

การเสริมประสิทธิภาพปุ๋ยหมักและปุ๋ยมูลไส้เดือนด้วยจุลินทรีย์ตรึงในโตรเจน เพื่อยกระดับศักยภาพการให้ธาตุอาหารพืช

Bioaugmentation of Compost and Vermicompost with Nitrogen-Fixing Bacteria for Improved Plant Nutrient Supply

ปัญญา ชัยรัตนพานิช^{1,2} ปิยะวดี สราภิรมย์¹ พงศธร กองแก้ว¹ ชรินรัตน์ พรรณะ³ ทิพย์ธัญญา ศรีโยพล³ ศิริน เรื่องศิริ³ ทรงกลด พลพวก⁴ มัลลิกา ธีระกุล⁵ จักรกฤช ศรีละออ⁴ ทัย กาบบัว¹ และ ทรงวิทย์ หงสประภาส¹.8*

Panya Chairattanaphanich¹,², Piyawadee Saraphirom¹, Pongsathorn Kongkaew¹, Charinrat Panna³,

Thipthanya Sriyophon³, Sirin Ruangsiri³, Songklot Phonphuak⁴, Mullika Teerakun⁵,

Chakkrit Sreela-or⁴, Thai Kabbua¹ and Throngvid Hongsaprabhas¹,8*

¹คณะวิทยาศาสตร์และเทคโนโลยี มหาวิทยาลัยราชภัฏมหาสารคาม จังหวัดมหาสารคาม
 ²บริษัท กรีนเดลี่ ฟู้ดส์ จำกัด จังหวัดปทุมธานี
 ³คณะครุศาสตร์ มหาวิทยาลัยราชภัฏมหาสารคาม จังหวัดมหาสารคาม
 ⁴คณะวิทยาการจัดการ มหาวิทยาลัยราชภัฏสุรินทร์ จังหวัดสุรินทร์
 ⁵คณะเทคโนโลยีการเกษตร มหาวิทยาลัยภาชสินธุ์ จังหวัดกาฬสินธุ์
 ⁴คณะเทคโนโลยีการเกษตรและอาหาร มหาวิทยาลัยราชภัฏพิบูลสงคราม จังหวัดพิษณุโลก
 ³คณะศิลปศาสตร์และวิทยาศาสตร์ มหาวิทยาลัยราชภัฏศรีสะเกษ จังหวัดศรีสะเกษ
 ³บริษัท สหภัณฑ์ เช็นจูรี่ จำกัด กรุงเทพมหานคร

¹Faculty of Science and Technology, Rajabhat Maha Sarakham University, Maha Sarakham Province
 ²Greendeli Foods Co., Ltd., Pathum Thani Province
 ³Faculty of Education, Rajabhat Maha Sarakham University, Maha Sarakham Province
 ⁴Faculty of Management Sciences, Surindra Rajabhat University, Surin Province
 ⁵Faculty of Agricultural Technology, Kalasin University, Kalasin Province
 ⁶Faculty of Food and Agricultural Technology, Pibulsongkram Rajabhat University, Phitsanulok Province
 ⁷Faculty of Liberal Arts and Science, Sisaket Rajabhat University, Sisaket Province
 ⁸Sahapan Century Co., Ltd., Bangkok

*Corresponding Author: throngvid.ho@rmu.ac.th

Received: February 20, 2025 Revised: June 08, 2025 Accepted: June 18, 2025

บทคัดย่อ

การศึกษานี้มีวัตถุประสงค์เพื่อยกระดับคุณสมบัติของปุ๋ยหมักและเวอร์มิคอมโพสต์ให้เป็นปุ๋ยชีวภาพที่มีคุณภาพสูง โดยประเมินปุ๋ย 4 สูตร ได้แก่ (1) ปุ๋ยหมักจากเศษผักและผลไม้ผสมมูลสัตว์ หมักในถังหมุนเป็นเวลา 42 วัน ก่อนทำให้แห้ง และร่อน (2) ปุ๋ยมูลใส้เดือนย่อยสลายโดยใส้เดือนสายพันธุ์ Eudrilus eugeniae เป็นเวลา 10 วัน แล้วนำไปทำให้แห้งและ ร่อน (3) ปุ๋ยหมักสูตรที่ (1) เสริมด้วยแบคทีเรีย Azotobacter vinelandii ซึ่งมีความสามารถในการ ตรึงในโตรเจน และ (4) ปุ๋ยมูลใส้เดือนสูตรที่ (2) เสริมด้วยแบคทีเรีย A. vinelandii ประสิทธิภาพของปุ๋ยทั้งหมดได้รับการประเมินโดยวิเคราะห์ สมบัติทางกายภาพและเคมี เปรียบเทียบมาตรฐานปุ๋ยอินทรีย์ของประเทศไทย (พ.ศ. 2548) พบว่าทุกสูตร มีคุณลักษณะตรง ตามเกณฑ์มาตรฐาน ได้แก่ ขนาดเม็ดปุ๋ยไม่เกิน 5×5 มิลลิเมตร ความชื้นและสารระเหยต่ำกว่าร้อยละ 30 ปริมาณอินทรียวัตถุ สูงกว่าร้อยละ 3.5 ปริมาณเศษหินต่ำกว่าร้อยละ 1 และปลอดจากสิ่งปนเปื้อน เช่น พลาสติก แก้ว โลหะ และวัสดุมีคม

