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Waste and Biomass Valorization

ISSN 1877-2641
Volume 11
Number 12

Waste Biomass Valor (2020)
11:6483-6497
DOI 10.1007/s12649-019-00899-0

Springer

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https://doi.org/10.1007/s12649-019-00899-0

ORIGINAL PAPER

Vermiremediation of Urban and Agricultural Biomass Residues for Nutrient Recovery and Vermifertilizer Production

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Received: 24 August 2019 / Accepted: 18 November 2019 / Published online: 11 December 2019

Abstract

Purpose Recovering the nutrients from organic solid residues is an ever demanding approach in terms of eco-friendly management. In this study, the efficiency of the earthworms, *Perionyx ceylanensis* and *Perionyx excavatus* in recycling and recovering plant nutrients from urban and agricultural biomass residues was investigated.

Methods Vermicomposting of four different types of organic biomass resources in combination with cowdung (1:1) for 50 days was conducted using the earthworms, *P. ceylanensis,* and *P. excavatus*. Physico-chemical, microbiological and maturity characteristics of the final product were analyzed.

Results Results showed a decrease in pH, organic matter content (OMC), total organic carbon (TOC), cellulose, lignin, carbon to nitrogen (C/N) and carbon to phosphorus (C/P) ratios, and an increase in total Kjeldahl nitrogen (TKN), phosphorus (TP), potassium (TP), calcium and sodium in vermicomposts over composts (prepared without earthworms). TKN increase in vermicomposts over composts was insignificant between *P. ceylanensis* and *P. excavatus,* while it was significant for TP, TK, and total microbial population ($P < 0.05$). The reduction of TOC, OMC, C/N, and C/P ratios were significantly higher in vermicomposts recovered from *P. ceylanensis* than *P. excavatus*. The results clearly indicated that physico-chemical and microbiological changes are dependent on the nature of organic resources and earthworm species used. The seed germination tests with black gram and sorghum showed that the vermicomposts were mature and non-phytotoxic.

Conclusion *P. ceylanensis* is equally efficient in recovering nutrients from organic biomass residues to that of a widely used, *P. excavatus*. Therefore, both *P. ceylanensis* and *P. excavatus* are efficiently used for recovering nutrients from urban and agricultural biomass residues and for vermifertilizer production.

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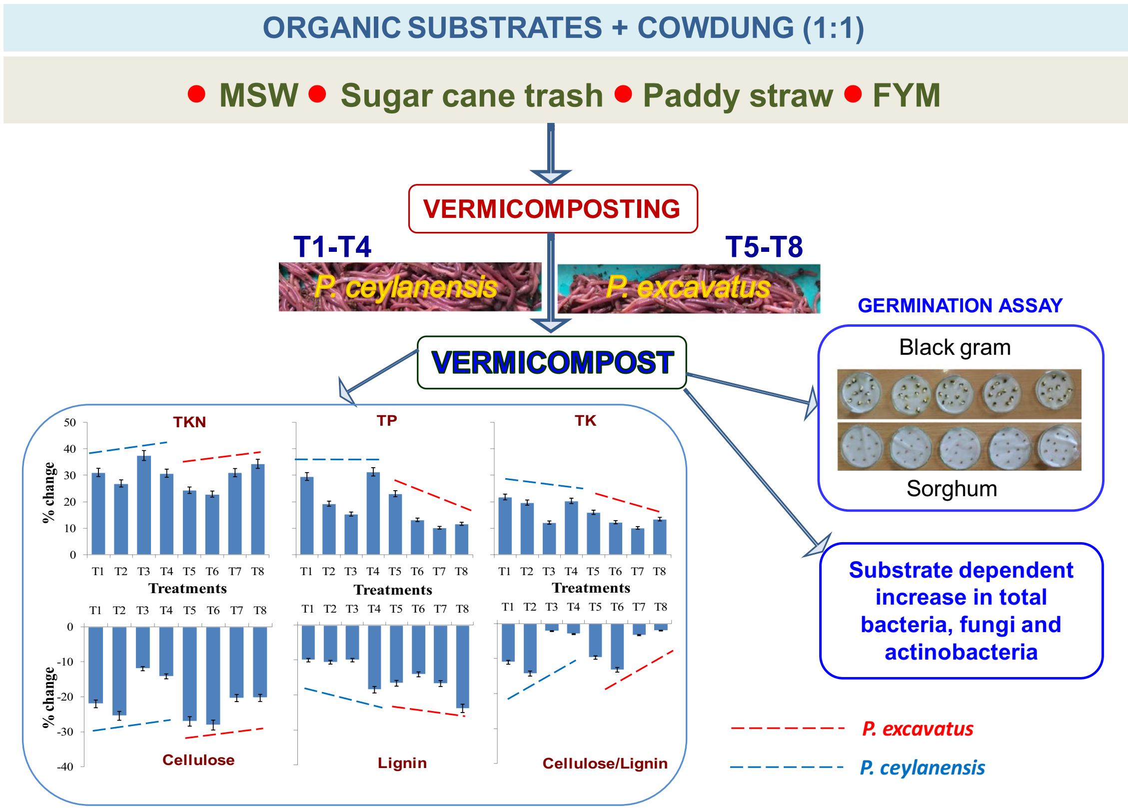
Graphic Abstract



Keywords Vermicomposting · Earthworms · Organic waste recycling · Microbial population · Nutrient recovery · C/N ratio

Statement of Novelty

Vermiremediation of organic wastes is a low-cost, eco-friendly technology that can be utilized for the recovery of plant nutrients from organic waste residues into valuable vermicompost. The study has been carried out with a strong belief that this attempt might explore the possibility of resource (nutrients) recovery through vermiremediation with the epigeic earthworms, *Perionyx ceylanensis* and *Perionyx excavatus* for a cleaner environment and sustainable agriculture. Comparative studies are essential to select efficient vermicomposting species; hence the study compared the *P. ceylanensis* with that of the widely used *P. excavatus* under similar conditions. The results obtained showed a potential future for *P. ceylanensis* on par with *P. excavatus* in recovering nutrients from waste residues and nutrient-rich vermicompost production.

Introduction

The generation of organic solid wastes is increasing day-by-day and poses severe concern towards sustainable management and resource recovery. *Per capita* generation of municipal solid waste exceeded 0.67 kg/day in India with an alarming yearly increase of around 5% posing disposal and environment-related issues [1, 2]. India is one of the largest sugarcane producers where it generates 5–8 tonnes of sugarcane trash per hectare of cultivation, which normally is burnt in the field itself resulting in nutrient loss and atmospheric pollution [3]. Being the second-largest country in the production of paddy, India generates around 130 million tonnes of paddy straw and about 50% is utilized for fodder and remaining quantities are burnt and a meager portion is used for other purposes [4]. Despite a vast potentiality of rice straw as a renewable energy source, a considerable portion is field burned due to practical difficulties [5]. Microbial composting and vermicomposting are found to be the suitable options to sustainably recover nutrients from paddy straw [6, 7]. The availability of these organic residues and their indiscriminate disposal needs to be considered in terms of environmental sustenance. Therefore attracts the attention of coming up with an environmental-friendly, economically viable and socially sustainable solid waste management technique [8]. Vermitechnology is one of such technologies proposed and utilized for the safe management of a variety of organic solid wastes [9, 10]. Vermitechnology or vermicomposting technology employs earthworms for the management of organic waste residues originating from various activities of human beings. The earthworms are the biocatalytic and detoxifying agents in the waste conversion process which ends up with waste minimization and production of valuable vermicompost by the way of positive influence on crop growth and yield [11–13]. Despite, vermicomposting—a biochemical transformation process supports the recovery of nutrients from a variety of organic substrates to a great extent, resulting in vermifertilizer production [14].

