

Article

The Overlooked Decomposers: Effects of Composting Materials and Duration on the Mesofauna Mediating Humification

Felix Matheri ^{1,2,*} , Nehemiah Ongeso ³ , David Bautze ⁴ , Steven Runo ², Maina Mwangi ², AnneKelly Kambura ⁵, Edward Karanja ¹, Chrysantus Tanga ¹  and Milka Kiboi ⁴ 

¹ International Centre for Insect Physiology and Ecology (icipe), Nairobi P.O. Box 30772-00100, Kenya; ekaranja@icipe.org (E.K.); ctanga@icipe.org (C.T.)

² Department of Biochemistry, Microbiology and Biotechnology, Kenyatta University (KU), Nairobi P.O. Box 43844-00100, Kenya; smruno@gmail.com (S.R.); mmmainam@gmail.com (M.M.)

³ Institute of Ecology and Evolution, University of Bern, Baltzerstrasse 6, 3012 Bern, Switzerland; nehemiahongeso@gmail.com

⁴ Department of International Cooperation, Research Institute of Organic Agriculture (FiBL), Ackerstrasse 113, Postfach 219, 5070 Frick, Switzerland; david.bautze@fibl.org (D.B.); milka.kiboi@fibl.org (M.K.)

⁵ Department of Agricultural Sciences, Taita Taveta University (TTU), Voi P.O. Box 635-80300, Kenya; annekambura@ttu.ac.ke

* Correspondence: fmatheri@icipe.org

Abstract: Compost fauna act by releasing various enzymes that break down organic matter into a stable, agriculturally useful products. Mesofauna are the least studied compared to micro- and macrofauna, with the existing studies relying on classical methods such as morphological identification, essentially leaving out cryptic taxa. We sought to evaluate the ecological response of the mesofauna community to different composting materials and durations. Total mesofauna community 18S rRNA was purified in triplicate from lantana-based, tithonia-based, grass-based, and mixed (lantana + tithonia + grass)-based compost heaps after 21, 42, 63, and 84 days of composting and sequenced using the Illumina Miseq platform. Before performing statistical data analysis, we used the Divisive Amplicon Denoising Algorithm version 2 workflow for bioinformatic analyses. The composting duration, but not the composting materials, significantly influenced the total population and composition of the mesofauna communities. The composting materials and duration significantly affected the dispersion and uniqueness of the compost mesofauna communities. Canonical correspondence analysis of the compost's physical–chemical and biological states showed a significant influence of the materials on the mesofauna community colonization capacity. The mesofauna communities had a significant response to the composting duration. This, therefore, presents them as valuable tools for understanding the temporal evolution of compost.

Keywords: sub-Saharan Africa; cattle manure; metagenomics; ecology; fauna; ecosystems



Citation: Matheri, F.; Ongeso, N.; Bautze, D.; Runo, S.; Mwangi, M.; Kambura, A.; Karanja, E.; Tanga, C.; Kiboi, M. The Overlooked Decomposers: Effects of Composting Materials and Duration on the Mesofauna Mediating Humification. *Sustainability* **2024**, *16*, 6534. <https://doi.org/10.3390/su16156534>

Academic Editor: Anna De Marco

Received: 19 June 2024

Revised: 25 July 2024

Accepted: 27 July 2024

Published: 30 July 2024



Copyright: © 2024 by the authors. Licensee MDPI, Basel, Switzerland. This article is an open access article distributed under the terms and conditions of the Creative Commons Attribution (CC BY) license (<https://creativecommons.org/licenses/by/4.0/>).

1. Introduction

Sustainable soil fertility enhancement relies on recycling locally available renewable resources and techniques such as composting, mulching, biomass transfer, and green manuring [1]. Compost is superior to other single input uses [2,3] since it supplies the soil with important nutrients and organisms that support nutrient cycling [4,5]. There are several methods of composting, and thermophilic composting is widely accepted. This is because the technique can sanitize pathogens and weeds in raw manure, which is not achievable through other composting methods [6,7]. The process involves the controlled cycling of organic wastes through the mediation of litter chemistry and decomposition communities to produce organic fertilizers, dissipating temperature [8]. The decomposition communities are categorized as microfauna, mesofauna, and macrofauna, which all uniquely contribute to the composting process through rate-limiting functions [9,10]. However, mesofauna

are the least studied decomposition biota, with most existing studies relying on classical methods [11–13].

Mesofauna are a small invertebrate group, measuring 0.2 to 2.0 mm, and are ubiquitous in diverse ecosystems. They are the higher organisms based on size and trophic levels in ecosystems, with some belonging to feeding guilds that rely on microfauna [9,14,15]. The group is generally regarded as an intermediate taxonomic group since they are larger than microfauna such as bacteria and protozoa but smaller than macrofauna such as earthworms and Diptera [16]. They participate in ecosystem turnover by fragmenting organic matter, thus accelerating microbial decomposition [17]. Through this, mesofauna communities cycle soil nutrients and facilitate below-ground biodiversity, enhancing soil health. Among the major mesofauna taxa in the ecosystems are mites, pauropods, symphylans, proturans, and springtails, all mediating various roles in their environments [17–19].

The mesofauna communities are reciprocally influenced by the prevailing physical, biogeographical, and chemical environmental conditions, impacting their composition and functional capacity [19]. Consequently, ecosystem modifications contribute substantially to deep changes in biogeochemical properties and the mesofauna population and functions. This presents mesofauna as an ideal tool for establishing the maturity and quality of compost due to its direct functional sensitivity and correlation with the environment [13]. Understanding which specific mesofauna species participate in ecosystem processes will contribute to optimizing composting techniques and maximizing their benefits. Moreover, most studies on compost fauna have focused on municipal wastes and manure-only decomposition. This leaves out the farm-yard manure composting that involves Supplemental Materials such as tithonia and dry maize stalks.

Despite prokaryotes and fungi receiving much scientific attention and being regarded as main decomposers, materials such as lignin are difficult for these organisms to break down, requiring larger organisms such as mesofauna [16,19,20]. Mesofauna also play host to microorganisms by harboring them in their body systems, e.g., as the gut microbiome, where they play symbiotic roles [21]. Notwithstanding their ecological importance, a comprehensive and systematic assessment of the community structure and function of the mesofauna is still lacking.

Therefore, we conducted this study to evaluate (i) the ecological response of the mesofauna community to composting substrates and the temporal shifts in their physical–chemical state and (ii) their role in stimulating decomposition using next-generation sequencing.

2. Materials and Methods

2.1. Research Site and Treatments

We set the field experiment at the SysCom project Kenya (Farming Systems Comparison in the Tropics; Kenya) trial site at Thika, Kenya (01°0.231' S 37°04.747' E), between September 2020 and December 2020 [22,23], www.system-comparison.fibl.org (accessed on 14 April 2024)).