ค่าความเป็นกรด-ด่าง (pH) อยู่ระหว่าง 8.00-8.55 และค่าการนำไฟฟ้า (EC) อยู่ในช่วง 1.97-2.55 dS/m ทั้งนี้วัสดุอินทรีย์ ในทุกสูตรถูกย่อยสลายมากกว่าร้อยละ 90 แสดงถึงความสมบูรณ์ของกระบวนการผลิต ผลการเติมแบคทีเรีย A. vinelandii ซึ่งเป็นแบคทีเรียตรึงในโตรเจน แสดงให้เห็นถึงการยกระดับคุณภาพของปุ๋ยอย่างชัดเจน โดยในปุ๋ยหมักพบว่าอินทรียวัตถุเพิ่ม จากร้อยละ 17.33 เป็น 22.33 ขณะที่ในปุ๋ยมูลไส้เดือนเพิ่มจากร้อยละ 27.61 เป็น 28.66 ปริมาณไนโตรเจนในปุ๋ยหมักเพิ่ม จากร้อยละ 0.87 เป็น 1.12 และในปุ๋ยมูลไส้เดือนเพิ่มจากร้อยละ 1.38 เป็น 1.43 ฟอสฟอรัสในปุ๋ยหมักเพิ่มจากร้อยละ 0.068 เป็น 0.108 และในปุ๋ยมูลไส้เดือนเพิ่มจากร้อยละ 0.188 เป็น 0.240 ส่วนโพแทสเซียมในปุ๋ยหมักเพิ่มจากร้อยละ 0.16 เป็น 0.21 และในปุ๋ยมูลไส้เดือนเพิ่มจากร้อยละ 1.39 เป็น 2.13 โดยสรุปการศึกษานี้นำเสนอแนวทางที่มีศักยภาพในการ ยกระดับกระบวนการผลิตปุ๋ยหมัก ด้วยการใช้จุลินทรีย์ที่มีคุณสมบัติทางชีวภาพอย่าง A. vinelandii เพื่อเปลี่ยนของเหลือทิ้ง ทางชีวภาพให้เป็นปุ๋ยชีวภาพคุณภาพสูงที่อุดมด้วยธาตุอาหาร ส่งเสริมความยั่งยืนในภาคการเกษตรได้อย่างเป็นรูปธรรม

คำสำคัญ: การเสริมจุลินทรีย์, ปุ๋ยอินทรีย์, Azotobacter vinelandii, ปุ๋ยมูลไส้เดือน

Abstract

This study aimed to enhance the properties of compost and vermicompost by transforming them into biofertilizers. Four types of fertilizers were evaluated: (1) compost prepared from fruit and vegetable scraps mixed with manure, fermented in a rotating tumbler for 42 days, then dried and sieved; (2) vermicompost produced from fruit and vegetable scraps decomposed by *Eudrilus eugeniae* earthworms for 10 days, then dried and sieved; (3) compost from (1) supplemented with Azotobacter vinelandii, a nitrogen-fixing bacterium; and (4) vermicompost from (2) enriched with A. vinelandii. All fertilizers were assessed for maturity using both physical and chemical analyses, and their performance was compared to Thai organic fertilizer standards. All formulations contained granules smaller than 5×5 mm, moisture and volatile matter below 30%, and organic matter (OM) content above 3.5%. Rock and gravel content was under 1%, and all samples were free from contaminants such as plastic, glass, metals, and sharp materials. The pH ranged from 8.00 to 8.55, with electrical conductivity (EC) between 1.97 and 2.55 dS/m. More than 90% of the material was decomposed, and all fertilizer types exceeded the 2005 standards set by the Department of Agriculture. Bioaugmentation with nitrogen-fixing bacteria significantly improved nutrient composition. OM increased from 17.33% to 22.33% in compost, and from 27.61% to 28.66% in vermicompost. Nitrogen content increased from 0.87% to 1.12% in compost and from 1.38% to 1.43% in vermicompost. Phosphorus content increased from 0.068% to 0.108% in compost and from 0.188% to 0.240% in vermicompost. Potassium content increased from 0.16% to 0.21% in compost and from 1.39% to 2.13% in vermicompost. In conclusion, this study provides a valuable framework for improving composting efficiency and converting organic waste into high-quality biofertilizers. The findings highlight the potential of microbial enrichment in enhancing the quality and nutrient composition of compost for agricultural applications.

Keywords: Bioaugmentation, Organic fertilizer, Azotobacter vinelandii, Vermicompost

1. Introduction

The increasing global demand for sustainable agricultural practices has accelerated the development of organic fertilizers as environmentally friendly alternatives to synthetic chemicals. Compost and vermicompost, derived from biodegradable waste, are widely recognized for their ability to improve soil fertility, structure, and microbial diversity (Chen et al., 2020; Manzoor et al., 2024). However, the nutrient content and quality of these organic fertilizers can vary significantly depending on the type of raw materials and composting conditions (Yang et al., 2021; Chaudhary et al., 2023).

The food processing industry generates substantial amounts of fruit and vegetable waste that, if improperly managed, contribute to environmental pollution and greenhouse gas emissions (Wei et al., 2017; Genetu, 2024). Converting these organic residues into compost aligns with the principles of the circular economy by creating value-added products while reducing environmental impact (Petraglia et al., 2019; Sial et al., 2024). Among various composting techniques, vermicomposting using earthworms such as *Eudrilus eugeniae* offers advantages including faster decomposition and enhanced nutrient content in the final product (Kaur, 2020; Oyege & Balaji Bhaskar, 2023; Sharma & Garg, 2019).

To further enhance the agronomic value of compost, bioaugmentation using nitrogen-fixing bacteria such as *Azotobacter vinelandii* has been proposed. These microorganisms improve the availability of essential nutrients, stimulate plant growth, and increase the efficiency of compost as a biofertilizer. *Azotobacter* sp. is a nitrogen-fixing bacterium that plays an important role in the agricultural ecosystem. It can extract nitrogen from the air and convert it into a form that plants can use, reducing dependence on chemical fertilizers and increasing soil fertility. Therefore, using *Azotobacter* sp. in biofertilizers is a sustainable approach to increase crop yields. The properties of *Azotobacter* sp. in biofertilizers include nitrogen fixation from atmospheric nitrogen, production of plant growth promoters (PGPs) such as auxins, gibberellins, and cytokinins, enhancement of soil fertility by improving soil structure and promoting the growth of beneficial microorganisms, improving the absorption of nutrients such as phosphorus and potassium by plants, and producing extracellular polysaccharides (EPS), which help improve the physical properties of the soil. Therefore, using *Azotobacter* sp. in biofertilizers is an approach that promotes environmentally friendly agriculture and helps plants grow more robustly (Sumbul et al., 2020; O'Callaghan et al., 2022; Martin del Campo et al., 2022).

According to the Thai Organic Fertilizer Standards (TAS 2005; TIS 2731-2009), organic fertilizers must meet specific criteria: moisture and volatile matter must be below 30%, organic matter content should exceed 20% by dry weight, and the minimum required levels of total nitrogen (N), phosphorus (P), and potassium (K) are 1.0%, 0.1%, and 0.5%, respectively. Additionally, fertilizers should be free from contaminants such as plastics, metals, and pathogenic microorganisms (Department of Agriculture, 2005; Thai Industrial Standards Institute, 2009).

