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ORIGINAL ARTICLE

Production and analytical profiling of *Azolla*-augmented vermicompost derived from banana leaf biomass

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Abstract

In southern India, the widespread cultivation of bananas results in substantial agrowaste, particularly banana leaves, posing significant disposal challenges. Vermicomposting these banana leaves offers an efficient solution, transforming organic waste into nutrient-rich fertilizer and promoting sustainable solid waste management. This study examined 16 compositions of banana leaf biomass (BLB) and cow dung (CD) at ratios of 1:1, 2:1, 3:1, and 4:1, with *Azolla* amendments of 0%, 10%, 20%, and 30%. Vermicomposting was performed for 75 days using *Eudrilus eugeniae.* Growth and biomass production of *E. eugeniae* were high in the compositions amended with *Azolla* in a 1:1 ratio of BLB: CD with 30% *Azolla*. Whereas, peak changes in electrical conductivity (1.64 dS/m), organic carbon (29.75%), total Kjeldahl nitrogen (2.27%), and total phosphorus (1.17%) were observed in the vermicompost of 1:1 BLB: CD substrate enriched with 20% *Azolla*, surpassing the nutrient levels recorded in all other treatments. A lower C/N ratio (13.12) and C/P ratio (24.37) was observed in BLB: CD (1:1) + 30% *Azolla* and BLB: CD (3:1) + 30% *Azolla*, respectively. The introduction of a 20% −30% *Azolla* supplement to a banana leaf biomass and cow dung vermicompost formulation facilitated substantial enhancements, leading to an amplified nutrient density and an optimized composting procedure.

Keywords Vermicomposting · Banana leaf waste · *Azolla* amendment · *Eudrilus eugeniae*

Introduction

Bananas are a vital component of the global fruit basket, being the oldest tropical fruit and the fourth most traded commodity worldwide. They serve not only as a staple food and cash crop but also as a primary source of food energy for millions of people who rely on bananas and plantains. The banana is an affordable, nutritious fruit rich in carbohydrates, making it accessible to people from all walks of life [1]. Over the past five years, banana production has increased by 32%, making it an increasingly popular crop among farmers. Tamil Nadu ranks as the fifth-largest state in banana production [1]. In India, banana leaves are utilized and discarded through a variety of methods, such as in serving food, wrapping, rituals, mulching, as biodegradable plates, and for decorative. Although banana leaves serve

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numerous purposes, they eventually become waste, which can be repurposed via composting to produce a fertilizer rich in nutrients. Banana leaves as fertilizer also improve soil texture, water holding capacity and promote plant growth [2]. In most states of India, banana waste biomass, particularly the pseudostem, is treated as waste. Farmers spend approximately ₹8,000 to ₹10,000 per hectare solely on its disposal [3]. Managing banana residue effectively remains a challenge for farmers, but also presents significant opportunities for recycling and reuse in subsequent crops. The rising costs of chemical fertilizers, coupled with their negative impact on soil properties, have prompted the adoption of organic manures in crop cultivation [4].

Through vermicomposting, discarded banana foliage is transformed into nutrient-dense, granular compost, acting as a superior natural soil enhancer. This process enhances the compost with a variety of nutrients and beneficial components [5]. The process of vermicomposting using earthworms to decompose organic waste is experiencing significant advancements that are enhancing its efficiency and applications. There has been increased research into the types of organic waste that can be effectively processed by worms. Urban agriculture has a growing interest in the potential of vermicomposting. Urban farms and community gardens are adopting vermicomposting to enhance soil fertility and support sustainable food production. Vermicomposting not only converts waste into valuable compost but also helps mitigate methane emissions—a significant contributor to climate change. The combined activity of microbes and enzymes in the earthworm gut helps in breaking down complex pollutants and converting them into safe biofertilizers [6]. Essentially, earthworms are vital recyclers, optimizing the composting process and yielding superior compost. Through their ceaseless activity, they catalyse the rapid transformation of organic matter, meticulously refine the soil's structure, and generously endow the compost with a rich tapestry of essential nutrients. Vermicomposting can also be effectively applied in industries to stabilize and reduce the volume of heavy metal-containing by-products [7].

Studies highlight the versatility and efficiency of *Eudrilus eugeniae* in vermicomposting various organic wastes, including crop residues, cattle shed waste, and rice husk, resulting in the production of nutrient-enriched organic fertilizers suitable for sustainable agriculture [8, 9]. Studies have also explored its potential for bioremediating soils contaminated with heavy metals through composting [10]. *E. eugeniae*, commonly known as the African Night crawler, prefers slightly lower temperatures for composting than other earthworm species. Their composting efficiency is subject to specific environmental constraints: a temperature range of 20 °C to 30 °C (68 °F to 86 °F) and a developmental period of 40 to 60 days to reach sexual maturity. Studies have reported the efficiency of *E. eugeniae* in breaking down the complex cellulose and high lignin content in banana waste due to the presence of higher levels of cellulase and amylase when compared to other common species of earthworms used in vermicomposting [11]. The vermicompost produced by *E. eugeniae* exhibits improved physical properties, including increased porosity and aeration, which facilitate better water retention and root penetration in soil [12].

The use of cow dung is essential for enhancing earthworm activity and reproduction in vermicomposting. Previous studies demonstrated that cow manure creates a supportive habitat for earthworms, resulting in greater biomass accumulation and enhanced compost quality [13]. The research also reported notable declines in organic carbon and the carbon-to-nitrogen ratio, accompanied by a rise in total nitrogen content—indicators of efficient nutrient recovery. *Azolla* is used as an enriching factor in the vermicomposting of banana leaves. The study of *Azolla* as a biofertilizer stated that due to its specialized symbiotic alliance with the nitrogen-fixing Cyanobacteria, *Anabaena azollae*, the petite water fern, *Azolla*, constitutes an exceptionally abundant natural source of nitrogen [14]. *Azolla* contributes significantly to improving soil quality by enriching it with organic matter and facilitating the release of essential cations like magnesium, calcium, and sodium. According to [15], its incorporation can enhance total nitrogen levels, available phosphorus, and exchangeable potassium in the soil, thereby boosting nitrogen uptake in rice crops. Experiments reveal that integrated nitrogen management involving the use of urea, vermicompost, and *Azolla* significantly improved both macronutrient uptake and the micronutrient content of rice grains [16]. *Azolla* biomass decomposition in paddy fields can release a significant amount of nitrogen (75–80% of the total accumulated nitrogen) for plant uptake. It can be used as a ready source of nitrogen for different cultivation, mainly for rice under sub-tropical conditions [17]. *Azolla* is highly beneficial due to its rich nutrient profile, making it an excellent organic fertilizer and biofertilizer. In addition to organic matter, it also adds ions such as magnesium, calcium, and sodium to the soil [18]. *Azolla* has the potential to absorb elements like Fe, Zn, Cu, and Mn from the water in which it grows, and the decomposition of *Azolla* during vermicomposting releases these elements in a bioavailable form [19].

Another major nutritional composition of *Azolla* is the protein: 25–35% (dry weight basis), mineral content: 10–15%, and about 10% of amino acids, bioactive substances, and biopolymers [20]. Agrowaste alone sometimes remains recalcitrant to biodegradation in a compost vessel as they have a high carbon-to-nitrogen (C: N) ratio, low moisture content, and contains lignocellulosic compounds that are resistant to easy breakdown. The addition of cow dung acclimatizes the earthworm to initiate composting with cow dung and then initiates the vermicomposting of agro wastes [21]. The present study aims to develop an efficient vermicomposting process for banana leaf waste, a common but resistant organic material, by utilizing *E. eugeniae*. Cow dung and *Azolla* are incorporated as co-substrates to enhance the decomposition process. The research evaluates the influence of these combinations on *E. eugeniae* activity, including growth and reproduction. It conducts detailed physicochemical analyses of the resulting vermicompost—such as pH, organic carbon, nitrogen, phosphorus, and potassium levels—to assess its quality, thereby exploring its potential as an eco-friendly solution for organic waste management and sustainable agriculture.

Materials and methods

Collection and characterization of raw materials

Banana leaf biomass from *Musa x paradisiaca* L. (AAB group), commonly known as "Nendran" was collected from a banana plantation in Payyanur, Kannur, Kerala, India. After cleaning and removing organic debris and residues, the leaves were cut into sections measuring 2–3 cm were prepared for subsequent processing. Urine-free, fresh cow dung was collected from a local farm to improve composting. *Azolla pinnata* was collected from the College of Agriculture, Padannakad, Kannur. Mature clitellate earthworms (*E. eugeniae*) were collected from Sreevalsam Agri Farm in Pilathara, Kannur.