Our treatment selection was based on the standard composting practices in the tropics (Table 1). Cattle manure was sourced from a zero-grazing dairy unit close to the trial site, while dry maize stalks and green matter (tithonia, lantana twigs, and grass clippings) were obtained from the site and nearby farms. Each compost treatment was set under a composting shade in three replicates. Layering of the materials was performed as per the standard practice in sub-Saharan Africa [23]. Small dry twigs and pebbles were first laid on a flat surface under the composting shade. This was followed by a layer of chopped dry maize stalks, then a layer of cow dung manure. We finally added a layer of green materials (lantana, tithonia, grass, or a mixture for the specific treatments). At the beginning of composting, the moisture content of each heap was adjusted to approximately 60%. Compost heap aeration was performed by turning it every four days during the first 20 days, then weekly until 84 days of composting.

Table 1. The experiment treatments and respective compost feedstock combination.

Treatment	Main Composting Materials
Lantana-based compost	Fresh cow dung manure, chopped dry maize stalks, chopped lantana twigs (4:2:1 w/w)
Tithonia-based compost	Fresh cow dung manure, chopped dry maize stalks, chopped tithonia twigs (4:2:1 w/w)
Grass-based compost	Fresh cow dung manure, chopped dry maize stalks, chopped grass clippings (4:2:1 w/w)
Lantana + tithonia + grass-based compost [Mixed]	Fresh cow dung manure, chopped dry maize stalks, chopped * (lantana + tithonia + grass) (4:2:1 w/w)

* Equal amounts of grass clippings, tithonia, and lantana twigs were used to prepare the mixed treatment.

2.2. Evaluation of the Physical–Chemical Evolution of Compost

Daily temperature monitoring was performed using a compost thermometer (model: WIKA 110824862-EN 13190; Louisville, KY, USA) at three points on the individual compost heaps, as described by Matheri et al. [23]. The pH, moisture, total Kjeldahl nitrogen content, total organic carbon content, and Olsen phosphorous content were analyzed gravimetrically according to the protocols of Okalelebo et al. and Adamtey [24,25].

2.3. Sample Collection and Total Compost DNA Extraction

Compost samples were collected from five points on each pile on the 21st, 42nd, 63rd, and 84th day. We used day 21 of composting as the baseline since the composting materials (maize stovers, cow dung manure, and lantana/grass/tithonia) were not sufficiently homogenized during the earlier days of composting and therefore would have biased the sampling. A top-to-bottom sampling approach was used with sharp pre-sterilized shovels (under 70% ethanol). The samples were then transported on ice to the sample room and stored at -20°C pending total DNA extraction.

Total compost microbial DNA extraction from the samples was performed at the Kenyatta University plant transformation laboratories in Nairobi, Kenya. We used the PureLink™ Microbiome DNA Purification Kit (Catalog number: A29790, Thermo Scientific, Waltham, MA, USA) according to the manufacturer's instructions; www.thermofisher.com/ke/en/home/life-science, (accessed on 14 April 2024). DNA quality and concentrations were measured using a 2% agarose gel and a NanoDrop (Maestro-gen) before shipping them on dry ice to the Molecular Research DNA Lab (www.mrdnalab.com, (accessed on 14 April 2024). Shallowater, TX, USA) for sequencing.

2.4. Compost Mesofauna Microbiome Amplification

The 18S PCR primer sets EUK1391F (5'GTACACACCGCCCGTC-3') and EukBr (5'-TGATCCTTCTGCAGGTTACCTAC-3') [26] were used for the amplification of non-fungal eukaryotic DNA which includes mesofauna DNA. Eukaryotic DNA amplification was performed using the HotStarTaq Plus Master Mix Kit (Qiagen, Germantown, MD, USA). We set the PCR conditions to be 30 cycles of 95°C for 30 s, 53°C for 40 s, and 72°C for 1 min, with a final elongation step at 72°C for 10 min. The PCR amplicons were then visually quantified using a 2% agarose gel [23]. Equimolar quantities of the obtained eukaryotic PCR amplicons were multiplexed using unique indices, pooled, and sequenced using Illumina MiSeq next-generation technology at MR DNA (www.mrdnalab.com, (accessed on 14 April 2024) Shallowater, TX, USA). We then trimmed the barcodes and amplicon primer sequences. De-noising and filtering of low-quality sequences with reads <300 base pairs were performed after phred20-based quality trimming, while sequences with ambiguous base calls and homopolymer runs (exceeding 5 bp) were removed [27]. The raw 18S sequences were submitted to the NCBI sequence archive under the accession number PRJNA822850 (<https://www.ncbi.nlm.nih.gov/sra/PRJNA822850> (accessed on 14 April 2024)).

2.5. Bioinformatics and Sequence Data Statistical Analyses

The raw 18S amplicon sequences obtained from the Illumina MiSeq platform were first assessed for sequence quality using FastQC (v 0.11.6). Low-quality reads and primers were then trimmed using cutadapt (v.4.1). Subsequently, the filtered reads underwent preprocessing utilizing the Divisive Amplicon Denoising Algorithm 2 (DADA2) v 1.26.0 [28]. After that, the amplicon sequence variants' (ASVs') taxonomic classification was inferred using the pr2 protist pre-trained database [29]. The filtered ASV abundance matrix, taxonomy table, and metadata were merged into a phyloseq object, facilitating the determination of differential mesofauna abundance using the phyloseq (v1.41) package [30].

Mesofauna alpha diversity metrics were computed and tested for normality and significance using the alpha function of the microbiome (v 1.22.0) package. Permutational Multivariate Analysis of Variance (PERMANOVA) and betadisper were used to compare mesofauna population distributions across different compost treatments and composting days. The metagMisc (v.0.04) package aided in the visualization of the phyloseq object, presenting mesofauna relative abundances and percentages across compost treatments and composting days. The beta diversity analysis utilized Principal Coordinate Analysis (PCoA) to elucidate the contribution of compost treatments and composting duration to mesofauna variation. Shared mesofauna communities were determined through Venn diagrams using Euler (v7.0.0). Lastly, we utilized Canonical Correlation Analysis (CCA) following a step-wise model-choosing procedure to identify and measure associations between mesofauna distribution and the abiotic status of the compost [31].

3. Results

We obtained 28,253,246 paired reads spanning the V7–V8 variable regions of eukaryotic 18s rRNA. The DADA2 preprocessing led to 19,109,631 reads and 1159 ASVs for taxonomic assignment. From the analysis, 1158 species of eukaryotes were determined to facilitate the decomposition of organic matter and were influenced by the different sources of green composting materials (Supplementary File S1).

3.1. Alpha Diversity of Mesofauna Community under the Influence of Composting Materials and Duration

We observed significant differences in the mesofauna populations (Chao) between the different sampling time points (Kruskal–Wallis chi-squared = 11.517, $p = 0.009236$) (Figure 1). However, there were no significant differences in all other alpha diversity metrics among the composting treatments and durations.

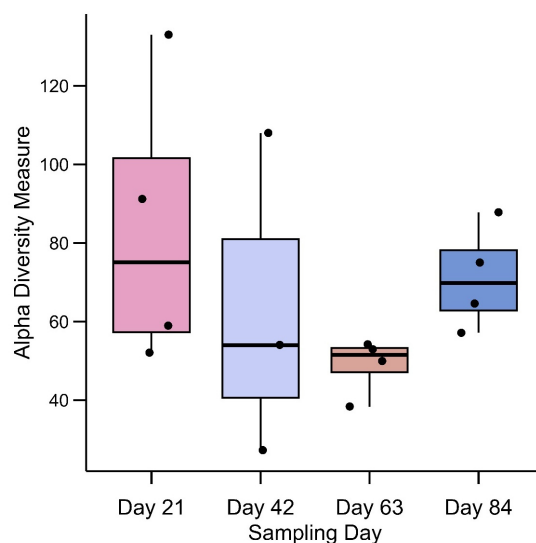


Figure 1. Alpha diversity (Chao1) of mesofauna communities under the influence of composting duration.

