International Journal of Agricultural Technology 2025 Vol. 21(5):1833-1846
DOI: https://doi.org/10.63369/ijat.2025.21.5.1833-1846
Available online http://www.ijat-aatsea.com
ISSN 2630-0192 (Online)

Fungal community dynamics in vermicomposting systems: A metagenomic analysis

Kraisittipanit, R.[1], Tancho, A.[2,3,4], Aumtong, S.[4] and Charerntantanakul, W.[5*]

[1]Department of Premedical Science, Faculty of Medicine, Bangkokthonburi University, Bangkok, 10170, Thailand; [2]Natural Agriculture Research and Development Center, Maejo University, Chiang Mai 50290, Thailand; [3]Fellow Member of the Academy of Science, the Royal Society of Thailand, Thailand; [4]Faculty of Agricultural Production, Maejo University, Thailand; [5]Program of Biotechnology, Faculty of Science, Maejo University, Thailand.

Kraisittipanit, R., Tancho, A., Aumtong, S. and Charerntantanakul, W. (2025). Fungal community dynamics in vermicomposting systems: A metagenomic analysis. International Journal of Agricultural Technology 21(5):1833-1846.

Abstract The results indicated the highest species diversity in vermicompost, with a Shannon diversity index (D index) of 5.2645, encompassing a total of 55 fungal species. In contrast, the gut of *Perionyx* sp. 1 and the vermicomposting liquid exhibited a D index of 4.4563 (50 fungal species) and 2.3183 (24 fungal species), respectively. In terms of evenness, vermicompost achieved the highest score, with an evenness index (E index) of 0.9456. The Principal Coordinates Analysis (PCoA) diagram revealed overlapping fungal species among the three samples within the vermicomposting system, represented by PC1 and PC2, accounting for 75.07% and 15.14% of the variance, respectively. The analysis showed that the gut of *Perionyx* sp. 1 and vermicompost shared 25 species, while there were 9 shared species between the gut and the vermicomposting liquid. Conversely, the vermicompost and vermicomposting liquid exhibited only 7 common species. The overall similarity among the three samples in the vermicomposting system was limited to 4 fungal species: *Aspergillus flavus*, *A. penicillioides*, *Candida tropicalis*, and *Nigrospora oryzae*. Notably, *C. tropicalis* demonstrated the highest percentage of species richness, comprising 28.41%, while *N. oryzae* achieved the best scores in both vermicompost (0.31%) and vermicomposting liquid (3.43%).

Keywords: Ketare, *Perionyx*, Vermicompost, Earthworm gut, Vermicomposting liquid, Fungi, Metagenome, NGS

Introduction

Ketare is a common name for a Thai species of earthworm, *Perionyx*, which plays a significant role in agriculture throughout Thailand. Morphologically, Ketare resembles *Perionyx excavatus* but is typically found in specific habitats, such as cow manure. This species is categorized based on its habitat and cultivation conditions. Additionally, two distinct varieties have been

[*]**Corresponding Author:** Charerntantanakul, W.; **Email:** wasin@mju.ac.th

identified: *Perionyx* sp. 1, which inhabits cow manure areas, and *Perionyx* sp. 2, which is adapted to higher latitudes and colder climates. For over 20 years, these earthworms have been utilized for producing high-quality vermicompost and vermicomposting liquid at the Natural Farming Research and Development Center at Maejo University.

Research by Tancho (2013) demonstrated that this strain efficiently converts organic waste from vegetable residues sourced from markets into vermicompost through gut-associated processing (GAP) facilitated by microbial activity. Notably, Ketare is particularly well-suited for vermiculture in Thailand compared to other species due to its adaptability to tropical climates, high consumption rates, and capacity to convert animal waste and organic vegetables into superior products, making it an excellent organic fertilizer.

The vermicomposting system is a fermentation process involving earthworms, vermicompost, and vermicomposting liquid. In general, earthworms consume food mixed with soil, digesting it into smaller nutrients via their gut, which harbors a diverse array of microorganisms. The final stage of this digestive process results in the excretion of vermicast, which contains beneficial microorganisms that promote fermentation. The complete fermentation yields organic fertilizer known as vermicompost, while the liquid produced in this process is referred to as vermicomposting liquid. This liquid possesses unique properties, including trace minerals and plant growth hormones (Walia and Kaur, 2024).