Table 1 Experimental protocol for vermicomposting analysis

Compositions	Azolla proportions	Experimental treatments
Compositions 1 (1:1)		
BLB + CD	Devoid of *Azolla*	C1A
BLB + CD	10%	C1B
BLB + CD	20%	C1C
BLB + CD	30%	C1D
Compositions 2 (2:1)		
BLB + CD	Devoid of *Azolla*	C2A
BLB + CD	10%	C2B
BLB + CD	20%	C2C
BLB + CD	30%	C2D
Compositions 3 (3:1)		
BLB + CD	Devoid of *Azolla*	C3A
BLB + CD	10%	C3B
BLB + CD	20%	C3C
BLB + CD	30%	C3D
Compositions 4 (4:1)		
BLB + CD	Devoid of *Azolla*	C4A
BLB + CD	10%	C4B
BLB + CD	20%	C4C
BLB + CD	30%	C4D

Primary processing of banana leaf biomass

Plastic trays were prepared with drainage holes in the bottom. The cut banana leaves were transferred into the trays and were watered intermittently, and excess water drained out through the pores. To control solar exposure and minimize water depletion, the trays were placed in a shaded location. The study was carried out at Payyanur, Kannur, Kerala, where the temperature during the experimental period ranged from a minimum of 24 °C to a maximum of 28 °C. The relative humidity remained between 89% and 91% reflecting the climatic conditions of the late monsoon.

The leaves were pre-composted for 30 days to make it suitable for consumption by *E. eugeniae*. Precomposting banana leaves for 30 days is crucial, and was carried out based on established techniques for lignin-rich plant wastes. It was reported that precomposting brings partial breakdown of lignin and cellulose of banana leaves, removes the volatile gases, and maintains a mesophilic condition. Precomposting provides a stabilized substrate for the optimal growth and decomposition by *E. eugeniae* [22, 23]. Table 1 outlines the composition of pre-composted banana leaf biomass amalgamated with cow manure and amended with *Azolla*.

Vermicomposting experiments

To evaluate the effects of vermicomposting various substrates of different combinations of banana leaf biomass (BLB), cow dung (CD), and *Azolla* (AZ) from (C1A to C4D) in 16 experimental grow bags. *Azolla* was shade-dried for 48 h before the addition into the substrate mix, as it maintains the moisture level and avoids rapid fermentation, resulting in a temperature hike in the bin. The shade-drying of *Azolla* in turn balances the C: N ratio in the vermibed [24].

Each grow bag had 2 kg of samples into which 25 randomly selected *E. eugeniae* of medium size were placed. *E. eugeniae* were utilized to decompose organic matter and produce vermicompost in a controlled environment. Composting bags were maintained at a consistent temperature of 25 ± 3 °C and left undisturbed to enhance the vermicomposting process. The grow bags were kept in a permanently shaded area (porch) with high natural ventilation. Additionally, the moisture content was monitored daily and maintained at 60–70% by sprinkling water, which provided evaporative cooling within the substrate. The temperature of the Vermibed was checked at regular intervals using a thermometer to ensure that the temperature remains at the mentioned optimal range. After the study, *E. eugeniae* counts, weights, and cocoon numbers were documented. The resulting vermicompost was dried, sieved, and stored for subsequent analysis and characterization.

Growth and reproductive potential of *E. eugeniae*

The growth of *E. eugeniae* was determined by measuring the biomass gain and final population during vermicomposting. The *E. eugeniae* biomass was weighed using a digital electronic balance, and the number of clitellate adult worms was counted after the study. After 75 days of vermicomposting, the final biomass and the total count of worms were recorded for each experimental treatment.

Physicochemical characterization and evaluation of vermicomposting

Standardized analytical protocols were utilized to ascertain the pH, electrical conductivity (EC), total organic carbon (TOC), major nutrient composition (total NPK), and trace element/heavy metal burden (Cu, Fe, Mn, Zn, Cd, Pb, Cr) within the produced vermicompost across varied formulations. Samples were collected at 15-day intervals (15, 30, 45, 60, and 75 days) and analysed for physicochemical properties. The pH was measured using a digital pH meter (Elico), and the electrical conductivity was measured using a conductivity bridge (Elico). Total organic carbon contents of the samples were analyzed according to the potassium dichromate oxidation method [25]. Total Kjeldahl nitrogen (TKN) was estimated with the Micro Kjeldahl method . Total phosphorus, potassium, and sodium, calcium, magnesium, zinc, copper and iron were analyzed using standard methods : Phosphorus and iron were estimated using the spectrophotometric method, while potassium and sodium were analyzed using the photometric method. Calcium and magnesium were estimated by the acid digestion method, while zinc and copper estimated by the Atomic Absorption Spectrophotometric method. Manganese content was estimated by spectrophotometric method [26].

Statistical analysis

Data for vermicompost characteristics and *E. eugeniae* growth are presented as the mean ± standard deviation of three replicates. To evaluate changes over time, physico-chemical parameters measured at 0, 15, 30, 45, 60, and 75 days were statistically analyzed using SPSS software. Initial and final values physico-chemical parameters, worm growth were biomass were compared at a 5% significance level using Duncan's Multiple Range Test (DMRT).

Results and discussion

The changes of pH, EC, TOC, TKN, TP, TK, C/N ratio, and C/P ratio from the initial (0 day) to final vermicompost (75 days) are given in Tables 2 and 3 along with the percent change of each parameter over control (Fig. 1). The pH changes in vermicompost, compared to control substrates, was minimal, with the final pH of 6.85 ± 0.43 in the 75th day sample of C3B (3:1 BLB: CD with 10% AZ), 6.87 ± 0.44in the 75th day sample of C3C (3:1 BLB: CD with 20% AZ). Figure 2(A-D) represents the pH in the samples. *E. eugeniae* are neutrophilic and prefer neutral pH. Nutrient availability depends on the pH of the medium [27]. As composting proceeds, the microbial action brings about the production of organic acids and carbon dioxide results in the lowering

Table 2 Initial physicochemical characteristics of banana leaf waste substrate combinations by vermicomposting with *E. eugeniae* (Refer Table 1 for treatment combinations). Values are mean±standard deviation

Treatment	pH	EC (dS/m)	TOC (%)	TKN (%)	TP (%)	TK%	*C/N* ratio	*C/P* ratio
C1A	7.79 ± 0.51	0.94 ± 0.06	39.46 ± 2.57	1.02 ± 0.12	0.73 ± 0.06	1.27 ± 0.11	38.69 ± 2.23	54.01 ± 3.28
C1B	7.23 ± 0.48	1.08 ± 0.07	40.67 ± 2.54	1.13 ± 0.10	0.74 ± 0.08	1.27 ± 0.14	35.99 ± 2.17	54.95 ± 3.17
C1C	7.43 ± 0.45	1.22 ± 0.08	42.92 ± 2.81	1.25 ± 0.15	0.74 ± 0.07	1.30 ± 0.13	34.36 ± 2.31	57.99 ± 3.42
C1D	7.35 ± 0.48	1.20 ± 0.08	44.11 ± 2.76	1.20 ± 0.11	0.72 ± 0.06	1.33 ± 0.15	36.78 ± 2.18	61.19 ± 3.67
C2A	7.58 ± 0.44	0.83 ± 0.06	42.62 ± 2.78	0.93 ± 0.10	0.80 ± 0.08	1.16 ± 0.11	45.74 ± 2.78	53.42 ± 3.56
C2B	7.25 ± 0.50	0.97 ± 0.06	44.38 ± 2.77	1.06 ± 0.08	0.80 ± 0.09	1.19 ± 0.14	41.68 ± 2.84	55.63 ± 3.67
C2C	7.22 ± 0.48	1.11 ± 0.08	46.33 ± 2.84	1.12 ± 0.09	0.82 ± 0.06	1.18 ± 0.13	17.97 ± 2.67	56.69 ± 3.42
C2D	6.87 ± 0.43	1.27 ± 0.09	47.92 ± 2.93	1.33 ± 0.11	0.83 ± 0.06	1.21 ± 0.15	36.08 ± 2.52	57.97 ± 3.53
C3A	7.72 ± 0.49	0.92 ± 0.08	39.86 ± 2.52	1.02 ± 0.11	0.72 ± 0.07	1.28 ± 0.13	39.09 ± 2.47	55.02 ± 3.48
C3B	7.17 ± 0.45	1.06 ± 0.07	41.08 ± 2.60	1.11 ± 0.11	0.73 ± 0.08	1.28 ± 0.13	37.11 ± 2.35	55.97 ± 3.54
C3C	7.37 ± 0.47	1.20 ± 0.08	43.36 ± 2.74	1.22 ± 0.13	0.73 ± 0.08	1.31 ± 0.14	35.43 ± 2.24	59.06 ± 3.74
C3D	7.29 ± 0.46	1.18 ± 0.07	44.56 ± 2.82	1.25 ± 0.13	0.71 ± 0.07	1.34 ± 0.14	35.71 ± 2.26	62.32 ± 3.95
C4A	7.52 ± 0.48	0.81 ± 0.05	43.05 ± 2.73	0.91 ± 0.09	0.79 ± 0.08	1.17 ± 0.12	47.16 ± 2.99	54.41 ± 3.44
C4B	7.19 ± 0.45	0.95 ± 0.06	44.83 ± 2.84	0.90 ± 0.09	0.79 ± 0.08	1.20 ± 0.12	49.63 ± 3.14	56.66 ± 3.59
C4C	7.16 ± 0.45	1.08 ± 0.07	46.79 ± 2.96	0.95 ± 0.10	0.81 ± 0.08	1.19 ± 0.12	49.16 ± 3.11	57.75 ± 3.66
C4D	7.03 ± 0.45	1.25 ± 0.08	48.40 ± 3.06	1.14 ± 0.12	0.82 ± 0.08	1.22 ± 0.13	42.53 ± 2.69	59.04 ± 3.74