Therefore, the objective of this study was to evaluate the effects of microbial enrichment using *A. vinelandii* on the physicochemical properties of compost and vermicompost produced from fruit and vegetable waste. The study aimed to compare the resulting biofertilizers with national organic fertilizer

standards in order to assess their potential for agricultural application.

2. Materials and methods

2.1 Preparation of compost and vermicompost from vegetable and fruit waste

2.1.1 Material preparation

Fruit and vegetable scraps were collected from local fresh markets in Maha Sarakham, Thailand. To reduce variability and ensure sample uniformity, the waste was limited to commonly available items such as cabbage, carrot, papaya peel, and banana peel. All materials were collected within 24 hours of discard, chopped into pieces (2–5 cm), and stored in sealed plastic containers at 4°C to prevent premature decomposition prior to composting.

Cattle manure was obtained from a small-scale organic dairy farm in the same area. Fresh manure was aged under shade for 7 days to reduce ammonia content and partially stabilize the material before mixing. It was stored under ambient conditions, protected from sunlight and rain, until use.

The initial composting mixture was prepared by blending fruit and vegetable scraps with cattle manure in a weight ratio of 2:1. The carbon-to-nitrogen (C/N) ratio of the mixture was calculated based on standard values for total carbon and nitrogen content of each material, as reported by Tiquia and Tam (2002). The calculated initial C/N ratio ranged between 25:1 and 30:1, which is considered optimal for aerobic composting (Lv et al., 2018). This range supports rapid microbial activity while minimizing nitrogen losses through volatilization.

2.1.2 Composting process

Composting was conducted in a 160-liter rotating compost bin, designed to enhance the efficiency of the composting process (Matheri et al., 2018). The composting process was prepared using the following steps: 1) chopping the prepared vegetable and fruit waste into small pieces to accelerate decomposition; 2) layering the materials in the bin, alternating between the vegetable and fruit processing waste, cattle manure, and dry leaves; 3) Spraying bio-extract or EM solution (effective microorganisms) evenly over each layer; 4) moistening the compost pile by spraying water, ensuring a moisture level of 50-60%; 5) Turning the compost by rotating the bin 3-5 times every 3-5 days to increase oxygen availability and accelerate decomposition; and 6) monitoring the compost for 30 days, checking temperature, pH and odor to track fermentation progress. The steps in preparing compost are shown in Figure 1.

2.1.3 Vermicomposting process

The vermicomposting process was conducted using a cement block worm bed for vermiculture. The bedding material was prepared by soaking cattle manure (4 parts) in water for one week. A mixture of fine rice bran (1 part), microbial solution (1 part), molasses (1 part), and water (3 parts) was then thoroughly combined and fermented for one week. After fermentation, 30 kg of the prepared mixture was added to the worm bed, and the moisture level was adjusted to 70-80%, following the method of Ferreira Ponciano Ferraz et al. (2020). Mature earthworms were selected and introduced into the rearing beds at a rate of 50 grams per bed or 1 kilogram per square meter of rearing area. The worms were fed 1

kilogram of organic waste, including Chinese cabbage leaves, cabbage leaves, and pineapple peels, every week. The rearing area was rotated once a week to facilitate decomposition, and water was applied twice per week to maintain adequate moisture, ensuring it remained damp but not overly wet. This process was maintained for two weeks. The collection of vermicast (worm castings) was performed by harvesting from the surface of the bedding material, where the castings appeared as dark brown, delicate, and crumbly granules. The vermicomposting was then air-dried in a shaded area for 3-5 days before being sieved using a screening tray to obtain a refined final product (Figure 2).



Figure 1. Steps in the composting process using a 160-liter rotating bin. (a) Preparation and layering of compost materials, including fruit and vegetable waste, cattle manure, and dry leaves. (b) Aerobic fermentation and regular rotation of the compost bin to enhance decomposition.

(c) Final compost product after 42 days of fermentation.

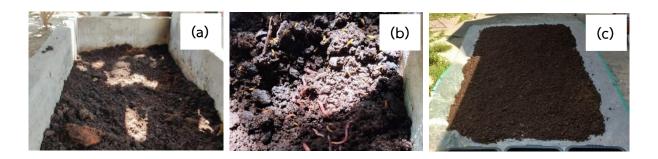


Figure 2. Vermicomposting process using a cement-block worm bed. (a) Preparation of bedding material with fermented cattle manure and rice bran mixture. (b) Introduction of *E. eugeniae* earthworms and weekly feeding with vegetable waste. (c) Collection and refinement of vermicast after 10–14 days.

2.1.4 Moisture control and EM application

To enhance microbial activity and accelerate decomposition during composting, an effective microorganism (EM) solution was prepared at a concentration of 1 liter of EM per 10 liters of clean water. The solution was sprayed evenly onto each compost layer during the layering process. Moisture content of the composting mixture was carefully maintained within the optimal range of 50–60%, which is suitable for aerobic microbial activity. Moisture was monitored regularly and adjusted as needed by spraying water to ensure uniform moisture distribution throughout the composting material.



2.1.5 Microbial inoculation and application ratio

For microbial enrichment, A. vinelandii was cultured in nitrogen-free liquid medium at 37°C for 5 days and then diluted to an optical density (OD₆₀₀) of 1.0, corresponding to approximately 1.2×10^8 CFU/mL. A standardized volume of 100 mL of this bacterial suspension was applied per 1 kg of semi-dry compost or vermicompost. This inoculation rate, based on published protocols and preliminary trials, was selected to ensure effective microbial colonization. The suspension was evenly mixed into the matured compost or vermicompost to achieve uniform distribution.

2.1.6 Replication and experimental units

All treatments were performed in triplicate (n = 3) to allow for statistical analysis. Each replicate represented an independently prepared composting or vermicomposting unit, processed under identical environmental and procedural conditions. This design ensured the consistency and reliability of the experimental data across treatments. Consequently, a total of 12 independent experimental units (4 treatments \times 3 replications) were prepared.