3.2. PERMANOVA and Species Composition of Compost Mesofauna

The overall mesofauna community composition was substantially (36.0%) attributable to composting duration ($R^2 = 0.350$, $F = 1.974$, $p = 0.0004$) but not to differences in dispersion. However, there were no significant differences in dispersion among the composting treatments.

There was preferential colonization of specific mesofauna species on the different composting materials. For instance, *Pergamasus canestrinii* displayed a higher abundance in the lantana (45.4%) and tithonia (28.6%) compost regimes compared to the grass (2.5%) and mixed (3.5%) regimes. Similarly, *Lepidocyrtus paradoxus* showed elevated percentages in the mixed (10.0%) and tithonia (25.5%) compost regimes compared to the grass (2.5%) and lantana (2.7%) regimes (Figure 2a, Supplementary File S2). The diversity and dynamics of the mesofauna populations across the sampling time points showed that *Cryptopygus antarcticus* dominated, representing 31.9% of the total mesofauna population on the 21st day, followed by a significant increase to 69.3% on day 42 and gradually declining to 2.5% by the 84th day. *Lepidocyrtus paradoxus* showed a minor presence on day 21 (0.2%) but experienced a substantial surge to 46.9% by day 84. *Sminthurides aquaticus* displayed fluctuations in abundance, peaking at 25.9% on day 42 and gradually declining afterward. Some species, including *Aphodanoetus teinophallus*, *Cosmoglyphus* spp., and *Culicoides variipennis*, exhibited sporadic occurrences with minimal representation across the sampling time points (Figure 2b, Supplementary File S3).

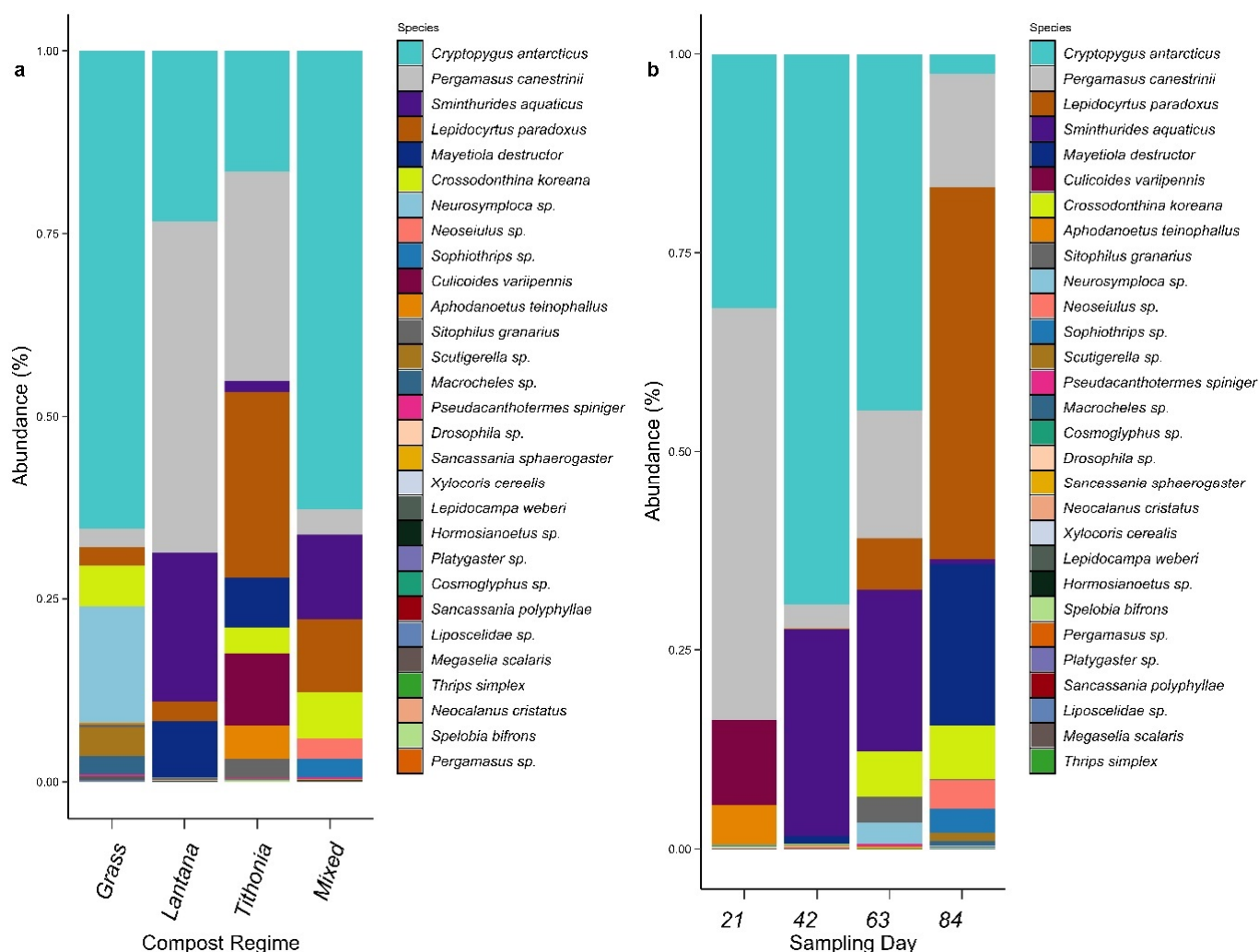


Figure 2. Top 30 mesofauna species facilitating degradation of the green nitrogenous feedstock. (a) mesofauna species diversity by compost type and (b) composting day.

3.3. Distribution and Uniqueness of Compost Mesofauna Communities

The Bray–Curtis dissimilarity-based analysis showed a significant (45.9%) distinction among the composting treatments and time points ($p \leq 0.001$). The mesofauna community distinction was highly variable in the lantana-based compost and on the 84th day of composting (Figure 3a,b). The different compost treatments had more shared species and fewer unique ones while the different composting durations bore more unique mesofauna species. Overall, the composting durations had more distinct ASVs compared to the composting treatments. The lantana-based compost had the highest number of unique ASVs among the treatments, while day 21 had the most unique ASVs among the composting durations (Figure 3c,d).