The earthworms like *Eudrilus eugeniae*, *Perionyx excavatus*, and *Polypheretima elongata* are employed for waste stabilization in tropical regions of the world [15]. The role of *P. excavatus* in waste management has been well established after the validation of its efficiency [16]. The growth, reproduction, the suitability of vermiconversion of a wide variety of organic wastes, production of vermicompost and the effect of vermicompost on plant growth and yield has been well established under tropical climates [17–19]. These pioneer studies have opened the way for the successful vermiconversion of rubber leaf litters [20], agriculture wastes [21], distillery industry sludge [22] pressmud [23], Java citronella biomass [24], kitchen vegetable waste and paddy straw in earthworm consortium [25], paper mill sludge [26], seaweeds [27], and other wastes by the earthworm, *P. excavatus*. Apart from *P. excavatus,* another Asian epigeic earthworm, *P. ceylanensis* is narrowly reported for its efficiency in vermicomposting [28–31].

The comparative studies on vermicomposting of various organic solid wastes for different species of earthworms are necessary to ascertain the efficiency in terms of physico-chemical and microbiological characteristics of vermicompost, nutrient recovery, the change in C/N ratio, mineralization of nutrients, duration of vermicomposting, growth and reproduction of selective species on par with widely used species. Even more, the comparison of composting and vermicomposting of organic solid wastes such as MSW is helpful in evaluating enhanced nutrient contents in the final product [32]. The studies conducted by Suthar and Singh [22] and Khwairakpam and Bhargava [23] insist that the comparative performance of earthworm species in monoculture or polyculture is essentially required since waste minimization and compost characteristics are dependent on organic waste inputs and the type of earthworm species employed. The better efficiency of *P. ceylanensis* was established in comparison with *Lampito mauritii* in vermicomposting of agro-industrial residue, leaf litter and a weed [31]. It is a well-known fact that the epigeic group of earthworms is well suited for the vermicomposting process. *P. ceylanensis,* being an epigeic species showed its enhanced vermiconversion efficiency than that of an anecic earthworm species, *L. mauritii* which has restricted utility in vermicomposting. Nevertheless, the suitability and waste conversion efficiency of *P. ceylanensis* has not been tried with the epigeic tropical earthworms like *P. excavatus*. The comparative studies of these two tropical earthworm species may provide an insight into their extensive deployment in vermicomposting in other parts of the world with tropical climates. Keeping this in view, the present study has been carried out to drive out the competence of *P. ceylanensis* and *P. excavatus* in vermiremediation of agricultural residues and municipal solid waste as a clean technology for waste minimization and nutrient recovery in a sustainable manner.

Materials and Methods

Vermicomposting

Four different organic wastes, source-separated municipal solid waste (MSW) from dumping yard of Karaikudi town, sugarcane trash (ST), paddy straw (PS) and farmyard manure (FYM) were collected from a private dairy farm in Karaikudi, Tamil Nadu, for the present study. The main source of FYM was cowdung and organic litters. The cowdung was collected from a local cow shed and air-dried. The MSW, ST, and PS were chopped into 3–5 cm size and separately subjected to pre-decomposition in rectangular draining cement tanks by sprinkling water and turning for 15 days. Each pre-decomposed organic waste was mixed with cowdung in 1:1 ratio (2 kg + 2 kg) on a dry weight basis and filled in plastic containers of 45 × 35 × 15 cm size in triplicates. The ratio of organic waste + cowdung (1:1) was used as this ratio was found suitable for *P. ceylanensis* and *P. excavatus* [26, 27, 31]. The vermibeds were maintained with adequate water to hold 60–70% moisture content and allowed 24 h for stabilization. The clitellate adult earthworms, *P. ceylanensis* and *P. excavatus* originally obtained from the vermiculture units, Department of Biology, Gandhigram Rural Institute, Gandhigram, Tamil Nadu, India, mass cultured in cowdung were used for vermicomposting treatments (T1-T8) as detailed in Table 1. After 24 h of stabilization of vermibed substrates, *P. ceylanensis* and *P. excavatus* were introduced into T1-T4, and T5-T8 respectively and maintained for 50 days. The substrates in treatments T1-T8 without earthworms were also maintained to compare the physico-chemical and microbiological changes with and without earthworms.

Table 1 Vermicomposting treatments

Treatments	Earthworms* used (no./vermibed)	Average weight of worms ± SD (mg)	Organic substrate mix** (1:1, w/w)
T1	*P. ceylanensis* (50)	573.72 ± 54.45	MSW + CD
T2	*P. ceylanensis* (50)	545.55 ± 44.73	ST + CD
T3	*P. ceylanensis* (50)	568.60 ± 49.59	PS + CD
T4	*P. ceylanensis* (50)	583.97 ± 41.81	FYM + CD
T5	*P. excavatus* (50)	624.95 ± 59.31	MSW + CD
T6	*P. excavatus* (50)	638.26 ± 56.40	ST + CD
T7	*P. excavatus* (50)	630.07 ± 50.56	PS + CD
T8	*P. excavatus* (50)	647.48 ± 61.26	FYM + CD

The data represent the mean ± standard deviation of three replicates

MSW municipal solid waste, *ST* sugarcane trash, *CD* cowdung, *PS* paddy straw, *FYM* farmyard manure

*Clitellate adults; ** 2 kg + 2 kg

Analysis of Physico-chemical Characteristics

The initial organic substrate mix, final compost (compost, recovered from treatments without earthworms) and vermicompost (vermicompost, recovered from treatments with earthworms) from each treatment were collected after 50 days and subjected to physico-chemical analyses following standard procedures. The final materials were sieved through 2 mm sieve to exclude worms and cocoons, and used for the analyses. The pH and electrical conductivity (EC) were analyzed in 1:10 (w/v) water suspension respectively using pH meter and electrical conductivity meter. The organic matter content (OMC) was calculated by deducing ash content while the method of Walkley and Black [33] was used to estimating total organic carbon (TOC). The total Kjeldahl nitrogen (TKN), total phosphorus (TP), total potassium (TK), calcium (Ca) and sodium (Na) were analyzed adopting the methods as described by Tandon [34]. The cellulose and lignin contents were estimated using the method of Chesson [35] and Updegraff [36], respectively. The ratios of TOC to TKN (C/N ratio), TOC to TP (C/P ratio), and cellulose to lignin (cellulose/lignin ratio) were calculated by deducing method. The initial physico-chemical characteristics of the organic substrate mixtures are given in Table 2. The pH of the initial substrates ranged from 7.38 to 7.95 with an average EC of 1.82 dS m^{-1}. The C/N ratio of 41.32, 48.61, 57.57 and 38.97 was recorded in the initial substrate cowdung mixtures (1:1) of MSW, ST, PS, and FYM.

Enumeration of Total Colony Forming Units of Microorganisms

The total colony forming units (CFU) of bacteria, fungi, and actinobacteria in the treatments on commencement of the experiment (initial), and after 50 days (compost and vermicompost) were enumerated by standard plate count method [37]. For the preparation of the stock solution, 1 g of each sample in a sterile conical flask containing 9 ml of distilled water was added and vortexed for 30 min. By using this stock, different dilutions were prepared from 10^{-1} to 10^{-7} with sterile distilled water. The diluted sample (1 ml) was poured into Petri plates containing the respective culture medium. The media used were: nutrient agar for bacteria,

Table 2 Initial physico-chemical composition of organic substrates used for vermicomposting (0 day)