2.2 Preparation of nitrogen-fixing microorganisms

A. vinelandii was initially cultured on nitrogen-free agar medium at 37°C for 5 days. The bacterial cells were then transferred to a nitrogen-free liquid medium for suspension preparation. The cultivation was conducted in 500 mL conical flasks containing 250 mL of medium, incubated at 37°C for 5 days under controlled conditions (adapted from Saraphirom et al., 2024). The suspension of nitrogen-fixing bacteria was diluted in saline solution to be used as a bacterial inoculum for augmenting organic fertilizers used in this study (Figure 3).

2.3 Experimental design

The independent variable was the bioaugmentation of compost and vermicompost with *A. vinelandii* to improve nutrient content, organic matter composition, and fertilizer stability. The experiment was arranged in a Completely Randomized Design (CRD). Four types of organic fertilizers (Figure 3b) were examined: 1) compost, 2) vermicompost processed by *E. eugeniae*, 3) compost augmented with *A. vinelandii*, and 4) vermicompost augmented with *A. vinelandii*. The physical and chemical properties of these fertilizers were analyzed and compared with Thai organic fertilizer standards to evaluate their maturity and effectiveness.

2.4 Organic matter content and nutrient composition analysis

The analysis of pH, electrical conductivity (EC), organic matter percentage (%OM), and the concentrations of essential nutrients, including nitrogen, phosphorus, and potassium, was conducted using standard laboratory methods (adapted from Barney et al., 2020). Organic matter content was determined using the loss on ignition (LOI) method, in which dried samples were incinerated at 550°C for 4 hours, with %OM calculated based on weight loss. Nitrogen concentration was analyzed using the Kjeldahl method, where samples were digested with sulfuric acid (H₂SO₄) and a catalyst to convert organic nitrogen into ammonia (NH₃). The ammonia was then distilled and titrated to determine total nitrogen content. Phosphorus concentration was measured using spectrophotometry, applying Bray-1 extractant for low-pH samples and Olsen extractant for high-pH samples. The phosphorus content was quantified by measuring

light absorbance at 882 nm. Potassium concentration was determined using flame photometry, in which potassium was extracted from the sample using ammonium acetate (NH_4OAc) and analyzed by comparing readings with standard solutions.

2.5 Statistical analysis

All experimental data were analyzed using descriptive statistics and one-way analysis of variance (ANOVA) to determine the significance of differences among the four fertilizer treatments: T1 (compost), T2 (vermicompost), T3 (compost + A. vinelandii), and T4 (vermicompost + A. vinelandii). Data from each treatment were collected in triplicate (n = 3), and results were expressed as mean \pm standard deviation (SD). When significant differences were detected (p<0.05), Tukey's Honestly Significant Difference (HSD) post hoc test was performed to identify pairwise differences among treatment means. Statistical analyses were conducted using IBM SPSS Statistics (Version 26), and results were interpreted at a 95% confidence level.



Figure 3. The experimental design includes (a) the dilution of *A. vinelandii* cells, (b) 4 types of organic fertilizers used in this study.

3. Result

3.1 Composting temperature profile

The composting process exhibited a typical four-phase temperature pattern: mesophilic, thermophilic, cooling, and maturation. The highest temperature observed during the thermophilic phase was 50.33°C, which is within the optimal range for efficient organic matter decomposition and pathogen reduction. Over the 42-day composting period, temperature gradually decreased and stabilized near ambient levels, indicating completion of the composting cycle (Figure 4).

Although no formal statistical analysis was conducted to assess the significance of temperature changes across phases, the average temperature within each phase was calculated and expressed as mean \pm standard deviation (SD) to illustrate variation and consistency in the data. The mesophilic phase (day 1–5) exhibited an average temperature of $30.5 \pm 2.5^{\circ}$ C, increasing to $45.6 \pm 3.1^{\circ}$ C during the thermophilic phase (day 6–15). This was followed by a gradual decline during the cooling phase (days 16-30) with a mean of $35.0 \pm 1.8^{\circ}$ C, and finally stabilization in the maturation phase (days 31-42) at $26.1 \pm 0.4^{\circ}$ C. These

observations reflect microbial activity dynamics and thermal behavior throughout the composting process. However, caution should be exercised when interpreting phase differences, as the current study did not include statistical hypothesis testing to confirm significance between phases.

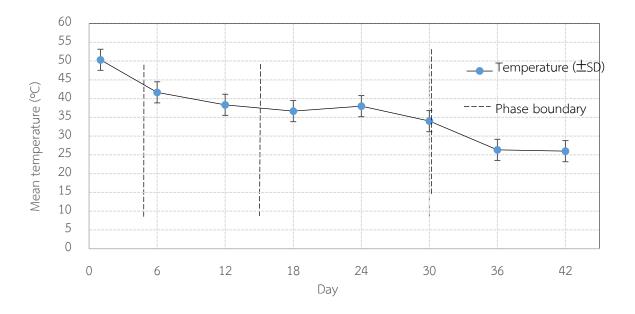


Figure 4. Mean compost temperature \pm SD over 42 days (n = 3), showing mesophilic, thermophilic, cooling, and maturation phases. Vertical dashed lines represent phase boundaries.

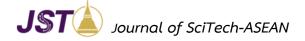
3.2 Evaluation of physicochemical properties of organic fertilizers compared to standard values

3.2.1 pH measurement

The pH values of the organic fertilizers were determined using a pH meter. The results indicated that at the final stage of composting (42 days), the pH values ranged between 8.0 and 8.55, which falls within the acceptable range for organic fertilizers according to the Thai Agricultural Standard (TAS 2005), as shown in Table 1. During the composting process, the pH typically starts at approximately 5.5 and gradually increases to around 7.0-8.0 due to microbial activity and the decomposition of organic matter. Over time, as the process stabilizes, the pH may decline to approximately 6.0-7.0, which is considered an optimal range for balanced compost.

3.2.2 Electrical conductivity (EC) measurement

The electrical conductivity (EC) of four different fertilizer formulations compost, compost mixed with beneficial microorganisms, vermicompost, and vermicompost mixed with beneficial organisms was analyzed. The results, presented in Table 1, showed that EC values ranged from 1.97 to 2.55 dS/m, indicating that all formulations met the recommended EC threshold of ≤ 10 dS/m (TAS 2005). These values confirm that the electrical conductivity of the tested organic fertilizers is within the acceptable limits for safe agricultural application.