Table 3 Physicochemical characteristics of final banana leaf waste substrate combinations by vermicomposting with *E. eugeniae* (Refer Table 1 for treatment combinations). Values are mean±standard deviation. Similar superscript letters between treatments of the parameters are not significant at $P<0.05$ by DMRT

Treatment	pH	EC (dS/m)	TOC (%)	TKN (%)	TP (%)	TK%	C/*N* ratio	C/*P* ratio
C1A	7.15±0.43^{a}	1.41±0.11^{cd}	31.18±2.15^{de}	1.81±0.17^{c}	0.84±0.10^{c}	1.39±0.13^{c}	17.26±1.45^{e}	37.33±2.57^{d}
C1B	6.97±0.47^{bc}	1.50±0.10^{b}	29.98±2.11^{e}	2.08±0.19^{b}	0.98±0.09^{b}	1.51±0.17^{ab}	14.41±1.21^{f}	30.56±2.25^{f}
C1C	7.00±0.38^{bc}	1.61±0.12^{a}	29.75±2.06^{e}	2.27±0.11^{a}	1.15±0.11^{a}	1.62±0.19^{a}	13.12±1.17^{f}	25.96±2.08^{g}
C1D	7.17±0.42^{a}	1.35±0.10^{d}	35.75±2.32^{a}	1.74±0.14^{c}	0.92±0.10^{b}	1.44±0.14^{c}	20.49±1.42^{de}	38.74±2.46^{d}
C2A	7.00±0.45^{b}	1.08±0.10^{e}	33.19±2.19^{c}	1.68±0.13^{cd}	0.87±0.08^{bc}	1.28±0.12^{e}	19.73±1.52^{d}	37.97±2.48^{d}
C2B	7.04±0.41^{b}	1.37±0.10^{d}	32.01±2.28^{cd}	1.78±0.16^{c}	0.85±0.09^{c}	1.36±0.14^{d}	17.97±1.57^{e}	37.89±2.51^{d}
C2C	7.11±0.42^{a}	1.43±0.11^{cd}	31.69±2.28^{d}	1.71±0.14^{c}	0.88±0.10^{bc}	1.41±0.16^{bc}	18.56±1.59^{e}	35.85±2.37^{de}
C2D	7.06±0.46^{b}	1.27±0.09^{d}	34.10±2.35^{bc}	1.53±0.12^{d}	0.84±0.11^{c}	1.31±0.12^{d}	22.25±1.63^{c}	40.82±2.38^{b}
C3A	7.09±0.45^{a}	1.39±0.12^{cd}	33.60±2.13^{cd}	1.47±0.15^{d}	0.82±0.08^{c}	1.39±0.14^{cd}	22.85±1.45^{c}	40.98±2.59^{b}
C3B	6.85±0.43^{c}	1.47±0.09^{c}	32.30±2.04^{c}	1.69±0.17^{cd}	0.96±0.10^{b}	1.50±0.16^{b}	19.07±1.21^{d}	33.54±2.12^{d}
C3C	6.87±0.44^{c}	1.58±0.10^{ab}	32.05±2.03^{c}	1.85±0.19^{c}	1.12±0.12^{a}	1.62±0.17^{a}	17.37±1.10^{e}	28.49±1.80^{f}
C3D	7.04±0.45^{b}	1.32±0.08^{d}	38.51±2.44^{a}	1.42±0.15^{e}	0.91±0.09^{bc}	1.44±0.15^{c}	27.13±1.72^{a}	42.52±2.69^{ab}
C4A	6.88±0.44^{c}	1.06±0.07^{e}	35.76±2.26^{b}	1.37±0.14^{ef}	0.86±0.09^{c}	1.28±0.13^{d}	26.12±1.65^{b}	41.68±2.64^{b}
C4B	6.91±0.44^{c}	1.35±0.09^{d}	34.49±2.18^{b}	1.45±0.15^{e}	0.83±0.09^{c}	1.36±0.14^{a}	23.79±1.51^{c}	41.59±2.63^{b}
C4C	6.98±0.44^{bc}	1.41±0.09^{cd}	34.14±2.16^{bc}	1.39±0.14^{ef}	0.87±0.09^{c}	1.41±0.15^{cd}	24.58±1.56^{bc}	39.35±2.49^{bcd}
C4D	6.93±0.44^{c}	1.25±0.08^{d}	36.73±2.33^{a}	1.25±0.13^{f}	0.82±0.08^{c}	1.31±0.14^{de}	29.45±1.86^{a}	44.81±2.84^{a}

of pH [28]. Cow dung activates the growth and activity of microbes in the compost. Substances added as amending material in vermicomposting along with cow dung plays a major role in the shift of pH [29]. The pH values of raw samples were 7.91±0.37, 7.79±0.41 and 7.64±0.29, respectively for BLB, CD and AZ. The percent change of pH in most of the treatments over initial values showed decrease to a maximum of 8.51% (Fig. 1A). The final pH varied between different treatments which ranged from 6.85 (C3B)−7.17 (C1D) and the difference in pH between C3B and C1D is significant ($P<0.05$) (Table 3).

The measurement of electrical conductivity provides a direct indication of the abundance of mobile ions, which are primarily derived from soluble salts present in a sample. EC increases as the composting proceeds, due to the release of K, NH_3, P, and other intermediates. The vermicompost, generated from a blend of weed biomass and cattle dung that had been pre-decomposed with mushroom spent straw, showed higher EC values between 1.63 and 2.35 dS/m [30]. In the present study, with banana leaf biomass and cow dung, the EC values range from 0.81 to 1.64 dS/m. The highest EC value, 1.64±0.12 dS/m, was observed in the 45th day of the C1C sample (1:1 BLB: CD dung with 20% *Azolla*). Lowest EC observed on the 0th day of the C4A sample (4:1BLB: CD with 0% *Azolla*). Figure 3 (E-H) gives the graphical representation of the EC values in four different vermicompost compositions. The EC values for raw samples recorded as BLB: 0.74±0.07 dS/m, CD: 0.79±0.07 dS/m, AZ:0.82±0.09 dS/m. These values indicate the nutrient stability in the substrate mixture. In C2D and C4D treatments no change in EC was observed while in other treatments, the change of EC over initial values ranged from 11.86 to 51.08% with a maximum change in C3A (Fig. 1B). The final EC varied between different treatments and the difference in EC of C3A and C1C is not significant ($P>0.05$) however significant between other treatments (Table 3).

