

Table 5
Cellulose, lignin, cellulose: lignin ratio and cellulose: nitrogen ratio in different treatments after vermicomposting (50 days).

Treatments	Cellulose (%)		% Change	Lignin (%)		% Change
	Compost	Vermicompost		Compost	Vermicompost	
T1	12.03 ± 1.10	09.32 ± 0.56	−22.53	15.26 ± 1.42	12.64 ± 1.03	−17.17
T2	24.65 ± 1.96	18.75 ± 0.91	−23.94	22.61 ± 1.36	20.30 ± 1.09	−10.22
T3	19.14 ± 1.20	15.21 ± 0.90	−20.53	24.97 ± 1.65	19.58 ± 0.94	−21.59
T4	21.56 ± 1.43	17.14 ± 0.73	−20.50	20.85 ± 1.52	17.00 ± 0.09	−18.47
ANOVA	*F-value*	*P-value*		*F-value*	*P-value*	
Between treatments	51.63349	0.00442*		28.69695	0.010384	
Between end products$	41.28827	0.00764*		25.62009	0.014884	
Treatments	**Cellulose: lignin ratio**		**% Change**	**Cellulose: nitrogen ratio**		**% Change**
	Compost	**Vermicompost**		**Compost**	**Vermicompost**	
T1	0.79 ± 0.05	0.74 ± 0.06	−06.33	10.60 ± 1.10	6.38 ± 0.35	−39.81
T2	1.09 ± 0.04	0.92 ± 0.04	−15.60	24.85 ± 1.23	13.90 ± 0.98	−44.06
T3	0.77 ± 0.04	0.78 ± 0.04	−01.30	30.08 ± 1.65	21.05 ± 0.85	−30.01
T4	1.03 ± 0.07	1.01 ± 0.03	−01.94	26.06 ± 2.45	12.98 ± 0.13	−50.19
ANOVA	*F-value*	*P-value*		*F-value*	*P-value*	
Between treatments	12.63052	0.032982*		14.14313	0.028231*	
Between end products$	2.124498	0.24102 ^{NS}		24.30363	0.015999*	

The data represent the mean ± standard deviation of three replicates. '*' and 'NS' in P-values indicate significant and non-significant difference at P < 0.05 by Two-way ANOVA, respectively. $ - refers to final compost and vermicompost obtained without and with *E. eugeniae*.