Parameters	Organic substrates (1:1) **			
	MSW + CD	ST + CD	PS + CD	FYM + CD
pH	7.38 ± 0.33	7.45 ± 0.45	7.57 ± 0.41	7.95 ± 0.59
EC (dS m^{-1})	1.98 ± 0.11	1.56 ± 0.09	1.83 ± 0.15	1.89 ± 0.14
TOC (%)	32.23 ± 1.90	34.51 ± 3.20	39.72 ± 2.76	35.85 ± 2.07
OMC (%)	55.56 ± 2.45	59.50 ± 3.27	68.48 ± 4.19	68.81 ± 4.12
TKN (%)	0.78 ± 0.04	0.71 ± 0.05	0.69 ± 0.03	0.92 ± 0.07
TP (%)	0.45 ± 0.03	0.40 ± 0.03	0.39 ± 0.02	0.59 ± 0.04
TK (%)	0.61 ± 0.03	0.52 ± 0.04	0.56 ± 0.03	0.78 ± 0.04
Ca (%)	0.78 ± 0.06	0.75 ± 0.04	0.80 ± 0.03	0.69 ± 0.05
Na (%)	0.41 ± 0.03	0.34 ± 0.01	0.31 ± 0.01	0.37 ± 0.01
Cellulose (%)	23.15 ± 1.66	25.43 ± 0.21	26.06 ± 1.33	22.71 ± 1.49
Lignin (%)	25.73 ± 1.04	27.28 ± 1.35	28.00 ± 2.03	23.15 ± 1.22
C/N ratio	41.32 ± 1.21	48.61 ± 2.53	57.57 ± 4.92	38.97 ± 1.15
C/P ratio	71.62 ± 4.37	86.28 ± 5.39	101.85 ± 8.10	60.76 ± 4.58
Cellulose/lignin ratio	0.90 ± 0.05	0.93 ± 0.04	0.93 ± 0.05	0.98 ± 0.04

The data represent the mean ± standard deviation of three replicates

MSW municipal solid waste, *ST* sugarcane trash, *CD* cowdung, *PS* paddy straw, *FYM* farmyard manure

**2 kg + 2 kg on dry weight basis

Martin's Rose Bengal agar for fungi and Kenknight's agar for actinobacteria with three replicates per sample. The colonies developed in Petri plates ranging from 30 to 300 in were considered for enumeration. The results of total microbial enumeration were $\log_{10}$ transformed and expressed as $\log_{10}$ CFU g^{-1}.

Seed Germination Assay

To assess the maturity of vermicompost, and phytotoxicity of vermicomposts if any, seed germination assay tests were conducted using the seeds of black gram [*Vigna unguiculata* (L.) Walp.], and sorghum [*Sorghum bicolor* (L.) Moench] following the method adopted by Karmegam et al. [38]. The seeds of black gram and sorghum were obtained from Arunai Agro-agencies, Salem and good quality seeds were picked up by visual examination. Prior to germination assay, the seeds were surface sterilized with 2% mercuric chloride, then rinsed with distilled water and the moisture was removed with blotting paper. To each Petri plate containing 6 ml of 1:10 w/v compost extract over the Whatman No. 1 filter paper, 10 seeds were carefully laid and kept for 10 days, and the germination of seeds was noted every day. Five lots were maintained for each plant seeds. The method of UAC [39] for the calculation of germination percentage and Benech Arnold et al. [40] for finding out the germination index were consulted.

Statistical Analysis of Results

The results obtained were expressed as mean ± standard deviation (SD). The physico-chemical characteristics of the initial organic material mix, compost and vermicompost were statistically interpreted using ANOVA. The statistical significance of various treatments was assessed by one-way analysis of variance (ANOVA) with SPSS version 18.0 (SPSS Inc., Chicago, USA). Tukey's honestly significant different (HSD) multiple comparison tests were performed at $P < 0.05$ significance level. Two-way ANOVA was used to find out the effect of different treatments with reference to physico-chemical parameters and total microbial population. The seed germination percentage and germination index recorded were also subjected to two-way ANOVA to find out the significance ($P < 0.05$) of difference within treatments and between compost and vermicompost.

Results and Discussion

pH and Electrical Conductivity

The pH showed a reduction, whereas EC showed an increment in both compost and vermicompost from the initial levels after 50 days of experimentation (Fig. 1). The change in pH was 1.79–7.56% in different treatments with earthworms (vermicompost) over the treatments without earthworms (compost) (Fig. 1a). However, the change of pH in vermicompost over compost between treatments and between earthworm species did not differ significantly ($P > 0.05$). A maximum of 7.56% reduction in pH was observed in T4, followed by T8 (6.96%). The pH in compost and vermicompost ranged from 7.33 to 7.68 and 7.14–7.32 respectively. The reduction of pH during vermicomposting is a common feature as the decomposition process results in the production of organic acids. In general, the aerobic degradation of cellulosic substances results in the production of many organic acids and intermediates which lead to the shift in pH of vermicompost [25, 41]. A similar decrease in pH during vermicomposting of pressmud and seaweeds using *P. ceylanensis* and *P. excavatus* respectively have been reported [27, 28]. The reduction in pH of vermicompost than compost indicates that the decomposition of organic materials assisted with the mineralization process was effectively carried out by earthworms.

The percentage increase of EC in vermicompost over compost was ranged between 5.91 and 16.73% and the increment was significant at $P < 0.05$ (Fig. 1b). The EC in vermicompost obtained using *P. ceylanensis* was in the order of 2.45 > 2.39 > 2.11 > 1.92 dS/m respectively in T4 > T1 > T3 > T2, while it was 2.09 > 2.04 > 1.91 > 1.63 dS/cm, respectively in compost recovered from T1 > T4 > T3 > T2. The increase of EC in vermicompost over compost was significant ($P < 0.05$) in all the treatments and the difference of EC in respective treatments between *P. ceylanensis* and *P. excavatus* did not differ significantly ($P > 0.05$). The increase of EC in vermicompost is mainly attributed to the loss of weight of organic matter and the release of mineral salts like phosphate and potassium [23].

Organic Matter Content and Total Organic Carbon

The OMC in compost of T1, T2, T3, and T4 was 51.79 ± 3.25, 55.39 ± 3.82, 60.82 ± 4.57 and 54.08 ± 3.56%, respectively, while it was 44.41 ± 3.23, 45.63 ± 3.81, 51.12 ± 4.05 and 41.07 ± 2.98% respectively in vermicompost of T1, T2, T3 and T4 recovered from the treatments with *P. ceylanensis*. The OMC was 46.72 ± 2.45, 50.43 ± 3.07, 54.82 ± 3.69 and 43.10 ± 2.32% in the vermicompost recovered from the treatments with *P. excavatus* (Fig. 1c). The percentage decrease of OMC in vermicompost over compost ranged between 9.83 and 31.67%, and the decrease was significantly higher in all the treatments ($P < 0.05$) and the decrease in OMC content in vermicompost and compost between *P. ceylanensis* and *P. excavatus* also differ significantly. Similar to that of OMC, TOC also showed a