Carbon source is the major source of energy for microbes and even plants. Utilization of carbon by increasing microbial population reveals a decrease in the TOC percentage. Respiration and assimilation, as reported earlier [31], are key mechanisms through which *E. eugeniae* accelerate the transformation of organic carbon. The present analytical study on banana leaf biomass and cow dung vermicomposting amended with *Azolla* has shown a reduction in the percentage of TOC as the vermicomposting proceeds. Minimum percentage of TOC 29.75±2.06% was reported on the 75th day sample from the C1C sample (1:1 BLB: CD with 20% *Azolla*). TOC records for raw materials are; BLB: 44.84±2.64%, CD: 46.45±2.32% and AZ%: 47.08±2.58. The TKN value in most of the samples was initially low on the 0th day, but it increased as the composting process progressed. By the 45th day of composting, all samples exhibited higher TKN values, reaching their peak on the final sample collected on the 75th day. The sample from C1C (1:1 BLB: CD with 20% *Azolla*) on the 75th day showed the highest TKN value, reaching 2.27±0.11% (Fig. 3A-D). TKN values of raw materials are- BLB:0.80±0.10%, CD: 0.89±0.09% and AZ:2.2±0.9%, and AZ as a nitrogen fixer, is rich in nitrogen, whereas banana leaves biomass and cow provide the structural carbon. The TK and TP values in all the samples during the 0th day were low. It increased as composting proceeded, reaching a peak on the 45th day, followed by a decrease as given in Fig. 3 (E-H) and Fig. 3 (I-L), respectively. The peak TK and TP values were observed in the 45th-day samples of C1C (1:1 ratio of BLB: CD with 20% *Azolla*) and C1C (1:1 ratio of BLB: CD with

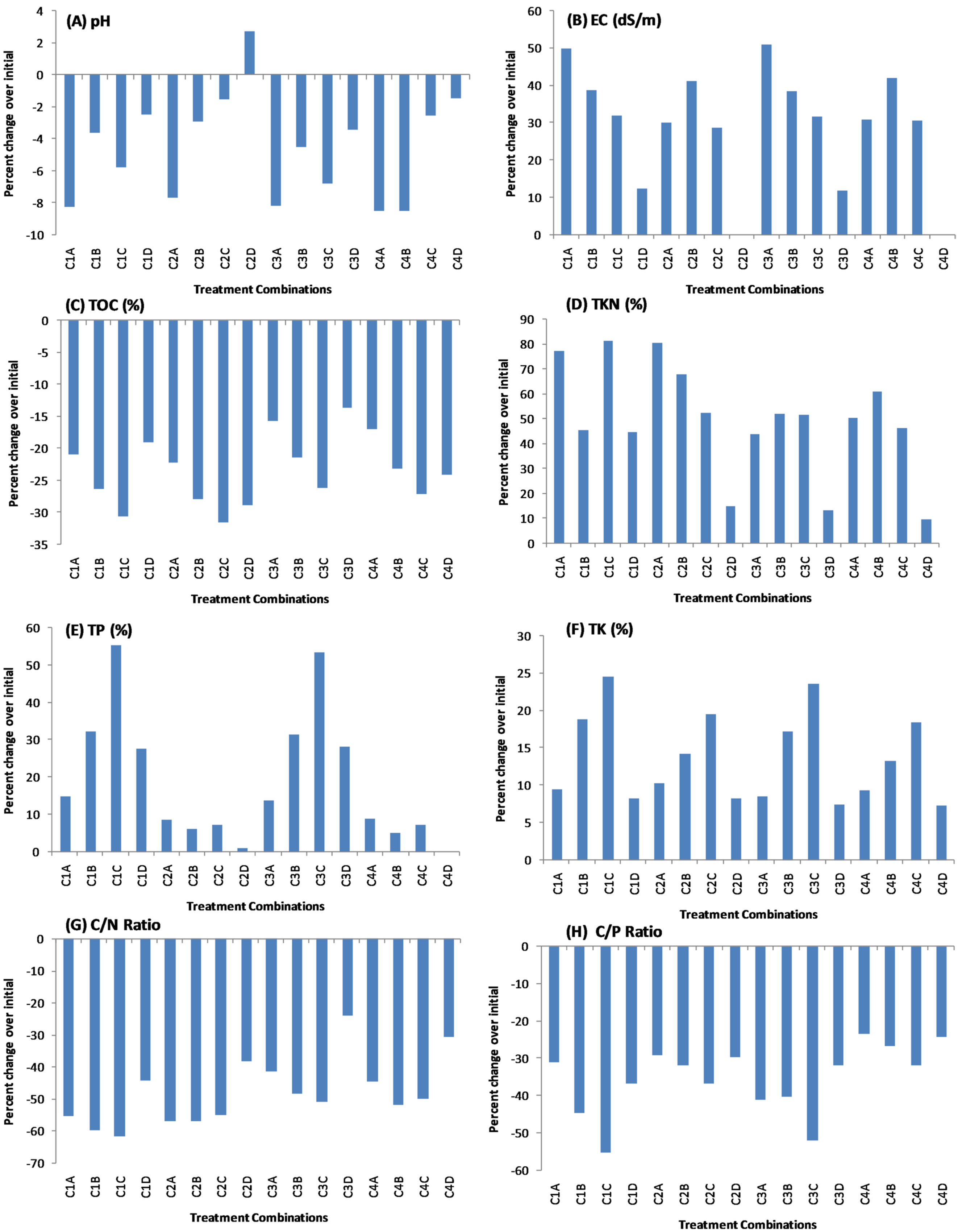

Fig. 1 Percent change of physicochemical characteristics of banana leaf waste substrate combinations by vermicomposting with *E. eugeniae* (Refer Table 1 for treatment combinations). Negative sign indicates decline; Positive sign indicates increment

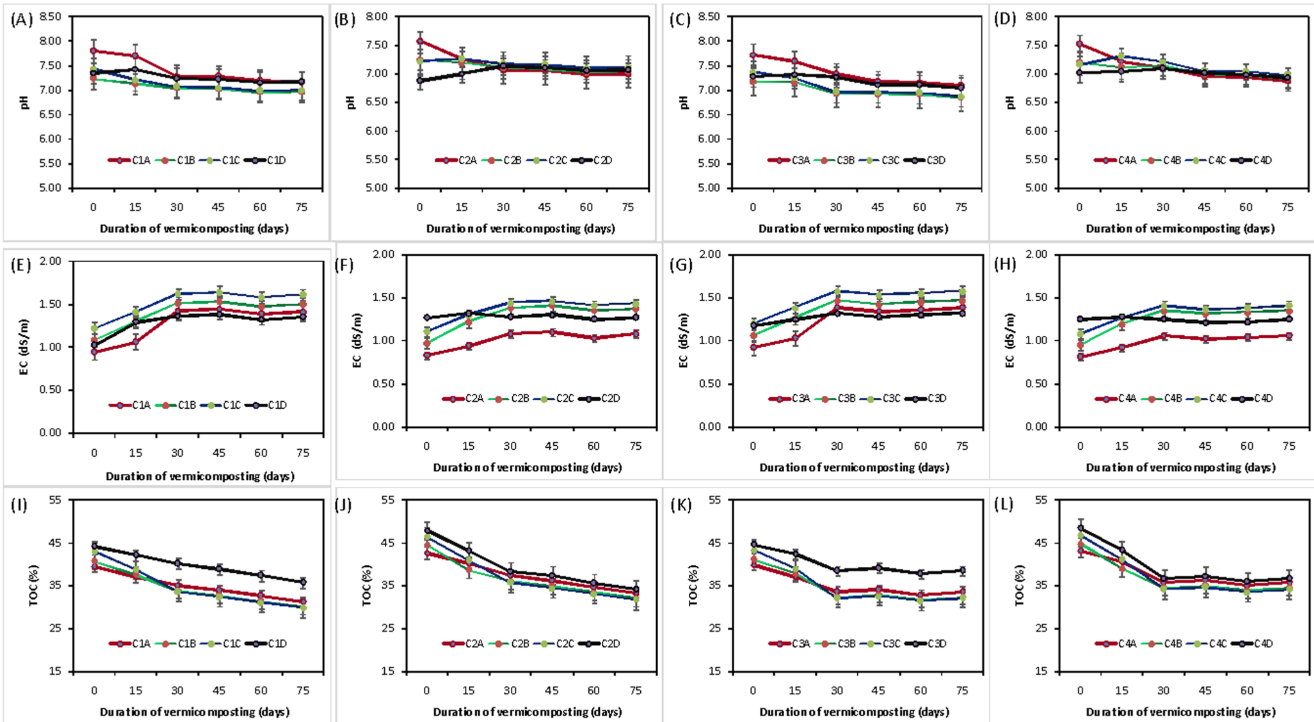

Fig. 2 Changes in pH (ABCD), EC (EFGH) and TOC (IJKL) contents during vermicomposting of different vermibed combinations. Values are mean ± standard deviation (Refer to Table 1 for vermibed combinations)