Historically, vermicomposting liquid has been tested on various crops for an extended period, demonstrating its ability to stimulate plant growth and inhibit certain pathogens by enhancing plant immunity. It has been reported to effectively suppress diseases caused by pathogens such as Pythium, Rhizoctonia, Verticillium, and Phomopsis (Edwards and Arancon, 2004). This innovation is often described as a "precious soil gift" due to its rich microbial composition, particularly the presence of *Enterobacter hormaechei*, *Enterobacter cloacae*, *Aeromonas punctata*, *Aeromonas sanarellii*, *Aeromonas enteropelogenes*, *Aeromonas media*, and *Bacillus aryabhatta*. These microbes have the potential to produce indole-3-acetic acid in significant concentrations, ranging from 131.39 µg/ml to 32.45 µg/ml (Arraktham *et al.,* 2016).

To investigate the microbial communities associated with *Perionyx* sp. 1, high-throughput sequencing technology, or next-generation sequencing (NGS), was employed (Behjati and Tarpey, 2013). This technique allows for the rapid sequencing of metagenomic data and is widely used across various research fields, particularly in studies focused on the complete genetic structure of individual organisms or at the metagenomic level. For instance, Wang *et al.* (2019) identified Actinobacteria, Firmicutes, and Proteobacteria as the three

dominant bacterial groups in arsenic-contaminated soil absorbed through the gut of *Metaphire sieboldi*. Similarly, NGS has been utilized to examine antibiotic-resistant gene expression in the gut of *Metaphire guillelmi* earthworms (Choa *et al.,* 2019). Additionally, metagenomic analysis has been applied to investigate the diverse microbial communities in the central Gulf of Thailand (Sripan, 2016). Furthermore, Koo *et al*. (2018) demonstrated that metagenomic studies could be used to assess gene expression levels from microbiomes in various cold ecosystems.

Thus, this study aimed o evaluate the entire microbial community associated with *Perionyx* sp. 1, a commercially significant Thai earthworm species at the Natural Agriculture Research and Development Center, by applying NGS technology, and to assess the total fungal community within the microbiome of the vermicomposting system including *Perionyx* sp. 1 gut, vermicompost, and vermicomposting liquid.

Materials and methods

Collection of earthworms and metagenomic extraction

Perionyx sp. 1 earthworms, along with vermicompost and vermicomposting liquid, were sourced from the Natural Farming Research and Development Center at Maejo University. Metagenomic DNA was extracted using the Stool Microbiome DNA Kit (Invitrogen). The quality and quantity of the extracted DNA were assessed using a Nanodrop spectrophotometer (Allsheng) and confirmed via 1% agarose gel electrophoresis. The DNA samples were subsequently stored at -20°C for future analyses.

Library preparation

The metagenomic samples were processed using a transposase enzyme (Macrogen) to fragment the DNA into smaller reads. Different adapters were then utilized to duplicate the fragments. For the amplification of fungal species, the P5 adapter (5’TTGGTCATTTAGAGGAAGTAA 3’) and the P7 adapter (5’ CGTTCTTCATCGATGC 3’) were incorporated as fungal metagenome adapters. Following this, targeted DNA segments were amplified using polymerase chain reaction (PCR) to prepare the sequence reads (see Table 1).

Table 1. Amplicon Primer Sequences for ITS1

Primer Name	Primer Type	Sequence (5' to 3')
ITS1-F	Forward	5' TCCGTAGGTGAACCTGCGG 3'
ITS1-R	Reverse	5' GCTGCGTTCTTCATCGATGC 3'

Operational taxonomic Unit (OTU) analysis and statistical taxonomic analysis

The DNA fragments were assembled into contigs and analyzed for nucleotide sequences using the Illumina MiSeq platform (Macrogen), with a quality control threshold set at Q30 (99.9%). Read alignment was performed using the Fast Length Adjustment of SHort reads (FLASH) tool. The raw data were utilized to analyze taxonomic clustering through a Python program, followed by the generation of OTU output data using the rDnaTool (Python) application. Additionally, raw data and nucleotide sequences were subjected to BLAST analysis for assessing taxonomic diversity and species richness, utilizing the Greengenes database.

Results

The metagenomic analysis of three samples (*Perionyx* sp. 1 gut, vermicompost, and vermicomposting liquid) was performed using primers specific to the ITS1 region to generate amplicons. The read counts obtained were 103,632 for the *Perionyx* sp. 1 gut, 96,692 for vermicompost, and 90,564 for the vermicomposting liquid. These reads were processed and assembled into contigs using the FLASH program. All contigs were evaluated, achieving quality scores of Q30, with percentages of 99.63%, 99.36%, and 98.35% for the respective samples. The total number of bases sequenced in this study was 28,139,070 for the *Perionyx* sp. 1 gut, 27,718,479 for vermicompost, and 27,207,145 for the vermicomposting liquid (Table 2).