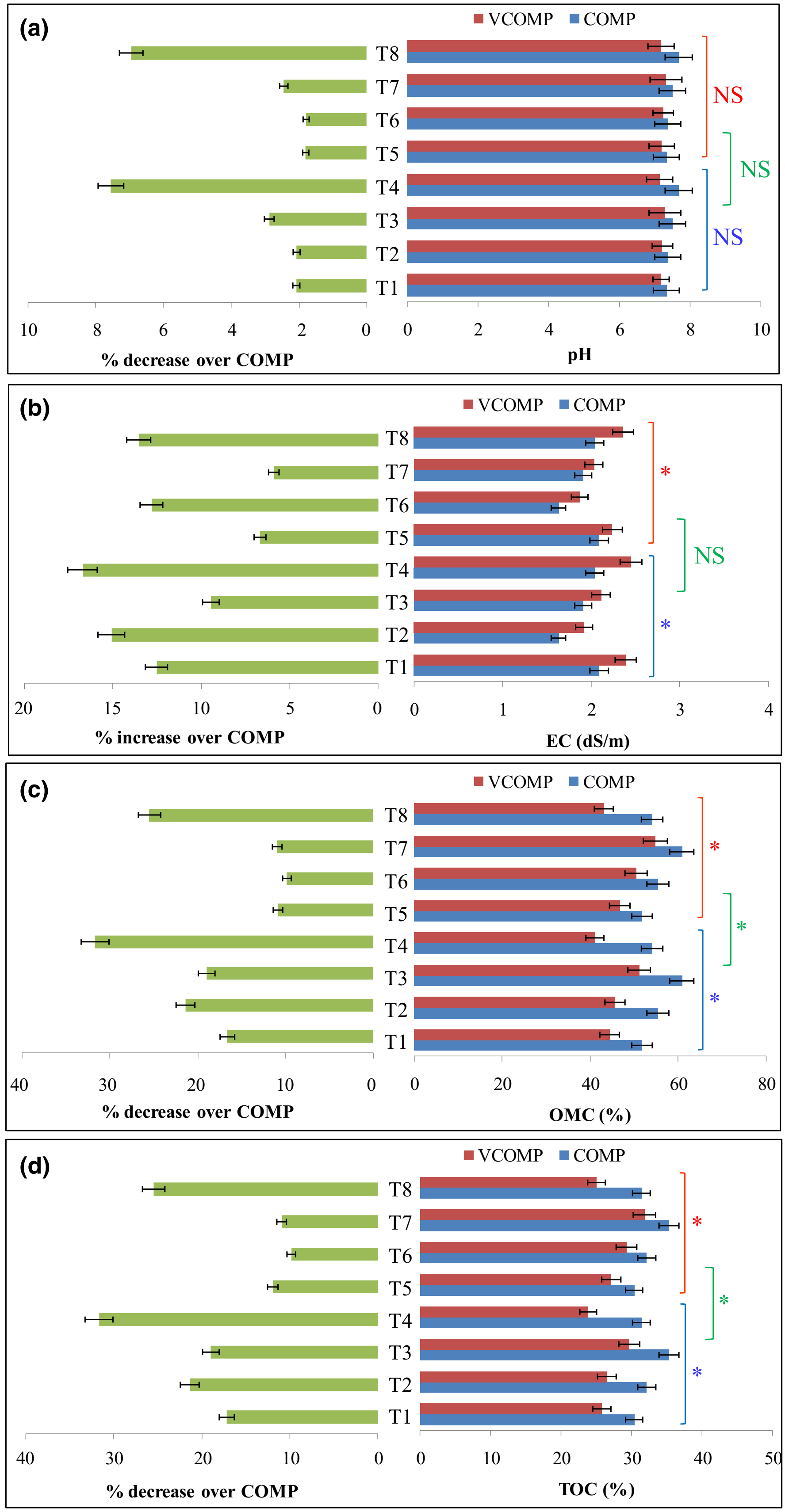


Fig. 1 **a** pH, **b** EC, **c** OMC and **d** TOC in different treatments after 50 days with and without earthworms. The data represent the mean ± standard deviation (error bars) of three replicates. '*' and 'NS' in P-values indicate significant and non-significant difference at P < 0.05 by Two-way ANOVA, respectively. *COMP* compost recovered from the treatments without earthworms, *VCOMP* vermicompost recovered from the treatments with earthworms

reduction. The TOC content in compost and vermicompost ranged from 30.34 to 35.28% and 23.82–31.80% respectively (Fig. 1d). A maximum of 31.69% reduction of TOC was observed for vermicompost recovered from the treatments with *P. ceylanensis* over the compost in T4, followed by T2 (21.38%) > T3 (18.98) > T1 (17.17%). The TOC in vermicompost recovered from the treatments with *P. excavatus* over compost showed a maximum decrease in T4 (25.48%) followed by T1 (11.95%), T3 (10.94%) and T2 (9.84%). The reduction of TOC in vermicompost over compost respective treatments differ significantly by ANOVA ($P < 0.05$). The difference in TOC contents between treatments with *P. ceylanensis* and *P. excavatus* showed a significant difference ($P < 0.05$), where the percentage decrease was higher for the treatments with *P. ceylanensis*. The rapid degradation of organic materials during vermicomposting as respiratory activity of earthworms and microorganisms and the assimilation of carbon by the earthworms might have caused the reduction in TOC and OMC in vermicompost [42–44]. The present study clearly demonstrated that the assistance of earthworm action is important for quick degradation of organics in vermicompost which is significantly slower in compost produced without the earthworms. The enhanced degradation of organic substrates by the earthworms results in mineralization of nutrients, thereby help in recovering the nutrients.

Total Kjeldahl Nitrogen, Phosphorus, Potassium Contents

The major plant nutrients, TKN, TP and TK mineralization in composting and vermicomposting is much needed as the quality of the final composts relies on these nutrients. Table 3 shows the TKN, TP and TK contents of compost and vermicompost in different treatments over 50 days. The TKN, TP and TK contents in vermicompost of all the treatments were significantly higher than that of the compost which clearly shows that the activity of earthworms in organic materials favored the increase of major plant nutrients. A maximum of 1.95% TKN was recorded in T8 among the treatments with *P. excavatus*, followed by 1.85% in T4 vermicompost. The percentage increase of TKN ranged between 22.93 (T6) and 37.50 (T3) and the percentage change in TKN from compost to vermicompost was significant at $P < 0.05$ (Fig. 2a). Also, the variation of TKN between treatments (T1-T4) with *P. ceylanensis*, and between treatments (T5-T8) with *P. excavatus* was statistically significant ($P < 0.05$). However, the percentage of TKN in the vermicompost between *P. ceylanensis* and *P. excavatus* treatments did not differ significantly at $P < 0.05$. Consequently, the change in TKN followed uniformity in the treatments with *P. ceylanensis* and *P. excavatus*. It has been reported that 1.6–1.7 times increase in TKN has resulted in the earthworm inoculated beddings in comparison to control beddings without earthworms [22]. Khwairakpam and Bhargava [23] also reported 1.4–1.7 times and 1.7–2.1

Table 3 TKN, TP, and TK contents in different treatments after 50 days with and without earthworms

Treatments	TKN (%)		TP (%)		TK (%)	
	COMP	VCOMP	COMP	VCOMP	COMP	VCOMP
T1	0.93 ± 0.05	1.35 ± 0.06	0.50 ± 0.03	0.71 ± 0.04	0.68 ± 0.04	0.87 ± 0.05
T2	0.84 ± 0.04	1.15 ± 0.05	0.46 ± 0.03	0.57 ± 0.02	0.57 ± 0.03	0.71 ± 0.04
T3	0.80 ± 0.03	1.28 ± 0.04	0.44 ± 0.02	0.52 ± 0.02	0.62 ± 0.04	0.74 ± 0.04
T4	1.28 ± 0.06	1.85 ± 0.07	0.68 ± 0.04	0.84 ± 0.04	0.90 ± 0.05	1.13 ± 0.05
ANOVA	*F-value*	*P value*	*F-value*	*P-value*	*F-value*	*P-value*
Between composts[a]	66.563	0.0039*	24.00	0.0163*	46.865	0.0064*
Between treatments	22.807	0.0144*	19.1225	0.0185*	45.851	0.0053*
T5	0.93 ± 0.05	1.23 ± 0.03	0.50 ± 0.03	0.65 ± 0.03	0.68 ± 0.04	0.81 ± 0.04
T6	0.84 ± 0.04	1.09 ± 0.02	0.46 ± 0.03	0.53 ± 0.01	0.57 ± 0.03	0.65 ± 0.03
T7	0.80 ± 0.03	1.16 ± 0.04	0.44 ± 0.02	0.49 ± 0.01	0.62 ± 0.04	0.69 ± 0.03
T8	1.28 ± 0.06	1.95 ± 0.07	0.68 ± 0.04	0.77 ± 0.04	0.90 ± 0.05	1.04 ± 0.04
ANOVA	*F-value*	*P-value*	*F-value*	*P-value*	*F-value*	*P-value*
Between composts[a]	17.515	0.0249*	17.357	0.0252*	35.757	0.0094*
Between treatments	10.621	0.0417*	29.00	0.0102*	83.189	0.0022*
Between species	0.926	0.4069[NS]	30.00	0.0119*	56.333	0.0049*

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

[a]Refers to COMP and VCOMP; *COMP* compost recovered from the treatments without earthworms; *VCOMP* vermicompost recovered from the treatments with earthworms