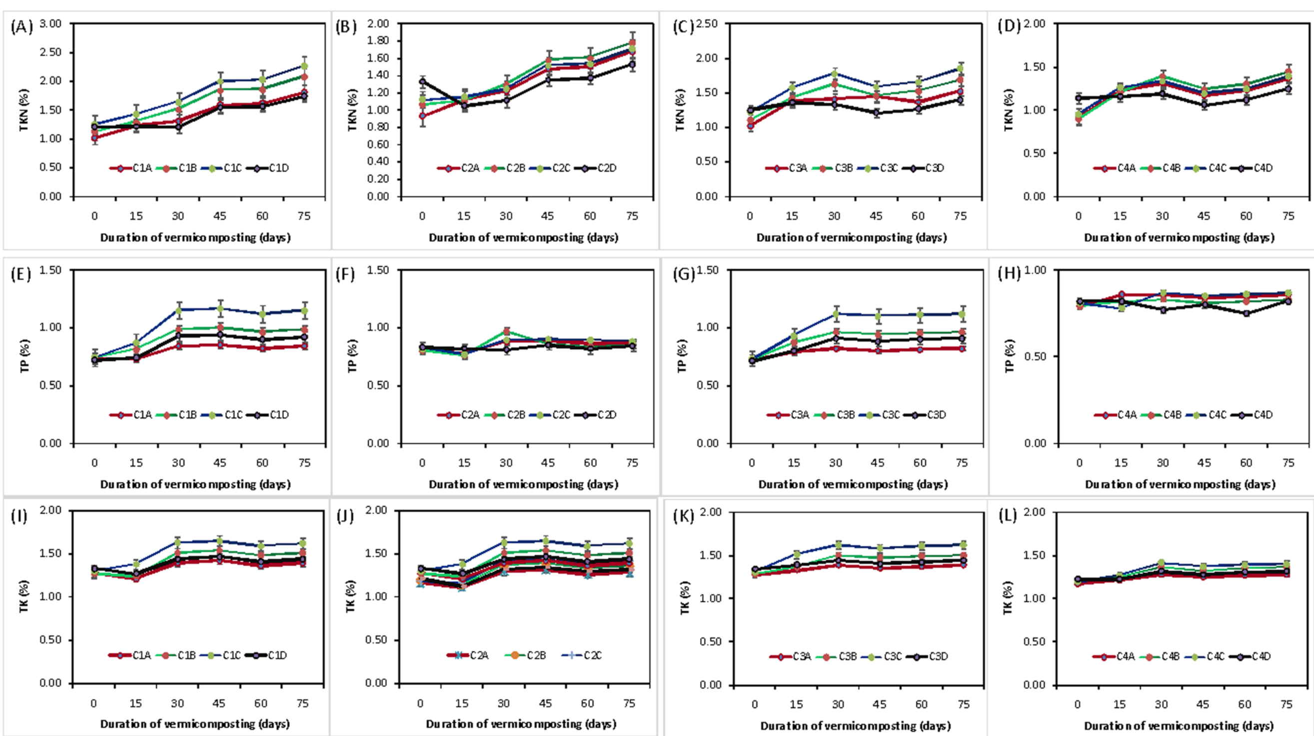

Fig. 3 Changes in TKN (ABCD), TP (EFGH) and TK (IJKL) contents during vermicomposting of different vermibed combinations. Values are mean ± standard deviation (Refer Table 1 for vermibed combinations)

20% *Azolla*), which recorded values of 1.65 ± 0.17% and 1.17 ± 0.13%, respectively. A drop in the TK value after the 45th day was attributed to the fact that, due to the unavailability of organic waste during the final stages, earthworms reassimilate phosphorus into their body during the final period of vermicomposting to overcome starvation. The loss of potassium is mostly due to the high relative humidity prevailing in the area during the vermicomposting, resulting in the leaching of potassium [32, 33]. The TK values recorded for raw samples were BLB: 1.13 ± 0.13%, CD: 1.16 ± 0.11%, AZ: 1.19 ± 0.14% and TP values were BLB: 0.82 ± 0.05%, CD: 0.83 ± 0.05%, AZ: 0.85 ± 0.04%. The percentage change of macronutrients TKN, TP and TK from their initial levels in C1C treatment was found to be higher which was significant in most of the C2, C3 and C4 treatments (Fig. 1D-F). The declining trend of TOC, C/N ratio and C/P ratio was also found to be higher in C1C treatment than other treatments implying that this treatment is most suitable for *E. eugeniae*.

There was an increase in the mineral nutrients such as Ca, Na, S, Mn, Cu, Fe, and Zn contents in the vermicompost (Table 4). The vermicompost with a 1:1 ratio BLB: CD amended with *Azolla* showed peak values in all these parameters at the 45th day of composting. The C/N ratios for the raw samples were as follows: BLB– 55.84 ± 2.18, CD – 51.98 ± 2.51, and AZ – 21.40 ± 2.18. The recorded C/N ratio of the banana leaf biomass after precomposting signifies effective softening of the leaf and making it more pliable. Further addition of cow dung and *Azolla* facilitates lowering the C/N ratio, providing an enhanced substrate for *Eudrilus eugeniae.*

A reduction in the C/N ratio of the vermicompost was noted (Fig. 4A-D), potentially resulting from the oxidative loss of carbon through respiratory processes and its assimilation by microbes within the *E. eugeniae* digestive tract. Previous studies have indicated that microbial mineralization of organic compounds enhances nitrogen and phosphorus content in vermicompost [5]. During vermicomposting, the transformation of organic phosphorus into plant-available forms is driven by phosphatase enzymes secreted in the *E. eugeniae* gut. These enzymes, along with the activity of phosphorus-solubilizing microorganisms, significantly enhance phosphorus bioavailability in the final compost. The C/N ratio ranged from 45.74 to 13.12, with the lowest value, 13.12 ± 1.17, recorded in the 75th day sample of C1C (1:1 BLB: CD with 20% *Azolla*). A reduction in both C/N and C/P ratios was observed within the vermicompost samples. The lowest C/P ratio, 25.96 ± 2.08, was observed in the 75th day sample of C1C (1:1 BLB: CD with 20% AZ).

Figure 4 (E-H) illustrates the changes in the C/P ratio in different combinations of the substrates during vermicomposting. Microbial mineralization, leading to nitrogen and phosphorus release, and the excretion of nitrogenous by-products are potential mechanisms contributing to the decline in C/P and C/N ratios [34]. The C/P ratios for the raw materials were as follows: BLB- 54.87, CW – 56.19, and AZ– 55.66. Previous studies on composting different substrates with earthworms reported an increase in the mineral nutrients such as Ca and P by the end of 45 days of vermicomposting and a gradual decrease in the values of pH, total organic carbon, C/N, and C/P ratios [35]. The vermicompost prepared from BLB and CD showed an increase in Ca, Na, S, Mn, Cu, Fe, and Zn contents in the vermicompost. Table 3 presents the values obtained for the changes in Ca, Na, S, Mn, Cu, Fe, and Zn during vermicomposting. C1C (1:1 BLB: CD amended with 20% *Azolla*) on 45th day showed the peak level of Ca (1.71%), C1D (1:1 BLB: CD amended with 30% *Azolla* on 30th and 45th day showed the peak level of Na (0.55%), C1C (1:1 BLB: CD amended with 20% *Azolla* on 45th day showed the peak level of Mg (0.74%), C3D (3:1 BLB: CD amended with 30% *Azolla*) on 15th day and C1D (1:1 BLB: CD amended with 30% *Azolla*) on 30th and 45th day showed the peak level of S (0.18%). The highest values of minerals obtained in the vermicompost are as follows: Cu (26.44 ppm) in the 45th day sample of C1C, Mn (21.14 ppm) in the 45th day sample of C2A, Fe (146.35 ± 0.41 ppm) in the 45th day sample of C1C, and Zn (43.32 ppm) in the initial (0 day) sample of C2D. The values recorded for Cu and Zn are far less when compared to the permissible limits of 300ppm for Cu and 1000 ppm for Zn. Similarly, Mn and Fe are also recorded in an acceptable range that is favorable for plant growth and development [36]. The addition of CD and biochar to the nitrogen-rich green manure plant improved the vermicompost quality and enhanced the organic matter decomposition [37]. The current results demonstrated significant improvements in vermicomposting when assisted by *Azolla*.

Biomass and reproduction of earthworm

Table 5 shows the final earthworm count across all 16 vermicomposting groups at the study's conclusion. Starting with 25 adult earthworms in each group, the vermicompost produced from banana leaf biomass waste and cow dung, when enhanced with *Azolla*, fostered a larger *E. eugeniae* population than the group lacking *Azolla*. This suggests that *Azolla*'s inclusion boosted *E. eugeniae* numbers, probably by improving the nutritional content and moisture levels within the vermicomposting blend. These factors play a major role in influencing its population.