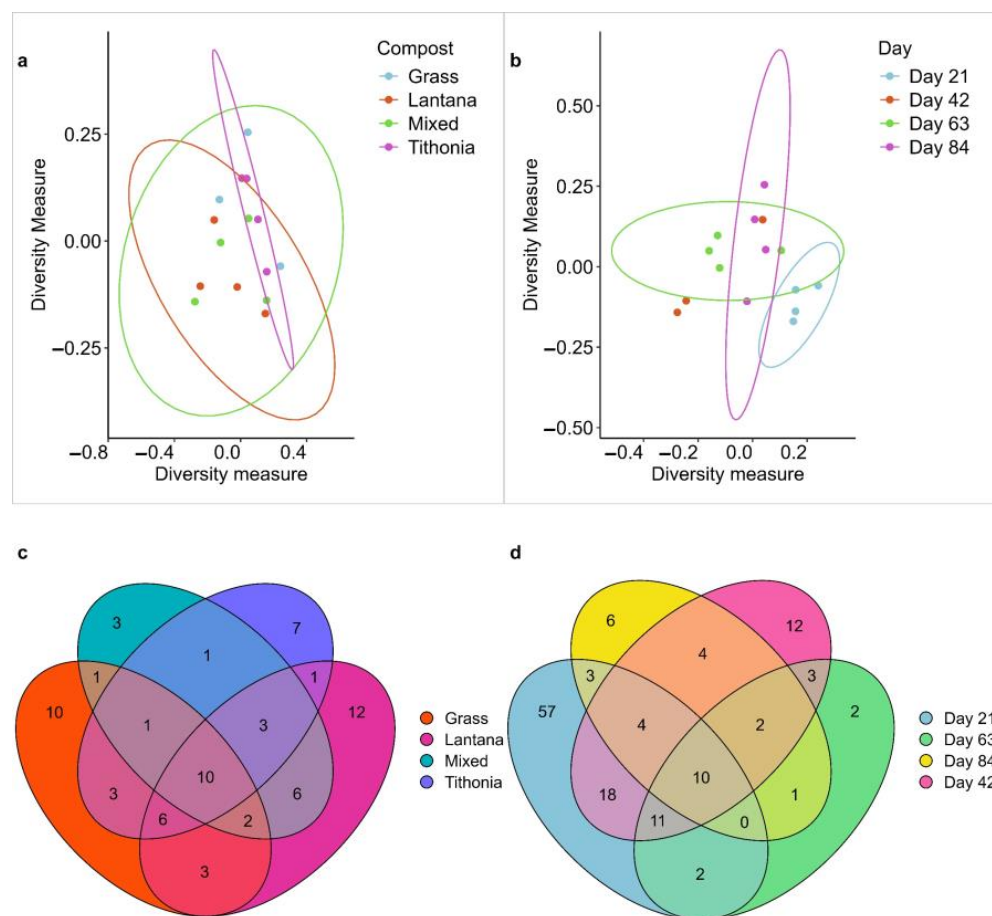


Figure 3. Principal Coordinate Analysis (PCoA) of eukaryotic community ellipse clustering indicating their distribution based on compost treatment (a) and composting day (b). Venn diagrams show the distribution of unique and shared ASVs within the various compost treatments (c) and composting days (d).

3.4. Influence of Environmental Factors on Compost Mesofauna Community Structure

The ordination biplots showed the compost abiotic characteristics that significantly influenced the mesofauna assemblages ($p = 0.001$). Temperature and organic carbon content produced the highest rate of change in the mesofauna communities. These physical–chemical elements and pH and total nitrogen, nitrate, and phosphorous contents significantly influenced the mesofauna community structure (Figure 4a). No groupings were observed based on composting materials, while the composting duration plot showed that ammonia, phosphorous, and total nitrogen contents were major community structure shapers after 21 days of composting. On the other hand, pH and nitrate content had a higher influence on the composition and abundance on the 42nd day of composting (Figure 4b).

The direction of the arrows indicates the direction of the maximum change of that variable. The length of the arrow is proportional to the rate of change.

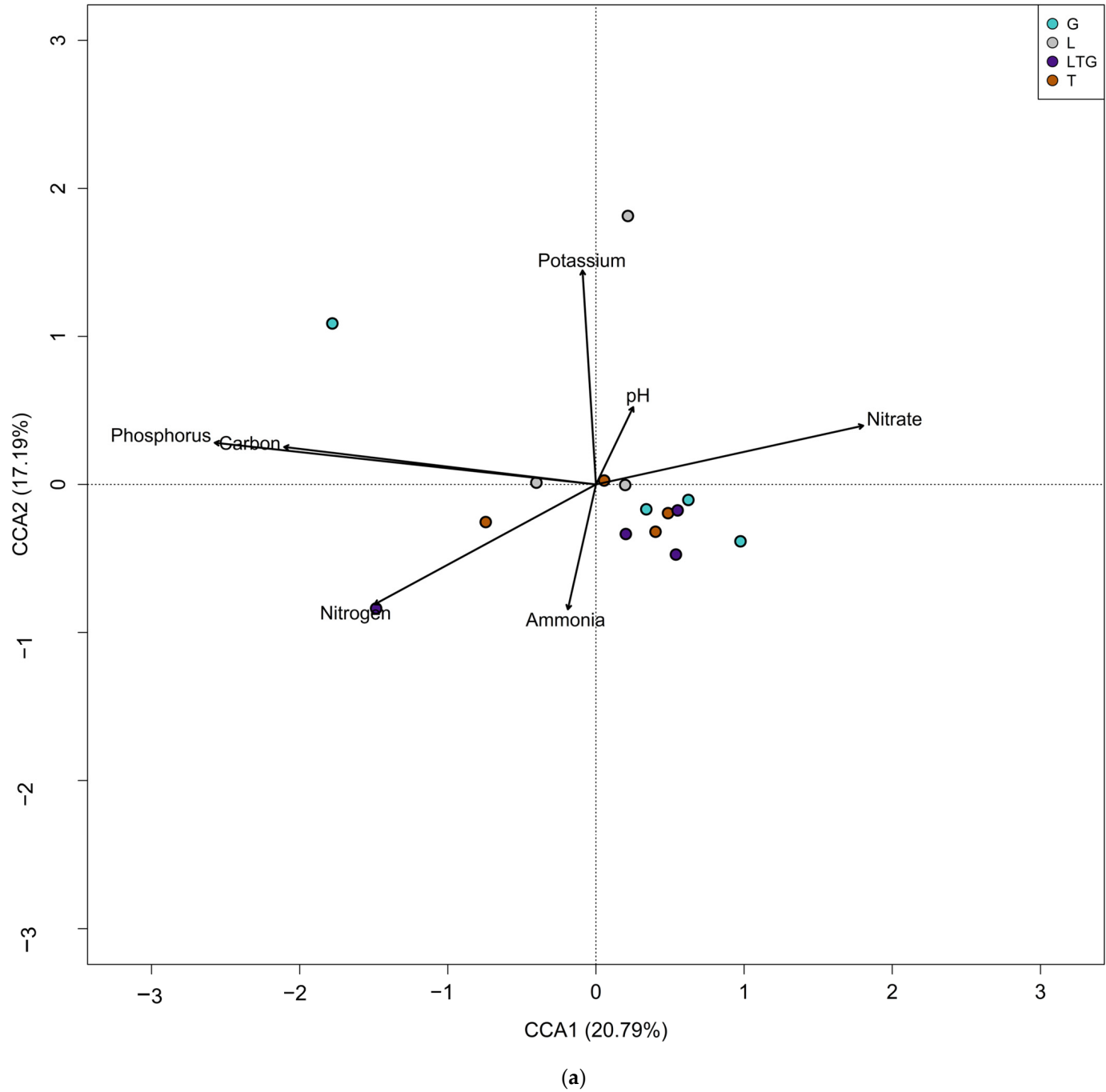


Figure 4. Cont.