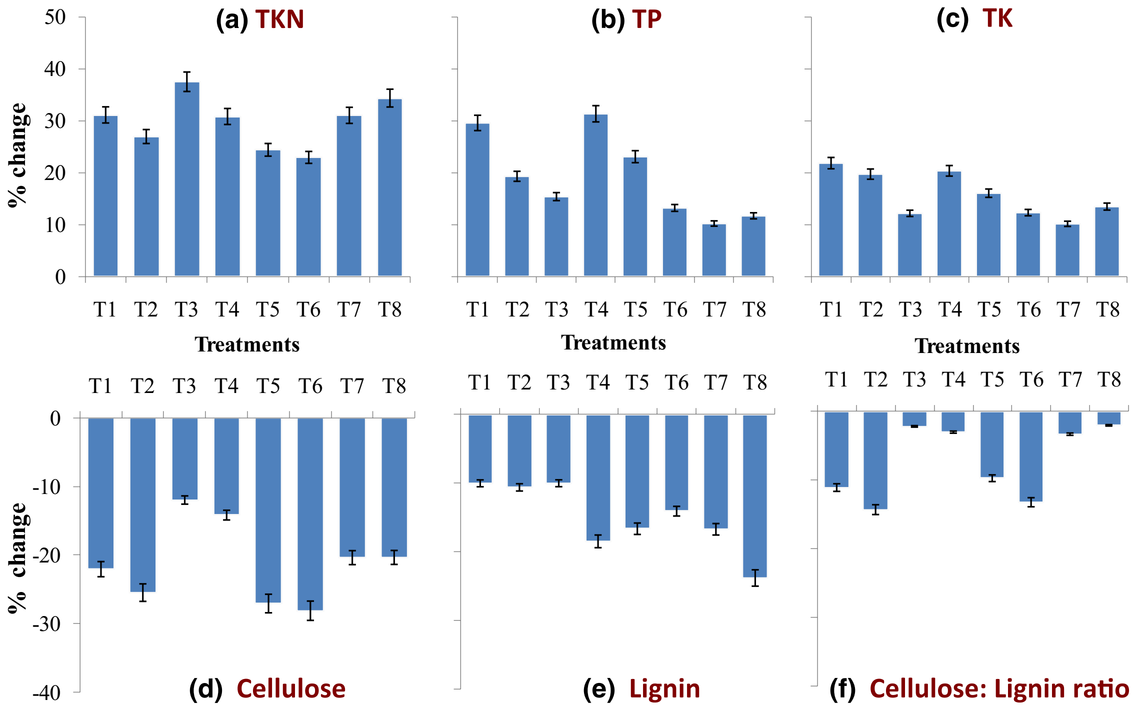


Fig. 2 Percentage change of **a** TKN, **b** TP, **c** TK, **d** Cellulose, **e** Lignin, and **f** Cellulose/Lignin ratio of vermicompost over compost in different treatments after 50 days. The data represent the mean ± standard deviation of three replicates. *COMP* compost recovered from the treatments without earthworms, *VCOMP* vermicompost recovered from the treatments with earthworms

times higher TKN in final materials than in the initial substrate during monoculture and polyculture vermicomposting respectively. The earthworms elevate the TKN levels in the vermicompost through the addition of excretory products, mucus, body fluid, enzymes and partly by decaying tissues of dead worms, also by the decrease of TOC in the substrate and mineralization of organic materials by the combined action of earthworms and microbes [7, 45]. The studies with municipal solid waste vermicomposting using *P. ceylanensis* [29] and seaweed vermicomposting using *P. excavatus* [27, 46] also revealed the same.

The TP contents showed a significant enhancement in vermicompost than compost and the incremental change in TP (10.20–31.33%) between treatments and between compost and vermicompost was found to be significant ($P < 0.05$) (Table 3; Fig. 2b). Unlike TKN, the TP change in vermicompost recovered from treatments with *P. ceylanensis* was higher than that of the treatments with *P. excavatus* which was statistically significant at $P < 0.05$. The TP content ranged between 0.52 and 0.84% in treatments with *P. ceylanensis*, whereas it was in the range of 0.49–0.77% for *P. excavatus*. These results indicate that the TP mineralization was much favored by *P. ceylanensis* compared to *P. excavatus*. The TK content in vermicompost (0.71–1.13%) vermicomposted by *P. ceylanensis* was significantly higher than that of the compost (0.57–0.90%). Similar results were also recorded for the vermicompost in the treatments with *P. excavatus* (Table 3). The percentage change in TK also followed the same trend (Fig. 2c) as to that of TP. The TK content in vermicompost recovered from treatments with *P. ceylanensis* was significantly higher than that of vermicompost with *P. excavatus*. The TK and TP contents of 9.9 and 25.7 g/kg, respectively were reported in 45 days vermicomposted pressmud substrates in polyculture systems [23]. An elevated level of TK and TP was observed in municipal solid waste after vermicomposting with *P. ceylanensis* [29]. The increased level of TP during vermicomposting might probably due to earthworm-gut derived phosphatase activity and also increased microbial activity in the vermicasts. The presence of a large number of microorganisms in the gut of earthworm might play an important role in increasing TP and TK contents in the process of vermicomposting [28, 45]. In terms of TK and TP increment in the present study, *P. ceylanensis* was found to be efficient than *P. excavatus* in all the treatments.

Cellulose, Lignin and Cellulose/Lignin Ratio

The cellulose, lignin, and cellulose/lignin ratio showed a decrease in vermicompost of all the treatments compared to respective compost, and the difference in the decrease was significant for cellulose and lignin ($P < 0.05$), while it was not significant for cellulose/lignin ratio ($P > 0.05$) (Table 4). T1, T2, T3, and T4 showed 18.02, 19.00, 21.50 and 17.95% in vermicompost with a percentage decrease of 22.08, 25.50, 12.00 and 14.20 respectively (Fig. 2d). The percent decrease of cellulose in vermicompost recovered from T5, T6, T7, and T8 was 27.09, 28.14, 20.40 and 20.37%, respectively, where the values were significantly higher than that of the treatments, T1, T2, T3, and T4. However, the decrease of cellulose between treatments in both *P. ceylanensis* and *P. excavatus* vermicompost did not differ significantly ($P < 0.05$). Overall the lignin content in vermicompost ranged from 17.33 to 23.65%, which was significantly ($P < 0.05$) lower than that of compost (21.46–26.02%) (Fig. 2e). The cellulose/lignin ratio was found between 0.81 and 0.99 in vermicompost treatments with earthworms, while it ranged between 0.90 and 0.96 in compost treatments without earthworms. The change in the cellulose/lignin ratio was in the range of 2.19 to 14.33% (Fig. 2f). The change in the cellulose/lignin ratio showed a non-significant ($P > 0.05$) difference between compost and vermicompost treatments, and between *P. ceylanensis* and *P. excavatus*, indicating that the change of cellulose and lignin in treatments with earthworms is uniform. The significant difference of cellulose/lignin ratio at $P < 0.05$ between compost and vermicompost indicates that the action of earthworms played a major role in the decrease of cellulose and lignin. Parthasarathi et al. [47] while vermicomposting cashew leaf litter incorporated with animal dung using the earthworm *P. excavatus* reported that there was a significant decrease in cellulose, hemicelluloses, and lignin contents in worm-worked substrates over worm-unworked and initial substrates. A maximum reduction of 17.6 and 11.9% lignin and cellulose, respectively during vermicomposting of pressmud, ST and biomethanated distillery effluent combinations employing the earthworm *P. excavatus* was reported [48]. The reduction of lignocellulosic materials is due to the degradative activity of earthworms and microflora [29] and the reduction rate of lignin and cellulose recorded in the present study is higher than that of the above findings which might probably due to the nature of organic wastes and bulking material used.

Calcium and Sodium Contents

The Ca and Na contents in vermicompost were higher than in compost and the difference was significant ($P < 0.05$). However, the change of Ca and Na brought out by *P. ceylanensis* and *P. excavatus* in respective treatments were insignificant ($P > 0.05$) (Fig. 3). This shows that the incremental shift followed a similar pattern in both the earthworm species (*P. ceylanensis* and *P. excavatus*). The change of Ca ranged between 12.50 and 16.16% for *P. ceylanensis* whereas it was in the range of 5.61–9.19% for *P. excavatus*. The increase of Ca and Na perhaps is linked with organic matter decomposition and assimilation rate which is dependent on earthworm species. This is apparently clear in the case of pressmud vermicomposting with *P. ceylanensis* where the results obtained showed 34.92 and 30.12% increase of Ca and Na, respectively [28].