3.3 Evaluation of organic matter content and nutrient composition

The study evaluated the organic matter content and nutrient composition (nitrogen, phosphorus, and potassium) of different organic fertilizers, including compost and vermicompost with microbial supplementation. The results were compared against the Thai organic fertilizer standard (TIS 2731-2009) (Sukitprapanon et al., 2020) to determine their compliance and effectiveness as organic fertilizers. Organic matter content plays a crucial role in improving soil structure and fertility. Table 2 showed variations among different treatments. Compost had the lowest %OM, indicating a lower degree of decomposition. Vermicompost processed by *E. eugeniae* exhibited a significantly higher %OM, suggesting enhanced organic matter stabilization. Compost enriched with *A. vinelandii* contained 22.33 ± 0.16%, indicating moderate improvement in organic matter content. Vermicompost supplemented with *A. vinelandii* had the highest %OM, demonstrating the combined benefits of vermicomposting and microbial enrichment.

Table 1. Evaluation of pH and electrical conductivity (EC) measurements of organic fertilizers compared to standard values

No.	Treatment	рН		[EC	
		TAS 2005	Observed	TAS 2005	Observed	
T1	Compost		8.50±0.05		2.55±0.05	
T2	Vermicompost processed by	_	8.40±0.05		2.05±0.04	
	E. eugeniae					
T3	Compost enriched with	5.50-8.50	8.00±0.04	<10 dS/m	2.46±0.04	
	A. vinelandii					
T4	Vermicompost supplemented	_	8.00±0.05		1.97±0.05	
	with A. vinelandii					

According to the Thai organic fertilizer standard, organic fertilizers should contain at least 20% organic matter. Based on this criterion:

- 1) The compost alone did not meet the requirement.
- 2) The compost enriched with A. vinelandii marginally met the requirement.
- 3) Both vermicompost treatments exceeded the minimum %OM standard.

The total nutrient content in terms of nitrogen (N), phosphorus (P), and potassium (K) was analyzed and compared to the Thai standard (Thammayod et al., 2025), which requires:1) total nitrogen (N) \geq 1.0%, 2) total phosphorus (P) \geq 0.1% and 3) total potassium (K) \geq 0.5%. The experimental results showed that the nitrogen content (N) found in the compost (0.87%) was slightly lower than the standard. In comparison, all vermicompost formulas of earthworm compost had nitrogen content that met or exceeded the standard of 1.0%. The vermicompost supplemented with *A. vinelandii* was found to have the highest nitrogen content, while the phosphorus (P) content found in the compost did not meet the standard. Meanwhile, the compost supplemented with *A. vinelandii* and both vermicompost formulas exceeded the minimum phosphorus standard. The potassium (K) content found in the compost and the compost supplemented

with *A. vinelandii* were significantly lower than the standard. Only the vermicompost processed by *E. eugeniae* met the standard, while the vermicompost supplemented with *A. vinelandii* was slightly below the specified level.

One-way ANOVA showed a statistically significant difference in organic matter (%OM) content among the four treatments (p<0.05). Post hoc analysis using Tukey's HSD test indicated that Treatment 3 (compost + A. vinelandii) had significantly higher %OM than Treatment 1 (control compost), while there was no significant difference between Treatment 2 (vermicompost) and Treatment 4 (vermicompost + A. vinelandii), suggesting that microbial enrichment had limited effect on vermicompost in terms of %OM.

Table 2. Evaluation of organic matter content and nutrient composition of organic fertilizers

No.	Treatment	%OM -	Nutrient composition (%)*		
			Nitrogen (N)	Phosphorus (P)	Potassium (K)
T1	Compost	17.33 ^d ±0.2752	0.87 ^d ±0.0138	0.07 ^d ±0.0020	0.16°±0.0010
T2	Vermicompost		1.38 ^b ±0.0105	0.19 ^b ±0.0030	1.391°±0.0418
	processed by	27.61 ^b ±0.2101			
	E. eugeniae				
Т3	Compost enriched with	22.33°±0.1589	1.12 ^c ±0.0079	0.11 ^c ±0.0045	0.14 ^d ±0.0032
	A. vinelandii	22.33 ±0.1309			
T4	Vermicompost		1.43°±0.0079	0.24 ^a ±0.0055	0.21 ^b ±0.0026
	supplemented with	28.66°±0.1589			
	A. vinelandii				

^{*}Different superscript letters (a, b, c, d) within each column indicate significant differences between treatments at p<0.05 according to Tukey's HSD test.

4. Discussion

4.1 Composting temperature

The composting process in this study followed the typical thermal stages—mesophilic, thermophilic, cooling, and maturation (Mahapatra et al., 2022). The peak temperature of 50.33°C falls within the effective range for pathogen reduction and organic matter degradation (Matheri, 2024). The rapid rise from 22–30°C to 50.33°C reflects mesophilic microbial metabolism (Mironov et al., 2023), while the subsequent thermophilic phase (down to 38.3°C) facilitated macromolecule breakdown, enhancing nutrient availability (Tiquia et al., 2002; Jiang et al., 2011). The gradual decline during cooling (36.6–34.0°C) and stabilization during maturation (26.3–26.0°C) indicate reduced thermophilic activity and compost readiness (Sharma et al., 2023; Matheri, 2024). These results confirm the effectiveness of composting, consistent with prior findings that emphasize the roles of microbial activity, oxygen, and moisture control (Ji et al., 2023). Process optimization should focus on maintaining adequate aeration and avoiding excess moisture, which can hinder microbial efficiency and lead to anaerobic conditions (Tran et al., 2024).



4.2 Physicochemical properties of organic fertilizers compared to standard values4.2.1 pH

The pH values of the organic fertilizers ranged from 8.00 to 8.55, all within the acceptable range set by the Thai Agricultural Standard (TAS 2005). This aligns with known composting trends, where an initially acidic pH (5.50–6.00) gradually increases due to microbial degradation and ammonia release during the thermophilic stage (Ryue et al., 2020; Sharma et al., 2023). The slightly lower pH in *A. vinelandii-*enriched compost (8.00) compared to the non-enriched version (8.50) may result from nitrogen fixation and balanced organic acid production (Barney, 2020). Vermicompost formulations, with or without *A. vinelandii*, maintained stable pH values around 8.00–8.40, consistent with mature compost (Tran et al., 2024). These values fall within the optimal pH range (6.00–8.50) for nutrient availability and microbial function (Mironov et al., 2023), confirming the agricultural suitability of all compost types tested in this study.