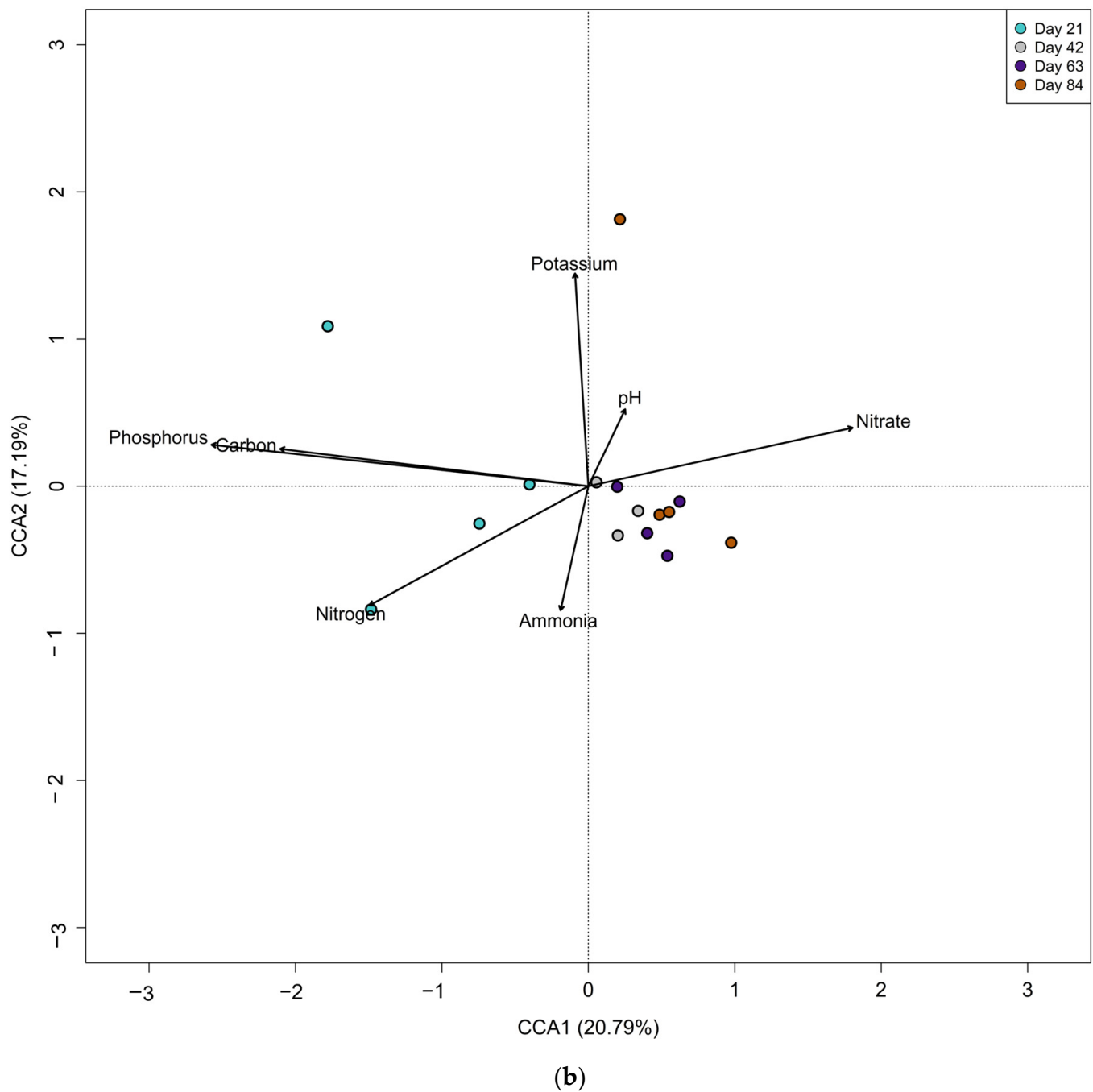


Figure 4. Canonical correspondence analysis (CCA) biplot of mesofauna communities and compost abiotic parameters that significantly shaped the communities and were influenced by the composting materials (a) and composting duration (b).

4. Discussion

Our results showed that the composting duration significantly influenced the number of species (Chao1) while the composting treatments (feedstock) did not. The high mesofauna population on day 84 of composting indicates that the compost environment was stable, allowing for mesophilic colonization. These organisms are generally heat-sensitive and have low loads during the thermophilic phase of composting [32]. All composting treatments conferred a similar mesofauna diversity and, therefore, are equally capable of enhancing ecosystem resilience through functional redundancy. The lack of influence of the treatment on the mesofauna species means they responded similarly, irrespective of the ma-

terials and duration of composting. Our findings corroborate the results of Barus et al. [33], who averred that adding organic matter has little immediate influence on the mesofauna, indicating that time was a factor in the ecological response of the community.

The top 30 mesofauna species reported in this study have also featured as the top taxa in other studies with diverse roles in nutrient cycling and phyto-protection. These taxa have been consistently shown to be in relatively higher abundance compared to other species, colonizing many different habitats such as grasslands, forest soils, and agricultural farms [34–36]. For example, *Macrocheles mites* are the dominant species in soil ecosystems, where they play a significant predatory role, thus providing an ecological balance for nematodes and dung beetles [37]. *Pergamasus canestrinii* is a species of mites that dominates forest soils, grazing lands, and decaying manures and is among the top 14 dominant species in many habitats [38,39] and is associated with breaking down organic matter and controlling microbial populations through predation [40].

The species *Lepidocyrtus paradoxus*, which was predominant in the tithonia-based compost, is a springtail that contributes to leaf litter decomposition through physical fragmentation [41]. They also regulate microbial population in ecosystems such as compost by grazing on fungi and bacteria, ultimately excreting nutrients such as nitrogen and phosphorous. This effectively positions *Tithonia diversifolia* as a superior compost amendment, encouraging flora that aid in nitrogen and phosphorous cycling. On the other hand, the absence of this species during the thermophilic phase (day 21) was due to the unsuitable high temperatures, limiting their survival [42,43]. The gradual increase beginning on day 42 and peaking on day 84 was commensurate with the gradual decline in pile temperatures, encouraging colonization and grazing of mesophilic micro-organisms which are preferential to the species. *Pergamasus canestrinii* is a soil mite that is common in leaf litter, contributing to nutrient cycling mainly through indirect decomposition of organic matter by relying on microorganism guilds [44]. The higher population of this species in the lantana-based compost is explained by the relatively high microbial population in this treatment that was observed by Matheri et al. [23]. The dominance of this species at day 21 of composting compared to other species points to its considerable adaptation to high temperatures. Moreover, this period has higher microbial activity, providing food guilds for the species. *Pergamasus canestrinii* and other mites have been reported to have resilience against high ecosystem temperatures through mechanisms such as the production of heat-stable enzymes, rapid reproduction, and burrowing into cooler pockets of the environment [45–47].