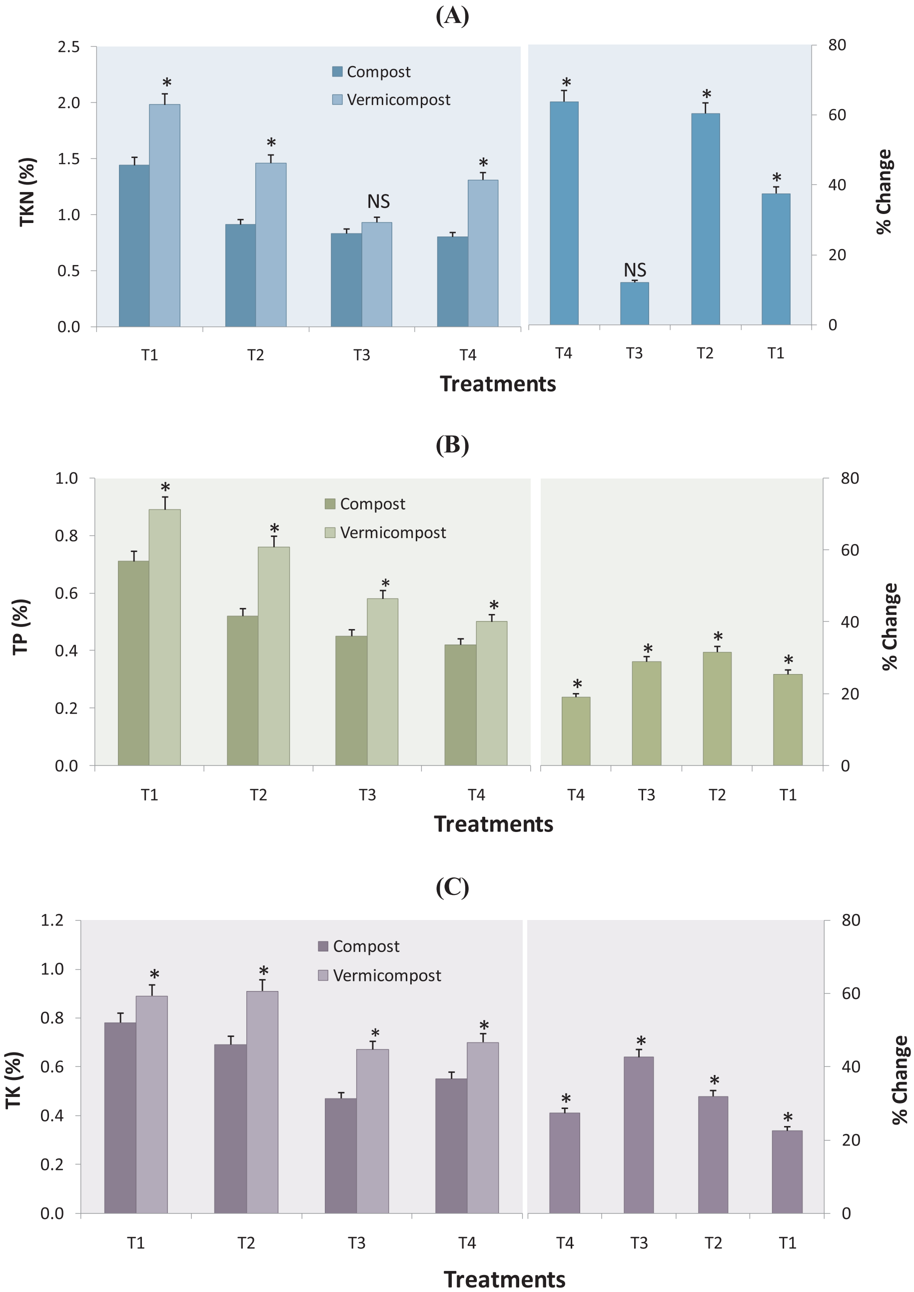

Fig. 1. (A) TKN, (B) TP and (C) TK contents in different treatments after vermicomposting (50 days). '*" and 'NS' indicate significant and non-significant difference by ANOVA at $P < 0.05$. Error bars indicate standard deviation.

for the same sample. The ratio of the percentage of cellulose to that of lignin (i.e. cellulose/lignin ratio) was calculated by dividing the percentage of cellulose estimated for the sample with the percentage of lignin estimated for the same sample. Similarly, the ratio of the percentage of lignin to that of nitrogen (i.e. lignin/nitrogen ratio) was also calculated by dividing the percentage of lignin estimated for the sample with the percentage of nitrogen estimated for the same sample. The percentage change (increase/decrease) of various physico-chemical parameters in the vermicompost over the worm-unworked compost was calculated.

2.3. Growth and reproduction of earthworms

In order to find out the growth and reproduction of the earthworms, a separate replica of experimental sets was carried out so as to avoid the disturbance to vermicomposting process. The vermibed substrate in each treatment was carefully screened for cocoons every 10 days until the termination of the study (50 days). The cocoons were collected and incubated in Petri dishes with respective substrate with 80% moisture content. The biomass and number of earthworms in the treatments were recorded. The growth rate, average cocoon production rate, percentage hatching success and number of hatchlings hatching from each cocoon were recorded following the method described by Karmegam and Daniel (2009).

2.4. Statistical analysis

The data obtained were subjected to statistical analysis using SPSS version 18.0 (SPSS Inc., Chicago, USA). All the data were expressed as mean ± standard deviation of three replicates. The differences in parameters between worm-unworked compost and worm-worked vermicompost were derived using Two-way ANOVA at $P < 0.05$ level and