E. eugeniae populations in different vermicompost combinations exhibited distinct patterns. The number of worms found in the 1:1 vermicompost combinations followed a descending order: C1C > C1B > C1D > C1A. In the

Table 4 Changes in calcium (Ca), sodium (Na), sulfur (S), manganese (Mn), copper (Cu), iron (Fe), and zinc (Zn) in the vermicompost (Refer Table 1 for treatment combinations)

	Ca (%)	Na (%)	Mg (%)	S (%)	Mn (ppm)	Cu (ppm)	Fe (ppm)	Zn (ppm)
0 Day Sample								
C3A	1.05 ± 0.11	0.32 ± 0.03	0.53 ± 0.06	0.17 ± 0.02	13.88 ± 1.43	16.52 ± 1.71	110.39 ± 4.78	39.10 ± 2.48
C3B	1.09 ± 0.11	0.34 ± 0.04	0.53 ± 0.06	0.16 ± 0.02	14.39 ± 1.49	16.68 ± 1.72	109.36 ± 4.74	38.28 ± 2.35
C3C	1.07 ± 0.11	0.33 ± 0.03	0.55 ± 0.06	0.16 ± 0.02	13.94 ± 1.44	16.64 ± 1.72	111.45 ± 4.83	39.18 ± 2.48
C3D	1.12 ± 0.12	0.40 ± 0.04	0.58 ± 0.06	0.17 ± 0.02	15.66 ± 1.62	17.26 ± 1.78	111.58 ± 4.83	38.98 ± 2.47
C4A	0.91 ± 0.09	0.36 ± 0.04	0.51 ± 0.05	0.14 ± 0.01	17.27 ± 1.78	19.81 ± 2.05	116.71 ± 5.05	42.56 ± 2.69
C4B	0.91 ± 0.09	0.35 ± 0.04	0.52 ± 0.05	0.13 ± 0.01	17.35 ± 1.79	18.02 ± 1.86	114.53 ± 4.96	42.27 ± 2.68
C4C	0.92 ± 0.10	0.39 ± 0.04	0.54 ± 0.06	0.15 ± 0.02	17.47 ± 1.81	19.18 ± 1.98	116.52 ± 5.05	40.15 ± 2.54
C4D	0.92 ± 0.09	0.38 ± 0.03	0.54 ± 0.04	0.15 ± 0.02	17.30 ± 1.79	20.402 ± 2.11	116.07 ± 5.03	42.96 ± 2.72
15 Days Sample								
C3A	1.17 ± 0.12	0.25 ± 0.03	0.57 ± 0.06	0.15 ± 0.02	14.80 ± 1.53	20.13 ± 2.08	114.51 ± 4.96	37.05 ± 2.35
C3B	1.16 ± 0.12	0.37 ± 0.04	0.63 ± 0.06	0.17 ± 0.02	15.92 ± 1.64	23.62 ± 2.44	118.95 ± 5.15	34.45 ± 2.18
C3C	1.38 ± 0.14	0.44 ± 0.05	0.61 ± 0.06	0.17 ± 0.02	14.62 ± 1.51	24.35 ± 2.52	136.66 ± 5.92	40.48 ± 2.56
C3D	1.24 ± 0.13	0.39 ± 0.04	0.59 ± 0.06	0.18 ± 0.02	16.00 ± 1.65	20.69 ± 2.14	119.29 ± 5.17	39.26 ± 2.48
C4A	0.96 ± 0.10	0.40 ± 0.04	0.58 ± 0.06	0.14 ± 0.01	19.18 ± 1.98	20.54 ± 2.12	120.81 ± 5.23	32.26 ± 2.04
C4B	0.93 ± 0.10	0.41 ± 0.04	0.60 ± 0.06	0.15 ± 0.02	18.36 ± 1.90	19.07 ± 1.97	118.59 ± 5.13	38.26 ± 2.42
C4C	0.94 ± 0.10	0.32 ± 0.03	0.59 ± 0.06	0.16 ± 0.02	18.94 ± 1.96	21.61 ± 2.23	122.36 ± 5.30	37.19 ± 2.35
C4D	0.90 ± 0.09	0.39 ± 0.04	0.57 ± 0.06	0.16 ± 0.02	18.25 ± 1.88	20.45 ± 2.11	117.27 ± 5.08	40.34 ± 2.55
30 Days Sample								
C3A	1.34 ± 0.14	0.22 ± 0.02	0.63 ± 0.06	0.13 ± 0.01	15.39 ± 1.59	21.45 ± 2.22	117.89 ± 5.10	35.04 ± 2.22
C3B	1.41 ± 0.15	0.24 ± 0.02	0.68 ± 0.07	0.14 ± 0.01	16.25 ± 1.68	23.96 ± 2.47	121.21 ± 5.25	33.58 ± 2.13
C3C	1.65 ± 0.17	0.32 ± 0.03	0.71 ± 0.07	0.13 ± 0.01	16.64 ± 1.72	25.44 ± 2.63	140.84 ± 6.10	39.06 ± 2.47
C3D	1.38 ± 0.14	0.53 ± 0.06	0.62 ± 0.06	0.17 ± 0.02	15.89 ± 1.64	21.35 ± 2.21	121.18 ± 5.25	39.26 ± 2.48
C4A	1.06 ± 0.11	0.42 ± 0.04	0.61 ± 0.06	0.12 ± 0.01	20.34 ± 2.10	21.07 ± 2.18	124.96 ± 5.41	32.46 ± 2.05
C4B	1.26 ± 0.13	0.36 ± 0.04	0.62 ± 0.06	0.13 ± 0.01	19.67 ± 2.03	21.64 ± 2.24	123.61 ± 5.35	36.70 ± 2.32
C4C	0.83 ± 0.09	0.34 ± 0.04	0.62 ± 0.06	0.15 ± 0.02	18.02 ± 1.86	22.67 ± 2.34	123.93 ± 5.37	36.85 ± 2.33
C4D	0.89 ± 0.09	0.40 ± 0.04	0.55 ± 0.06	0.14 ± 0.01	20.07 ± 2.07	21.66 ± 2.24	119.59 ± 5.18	38.25 ± 2.42
45 Days Sample								
C3A	1.31 ± 0.14	0.21 ± 0.02	0.61 ± 0.06	0.13 ± 0.01	15.00 ± 1.55	20.91 ± 2.16	114.95 ± 4.98	34.17 ± 2.16
C3B	1.38 ± 0.14	0.23 ± 0.02	0.66 ± 0.07	0.14 ± 0.01	15.85 ± 1.64	23.36 ± 2.41	118.18 ± 5.12	32.75 ± 2.07
C3C	1.61 ± 0.17	0.32 ± 0.03	0.70 ± 0.07	0.13 ± 0.01	16.22 ± 1.68	24.81 ± 2.56	137.32 ± 5.95	38.08 ± 2.41
C3D	1.35 ± 0.14	0.52 ± 0.05	0.60 ± 0.06	0.17 ± 0.02	15.49 ± 1.60	20.82 ± 2.15	118.15 ± 5.12	38.28 ± 2.42
C4A	1.03 ± 0.11	0.41 ± 0.04	0.59 ± 0.06	0.12 ± 0.01	19.83 ± 2.05	20.54 ± 2.12	121.83 ± 5.28	31.65 ± 2.00
C4B	1.23 ± 0.13	0.35 ± 0.04	0.60 ± 0.06	0.13 ± 0.01	19.17 ± 1.98	21.10 ± 2.18	120.52 ± 5.22	35.78 ± 2.27
C4C	0.81 ± 0.08	0.33 ± 0.03	0.60 ± 0.06	0.15 ± 0.02	17.57 ± 1.81	22.10 ± 2.28	120.83 ± 5.23	35.92 ± 2.27
C4D	0.86 ± 0.09	0.39 ± 0.04	0.54 ± 0.06	0.14 ± 0.01	19.57 ± 2.02	21.12 ± 2.08	116.60 ± 5.05	37.29 ± 2.36
60 Days Sample								
C3A	1.33 ± 0.14	0.22 ± 0.02	0.62 ± 0.06	0.13 ± 0.01	15.24 ± 1.57	21.25 ± 2.19	116.79 ± 5.06	34.71 ± 2.20
C3B	1.40 ± 0.14	0.24 ± 0.02	0.67 ± 0.07	0.14 ± 0.01	16.10 ± 1.66	23.73 ± 2.45	120.07 ± 5.20	33.27 ± 2.11
C3C	1.63 ± 0.17	0.32 ± 0.03	0.71 ± 0.07	0.13 ± 0.01	16.48 ± 1.70	25.20 ± 2.60	139.52 ± 6.04	38.69 ± 2.45
C3D	1.37 ± 0.14	0.53 ± 0.05	0.61 ± 0.06	0.17 ± 0.02	15.74 ± 1.63	21.15 ± 2.19	120.04 ± 5.20	38.89 ± 2.46
C4A	1.05 ± 0.11	0.42 ± 0.04	0.60 ± 0.06	0.12 ± 0.01	20.15 ± 2.08	20.87 ± 2.16	123.78 ± 5.36	32.15 ± 2.04
C4B	1.25 ± 0.13	0.36 ± 0.04	0.61 ± 0.06	0.13 ± 0.01	19.48 ± 2.01	21.44 ± 2.21	122.45 ± 5.30	36.36 ± 2.30
C4C	0.82 ± 0.08	0.34 ± 0.04	0.61 ± 0.06	0.15 ± 0.02	17.85 ± 1.84	22.46 ± 2.32	122.76 ± 5.32	36.50 ± 2.31
C4D	0.88 ± 0.09	0.40 ± 0.04	0.55 ± 0.06	0.14 ± 0.01	19.88 ± 2.05	21.46 ± 2.22	118.47 ± 5.13	37.89 ± 2.40
75 Days Sample								
C3A	1.34 ± 0.14	0.22 ± 0.02	0.63 ± 0.06	0.13 ± 0.01	15.39 ± 1.59	21.45 ± 2.22	117.89 ± 5.10	35.04 ± 2.22
C3B	1.41 ± 0.15	0.24 ± 0.02	0.68 ± 0.07	0.14 ± 0.01	16.25 ± 1.68	23.96 ± 2.47	121.21 ± 5.25	33.58 ± 2.13
C3C	1.65 ± 0.17	0.32 ± 0.03	0.71 ± 0.07	0.13 ± 0.01	16.64 ± 1.72	25.44 ± 2.63	140.84 ± 6.10	39.06 ± 2.47
C3D	1.38 ± 0.14	0.53 ± 0.06	0.62 ± 0.06	0.17 ± 0.02	15.89 ± 1.64	21.35 ± 2.21	121.18 ± 5.25	39.26 ± 2.48
C4A	1.06 ± 0.11	0.42 ± 0.04	0.61 ± 0.06	0.12 ± 0.01	20.34 ± 2.10	21.07 ± 2.18	124.96 ± 5.41	32.46 ± 2.05
C4B	1.26 ± 0.13	0.36 ± 0.04	0.62 ± 0.06	0.13 ± 0.01	19.67 ± 2.03	21.64 ± 2.24	123.61 ± 5.35	36.70 ± 2.32
C4C	0.83 ± 0.09	0.34 ± 0.04	0.62 ± 0.06	0.15 ± 0.02	18.02 ± 1.86	22.67 ± 2.34	123.93 ± 5.37	36.85 ± 2.33
C4D	0.89 ± 0.09	0.40 ± 0.04	0.55 ± 0.06	0.14 ± 0.01	20.07 ± 2.07	21.66 ± 2.24	119.59 ± 5.18	38.25 ± 2.42