4.2.2 Electrical conductivity (EC)

The electrical conductivity (EC) of the organic fertilizers ranged from 1.97 to 2.55 dS/m, well below the Thai standard limit of 10 dS/m (TAS 2005). EC reflects the concentration of soluble salts, which influences soil salinity and plant health (Gondek et al., 2020). All values observed in this study fall within the typical range for well-composted materials and pose no risk of salt toxicity (Younis et al., 2022). The highest EC (2.55 dS/m) was recorded in compost alone, likely due to elevated mineralization and ammonium levels. In contrast, the lowest EC (1.97 dS/m) in vermicompost with *A. vinelandii* suggests improved nutrient stabilization and reduced salt buildup from microbial activity (Zhang et al., 2024). These results align with recommendations indicating that EC values of 2.0–4.0 dS/m are ideal for compost use without inducing salinity stress (Suvendran et al., 2024). Therefore, all formulations in this study are confirmed as suitable for sustainable agricultural application.

4.3 Organic matter content and nutrient composition

Organic fertilizers are essential for improving soil fertility through the provision of organic matter (OM) and nutrients. According to the Thai Organic Fertilizer Standard (TAS 2005), a minimum of 20% organic matter (OM) is required. In this study:

- Compost alone fell below the standard, indicating a need for process optimization.
- Compost enriched with *A. vinelandii* slightly surpassed the threshold, likely due to microbial enhancement of OM stabilization rather than increased bulk content.
- Both vermicompost formulations exceeded the OM standard, consistent with findings that vermicomposting improves OM retention and nutrient availability (Sande et al., 2024).

The highest OM and nutrient levels were observed in vermicompost with *A. vinelandii*, confirming synergistic effects between earthworm activity and microbial inoculation. Earthworms and their microbiota enhance organic degradation while preserving stable compounds (Pereira et al., 2022), and contribute to the formation of humic substances that benefit soil structure and nutrient supply (Maffia et al., 2024). However, no significant difference in OM was found between vermicompost with and without *A. Vinelandii*, suggesting a performance ceiling in OM enhancement due to already optimal microbial activity.

The application of *A. vinelandii*—a nitrogen-fixing and phosphorus-solubilizing bacterium—also improved N and P contents (Mahmud et al., 2020; Panday et al., 2024). Still, the most consistent nutrient enhancement was from vermicompost-based treatments. These outcomes confirm that integrating microbial inoculants with vermicomposting can yield high-quality organic fertilizers suited for sustainable farming (Asadu et al., 2024; Dhayalan & Karuppasamy, 2021).

The broader use of microbial biofertilizers is supported by advances in microbiome engineering, which enable the development of resilient and efficient microbial consortia (Nadarajah & Abdul Rahman, 2023). Yet, field variability remains a challenge, necessitating robust formulations and diverse site trials (O'Callaghan et al., 2022). Continued research will be key to ensuring the reliability and scalability of these bio-based solutions.

4.4 Bioaugmentation of compost and vermicompost with *A. vinelandii* for enhanced nutrient content

Bioaugmentation of compost with beneficial microorganisms enhances nutrient content and bioavailability. Such biofertilizers—particularly those incorporating plant growth-promoting microorganisms (PGPMs)—increase essential nutrients (N, P, K), improve soil structure, enhance water retention, promote microbial diversity, and reduce reliance on chemical inputs. While compost, biochar, and PGPMs have individually shown agronomic benefits, their integrated use remains underutilized. Biochar, in particular, provides a stable, porous carrier that supports microbial viability (Bamdad et al., 2022). Organomineral fertilizers (OMFs) derived from organic-mineral blends also offer promise for circular agriculture, though adoption remains limited due to accessibility and market barriers (Bouhia et al., 2022).

In this study, compost and vermicompost enriched with *A. vinelandii* showed significantly increased total nitrogen and phosphorus, particularly in Treatment 3 (compost + *A. vinelandii*). The improvement in phosphorus likely results from phosphate-solubilizing activity via secretion of organic acids such as gluconic and 2-ketogluconic acids. Interestingly, potassium peaked in vermicompost alone (Treatment 2) but declined in Treatment 4 (vermicompost + *A. vinelandii*), possibly due to microbial antagonism or altered nutrient metabolism (Zhang et al., 2024).

The *A. vinelandii* strain, isolated from local agricultural soil, demonstrated strong growth in Burk's medium and formed a mucilaginous biofilm—a trait of nitrogen-fixing and phosphate-solubilizing organisms. Although this bacterium is also known to produce IAA, ammonia, and siderophores, these traits were not assessed here. Nonetheless, the observed outcomes align with earlier studies: Matheri (2024) reported improved compost nutrient stability following microbial inoculation, and Chaudhary et al. (2023) found enhanced phosphorus and potassium availability when compost was co-applied with PGPR and biochar.

Despite promising nutrient enhancements, this study did not assess direct plant growth impacts. Thus, potential agronomic benefits are inferred from literature and known microbial functions. Future work should include greenhouse or field trials to evaluate crop performance, nutrient uptake, and the impacts on soil health. Additionally, ensuring microbial viability during storage and application will be vital for successful scaling and field implementation of these bioaugmented fertilizers.

5. Conclusions and recommendations

5.1 Conclusions

This study evaluated the quality of compost and vermicompost formulations enriched with *A. vinelandii*, focusing on organic matter and nutrient content in comparison with the Thai Organic Fertilizer Standard (TAS 2005). The key findings are as follows:

- Traditional compost alone did not meet the required 20% organic matter threshold.
- Compost enriched with *A. vinelandii* showed marginal improvement, indicating limited effectiveness of microbial inoculation when used alone.
- Both vermicompost formulations exceeded the organic matter standard and provided superior nutrient profiles, affirming the advantages of vermicomposting.