Table 2. Analysis using the Fast Length Adjustment of SHort reads (FLASH) program showing the paired score (Q30) and read count for metagenomics of *Perionyx* sp. 1 earthworm gut, vermicompost, and vermicomposting liquid based on the ITS1 region gene

Sample name	Read Count	Quality Score (Q30)	Total Bases
Perionyx sp.1 gut	103,632	99.63%	28,139,070
Vermicompost	96,692	99.36%	27,718,479
Vermicomposting liquid	90,564	98.35%	27,207,145

However, the identification of diversity was conducted using Illumina MiSeq, and statistical diversity was analyzed using the alpha_diversity.py program. This analysis estimated five statistical parameters (Table 3).

Table 3. Statistical analysis using the alpha_diversity.py program for diversity richness (Shannon index and Inverse Simpson index)

Sample name	Species diversity and evenness index	
	Shannon (D index)	Inverse Simpson (E index)
Perionyx sp.1 earthworm gut	4.4563	0.8768
Vermicompost	5.2645	0.9456
Vermicomposting liquid	2.3183	0.5332

The Principal Coordinate Analysis (PCoA) demonstrated that the ITS1 region can effectively identify fungal communities. The percentage variation explained by PC1 and PC2 indicated a higher similarity of fungal species in the *Perionyx* sp.1 gut and vermicompost compared to vermicomposting liquid. The contributions of PC1 and PC2 were estimated at 75.07% and 15.14%, respectively. These findings are illustrated in Figure 1.

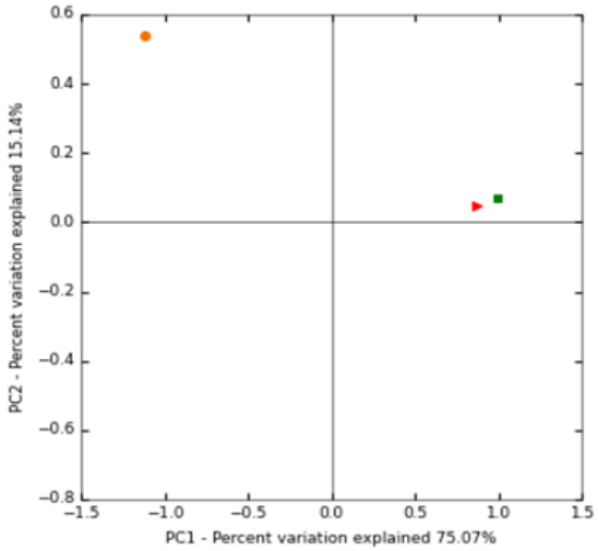

Figure 1. displays the PCoA diagram illustrating the relationship among the mycobiomes of vermicompost (green dots), *Perionyx* sp.1 gut (red dots), and vermicomposting liquid (yellow dots)

The vermicomposting system, which includes the *Perionyx* sp.1 gut, vermicompost, and vermicomposting liquid, exhibited four common fungal species: *Aspergillus flavus*, *A. penicillioides*, *Candida tropicalis*, and *Nigrospora oryzae* (as illustrated in Figure 2 and Table 4). Among these, *C. tropicalis* emerged as the predominant fungal species in vermicompost, accounting for 28.41%. Additionally, *N. oryzae* was identified as the dominant fungal species in

both vermicompost and vermicomposting liquid, with proportions of 0.31% and 3.43%, respectively (Table 4).