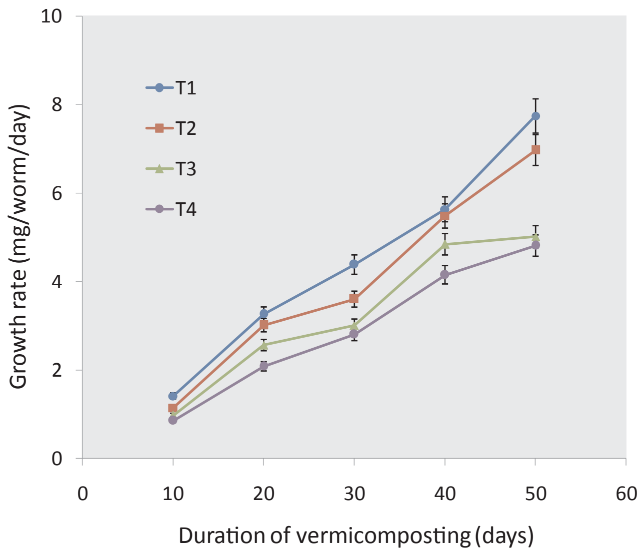

Fig. 2. Growth rate of *E. eugeniae* in vermicomposting treatments over a period of 50 days. Error bars indicate ± standard deviation.

the mean comparison was done by ANOVA ($P < 0.05$). The correlation of pH, EC, TOC, TKN and C/N ratio to that of worm growth and reproduction was analyzed by computing the data and the results obtained were tabulated with the level of significance between parameters.

3. Results and discussion

In the vermicomposting process, apart from various physico-chemical characteristics, C/N ratio is considered as central in terms of rate of decomposition, compost maturity, and worm growth and reproduction. In the present study, the initial C/N ratio was 23.91 (T1), 41.82 (T2), 48.23 (T3) and 69.81 (T4) with a wide variation from low-to-high levels of C/N ratio. The pH showed insignificant reduction in vermicompost in comparison with compost. The initial range of electrical conductivity in the initial substrates was 1.49–2.09 dS m^{-1}.

A maximum of 87.33% OMC was found in T4 followed by T3 > T2 > T1. The TKN, TP and TK contents in the initial T1 substrate was higher than the other treatments (Table 2). The pH of the vermicompost was decreased with a percentage change of 2.88 to 7.56 where the change was insignificant ($P < 0.05$) (Table 3). The decrease of pH during vermicomposting could be attributed to the joint action of earthworms and microorganisms leading to the production of organic acids (Boruah et al., 2019; Hussain et al., 2018). However, Singh and Kalamdhad (2016) reported increase of pH values after 45 days of vermicomposting of an invasive weed *Salvia natans* with *Eisenia fetida;* while the authors of the same study noticed decrease in pH values on 15th day. The increase or decrease of pH during vermicomposting could be due to the production of organic acids, substrate-dependent intermediate chemical substances, ammonium (NH^{4+}) ions and interaction of earthworms secretions (Singh and Kalamdhad, 2016; Vig et al., 2011). The pH range of vermicomposts obtained from different treatments is very close to neutral indicating the degradation of organic material mixture through humification process which is in conformity with the results of Devi and Khwairakpam (2020). The change of pH did not differ significantly between the treatments and also between the compost and vermicompost; while EC showed statistically significant difference between treatments ($P = 0.014$) and between vermicompost and compost. A maximum of 20.10% increase in EC was found in T1 followed by T3, T2, and T4 (Table 3). Due to mineralization process, the soluble salts are formed during vermicomposting which consequentially enhanced the level of EC. For instance, Ananthavalli et al. (2019a) reported 45.26–64.29% increase of EC in seaweed vermicomposts over the composts of respective substrates. A similar trend of increased EC in vermicompost has been well documented by earlier studies with different feedstock materials (Yadav and Garg, 2019; Yuvaraj et al., 2018, 2020). The percent decline of OMC was ranged between 12.21 and 19.68 in comparison with worm-unworked compost. The TOC was lower in T1 vermicompost (21.84%) followed by T3 (25.23%), T2 (27.91%) and T4 (30.75%). The variation in the change of TOC between treatments was highly significant ($P = 0.0027$) showing that the initial TOC content significantly influences vermicomposting (Table 3). The reduction of OMC and TOC during vermicomposting in the present study fall in line with the previous studies (Negi and Suthar, 2018; Prakash and Karmegam, 2010; Sharma and Garg, 2018). The reduction in TOC is principally due to the respiratory activity of earthworms and microorganisms, assimilation of carbon in tissues and cells where it ultimately reduced the level of OMC (Karmegam et al., 2019; Sharma and Garg, 2018).