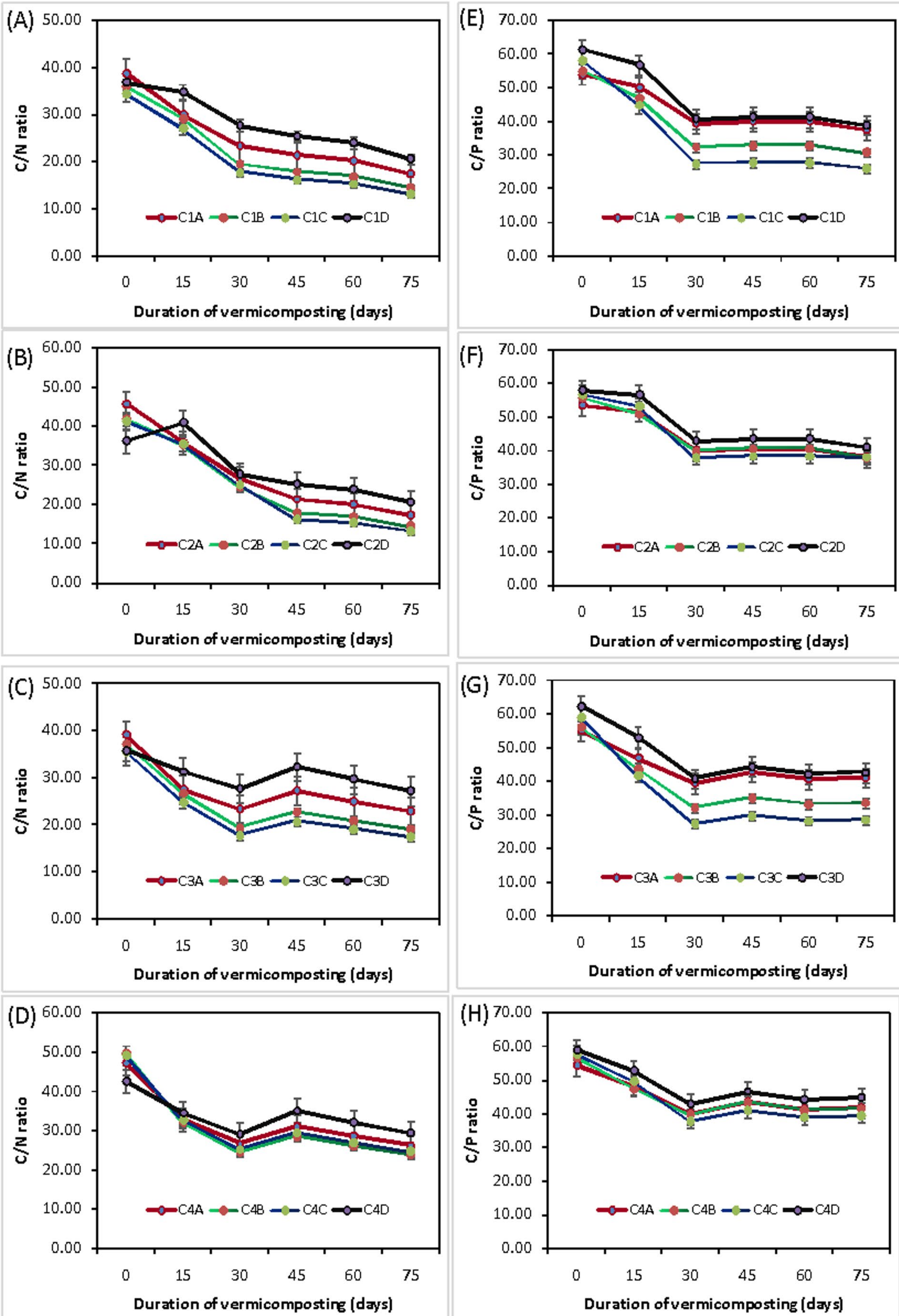
(A)
C/N ratio
50.00
40.00
30.00
20.00
10.00
0.00
C1A C1B C1C C1D
0 15 30 45 60 75
Duration of vermicomposting (days)
(E)
C/P ratio
70.00
60.00
50.00
40.00
30.00
20.00
10.00
0.00
C1A C1B C1C C1D
0 15 30 45 60 75
Duration of vermicomposting (days)
(B)
C/N ratio
60.00
50.00
40.00
30.00
20.00
10.00
0.00
C2A C2B C2C C2D
0 15 30 45 60 75
Duration of vermicomposting (days)
(F)
C/P ratio
70.00
60.00
50.00
40.00
30.00
20.00
10.00
0.00
C2A C2B C2C C2D
0 15 30 45 60 75
Duration of vermicomposting (days)
(C)
C/N ratio
50.00
40.00
30.00
20.00
10.00
0.00
C3A C3B C3C C3D
0 15 30 45 60 75
Duration of vermicomposting (days)
(G)
C/P ratio
70.00
60.00
50.00
40.00
30.00
20.00
10.00
0.00
C3A C3B C3C C3D
0 15 30 45 60 75
Duration of vermicomposting (days)
(D)
C/N ratio
60.00
50.00
40.00
30.00
20.00
10.00
0.00
C4A C4B C4C C4D
0 15 30 45 60 75
Duration of vermicomposting (days)
(H)
C/P ratio
70.00
60.00
50.00
40.00
30.00
20.00
10.00
0.00
C4A C4B C4C C4D
0 15 30 45 60 75
Duration of vermicomposting (days)