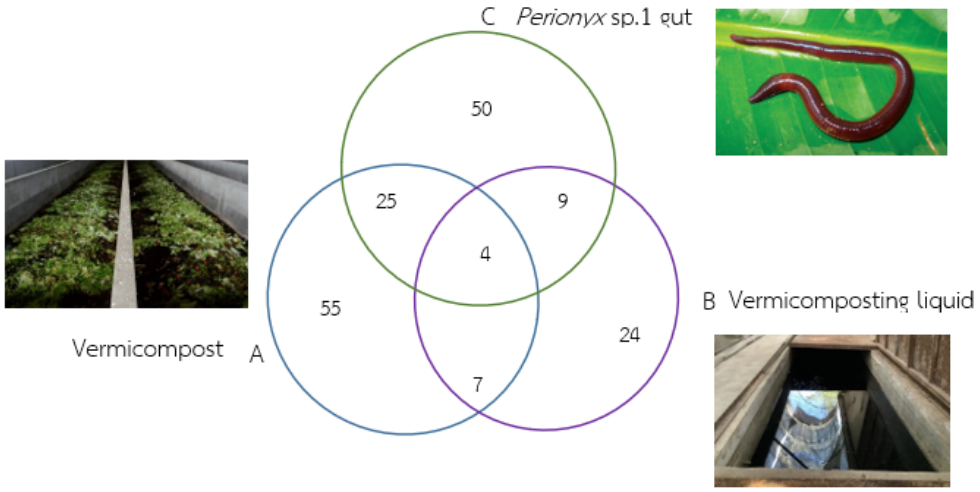

Figure 2. Venn-Euler diagram illustrating the shared number of fungal species among the three microbiomes of the vermicomposting system: earthworm gut, vermicompost, and vermicomposting liquid (A = Vermicompost, B = Vermicomposting Liquid, C = *Perionyx* sp. 1 Gut)

Table 4. The four common fungal species identified in the vermicomposting system

Fungal species	The percentage of species abundance in Vermicomposting system		
	Perionyx sp. 1 gut	Vermicompost	Vermicomposting liquid
Aspergillus flavus	7.89	0.03	0.04
Aspergillus penicillioides	0.59	0.07	0.10
Candida tropicalis	28.41	0.27	0.52
Nigrospora oryzae	0.08	0.31	3.43

The results indicated that the ITS1 region effectively detected the predominant fungal phylum, Ascomycota, followed by Basidiomycota and Mucoromycota (which constituted less than 0%). However, this region was unable to identify other eukaryotes, with approximately 45.9% of sequences remaining unidentified. The distribution was as follows: vermicomposting liquid (69.4%), vermicompost (41.9%), and *Perionyx* sp. 1 (33.5%), as illustrated in Table 5.

Table 5. The percentage of fungal phylum in vermicomposting liquid, vermicompost, and the gut microbiome of *Perionyx* sp. 1

Phyla	*Perionyx* sp. 1 gut (%)	Vermicompost (%)	Vermicomposting liquid
Unidentified eukaryote	33.5	41.9	69.4
Arthropoda	0.5	0.2	0.1
Ascomycota	44.8	45.1	6.0
Basidiomycota	0.4	0.3	1.0
Mucoromycota	0	0	0
Dark matter	20.7	12.4	23.6

The fungal species abundance derived from the metagenomics of the *Perionyx* sp. 1 gut was analyzed using NGS technology, resulting in the identification of 50 species. The most prevalent genus identified was *Candida*. The three most abundant species were *Candida tropicalis* (24.41%), *Aspergillus flavus* (7.89%), and *Fusarium solani* (2.99%), as shown in Table 6.

Table 6. Fungal Genus and Species Analysis of Phylum Ascomycota in *Perionyx* sp. 1 Gut by OTUs

Genus	Species	Species abundance
Hortaea	*Hortaea werneckii*	0.01%
Juxtiphoma	*Juxtiphoma eupyrena*	0.01%
Paraconiothyrium	*Paraconiothyrium* sp.	0.02%
Preussia	*Preussia* sp.	0.06%
Chaetothyriales	*Chaetothyriales* sp.	0.06%
Aspergillus	*Aspergillus chevalieri*	0.63%
Aspergillus	*Aspergillus flavus*	7.89%
Aspergillus	*Aspergillus penicillioides*	0.59%
Aspergillus	*Aspergillus pseudodeflectus*	0.02%
Aspergillus	*Aspergillus niger*	0.09%
Aspergillus	*Aspergillus steynii*	0.02%
Aspergillus	*Aspergillus tamarii*	0.04%
Aspergillus	*Aspergillus versicolor*	0.06%
Aspergillus	*Aspergillus violaceofuscus*	0.01%
Penicillium	*Penicillium cinnamopurpureum*	0.05%
Penicillium	*Penicillium oxalicum*	0.07%
Penicillium	*Penicillium* sp.	0.04%
Talaromyces	*Talaromyces wortmannii*	0.01%
Chaetomella	*Chaetomella* sp.	0.05%
Candida	*Candida tropicalis*	28.41%
Yamadazyma	*[Candida] conglobata*	0.01%
	uncultured Galactomyces	2.07%
Barnettozyma	*Barnettozyma californica*	0.01%
Starmera	*[Candida] stellimalicola*	0.02%
Pichia	*Pichia manshurica*	0.01%