Table 4 The contents of cellulose, lignin, and cellulose/lignin ratio in different treatments after 50 days with and without earthworms

Treatments	Cellulose (%)		Lignin (%)		Cellulose/lignin ratio	
	COMP	VCOMP	COMP	VCOMP	COMP	VCOMP
T1	22.00 ± 1.00	18.02 ± 1.01	24.35 ± 1.22	22.13 ± 1.47	0.90 ± 0.06	0.81 ± 0.03
T2	23.86 ± 1.30	19.00 ± 1.41	25.50 ± 1.79	23.05 ± 1.30	0.94 ± 0.05	0.82 ± 0.03
T3	24.08 ± 1.56	21.50 ± 1.33	26.02 ± 2.00	23.65 ± 1.52	0.93 ± 0.06	0.91 ± 0.04
T4	20.50 ± 1.33	17.95 ± 1.59	21.46 ± 1.48	18.11 ± 1.25	0.96 ± 0.06	0.99 ± 0.05
ANOVA	*F-value*	*P-value*	*F-value*	*P-value*	*F-value*	*P-value*
Between composts[a]	38.230	0.0085*	103.498	0.0020*	2.174	0.2368^{NS}
Between treatments	7.779	0.0629^{NS}	68.6533	0.0624^{NS}	2.3841	0.2471^{NS}
T5	22.00 ± 1.00	17.31 ± 1.22	24.35 ± 1.22	21.20 ± 1.16	0.90 ± 0.06	0.82 ± 0.04
T6	23.86 ± 1.30	18.62 ± 1.10	25.50 ± 1.79	22.35 ± 1.89	0.94 ± 0.05	0.83 ± 0.04
T7	24.08 ± 1.56	20.00 ± 1.00	26.02 ± 2.00	22.29 ± 2.00	0.93 ± 0.06	0.90 ± 0.05
T8	20.50 ± 1.33	17.03 ± 1.25	21.46 ± 1.48	17.33 ± 1.09	0.96 ± 0.06	0.98 ± 0.05
ANOVA	*F-value*	*P-value*	*F-value*	*P-value*	*F-value*	*P-value*
Between composts[a]	130.696	0.0014*	218.447	0.0007*	3.061	0.1785^{NS}
Between treatments	15.104	0.0558^{NS}	74.083	0.0622^{NS}	2.745	0.2145^{NS}
Between species	13.897	0.0636^{NS}	41.050	0.0717^{NS}	1.041	0.9999^{NS}

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

[a]Refers to COMP and VCOMP; *COMP* compost recovered from the treatments without earthworms, *VCOMP* vermicompost recovered from the treatments with earthworms

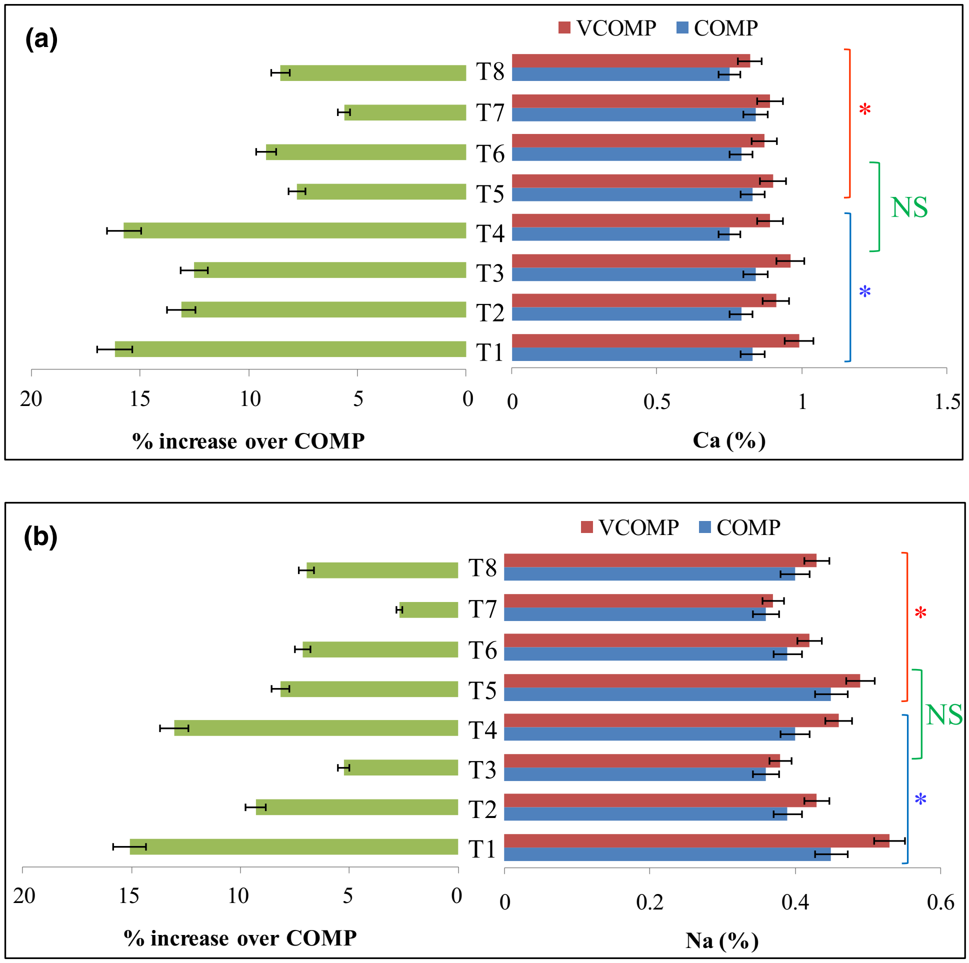


Fig. 3 **a** Ca and **b** Na contents in different treatments after 50 days with and without earthworms. The data represent the mean ± standard deviation (error bars) of three replicates. '*' and 'NS' in P-values indicate significant and non-significant difference at P < 0.05 by Two-way ANOVA, respectively. *COMP* compost recovered from the treatments without earthworms, *VCOMP* vermicompost recovered from the treatments with earthworms

C/N Ratio and C/P Ratio

C/N ratio is considered as an important parameter during composting and in composts. Along with the C/P ratio, the C/N ratio is considered as the key factor to determine the maturity of composts and their suitability to use for agricultural crops. In the present study, C/N and C/P ratios decreased tremendously in vermicompost than in compost in all the treatments (Table 5). A range of 66.15–90.41% reduction in C/N ratio for the treatments with *P. ceylanensis* and a range of 42.56–91.18% C/N ratio for *P. excavatus* was recorded, where the decrease between compost and vermicompost, and between treatments was significantly different (P < 0.05). The lowest C/N ratio of 12.82 was observed in T8 followed by T4 (12.88) > T1 (19.08) > T5 (22.03) > T2 (23.02) > T3 (23.16) > T6 (26.83) > T7 (27.41). The reduction in C/N ratio in treatments with *P. ceylanensis* and *P. excavatus* showed a statistically significant difference at P < 0.05 where the percent reduction was higher with *P. ceylanensis*. The C/P ratio also showed a similar trend of decrease to that of the C/N ratio where both the parameters were dependent on the reduction of TOC. As observed from Table 5, the percentage change of C/P was maximum in T1 (67.25%) followed by T4, T2, and T3 for *P. ceylanensis*, and C/P was maximum in T5 (45.55%) followed by T8, T6, and T7. The difference in the reduction of C/P between treatments, compost, and vermicompost, and between treatments with *P. ceylanensis* and *P. excavatus* was statistically significant at P < 0.05. The reduction of organic matter content and loss of CO_2 by respiratory activities of earthworms and microorganisms, and the mineralization of nutrients especially nitrogen are the influencing factors responsible for the narrow downtrend of C/N and C/P ratios which has been well established in previous studies with different organic wastes [10, 45]. More likely, the C/N ratio of the feedstock materials is decisive for nutrient enhancement and the activity of earthworms [49]. The results of the present study clearly indicate that the reduction of C/N and C/P ratios are dependent on the type of organic waste used as feedstock and the kind of earthworm species employed for vermiconversion.