Fig. 4 Changes in C/N ratio (ABCD) and C/P ratio (EFGH) during vermicomposting of different vermibed combinations. Values are mean ± standard deviation (Refer Table 1 for vermibed combinations)

2:1 combination, the number of worms was observed in the descending order: C2D > C2C > C2A > C2B. For the 3:1 combination, the order was C3C > C3B > C3A > C3D. Similarly, in the 4:1 combination, worm abundance followed the order: C4D > C4C > C4B > C4A. The highest quantity of *E. eugeniae* population in banana leaf biomass waste vermicompost was obtained in C1C (BLB: CD 1:1, amended with 20% AZ), and the lowest quantity was in C4A (BLB: CD 4:1, amended with 0% AZ). In combination ratio 1:1 with BLB: CD, the highest number of *E. eugeniae* was present in C1C amended with 20% *Azolla*, and the least number was observed in C1A amended with 0% *Azolla*. In combination ratio 2:1 with BLB: CD, the highest number of *E. eugeniae* was present in C2D amended with 30% *Azolla*, and the least number was observed in C2B amended with 10% *Azolla*. In combination ratio 3:1 with BLB: CD, the highest number was present in C3C amended with 20% *Azolla*, and the least number was observed in C3A amended with 0% *Azolla*. In combination ratio 4:1 with BLB: CD, the highest number of *E. eugeniae* was present in C4D amended with 30% *Azolla*, and the least number was observed in C3A amended with 0% *Azolla*. The suitability of an organic substrate as a food source for earthworms is largely determined by the density of the worm population and the accelerated breaking down of sludge during vermicomposting is mainly due to the increased aeration and waste turnover facilitated by earthworms [38, 39].

Vermibed amended with *Azolla* has favoured the weight gain in biomass. The nutrients in the biochar, amended vermibed groups, supported earthworm growth and weight gain, surpassing those in the groups without biochar [5]. In the 1:1 combination of BLB: CD, the biomass followed a descending order of C1C > C1B > C1D > C1A, with C1C exhibiting the highest biomass of 69.53 ± 1.47 g. In the 2:1 combination, the order was C2D > C2C > C2A > C2B, where C2D recorded the maximum biomass of 59.23 ± 1.76 g. The 3:1 combination showed a descending trend of C3C > C3B > C3A > C3D, with C3C having the highest biomass of 47.90 ± 0.78 g. Lastly, in the 4:1 combination, the biomass decreased in the order C4D > C4C > C4B > C4A, with C4C attaining the highest value of 45.12 ± 0.26 g. Studies on the effect of time on earthworm biomass reported that the initial phase lasted for 45 days and was marked by high microbial activity, with no significant changes observed in the populations of adult and juvenile earthworms [40]. In contrast, the final phase, spanning from the 45th to the 120th day, exhibited approximately a 45% reduction in microbial activity and the dry mass of vermicompost. Additionally, this phase saw a steep increase in the population of earthworm cocoons within the vermicompost.

Microbial communities are the primary drivers of organic matter's biochemical breakdown. While microbial activity is fundamental, earthworms enhance the process by physically fragmenting and conditioning the substrate, which in turn fosters microbial expansion due to increased surface area and modifies the substrate's biological functioning [41]. The vermicomposting process leads to a greater loss of dry matter in the waste as a result of the earthworm's growth and consumption. In the vermicomposting study using banana leaf biomass (BLB) and cow dung (CD) amended with *Azolla*, substrates were mixed to an initial total weight of 2 kg. After 75 days of vermicomposting, a reduction in the final compost weight was observed across all treatments. In the 1:1 BLB: CD combination, the final compost weight followed a descending order: C1A > C1C > C1B > C1D. For the 2:1 BLB: CD combination, the order was C2B > C2D > C2C > C2A. In the 3:1 combination, weights declined in the order: C3D > C3A > C3B > C3C. Finally, in the 4:1 BLB: CD composition, the descending order was C4A > C4C > C4B > C4D. Notably, the lowest final compost weight was recorded in C1D, which was amended with 30% *Azolla*. The C1 combinations, especially C1C treatment greatly supported the growth and reproduction of *E. eugeniae* significantly ($P < 0.05$) which might be attributed to the substrates suitability to *E. eugeniae*. The addition of CD and nitrogenous substrates like green manures influence the growth and reproduction of the earthworms by supplying nutrients and making the substrates suitable for vermiconversion [42, 43, 44].

Conclusions

The vermicomposting of banana leaves biomass with cow dung fortified with *Azolla*, utilizing *E. eugeniae*, yielded highly successful results. The incorporation of cow dung into vermicompost is essential for fostering initial microbial proliferation and sustaining *E. eugeniae* activity. Nutrient levels are substantially elevated through the incorporation of *Azolla* in diverse proportions, thereby accelerating the rapid mineralization of organic substrates in the mixture. The nutrient levels in the vermicompost, including TKN, TP, and TK, exhibited a statistically significant elevation relative to the initial substrates in all treatments. Meanwhile, the C/N and C/P ratios significantly decreased. Significantly, the incorporation of 20–30% *Azolla* resulted in a marked enhancement of nutrient content, most notably within the 1:1 blend of banana leaf biomass waste and cow dung (C1). Equal proportion of cow dung and banana leaf biomass amended with 20–30% *Azolla* was an excellent

Table 5 Initial and final values of earthworm biomass, earthworm population, and compost weight. Values are mean±standard deviation. Similar superscript letters between treatments of the parameters are not significant at $P < 0.05$ by DMRT

Treatment	Worm number		Worm biomass (g)		Compost weight (g)	
	Initial	Final	Initial	Final	Initial	Final
C1A	24 ± 0	57 ± 5 [bc]	24.33 ± 1.27	58.83 ± 3.63 [ab]	2000	671 ± 21 [d]
C1B	24 ± 0	65 ± 5 [b]	23.27 ± 1.63	67.87 ± 3.02 [a]	2000	540 ± 14 [h]
C1C	24 ± 0	74 ± 4 [a]	25.93 ± 1.72	69.53 ± 2.87 [a]	2000	615 ± 16 [f]
C1D	24 ± 0	64 ± 4 [b]	28.13 ± 1.21	67.2 ± 3.24 [a]	2000	425 ± 12 [i]
C2A	24 ± 0	52 ± 5 [c]	23.27 ± 1.53	41.97 ± 1.83 [c]	2000	604 ± 16 [fg]
C2B	24 ± 0	46 ± 3 [d]	24.7 ± 1.04	44.8 ± 2.78 [cd]	2000	795 ± 23 [a]
C2C	24 ± 0	55 ± 4 [c]	22.97 ± 1.32	54.77 ± 2.52 [b]	2000	643 ± 18 [e]
C2D	24 ± 0	58 ± 6 [b]	27.8 ± 1.64	59.2 ± 3.28 [ab]	2000	753 ± 17 [b]
C3A	24 ± 0	42 ± 4 [de]	25.12 ± 0.87	43.22 ± 2.04 [cd]	2000	623 ± 18 [f]
C3B	24 ± 0	48 ± 5 [d]	28.0 ± 0.74	46.06 ± 2.17 [c]	2000	611 ± 21 [f]
C3C	24 ± 0	51 ± 4 [c]	23.5 ± 1.03	47.9 ± 2.34 [c]	2000	598 ± 17 [g]
C3D	24 ± 0	43 ± 4 [de]	26.9 ± 1.08	38.6 ± 1.87 [d]	2000	702 ± 24 [c]
C4A	24 ± 0	31 ± 4 [e]	25.9 ± 1.22	39 ± 2.06 [d]	2000	684 ± 18 [c]
C4B	24 ± 0	35 ± 4 [e]	24.5 ± 1.27	40.89 ± 2.27 [cd]	2000	593 ± 15 [g]
C4C	24 ± 0	47 ± 4 [d]	251.1 ± 0.95	45.01 ± 2.34 [c]	2000	604 ± 21 [fg]
C4D	24 ± 0	49 ± 5 [cd]	25.3 ± 1.37	45.19 ± 2.74 [c]	2000	518 ± 13

vermibed that significantly influences both the earthworm's growth rate and the production of vermicompost.

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Declarations

Conflict of interest The authors declare that there is no conflict of interest regarding the publication of this article.

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