The species *Cryptopygus antarcticus* adapts to fluctuations in ecosystem temperature, moisture, and nutrients [44,48], hence its presence and dominance at all composting time points in our findings. This collembolan is native to undisturbed soils and has been reported among the most abundant mesofauna taxa in diverse ecosystems [49–52]. This taxon's thermal and moisture preferences explain its dominance in thermophilic compost environments, out-competing other species [51]. The population increases of *Mayetiola destructor* beginning on day 42 of composting suggest a potential response to maturation or changes in compost temporal conditions. This makes this species a feasible eco-indicator of compost maturity. Despite being a plant pathogen, this taxon has been reported to contribute to tilts in carbon metabolism and the C:N ratio by decreasing C compounds and increasing N compounds in plants [53].

The substantially higher unique species observed in the lantana-based treatment affirms the differential impact of composting material on the mesofauna community structure. Lantana has been reported to spur the recruitment of specialized decomposers [23] and, therefore, is suitable to mediate a more robust ecological response. Compared to other treatments, lantana is richer in secondary metabolites such as alkaloids and phenolics [54]. The greater number of shared species (ASVs) between the different composting durations compared to the different composting materials suggests a more significant influence of composting duration than the nature of the feedstock. The decomposition process naturally recruits certain organisms regardless of the available organic matter. These taxa are

generally opportunistic and indiscriminately feed on organic matter rather than selectively colonizing specific materials [17,55,56].

The abiotic factors explain how composting materials shape the major mesofauna community. We attributed the high influence of total nitrogen and carbon contents on the mesofauna communities to the labile nature of these elements. Some mesofauna species are part of a large nitrogen-cycling community in ecosystems. Therefore, significant changes in nitrogen during the composting process were responsible for their drastic changes [17]. Compost temperature is a principal driver of mesofauna assemblages due to their sensitivity to thermal fluctuations, which impacts their metabolic and reproductive capacities [57].

Additionally, temperature-mediated shifts in microbial activity indirectly influence mesofauna dynamics due to their role as hosts and precursor providers of microorganisms. These temporal shifts in communities are related to carbon levels, which are highly correlated with compost temperature [23,55,56,58]. Previous studies have also identified phosphorus as a rate-limiting element during litter decomposition, with a strong association with fauna [59]. This corroborates previous studies that showed that mesofauna are suitable predictors for ecosystem functioning, where the decomposition status was correlated with the properties of the materials [60,61].

5. Conclusions

The significant differences among the different composting time points uncover the dynamic response of mesofauna communities to temporal variations in organic matter during decomposition. This effectively positions the mesofauna community as a superior tool for fingerprinting different phases of compost succession over time. However, the composting treatments had less influence on the mesofauna communities compared to the composting duration. Our study points out the functional redundancy of composting substrates on the mesofauna community structure since they exhibit similar ecological responses to different materials.

Supplementary Materials: The following supporting information can be downloaded at: <https://www.mdpi.com/article/10.3390/su16156534/s1>, Supplementary File S1, Taxonomic composition; Supplementary File S2, Relative abundance Treatments; Supplementary File S3, Relative abundances duration.

Author Contributions: Conceptualization, F.M., A.K., M.M., E.K. and S.R.; methodology, F.M., N.O., A.K., M.M., E.K. and S.R.; software, F.M., N.O., C.T. and E.K.; validation, N.O., A.K., M.K. and D.B.; formal analysis, F.M. and N.O.; investigation, F.M., N.O. and E.K.; resources, M.M., S.R., D.B. and M.K.; data curation, F.M. and N.O.; writing—original draft preparation, F.M., N.O. and M.K.; writing—review and editing, F.M., N.O., D.B., C.T. and M.K.; visualization, F.M. and N.O.; supervision, A.K., M.M., M.K. and S.R.; project administration, E.K., D.B. and M.K.; funding acquisition, M.M., E.K., D.B. and M.K. All authors have read and agreed to the published version of the manuscript.

Funding: This research was partially funded by DAAD-Africa through the In-Country/In-Region Scholarship Programme Eastern Africa (grant no: 91712524). The study also received partial funding and technical support from the Long-Term Systems Comparison Program, which is financially supported by the Biovision Foundation, Coop Sustainability Fund, Liechtenstein Development Service (LED), and Swiss Agency for Development and Cooperation (SDC); Grant No. 1040. The funding sources had no role in the design and conduction of the study; collection, management, analysis, and interpretation of the data; preparation, review, or approval of the manuscript; and the decision to submit the manuscript for publication.

Institutional Review Board Statement: Not applicable.

Informed Consent Statement: Not applicable.

Data Availability Statement: The raw 18S sequences were submitted to the NCBI sequence archive under the accession number PRJNA822850 (<https://www.ncbi.nlm.nih.gov/sra/PRJNA822850>).

Acknowledgments: The authors wish to thank the Pan African Network for Bioinformatics Training (H3ABioNet) through the International Centre of Insect Physiology and Ecology (icipe) node for

providing the necessary computational infrastructure and invaluable support to bring this project to a good end. The authors acknowledge the support received from James Karanja during the fieldwork.

Conflicts of Interest: The authors declare no conflicts of interest. The funders had no role in the design of the study; in the collection, analyses, or interpretation of data; in the writing of the manuscript; or in the decision to publish the results.