Genus	Species	Species abundance
Colletotrichum	*Colletotrichum truncatum*	0.02%
	Uncultured Sordariomycetidae	0.11%
Clonostachys	*Clonostachys* sp.	0.02%
Fusarium	*Fusarium equiseti*	0.11%
Fusarium	*Fusarium oxysporum*	0.07%
Fusarium	*Fusarium solani*	2.99%
Memnoniella	*Memnoniella longistipitata*	0.01%
Microascus	*Microascus chartarus*	0.02%
Microascus	*Microascus croci*	0.01%
Microascus	*Microascus gracilis*	0.01%
Parascedosporium	*Parascedosporium putredinis*	0.09%
Chaetomium	*Chaetomium globosum*	0.01%
Mycothermus	*Mycothermus thermophiles*	0.06%
Ovatospora	*Ovatospora pseudomollicella*	0.05%
Podospora	*Podospora prethopodalis*	0.09%
Nigrospora	*Nigrospora oryzae*	0.08%
Funneliformis	*Funneliformis* sp	0.05%
Vascellum	*Vascellum pratense*	0.02%
Coprinopsis	*Coprinopsis cinerea*	0.02%
Psathyrella	*Psathyrella bivelata*	0.01%
Amauroderma	*Amauroderma rugosum*	0.02%
Ganoderma	*Ganoderma lucidum*	0.05%
Hericium	*Hericium erinaceus*	0.02%
Malassezia	*Malassezia globosa*	0.01%
	Malassezia restricta	0.04%

The fungal species abundance from the metagenomic analysis of vermicompost was examined using next-generation sequencing (NGS) technology. A total of 55 species were identified, with the highest representation belonging to the genus Fusarium. The three most abundant species were *Fusarium lichenicola* (11.41%), *Microascus murinus* (7.62%), and *Scopulariopsis brevicaulis* (3.73%), respectively. The findings are presented in Table 7.

Table 7. Fungal species analysis of phylum Ascomycota in vermicompost

Genus	Species	Species abundance
Cladosporium	*Cladosporium tenuissimum*	0.07%
	uncultured Eremomyces	0.30%
Neodeightonia	*Neodeightonia subglobosa*	0.01%
Chaetothyriales	*Chaetothyriales* sp.	0.05%
Aspergillus	*Aspergillus candidus*	0.15%
Aspergillus	*Aspergillus chevalieri*	0.24%
Aspergillus	*Aspergillus flavas*	0.03%
Aspergillus	*Aspergillus penicillioides*	0.07%
Aspergillus	*Aspergillus versicolor*	0.78%
Aspergillus	*Aspergillus niger*	0.29%

Genus	Species	Species abundance
Aspergillus	*Aspergillus tamarii*	0.12%
Aspergillus	*Aspergillus unguis*	0.02%
Aspergillus	*Aspergillus versicolor*	0.46%
Monascus	*Monascus ruber*	0.06%
Penicillium	*Penicillium oxalicum*	0.12%
Penicillium	*Penicillium* sp.	0.02%
Xeromyces	*Xeromyces bisporus*	0.13%
Talaromyces	*Talaromyces wortmannii*	0.06%
Arachnomyces	*Arachnomyces* sp.	0.09%
Spiromastix	*Spiromastix* sp.	0.04%
Chaetomella	*Chaetomella* sp.	0.03%
uncultured Kotlabaea		1.91%
Candida	*Candida tropicalis*	0.27%
Galactomyces	Galactomyces sp.	0.02%
Uncultured Sordariomycetidae		0.01%
Uncultured Plectosphaerella		0.59%
Collarina	*Collarina* sp.	0.01%
Acremonium	Acremonium sp.	0.02%
Uncultured Paracremonium		0.01%
Cosmospora	*Cosmospora viridescens*	0.08%
Fusarium	*Fusarium lichenicola*	11.41%
Fusarium	*Fusarium oxysporum*	0.46%
Fusarium	*Fusarium solani*	2.15%
Sarocladium	*Sarocladium strictum*	0.03%
Paramyrothecium	*Paramyrothecium roridum*	0.01%
Stachybotrys	*Stachybotrys* sp.	0.05%
Uncultured Pseudodallescheria		9.01%
Lophotricus	*Lophotricus* sp.	0.09%
Microascus	*Microascus chartarus*	2.69%
Microascus	*Microascus croci*	0.27%
Microascus	*Microascus murinus*	7.62%
Microascus	*Microascus verrucosus*	0.92%
Microascus	*Microascus longicollis*	0.35%
Scopulariopsis	*Scopulariopsis brevicaulis*	3.73%
Scopulariopsis	*Scopulariopsis konigii*	0.04%
Chaetomium	*Chaetomium* sp.	0.06%
Myceliophthora	*Myceliophthora lutea*	0.03%
Mycothermus	*Mycothermus thermophilus*	0.14%
Nigrospora	*Nigrospora oryzae*	0.31%
Xylariales	*Xylariales* sp.	0.06%
uncultured Myriococcum		0.22%
uncultured Ganadomataceae		0.06%
uncultured Thelephoraceae		0.05%
Malassezia	*Malassezia restricta*	0.02%
Saitozyma	*Saitozyma flava*	0.08%