Additionally, all tested fertilizers met the standard pH and electrical conductivity (EC) ranges, confirming their suitability for agricultural use. These results support the use of microbial enrichment, particularly in combination with vermicomposting, as a promising strategy for producing high-quality organic fertilizers. However, further validation under field conditions and long-term application is necessary to ensure stability, scalability, and practical effectiveness.

5.2 Recommendations

5.2.1 Optimize composting strategies

Improve raw material selection, aeration, and microbial inoculation protocols to enhance organic matter retention and microbial activity.

5.2.2 Study long-term nutrient dynamics

Future research should focus on nutrient release patterns, microbial succession, and crop responses to evaluate agronomic performance.

5.2.3 Evaluate performance in diverse soils

Field trials across various soil types are necessary to evaluate the impact on soil health, nutrient cycling, and yield sustainability.

5.2.4 Assess commercial scalability

Given the success of vermicompost formulations, future work should analyze production cost, market potential, and distribution models to support adoption in sustainable agriculture.

6. References

Asadu, CO., Ezema, CA., Ekwueme, BN., Onu, CE., Onoh, IM., Adejoh, T., & Emmanuel, UO. (2024). Enhanced efficiency fertilizers:

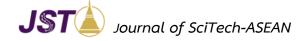
Overview of production methods, materials used, nutrients release mechanisms, benefits and considerations.

Environmental Pollution and Management, 1, 32-48.

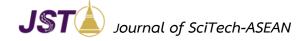
Bamdad, H., Papari, S., Lazarovits, G., & Berruti, F. (2022). Soil amendments for sustainable agriculture: Microbial organic fertilizers. Soil Use and Management, 38(1), 94-120.

Barney, BM. (2020). Aerobic nitrogen-fixing bacteria for hydrogen and ammonium production: current state and perspectives. Applied microbiology and biotechnology, 104(4), 1383-1399.

- Bouhia, Y., Hafidi, M., Ouhdouch, Y., Boukhari, MEME., Mphatso, C., Zeroual, Y., & Lyamlouli, K. (2022). Conversion of waste into organo-mineral fertilizers: current technological trends and prospects. Reviews in Environmental Science and Bio/Technology, 21(2), 425-446.
- Chaudhary, DK., Kim, J., Singh, R., Singh, SK., & Goel, R. (2023). Bioremediation of environmental wastes: The role of microorganisms. Frontiers in Agronomy, 5, 1183691.
- Chen, T., Zhang, S., & Yuan, Z. (2020). Adoption of solid organic waste composting products: A critical review. Journal of Cleaner Production, 272, 122712.
- Department of Agriculture. (2005). Organic fertilizer (TAS 9501-2005). Ministry of Agriculture and Cooperatives, Thailand.
- Dhayalan, V., & Karuppasamy, S. (2021). Plant growth promoting rhizobacteria in promoting sustainable agriculture. Global Journal of Environmental Science and Management, 7(3), 1-18.
- Ferreira Ponciano Ferraz, P., Araújo e Silva Ferraz, G., Leso, L., Klopčič, M., Rossi, G., & Barbari, M. (2020). Evaluation of the physical properties of bedding materials for dairy cattle using fuzzy clustering analysis. Animals, 10(2), 351.
- Genetu, A. (2024). Composting Technology for Municipal Solid Waste Management and Production of Organic Fertilizer. Adv Envi Wast Man Rec, 7(2), 1-13.
- Gondek, M., Weindorf, DC., Thiel, C., & Kleinheinz, G. (2020). Soluble salts in compost and their effects on soil and plants: A review. Compost Science & Utilization, 28(2), 59-75.
- Ji, Z., Zhang, L., Liu, Y., Li, X., & Li, Z. (2023). Evaluation of composting parameters, technologies and maturity indexes for aerobic manure composting: A meta-analysis. Science of The Total Environment, 886, 163929.
- Jiang, T., Schuchardt, F., Li, G., Guo, R., & Luo, Y. (2011). Effect of C/N ratio, aeration rate and moisture content on ammonia and greenhouse gas emission during composting. Journal of Environmental Sciences, 23(10), 1754-1760.
- Kaur, T. (2020). Vermicomposting: An effective option for recycling organic wastes. In Organic agriculture. London: IntechOpen.
- Lv, B., Zhang, D., Cui, Y., & Yin, F. (2018). Effects of C/N ratio and earthworms on greenhouse gas emissions during vermicomposting of sewage sludge. Bioresource technology, 268, 408-414.
- Maffia, A., Marra, F., Canino, F., Battaglia, S., Mallamaci, C., Oliva, M., & Muscolo, A. (2024). Humic Substances from Waste-Based Fertilizers for Improved Soil Fertility. Agronomy, 14(11), 2657.
- Mahapatra, S., Ali, MH., & Samal, K. (2022). Assessment of compost maturity-stability indices and recent development of composting bin. Energy Nexus, 6, 100062.
- Mahmud, K., Makaju, S., Ibrahim, R., & Missaoui, A. (2020). Current progress in nitrogen fixing plants and microbiome research. Plants, 9(1), 97.
- Manzoor, A., Naveed, MS., Ali, RMA., Naseer, MA., Maqsood, UH., Saqib, M., & Farooq, M. (2024). Vermicompost: A potential organic fertilizer for sustainable vegetable cultivation. Scientia Horticulturae, 336, 113443.
- Martin del Campo, JS., Rigsbee, J., Bueno Batista, M., Mus, F., Rubio, LM., Einsle, O., & Dos Santos, PC. (2022). Overview of physiological, biochemical, and regulatory aspects of nitrogen fixation in Azotobacter vinelandii. Critical Reviews in Biochemistry and Molecular Biology, 57(5-6), 492-538.
- Matheri, AN., Mbohwa, C., Ntuli, F., Belaid, M., Seodigeng, T., Ngila, JC., & Njenga, CK. (2018). Waste to energy bio-digester selection and design model for the organic fraction of municipal solid waste. Renewable and sustainable energy reviews, 82, 1113-1121.
- Matheri, F. (2024). Physico-chemical and biological evolution of thermophilic compost as influenced by nitrogenous feedstock and duration [Online]. Retrieved November 1, 2024, from: https://ir-library.ku.ac.ke/server/api/core/bitstreams/e3ab91e5-9181-4200-840c-07dac1304ac7/content.
- Mironov, V., Moldon, I., Shchelushkina, A., Zhukov, V., & Zagustina, N. (2023). Bio-Drying of Municipal Wastewater Sludge: Effects of High Temperature, Low Moisture Content and Volatile Compounds on the Microbial Community. Fermentation, 9(6), 570.