References

1. Wezel, A.; Herren, B.G.; Kerr, R.B.; Barrios, E.; Gonçalves, A.L.; Sinclair, F. Agroecological principles and elements and their implications for transitioning to sustainable food systems. A review. *Agron. Sustain. Dev.* **2020**, *40*, 40. [CrossRef]
2. Kumar, S.; Meena, R.S.; Jhariya, M.K. Resources Use Efficiency in Agriculture. *Resour. Use Effic. Agric.* **2020**, *1*, 1–760.
3. Rahman, M.M.; Alam, M.S.; Kamal, M.Z.U.; Rahman, G.K.M.M. Organic Sources and Tillage Practices for Soil Management. In *Resources Use Efficiency in Agriculture*; Springer: Singapore, 2020; pp. 283–328. Available online: https://link.springer.com/chapter/10.1007/978-981-15-6953-1_9 (accessed on 10 April 2024).
4. Pérez-Piqueres, A.; Edel-Hermann, V.; Alabouvette, C.; Steinberg, C. Response of soil microbial communities to compost amendments. *Soil Biol. Biochem.* **2006**, *38*, 460–470. [CrossRef]
5. Masters-Clark, E.; Shone, E.; Paradelo, M.; Hirsch, P.R.; Clark, I.M.; Otten, W. Development of a defined compost system for the study of plant-microbe interactions. *Sci. Rep.* **2020**, *10*, 7521. [CrossRef] [PubMed]
6. Lepesteur, M. Human and livestock pathogens and their control during composting. *Crit. Rev. Env. Sci. Technol.* **2022**, *52*, 1639–1683. [CrossRef]
7. Shelly, P.; Neemisha; Sharma, S. Modification in the Composting Environment through Additives. *Commun. Soil Sci. Plant Anal.* **2022**, *53*, 2141–2155. [CrossRef]
8. Nguyen, T.P.; Koyama, M.; Nakasaki, K. Effects of oxygen supply rate on organic matter decomposition and microbial communities during composting in a controlled lab-scale composting system. *Waste Manag.* **2022**, *153*, 275–282. [CrossRef] [PubMed]
9. Gongalsky, K.B. Soil macrofauna: Study problems and perspectives. *Soil Biol. Biochem.* **2021**, *159*, 108281. [CrossRef]
10. Pant, M.; Negi, G.C.S.; Kumar, P. Macrofauna contributes to organic matter decomposition and soil quality in Himalayan agroecosystems. *India. Appl. Soil Ecol.* **2017**, *120*, 20–29. [CrossRef]
11. Miller, J.J.; Battigelli, J.P.; Beasley, B.W.; Drury, C.F. Response of Soil Mesofauna to Long-Term Application of Feedlot Manure on Irrigated Cropland. *J. Environ. Qual.* **2017**, *46*, 185–192. [CrossRef]
12. Geisen, S.; Briones, M.J.; Gan, H.; Behan-Pelletier, V.M.; Friman, V.P.; de Groot, G.A.; Hannula, S.E.; Lindo, Z.; Philippot, L.; Tiunov, A.V.; et al. A methodological framework to embrace soil biodiversity. *Soil Biol. Biochem.* **2019**, *136*, 107536. [CrossRef]
13. Steel, H.; Bert, W. Biodiversity of compost mesofauna and its potential as an indicator of the composting process status. *Dyn. Soil Dyn. Plant* **2012**, *5*, 45–50.
14. Dervash, M.; Bhat, R.; Mushtaq, N.; Singh, V. Dynamics and importance of soil mesofauna. *Int. J. Adv. Res. Sci. Eng.* **2018**, *7*, 2010–2019.
15. Schröder, P. Mesofauna. In *Perspectives for Agroecosystem Management: Balancing Environmental and Socio-Economic Demands*; Elsevier: Amsterdam, The Netherlands, 2011; pp. 293–306.
16. Frouz, J. Effects of soil macro- and mesofauna on litter decomposition and soil organic matter stabilization. *Geoderma* **2018**, *332*, 161–172. [CrossRef]
17. Song, X.; Wang, Z.; Tang, X.; Xu, D.; Liu, B.; Mei, J. The contributions of soil mesofauna to leaf and root litter decomposition of dominant plant species in grassland. *Appl. Soil Ecol.* **2020**, *155*, 103651. [CrossRef]
18. Gonçalves, F.; Carlos, C.; Crespo, L.; Zina, V.; Oliveira, A.; Salvação, J. Soil arthropods in the douro demarcated region vine-yards: General characteristics and ecosystem services provided. *Sustainability* **2021**, *13*, 7837. [CrossRef]
19. Potapov, A.M.; Beaulieu, F.; Birkhofer, K.; Bluhm, S.L.; Degtyarev, M.I.; Devetter, M. Feeding habits and multifunctional classification of soil-associated consumers from protists to vertebrates. *Biol. Rev.* **2022**, *97*, 1057–1117. [CrossRef]
20. Coleman, D.C.; Geisen, S.; Wall, D.H. Soil fauna: Occurrence, biodiversity, and roles in ecosystem function. In *Soil Microbiology Ecology and Biochemistry*, 5th ed.; Elsevier: Amsterdam, The Netherlands, 2024; pp. 131–159.
21. Sun, M.; Chao, H.; Zheng, X.; Deng, S.; Ye, M.; Hu, F. Ecological role of earthworm intestinal bacteria in terrestrial environments: A review. *Sci. Total Environ.* **2020**, *740*, 140008. [CrossRef]
22. Taguiling, L.G. Quality improvement of organic compost using green biomass. *Eur. Sci. J.* **2013**, *9*, 319–341.
23. Matheri, F.; Kambura, A.K.; Mwangi, M.; Ongeso, N.; Karanja, E.; Adamtey, N. Composition, structure, and functional shifts of prokaryotic communities in response to co-composting of various nitrogenous green feedstocks. *BMC Microbiol.* **2023**, *23*, 50. [CrossRef]
24. Adamtey, N. Evaluation of Agricultural and Agro-industrial Residues for Composting for Agricultural Use in Ghana (A Case Study in the Kwaebibirem District). Master's Thesis, University of Ghana, Accra, Ghana, 2005.
25. Okalebo, J.R.; Gathua, K.W.; Woomer, P.L. *Laboratory Methods of Soil and Plant Analysis: A Working Manual*, 2nd ed.; Sacred Africa: Nairobi, Kenya, 2002; Volume 21, pp. 25–26.
26. Amaral-Zettler, L.A.; McCliment, E.A.; Ducklow, H.W.; Huse, S.M. A method for studying protistan diversity using massively parallel sequencing of V9 hypervariable regions of small-subunit ribosomal RNA genes. *PLoS ONE* **2009**, *4*, e6372. [CrossRef]