The final C/N ratio in vermicompost of all the treatments was significantly lower than that of worm-unworked compost (Table 4). The decline in the C/N ratio followed uniformity in all the treatments where the deviation of changes in C/N between treatments differs significantly. The lowest C/N of 11.03 was recorded in the T1 vermicompost followed by T2, T3, and T4, indicating that the initial C/N ratio is important factor for the final C/N ratio. As the reduction of TOC is the major factor for the reduction of C/N ratio, it was also reflected in C/P ratio. A range of 29.57–43.02% reduction in C/P ratio in vermicompost with that of compost was observed with a maximum in T1 (Table 4). The cellulose content in all the treatments with *E. eugeniae* showed significant reduction of 20.50 to 23.94% while it was 10.22 to 21.59% for lignin (Table 5). The cellulose: lignin ratio in vermicompost was well within 1.0. The percent decrease of cellulose: nitrogen ratio was found to be higher (30.01 to 50.19%) indicating that cellulose degradation plays a major role in the reduction of C/N ratio. The difference in cellulose: nitrogen ratio between vermicompost and compost was significant ($P = 0.016$). The reduction of C/N ratio was higher in vermicomposts than the composts. A maximum reduction of 48.80% C/N ratio was recorded in the vermicompost harvested from T4 followed by T2 (43.81%), T1 (38.14%) and T3 (28.30%). Different range of decrease of C/N ratio during vermicomposting has been reported by earlier studies. Zhi-wei et al. (2019) reported 58.55 to 71.96% reduction of C/N ratio for rice straw and kitchen waste vermicompost. Boruah et al. (2019) observed a higher rate of 91.10% reduction in C/N ratio during citronella bagasse and paper mill sludge vermicomposting employing *E. fetida*. A range of 41.5–48.4% reduction in C/N ratio was found for vermicomposting of organic solid wastes with *E. eugeniae* (Soobhany et al., 2015). The variation in the reduction of C/N ratio in vermicompost different biowaste materials could be attributed to the rate of earthworm activity and reproduction because of the food preference and favorable C/N ratio in the initial substrate. In general, the ratio of C/N is used to assess the mineralization of organic matter and is considered as an important indicator of compost maturity. The release of CO_2 by respiratory activity, addition of nitrogenous worm excretion and the earthworm gut-associated bioactivity have been found to be coupled with the reduction of C/N ratio in the vermicompost (John Paul et al., 2011; Alidadi et al., 2016; Sharma and Garg, 2018).

The incremental change of TKN in vermicompost was significantly higher than the compost obtained without the earthworms (Fig. 1A). In terms of final TKN, T1 showed the highest value of 1.98% followed by T2, T4, and T3; whereas, in terms of percent change, T3 was higher (63.75%) followed by T2 (60.37%). The TKN contents in the final vermicompost are significantly higher ($P < 0.05$) than in respective composts excepting T3. Overall, the TKN increase in vermicompost ranged from 12.05 to 63.75% over compost was found in the present study corroborates with the earlier reports by Ananthavalli et al. (2019b), Sharma and Garg (2018) and Yuvaraj et al. (2020). The TP in vermicompost ranged between 0.50 and 0.89% with a significant