Table 5 C/N and C/P ratios in different treatments after 50 days with and without earthworms

Treatments	C/N ratio		% decrease over COMP	C/P ratio		% decrease over COMP
	COMP	VCOMP		COMP	VCOMP	
T1	32.62 ± 1.90	19.08 ± 0.92	70.96	60.68 ± 4.70	36.28 ± 2.33	67.25
T2	38.25 ± 2.33	23.02 ± 1.43	66.15	69.85 ± 5.04	46.44 ± 3.19	50.40
T3	44.10 ± 2.95	23.16 ± 1.38	90.41	80.18 ± 5.91	57.02 ± 4.25	40.61
T4	24.51 ± 1.12	12.88 ± 0.50	90.29	46.13 ± 2.56	28.36 ± 1.46	62.65
ANOVA	*F-value*	*P-value*		*F-value*	*P-value*	
Between composts[a]	58.334	0.0047*		219.969	0.0007*	
Between treatments	10.526	0.0422 *		80.213	0.0023*	
T5	32.62 ± 1.90	22.03 ± 1.30	48.17	60.68 ± 4.70	41.69 ± 2.73	45.55
T6	38.25 ± 2.33	26.83 ± 1.92	42.56	69.85 ± 5.04	55.19 ± 3.61	26.56
T7	44.10 ± 2.95	27.41 ± 1.99	60.89	80.18 ± 5.91	64.90 ± 4.25	23.54
T8	24.51 ± 1.12	12.82 ± 0.85	91.18	46.13 ± 2.56	32.47 ± 1.93	42.06
ANOVA	*F-value*	*P-value*		*F-value*	*P-value*	
Between composts[a]	82.839	0.0028*		181.001	0.0009*	
Between treatments	29.066	0.0102*		152.177	0.0008*	
Between species	7.952	0.0468*		36.999	0.0089*	

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

[a]Refers to COMP and VCOMP; *COMP* compost recovered from the treatments without earthworms; *VCOMP* vermicompost recovered from the treatments with earthworms

Total Microbial Population

The total microbial population (bacteria, fungi, and actinobacteria) in compost and vermicompost of all the treatments increased from initial levels (Fig. 4). Though the compost of all treatments showed an increased microbial population, worm-worked vermicompost showed highly a significant difference. The range of the total bacterial population in vermicompost recovered from the treatments with *P. ceylanensis* was 8.05–8.18 $\log_{10}$ CFU g^{-1}, while it was 8.01–8.15 $\log_{10}$ CFU g^{-1} in vermicompost recovered from the treatments with *P. excavatus* (Fig. 4a). The overall range of total fungal and actinobacterial population was found to be in the range of 4.84–4.96 $\log_{10}$ CFU g^{-1} and 5.92–6.17 $\log_{10}$ CFU g^{-1} respectively (Fig. 4b and c). The difference in the increase of bacterial, fungal and actinobacterial population between compost and vermicompost, between treatments, and between the treatments with *P. ceylanensis* and *P. excavatus* was significant at $P < 0.05$ by two-way ANOVA. The increase of total microbial population in vermicompost of all the treatments in the present study is due to the activity of the earthworms, *P. ceylanensis* and *P. excavatus* which varied with reference to the type of organic wastes and earthworm species used. It has been reported that the abundance of the dominant microbiota varied according to earthworm density, indicating that earthworms are the drivers of the change in the microbial composition [50]. Accordingly, the vermicomposting studies with *P. ceylanensis* and *P. excavatus,* irrespective of organic substrates, influenced the increase of total bacterial, fungal and actinobacterial population [26, 27, 29]. Interestingly, the use of earthworm consortia tremendously increased the microbial population including N-fixing and P-solubilizing bacteria [25].

Seed Germination Assay

The seed germination and initial growth phase of crop plants in response to vermicompost application are dose-dependent and species-specific [51]. In the present study, the germination percentage of black gram and sorghum in worm un-worked compost and worm worked vermicompost of four different organic residues by two species of earthworms clearly showed that the vermicompost had higher germination percentage than composts in 50 days (Fig. 5a and b). The germination percentage of black gram ranged between 71 and 90 in composts where the maximum was observed in the compost obtained from farmyard manure (T4 and T8). The germination percentage in vermicomposts of T1-T4 by *P. ceylanensis* and T5-T8 by *P. excavatus* were higher than the respective composts which ranged from 96 to 98% for black gram (Fig. 5a) and 98–100% for sorghum (Fig. 5b). The germination percentage between treatments did not differ significantly ($P > 0.05$) while statistically significant difference ($P < 0.05$) was observed between worm un-worked composts and vermicomposts of all the treatments. Among the worm un-worked composts T4 and T8 showed 90% germination of BG and 92% germination of sorghum.

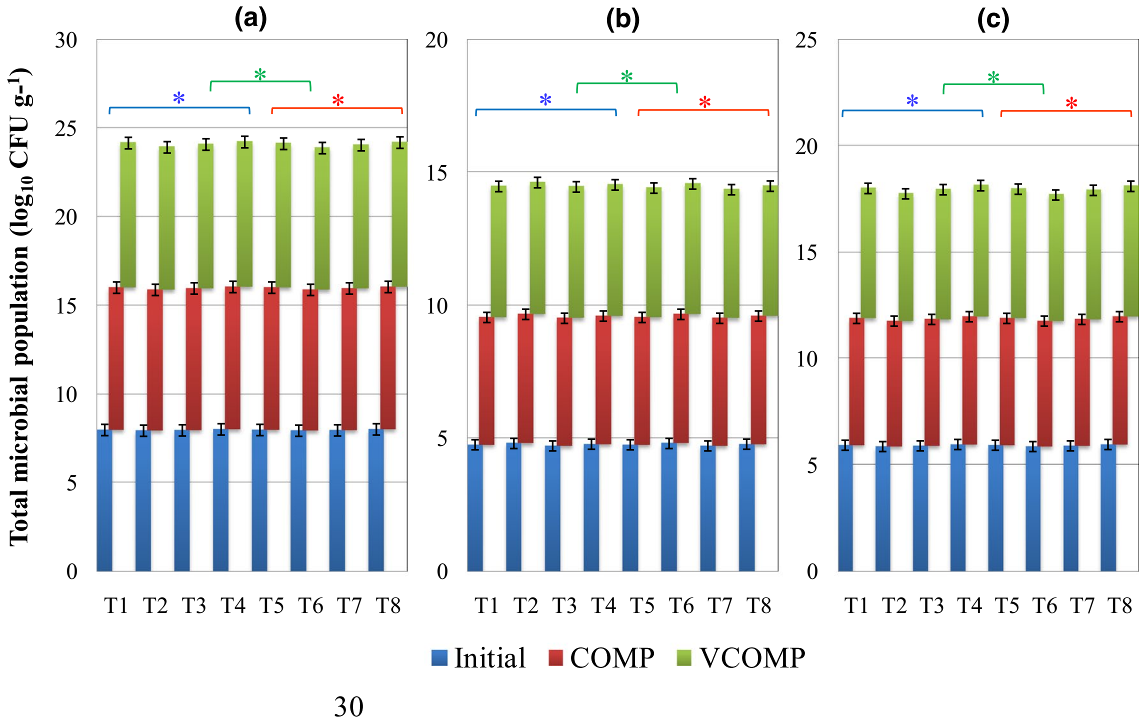


Fig. 4 Initial and final **a** total bacterial, **b** total fungal and **c** total actinobacterial population in different treatments. Values expressed are $\log_{10}$ transformed mean ± standard deviation (error bars) of three replicates. '*' and 'NS' in P-values indicate significant and non-significant difference at $P < 0.05$ by Two-way ANOVA, respectively. *COMP* compost recovered from the treatments without earthworms; *VCOMP* vermicompost recovered from the treatments with earthworms