- Muschler, RG. (2016). Agroforestry: essential for sustainable and climate-smart land use. Tropical forestry handbook, 2, 2013-2116.
- Nadarajah, K., & Abdul Rahman, NSN. (2023). The microbial connection to sustainable agriculture. Plants, 12(12), 2307.
- O'Callaghan, M., Ballard, RA., & Wright, D. (2022). Soil microbial inoculants for sustainable agriculture: Limitations and opportunities. Soil Use and Management, 38(3), 1340-1369.
- Oyege, I., & Balaji Bhaskar, MS. (2023). Effects of vermicompost on soil and plant health and promoting sustainable agriculture. Soil Systems, 7(4), 101.
- Panday, D., Bhusal, N., Das, S., & Ghalehgolabbehbahani, A. (2024). Rooted in nature: The rise, challenges, and potential of organic farming and fertilizers in agroecosystems. Sustainability, 16(4), 1530.
- Pereira, MMA., Moraes, LC., Mogollón, MCT., Borja, CJF., Duarte, M., Buttrós, VHT., & Dória, J. (2022). Cultivating biodiversity to harvest sustainability: Vermicomposting and inoculation of microorganisms for soil preservation and resilience. Agronomy, 13(1), 103.
- Petraglia, A., Cacciatori, C., Chelli, S., Fenu, G., Calderisi, G., Gargano, D., & Carbognani, M. (2019). Litter decomposition: effects of temperature driven by soil moisture and vegetation type. Plant and Soil, 435, 187-200.
- Ryue, J., Lin, L., Kakar, FL., Elbeshbishy, E., Al-Mamun, A., & Dhar, BR. (2020). A critical review of conventional and emerging methods for improving process stability in thermophilic anaerobic digestion. Energy for Sustainable development, 54, 72-84.
- Sande, TJ., Tindwa, HJ., Alovisi, AM. T., Shitindi, MJ., & Semoka, JM. (2024). Enhancing sustainable crop production through integrated nutrient management: a focus on vermicompost, bio-enriched rock phosphate, and inorganic fertilisers—a systematic review. Frontiers in Agronomy, 6, 1422876.
- Saraphirom, P., Namsena, P., Teerakun, M., Phonphuak, S., Chanprathak, A., Dakaew, S., & Phonphuak, N. (2024). Development of fired clay granules as eco-friendly substances for nitrogen-fixing bacterial cell. GEOMATE Journal, 27(120), 110-121.
- Sharma, K., & Garg, VK. (2019). Vermicomposting of waste: a zero-waste approach for waste management. In Sustainable resource recovery and zero waste approaches (pp. 133-164). New York: Elsevier.
- Sharma, P., Bano, A., Singh, SP., Srivastava, SK., Singh, SP., Iqbal, HM., & Varjani, S. (2023). Different stages of microbial community during the anaerobic digestion of food waste. Journal of Food Science and Technology, 60(8), 2079-2091.
- Sial, TA., Rajpar, I., Khan, MN., Ali, A., Shan, M., Rajput, AB., & Shah, PAN. (2024). Impact of Fruit and Vegetable Wastes on the Environment and Possible Management Strategies. In Planet Earth: Scientific Proposals to Solve Urgent Issues (pp. 307-330). Cham: Springer International Publishing.
- Sukitprapanon, TS., Jantamenchai, M., Tulaphitak, D., & Vityakon, P. (2020). Nutrient composition of diverse organic residues and their long-term effects on available nutrients in a tropical sandy soil. Heliyon, 6(11), e05601.
- Sumbul, A., Ansari, RA., Rizvi, R., & Mahmood, I. (2020). Azotobacter: A potential bio-fertilizer for soil and plant health management. Saudi journal of biological sciences, 27(12), 3634-3640.
- Suvendran, S., Johnson, D., Acevedo, M., Smithers, B., & Xu, P. (2024). Effect of Irrigation Water Quality and Soil Compost Treatment on Salinity Management to Improve Soil Health and Plant Yield. Water, 16(10), 1391.
- Thai Industrial Standards Institute. (2009). TIS 2731-2009: Organic fertilizer. Ministry of Industry, Thailand. (in Thai)
- Thammayod, A., Roschat, W., Leelatam, T., Phewphong, S., Maneerat, B., Leelatam, N., & Promarak, V. (2025). Development of High-Quality Organic Fertilizer from Biomass Waste and the Application of Biochar in Local Areas of Sakon Nakhon Province, Northeastern Thailand. Trends in Sciences, 22(2), 9105-9105.
- Tiquia, SM., & Tam, NF. (2002). Characterization and composting of poultry litter in forced-aeration piles. Process Biochemistry, 37(8), 869-880.
- Tran, HT., Binh, QA., Van Tung, T., Pham, DT., Hoang, HG., Nguyen, NSH., & Bolan, NS. (2024). A critical review on characterization, human health risk assessment and mitigation of malodorous gaseous emission during the composting process. Environmental Pollution, 124115.



- Wei, Y., Li, J., Shi, D., Liu, G., Zhao, Y., & Shimaoka, T. (2017). Environmental challenges impeding the composting of biodegradable municipal solid waste: A critical review. Resources, Conservation and Recycling, 122, 51-65.
- Yang, Z., Muhayodin, F., Larsen, OC., Miao, H., Xue, B., & Rotter, V. S. (2021). A review of composting process models of organic solid waste with a focus on the fates of C, N, P, and K. Processes, 9(3), 473.
- Younis, A., Ahsan, M., Akram, A., Lim, KB., Zulfiqar, F., & Tariq, U. (2022). Use of organic substrates in sustainable horticulture [Online]. Retrieved November 1, 2024, from: https://www.cabidigitallibrary.org/doi/10.1079/9781789248098.0009
- Zhang, G., Bai, J., Zhai, Y., Jia, J., Zhao, Q., Wang, W., & Hu, X. (2024). Microbial diversity and functions in saline soils: A review from a biogeochemical perspective. Journal of advanced research, 59, 129-140.