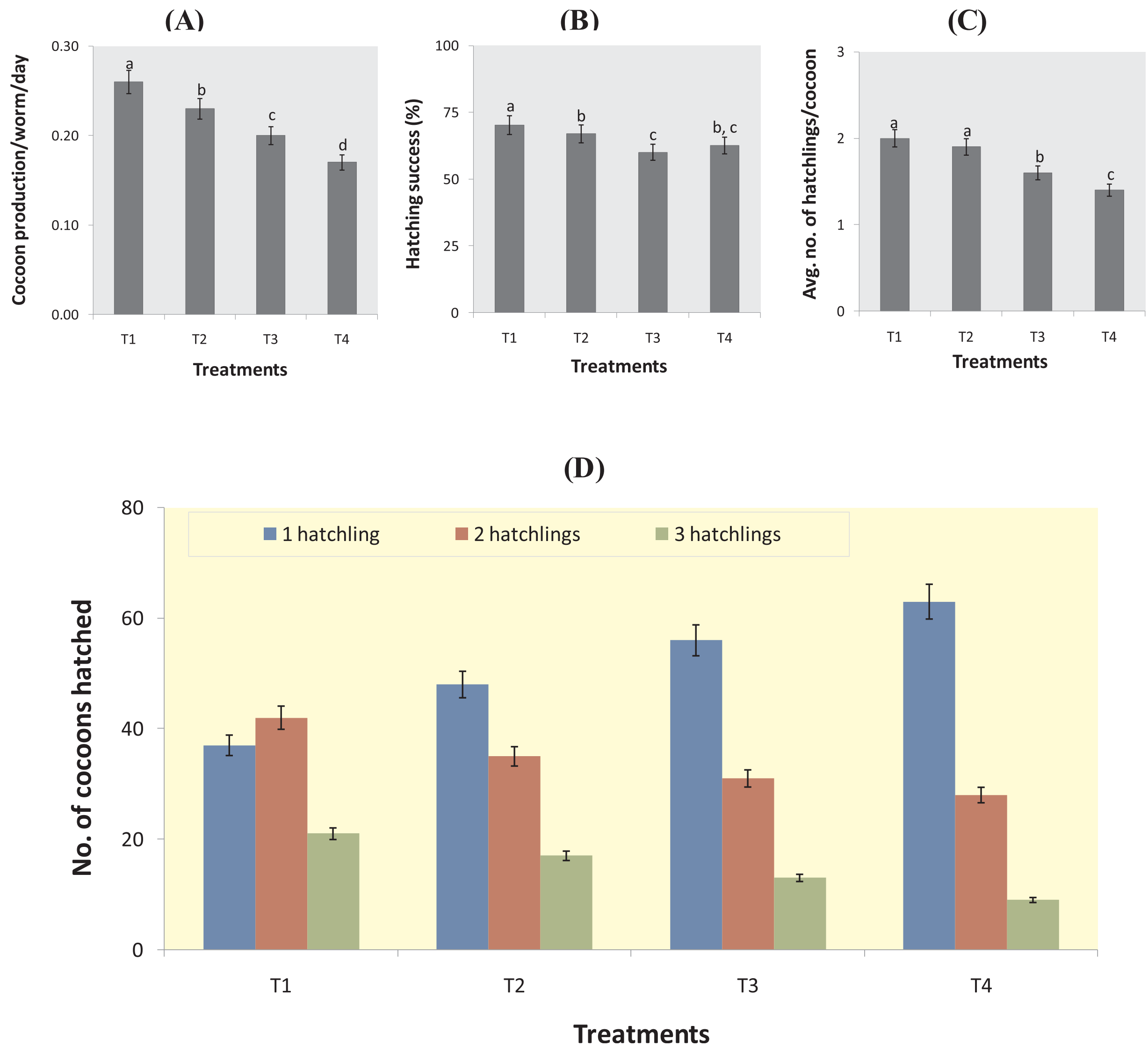

Fig. 3. Reproductive performance of *E. eugeniae* in different treatments over a period of 50 days. (A) Cocoon production, (B) Hatching success, (C) Mean number of hatchlings per cocoon and (D) Number of hatchlings per cocoon. Error bars indicate ± standard deviation. Treatment bars with different alphabets indicate significant difference at $P < 0.05$ by ANOVA.

Table 6
Correlation matrix showing the relationship of pH, EC, TOC, TKN and C/N ratio with growth and reproduction of *E. eugeniae*.

Parameters	pH	EC	TOC	TKN	C/N ratio	Growth rate	Cocoon production
pH	1						
EC	−0.510*	1					
TOC	0.803*	0.092	1				
TKN	−0.119	−0.583*	−0.616*	1			
C/N ratio	0.595*	0.301	0.937*	−0.852*	1		
Growth rate	−0.568*	−0.031	−0.768*	0.825*	−0.887*	1	
Cocoon production	−0.611*	−0.188	−0.899*	0.859*	−0.982*	0.958*	1

* Significant at $P < 0.01$.