The germination index for black gram and sorghum in vermicomposts recovered from T1-T4 (*P. ceylanensis*) ranged 338–357 and 346–365 respectively, while it was 253–308 and 224–311 in worm un-worked composts (Fig. 5c and d). Among the treatments T5-T8, the vermicomposts had 343–362 and 352–369 germination indices respectively for black gram and sorghum. The higher germination index values in vermicomposts than composts indicates that the vermicompost is less phytotoxic and mature enough to be used as organic amendments to agronomic crops, and worm un-worked composts require some more time to attain maturity. As observed from Fig. 5c and d, the higher germination index of black gram and sorghum in vermicomposts obtained from *P. ceylanensis* (T1-T4) and *P. excavatus* (T5-T8) worked substrates was significantly different ($P < 0.05$) from that of the worm un-worked composts of the respective treatments. However, the variation in germination index values between treatments did not differ significantly ($P > 0.05$). The germination percentage and germination index are the parameters used to signify the maturity and phytotoxicity of organic composts and soils. It is evidently seen from the present study findings that the vermicomposts obtained from T1-T4 with *P. ceylanensis* and T5-T8 with *P. excavatus* were mature and less phytotoxic due to the fact that the vermicomposts produced by both *P. ceylanensis* and *P. excavatus* possessed suitable characteristics to be used as organic soil amendment for agricultural crop productivity. It has been reported that the vermicompost and its extractives enhance seed germination due to the presence of plant growth hormones and humic acids [52]. The improved germination by vermicompost has been observed in crop plants like maritime pine, tomato, lettuce, chilly, ladies finger, and cucumber [51–53]. The seed germination assay is best suited method to assess the maturity and phytotoxicity of vermicomposts of different origins [38, 54].

Conclusions

Based on the vermicomposting characteristics, *P. ceylanensis* was found to be an equally efficient converter of organic solid wastes in comparison with *P. excavatus.* Both the worms followed the parallel trend of changes in the vermibed substrates with variation in the efficiency. The pH, TOC, OMC, lignin, cellulose, C/N ratio, C/P ratio in vermicompost recovered from the treatments with *P. ceylanensis* and *P. excavatus* showed reduction over compost, while TKN, TP, TK, Ca, Na and microbial population showed an increase. In terms of significant change in the parameters of all the four kinds of organic

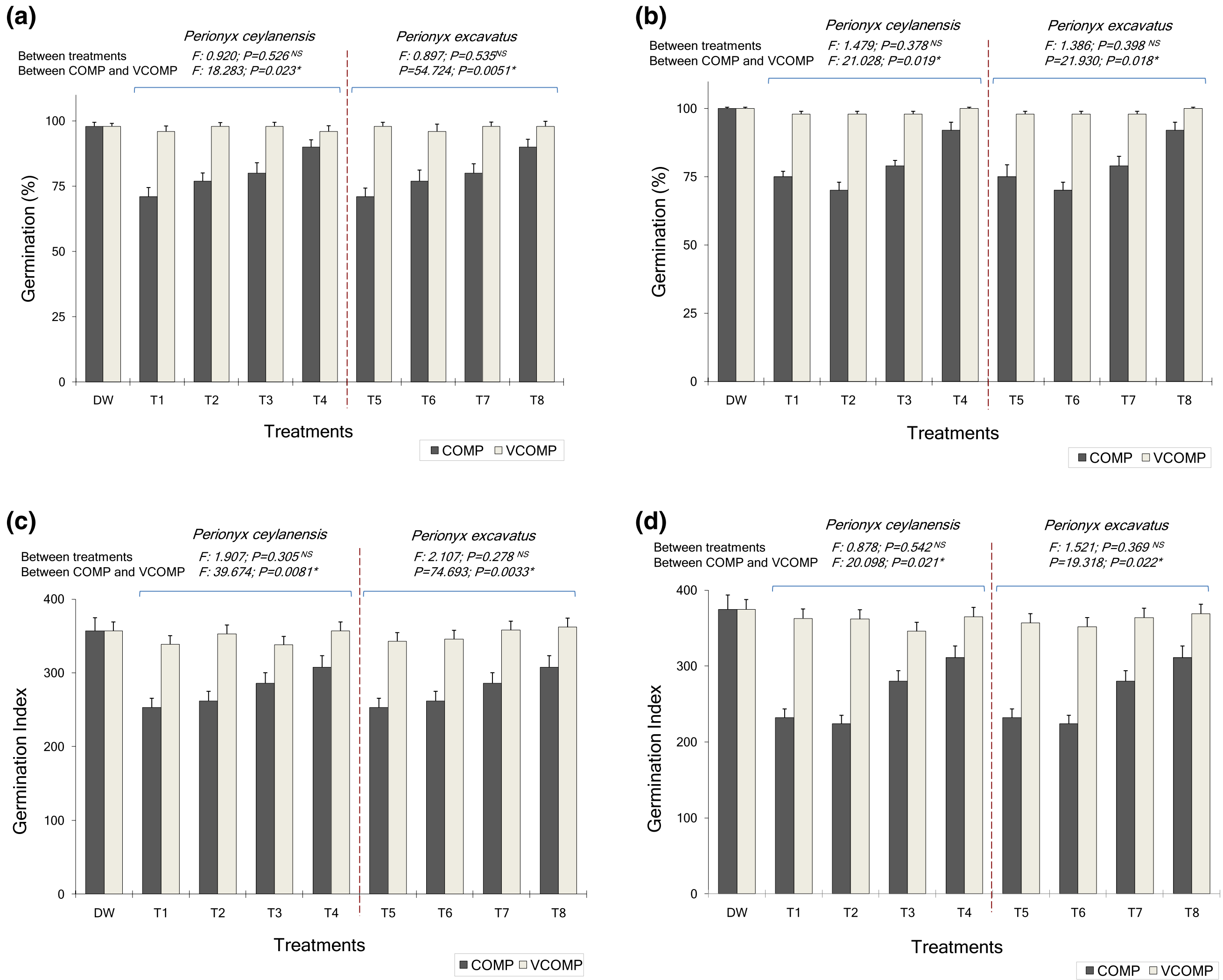


Fig. 5 Germination percentage and germination index of black gram and sorghum in composts of different organic residues obtained with and without earthworms. **a** and **b** Germination percentage of black gram and sorghum respectively; **c** and **d** Germination index of black gram and sorghum respectively; *DW* distilled water;. '*' and 'NS' in P-values indicate significant and non-significant difference at $P < 0.05$ by Two-way ANOVA, respectively. *COMP* compost recovered from the treatments without earthworms; *VCOMP* vermicompost recovered from the treatments with earthworms

substrates, MSW, ST, PS, and FYM during vermiremediation, *P. ceylanensis* was comparatively efficient in nature and can be utilized for organic waste management on par with *P. excavatus*. Both the earthworm species are suitable for nutrient recovery from organic solid residues in tropical climate. The vermicomposting characteristics of *P. ceylanensis* show the prospective utility of the species for organic waste recycling in terms of nutrient-rich vermicompost production. Also, the addition of cowdung to the organic substrates in 1:1 ratio is suitable for effective vermiremediation. The vermifertilizer produced from four different organic substrates had positive impact on seed germination of black gram and sorghum, hence can be suitably used as organic soil amendment for sustainable crop production. However, the change of heavy metals and their quantity in organic solid wastes and vermicompost offer further insight into the study for effective utilization of *P. ceylanensis* and *P. excavatus* in vermi-management of organic solid wastes.

Acknowledgements The first author, Dr. M. Biruntha, thankfully acknowledges RUSA Scheme Phase 2.0 grant [F-24-51/2014–U, Policy (TNMulti-Gen), Dept of Edn., Govt. of India, dated 09.10.2018] for supporting the work. The authors duly acknowledge Dr. (Mrs.) Thilagavathy Daniel, Professor (Retd.), Department of Biology, The Gandhigram Rural Institute (Deemed University), Gandhigram, Tamil Nadu, India for going through the manuscript and critical comments.

Compliance with Ethical Standards

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

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