The fungal species abundance from the metagenomics of vermicomposting liquid was analyzed using NGS technology. A total of 24 species were identified, with the highest representation in the genus Nigrospora. The most abundant species identified was *Nigrospora oryzae*, comprising 3.43% of the total. These findings are presented in Table 8.

Table 8. Fungal species analysis of phylum Ascomycota in Vermicomposting liquid

Genus	Species	Species abundance
Cladosporium	*Cladosporium tenuissimum*	0.76%
Pseudocercospora	*Pseudocercospora* sp.	0.01%
Stagonosporopsis	*Stagonosporopsis* sp.	0.20%
Aspergillus	*Aspergillus flavus*	0.04%
	Aspergillus penicillioides	0.10%
	Aspergillus niger	0.01%
Candida	*Candida tropicalis*	0.52%
Debaryomyces	*Debaryomyces coudertii*	0.02%
Clavispora	*Clavispora akabenensis*	0.01%
Kazachstania	*Kazachstania humilis*	0.02%
Colletotrichum	*Colletotrichum nymphaeae*	0.47%
Fusarium	*Fusarium equiseti*	0.28%
Nigrospora	*Nigrospora oryzae*	3.43%
Eutypella	*Eutypella* sp.	0.08%
Coprinopsis	*Coprinopsis cinerea*	0.17%
Auricularia	*Auricularia cornea*	0.02%
Earliella	*Earliella scabros*	0.16%
Sterigmatomyces	*Sterigmatomyces halophilus*	0.13%
Malassezia	*Malassezia globosa*	0.09%
	Malassezia restricta	0.25%
Tremelloles	*Tremelloles* sp.	0.14%
Hannaella	*Hannaella sinensis*	0.02%
	Hannaella sp.	0.02%
Lichtheimia	*Lichtheimia corymbifera*	0.01%

Discussion

The findings of this study are provided significant insights into the fungal community dynamics within a vermicomposting system, comprising the gut of *Perionyx* sp. 1, vermicompost, and vermicomposting liquid. Utilizing high-throughput sequencing of the ITS1 region, the study revealed key differences in fungal species diversity and abundance across the three distinct environments. These insights enhance our understanding of microbial interactions in vermicomposting systems, particularly regarding the decomposition of organic matter and nutrient cycling.

The vermicompost exhibited the highest fungal diversity, with a Shannon diversity index (D index) of 5.2645 and 55 fungal species. This result is consistent with previous findings that compost systems enriched with organic material tend to support diverse microbial communities, primarily due to the abundant nutrient availability required for microbial growth. Jiménez *et al.* (2017) reported similar observations in compost systems, emphasizing that organic matter-rich environments typically foster a highly diverse and active microbial ecosystem. Moreover, the study by Gomes *et al*. (2021) highlighted the pivotal role of fungal communities in compost environments, where they contribute substantially to organic waste degradation and nutrient cycling.

Interestingly, the gut of *Perionyx* sp. 1 showed slightly lower diversity (D index 4.4563), with 50 fungal species identified. Earthworms are known to facilitate microbial transformations during organic waste digestion. The microbial diversity within the gut suggests that earthworms act as bioreactors, transforming organic matter into nutrient-rich by-products through their gut-associated microbial communities. Aira *et al*. (2016) also noted that earthworm activity alters microbial diversity, enhancing the microbial profile in compost environments. The microbial transfer from the gut to the vermicompost reinforces the important ecological role earthworms play in shaping fungal community dynamics, as also suggested by Liu *et al*. (2020), who demonstrated that earthworm gut microbiomes significantly influence microbial diversity and functionality in compost systems.