percent change of 19.05–31.58% over compost (Fig. 1B). The TK content in vermicompost obtained from T1, T2, T3, and T4 was 0.89, 0.91, 0.67 and 0.70% with a respective percent change of 22.47, 31.88, 42.55 and 27.27 over relevant treatment composts (Fig. 1C). The incremental change of TP and TK in vermicomposts of all the treatments was significantly higher ($P < 0.05$) than the composts indicating that the shift in TP and TK during vermicomposting of biowastes followed uniformity irrespective of initial C/N ratio. However, the increase of TP and TK contents between vermicomposts varied in accordance with the initial contents. Several factors are reported to be responsible for the elevated levels of TKN contents in vermicompost, *viz.,* initial TKN, mucus secretion, TOC reduction and TKN addition by dead earthworms (Bhat et al., 2015; Boruah et al., 2019; Devi and Khwairakpam, 2020). The outcome of overall loss of dry matter proportionately increased the levels of TP and TK (Devi and Khwairakpam, 2020; Jain et al., 2018). In addition, the activities of phosphatases and enhanced microbial activity are possibly related to the higher levels of TP in vermicompost (Ghosh et al., 2018; Karmegam et al., 2019).

The worm growth and reproduction is an indication of the good vermicomposting process. The worm growth rate was higher in T1 than in the rest of the treatments (Fig. 2). The growth rate of *E. eugeniae* gradually increased until the end of vermicomposting period, i.e., 50th day. The highest growth rate of 7.74 ± 0.43 mg/worm/day was observed in T1 followed by T2 (6.67 ± 0.52) > T3 (5.01 ± 0.32) > T4 (4.81 ± 0.28) where it was 1.41 ± 0.06, 3.26 ± 0.19, 4.38 ± 0.35 and 5.63 ± 0.37 mg/worm/day respectively on 10, 20, 30 and 40 days of vermicomposting. The growth rate as depicted in Fig. 2 evidently illustrates the growth of *E. eugeniae* is dependent on the C/N ratio of the initial feedstock materials. The cocoon production rate, hatching success, number of hatchlings from each cocoon as in Fig. 3 clearly reveals that lower C/N ratio is responsible for enhanced activity, growth, and reproduction of earthworms. The highest cocoon production rate of 0.26/worm/day was recorded in T1 followed by T2 > T3 > T4. The cocoon production rate showed declining trend with the higher initial C/N ratio of the substrates and the C/N ratio between T1 and T2, T2 and T3, and T3 and T4 were significantly different at 5% level. The correlation of pH, EC, TOC, TKN and C/N ratio to that of growth rate and cocoon production of *E. eugeniae* is given in Table 6. The pH of the treatments showed positive influence over the C/N ratio while it was inversely related to the growth and cocoon production rate of *E. eugeniae* ($P < 0.01$). The EC did not show any reflective effect on worm growth and reproduction. The TOC values of the treatments had positive correlation with that of the C/N ratio while it had inverse relationship with growth rate and cocoon production ($P < 0.01$). It is reasoned from the present study that higher TOC content results in reduced growth and reproduction. The present study also reveals that TKN content in the treatments had significant impact on the final C/N ratio of the vermicompost. Similar to TOC, the C/N ratio showed highly significant inverse relationship indicating that its increase could have an unfavorable effect on growth and cocoon production rate of *E. eugeniae*. Further, it is apparently perceptible from the present study that the growth rate favored by the substrate results in higher cocoon production rate ($P < 0.001$). Parallel studies on vermicomposting also show variation in worm growth and reproduction which depend on the nature of initial feedstock materials (Ananthavalli et al., 2019a; Devi and Khwairakpam, 2020). More prominently, in the present study C/N ratio showed a highly significant correlation followed by TKN, TOC, and pH with that of the growth and reproductive performance of *E. eugeniae*.

4. Conclusion

The study clearly revealed that the physico-chemical parameters of vermicompost, pH, OMC, TOC, C/N, C/P, cellulose, lignin, cellulose: lignin ratio and cellulose:nitrogen ratio were found to decline with respect to the initial C/N ratio of the substrates. The incremental change in the nutrient contents in vermicompost over compost is indicative of compost maturity in presence of the earthworm, *E. eugeniae* which corresponded with that of the initial C/N ratio. Moreover, the worm growth rate and reproduction are having direct relevance with that of C/N ratio. From the results of the present study, it is clear that the initial vermibed substrates are to be properly mixed with bulking materials like cowdung so as to obtain nutrient-rich vermicompost.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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