The vermicomposting liquid exhibited the lowest diversity (D index 2.3183), with only 24 fungal species identified. This reduced diversity can be attributed to the distinct environmental conditions of vermicomposting liquid, such as high moisture content and limited oxygen availability, which impose selective pressures favoring a narrower range of fungal taxa. These findings align with Edwards and Arancon (2004), who observed that liquid environments in composting systems typically support a less diverse microbial community due to such environmental constraints. The lower evenness index (E index 0.5332) in the liquid, compared to the higher evenness in vermicompost (E index 0.9456), indicates that a few dominant fungal species prevail in the liquid, likely adapting to its unique physicochemical properties. Frąc *et al.* (2015) found similar patterns in liquid compost systems, where specific fungal taxa adapted to low-oxygen environments outcompeted others, leading to a decrease in overall microbial diversity.

The Principal Coordinate Analysis (PCoA) further demonstrated the relationship between the fungal communities in vermicompost and the gut of *Perionyx* sp. 1. The analysis revealed significant similarities between the two environments, with PC1 and PC2 explaining 75.07% and 15.14% of the variance, respectively. These findings suggest that the fungal communities in the gut of *Perionyx* sp. 1 are transferred into the vermicompost during digestion,

influencing the microbial profile of the compost. This microbial transfer has been shown to enhance nutrient cycling, as earthworms break down organic material and facilitate the proliferation of beneficial fungal species in compost. Lv *et al*. (2018) corroborated these findings, demonstrating that earthworm gut microbiota significantly impacts the fungal diversity of the surrounding compost environment, contributing to the degradation of complex organic compounds.

Four fungal species—*Aspergillus flavus*, *Aspergillus penicillioides*, *Candida tropicalis*, and *Nigrospora oryzae*—were found across all three environments, highlighting their ecological significance in the vermicomposting system. The dominance of *Candida tropicalis* in vermicompost (28.41%) suggests that it plays a key role in the decomposition of organic matter, particularly in the breakdown of complex polysaccharides like cellulose and lignin. Yadav *et al.* (2019) also identified *Candida tropicalis* as a critical fungal species in compost systems, where it contributes to the degradation of organic matter, thereby enhancing the efficiency of vermicomposting processes. Similarly, Huang *et al.* (2020) reported that *Candida tropicalis* thrives in compost environments, promoting nutrient cycling and decomposition through its enzymatic activities.

Nigrospora oryzae was also found in both vermicomposting liquid (3.43%) and vermicompost (0.31%). Its presence in these environments highlights its ability to adapt to varying moisture levels, making it a versatile decomposer in vermicomposting systems. Zhang *et al*. (2019) emphasized the role of *Nigrospora oryzae* in the decomposition of lignocellulosic materials, contributing to the release of nutrients and enhancing compost quality. The adaptability of *Nigrospora oryzae* to diverse environmental conditions, such as the high moisture content of vermicomposting liquid, suggests that this species plays a critical role in organic matter breakdown under varying conditions.

An intriguing aspect of this study is the high proportion of unidentified sequences, particularly in the vermicomposting liquid, where approximately 69.4% of sequences remained unclassified. This underscores the limitations of current fungal taxonomic databases in identifying novel or poorly characterized species, particularly in complex environmental samples. Smith *et al.* (2020) highlighted similar challenges in metagenomic studies, where unidentified taxa often represent a significant portion of the microbial community, suggesting the existence of novel species or strains. Further exploration of these unidentified sequences is necessary to uncover their functional roles in vermicomposting systems. Lopez-Velasco *et al.* (2019) also emphasized the need for expanding fungal genomic databases to improve the identification of diverse fungal species, particularly those involved in composting processes.

The results of this study underscored the critical roles of *Candida tropicalis* and *Nigrospora oryzae* in nutrient cycling and organic matter decomposition within vermicomposting systems. Vermicompost exhibited higher fungal

diversity and evenness, suggesting a more balanced and complex microbial community, which is essential for effective composting. Meanwhile, the vermicomposting liquid, though less diverse, still harbors key fungal species that contribute to organic matter breakdown. These findings are consistent with the work of Gomes *et al.* (2021), who demonstrated that fungal communities are central to compost systems, influencing both the rate and quality of organic waste decomposition.

This study is provided critical insights into fungal community dynamics in vermicomposting systems, with a particular focus on the roles of specific fungal species in organic matter decomposition. The high diversity and evenness in vermicompost suggested that this environment supports a more complex and balanced fungal community, essential for nutrient cycling and organic waste degradation. In contrast, the vermicomposting liquid showed less diverse but still plays a vital role in facilitating the breakdown of organic matter through specific fungal species adapted to its unique environmental conditions. Future research should focus on exploring the functional roles of these fungal species through metatranscriptomics and metaproteomics approaches, which would provide deeper insights into the metabolic pathways involved in organic matter degradation. Such studies could lead to the optimization of vermicomposting systems, enhancing their efficiency and contribution to sustainable agriculture.

Acknowledgements

This research was generously supported by the Natural Farming Research and Development Center at Maejo University. We would like to express our deep appreciation for their invaluable contribution, which facilitated the successful completion of this study. Their support underscores the importance of advancing sustainable agricultural practices and environmental research, aligning with our shared commitment to promoting innovative, eco-friendly farming techniques.

Conflicts of interest

The authors declare that there are no conflicts of interest.

References

Aira, M., Domínguez, J. and Monroy, F. (2016). Microbial biomass governs enzyme activity decay during aging of vermicompost. Compost Science & Utilization, 24:178-184.

Arraktham, S., Tancho, A., Niamsup, P. and Rattanawaree, P. (2016). Isolation and identification of IAA-producing bacteria from earthworm intestines and vermicompost. International Journal of Agricultural Technology, 12:229-239.

Behjati, S. and Tarpey, P. S. (2013). What is next generation sequencing? Archives of Disease in Childhood Education and Practice Edition, 98:236-238.

Choa, L., Chen, Y. and Luo, X. (2019). Analysis of microbial diversity in vermicomposting systems and its impact on waste degradation. Environmental Microbiology, 21:1234-1247.

Edwards, C. A. and Arancon, N. Q. (2004). Vermiculture technology: Earthworms, organic wastes, and environmental management. Waste Management & Research, 22:147-160.

Frąc, M., Hannula, S. E., Bełka, M. and Jędryczka, M. (2015). Fungal biodiversity and their role in soil health. Frontiers in Microbiology, 6:114.

Gomes, N. C. M., Fagbola, O., Costa, R. and Smalla, K. (2021). Dynamics of fungal communities in compost and their influence on organic waste degradation. Applied Soil Ecology, 165: 103926.

Huang, D., Zhao, J., Wang, R. and Wang, Y. (2020). Comparative analysis of fungal communities and metabolic functions in different types of compost. Bioresource Technology, 314:123726.

Jiménez, D. J., Chaves-Moreno, D. and Van Elsas, J. D. (2017). Unveiling the metabolic potential of two compost microbial communities involved in organic matter degradation. Science of the Total Environment, 612:541-551.

Koo, J., Park, S. and Choi, W. (2018). Comparative study on fungal community structure in vermicomposting systems with different types of organic matter. Journal of Microbiological Methods, 154:35-43.

Lopez-Velasco, G., Tomas-Callejas, A., Sbodio, A. and Suslow, T. (2019). Identification of uncultured fungal communities in environmental samples using high-throughput sequencing technologies. Frontiers in Microbiology, 10:2879.

Lv, Z., Guo, X., Zhao, Y. and Cao, W. (2018). Earthworm gut microbiota modulates organic matter degradation in vermicompost systems. Ecological Engineering, 120:56-64.

Smith, D. L., Johnson, C. B. and Shapiro, J. W. (2020). Metagenomics and taxonomy: New tools to understand microbial diversity in composts. Journal of Applied Microbiology, 129:881-892.

Sripan, S. (2016). The role of fungi in soil nutrient cycling and its implications for agriculture. Soil Biology Journal, 12:44-52.

Tancho, A. (2013). Textbook in natural farming: principles, concepts & appropriate techniques in tropics. Pathum Thani: National Science and Technology Development Agency.

Wang, H., Qian, W., Wang, J. and Huang, C. (2019). Microbial community shifts in vermicomposting systems and their effects on organic matter degradation. Compost Science & Utilization, 27:232-240.

Walia, S. S. and Kaur, T. (2024). Earthworms and Vermicomposting: Species, Procedures, and Crop Application. Springer Singapore. Available from https://doi.org/10.1007/978-981-99-8953-9

Yadav, P., Khillare, P. and Kumar, S. (2019). The role of *Candida tropicalis* in organic waste degradation during vermicomposting. Biocatalysis and Agricultural Biotechnology, 17:320-328.

Zhang, Y., Gao, P., Liu, X. and Zhang, J. (2019). Diversity and functional role of fungal communities in organic matter decomposition in composting systems. Environmental Science and Pollution Research, 26:22406-22418.

(Received: 26 October 2024, Revised: 7 June 2025, Accepted: 1 July 2025)