27. Reeder, J.; Knight, R. Rapidly denoising pyrosequencing amplicon reads by exploiting rank-abundance distributions. *Nat. Methods* **2010**, *7*, 668–669. [\[CrossRef\]](#) [\[PubMed\]](#)
28. Callahan, B.J.; McMurdie, P.J.; Holmes, S.P. Exact sequence variants should replace operational taxonomic units in marker-gene data analysis. *ISME J.* **2017**, *11*, 2639–2643. [\[CrossRef\]](#) [\[PubMed\]](#)
29. Guillou, L.; Bachar, D.; Audic, S.; Bass, D.; Berney, C.; Bittner, L. The Protist Ribosomal Reference database (PR2): A catalog of unicellular eukaryote Small Sub-Unit rRNA sequences with curated taxonomy. *Nucleic Acids Res.* **2013**, *41*, D597–D604. [\[CrossRef\]](#) [\[PubMed\]](#)
30. McMurdie, P.J.; Holmes, S. phyloseq: An R Package for Reproducible Interactive Analysis and Graphics of Microbiome Census Data. *PLoS ONE* **2013**, *8*, e61217. [\[CrossRef\]](#) [\[PubMed\]](#)
31. Matheri, F.; Kambura, A.K.; Mwangi, M.; Karanja, E.; Adamtey, N.; Wanjau, K. Evolution of fungal and non-fungal eukaryotic communities in response to thermophilic co-composting of various nitrogen-rich green feedstocks. *PLoS ONE* **2023**, *18*, e0286320. [\[CrossRef\]](#) [\[PubMed\]](#)
32. Li, W.; Zhu, L.; Wu, B.; Liu, Y.; Li, J.; Xu, L. Improving mesophilic anaerobic digestion of food waste by side-stream thermo-philic reactor: Activation of methanogenic, key enzymes and metabolism. *Water Res.* **2023**, *241*, 120167. [\[CrossRef\]](#) [\[PubMed\]](#)
33. Barus, J.; Meithasari, D.; Lumbanraja, J.; Sudarsono, H.; Hidayat, K.F.; Dermiyati. Soil mesofauna amount and diversity by return-ing fresh and compost of crops biomass waste in ultisols in-situ. *Biodiversitas* **2021**, *22*, 92–98.
34. Dombos, M. Collembola of loess grassland: Effects of grazing and landscape on community composition. *Soil Biol. Biochem.* **2001**, *33*, 2037–2045. [\[CrossRef\]](#)
35. Nitzu, E.; Nae, A.; Băncilă, R.; Popa, I.; Giurginca, A.; Plăiașu, R. Scree habitats: Ecological function, species conservation and spatial-temporal variation in the arthropod community. *Syst. Biodivers.* **2014**, *12*, 65–75. [\[CrossRef\]](#)
36. Bondarenko-Borisova, I.V.; Sandul, N.G. The fauna of springtails (Collembola) from the forest ecosystems of south-east Ukraine. In *Bulletin of Zoology*; National Academy of Sciences of Ukraine: Kyiv, Ukraine, 2002.
37. Grisez, C.; Perrin, W.; Begou, M.; Jay-Robert, P.; Jacquiet, P. An initial investigation of the predatory activity of the phoretic mites of dung beetles, *Macrocheles* sp. (Mesostigmata: Macrochelidae), on the gastrointestinal nematode of sheep *Haemonchus contortus* (Strongylida: Trichostrongylidae). *Biol. Control* **2023**, *185*, 105301. [\[CrossRef\]](#)
38. Ondrejková, K.; Fend'a, P. First records of three Pergamasinae species (Acari, Mesostigmata, Parasitidae) from Slovakia. *Check List* **2022**, *18*, 725–731. [\[CrossRef\]](#)
39. Manu, M.; Băncilă, R.I.; Onete, M. Effect of Grazing Management on Predator Soil Mite Communities (Acari: Mesostigmata) in Some Subalpine Grasslands from the Făgăraș Mountains—Romania. *Insects* **2023**, *14*, 626. [\[CrossRef\]](#) [\[PubMed\]](#)
40. Peschel, K.; Norton, R.A.; Scheu, S.; Maraun, M. Do oribatid mites live in enemy-free space? Evidence from feeding experiments with the predatory mite *Pergamasus septentrionalis*. *Soil Biol. Biochem.* **2006**, *38*, 2985–2989. [\[CrossRef\]](#)
41. Harta, I.; Simon, B.; Vinogradov, S.; Winkler, D. Collembola communities and soil conditions in forest plantations established in an intensively managed agricultural area. *J. For. Res.* **2021**, *32*, 1819–1832. [\[CrossRef\]](#)
42. Soto-Adames, F.N. Molecular phylogeny of the Puerto Rican *Lepidocyrtus* and *Pseudosinella* (Hexapoda: Collembola), a validation of Yoshii's "color pattern species". *Mol. Phylogenetics Evol.* **2002**, *25*, 27–42. [\[CrossRef\]](#) [\[PubMed\]](#)
43. Santeshwari, X.; Raghuraman, M.; Singh, J. Collembola (Insecta: Collembola) community from Varanasi and nearby regions of Uttar Pradesh, India. *J. Exp. Zool. India* **2015**, *18*, 571–577.
44. Leo, C.; Nardi, F.; Cucini, C.; Frati, F.; Convey, P.; Weedon, J.T. Evidence for strong environmental control on bacterial microbiomes of Antarctic springtails. *Sci. Rep.* **2021**, *11*, 2973. Available online: <https://www.nature.com/articles/s41598-021-82379-x> (accessed on 26 July 2024). [\[CrossRef\]](#)
45. Villani, M.G.; Allee, L.; Diaz, A.; Robbins, P.S. Adaptive strategies of edaphic arthropods. *Annu. Rev. Entomol.* **1999**, *44*, 233–256. [\[CrossRef\]](#)
46. Wehner, K.; Scheu, S.; Maraun, M. Resource availability as driving factor of the reproductive mode in soil microarthropods (Acari, Oribatida). *PLoS ONE* **2014**, *9*, e104243. [\[CrossRef\]](#)
47. Walter, D.E.; Proctory, H.C.; Walter, D.E.; Proctor, H.C. Mites in soil and litter systems. In *Mites: Ecology, Evolution & Behaviour*; Springer: Dordrecht, The Netherlands, 2013; pp. 161–228. [\[CrossRef\]](#)
48. Hawes, T.C.; Bale, J.S.; Worland, M.R.; Convey, P. Trade-offs between microhabitat selection and physiological plasticity in the Antarctic springtail, *Cryptopygus antarcticus* (Willem). *Polar Biol.* **2008**, *31*, 681–689. [\[CrossRef\]](#)
49. Everatt, M.J.; Convey, P.; Worland, M.R.; Bale, J.S.; Hayward, S.A.L. Heat tolerance and physiological plasticity in the Antarctic collembolan, *Cryptopygus antarcticus*, and mite, *Alaskozetes antarcticus*. *J. Therm. Biol.* **2013**, *38*, 264–271. [\[CrossRef\]](#)
50. Hayward, S.A.L.; Worland, M.R.; Convey, P.; Bale, J.S. Habitat moisture availability and the local distribution of the Antarctic Collembola *Cryptopygus antarcticus* and *Friesea grisea*. *Soil Biol Biochem.* **2004**, *36*, 927–934. [\[CrossRef\]](#)
51. Day, T.A.; Ruhland, C.T.; Strauss, S.L.; Park, J.H.; Krieg, M.L.; Krna, M.A. Response of plants and the dominant microarthropod, *Cryptopygus antarcticus*, to warming and contrasting precipitation regimes in Antarctic tundra. *Glob Chang. Biol.* **2009**, *15*, 1640–1651. [\[CrossRef\]](#)
52. Enríquez, N.; Tejedo, P.; Benayas, J.; Albertos, B.; Lucíañez, M.J. Collembola of Barrientos Island.; Antarctica: First census and assessment of environmental factors determining springtail distribution. *Polar Biol.* **2018**, *41*, 713–725. [\[CrossRef\]](#)
53. Zhu, L.; Liu, X.; Liu, X.; Jeannotte, R.; Reese, J.C.; Harris, M. Hessian Fly (*Mayetiola destructor*) Attack Causes a Dramatic Shift in Carbon and Nitrogen Metabolism in Wheat. *MPMI* **2007**, *21*, 70–78. [\[CrossRef\]](#) [\[PubMed\]](#)

54. Bhakta, D.; Ganjewala, D. Effect of Leaf Positions on Total Phenolics; Flavonoids and Proantho-cyanidins Content and Anti-oxidant Activities in *Lantana Camara* (L). *J. Sci. Res.* **2009**, *1*, 363–369. [[CrossRef](#)]
55. Cortet, J.; Joffre, R.; Elmholt, S.; Krogh, P.H. Increasing species and trophic diversity of mesofauna affects fungal biomass; mesofauna community structure and organic matter decomposition processes. *Biol. Fertil. Soils* **2003**, *37*, 302–312. [[CrossRef](#)]
56. Maraun, M.; Alphei, J.; Beste, P.; Bonkowski, M.; Bury, R.; Migge, S. Indirect effects of carbon and nutrient amendments on the soil meso- and microfauna of a beechwood. *Biol. Fertil. Soils* **2001**, *34*, 222–229. [[CrossRef](#)]
57. Schulte, P.M. The effects of temperature on aerobic metabolism: Towards a mechanistic understanding of the responses of ectotherms to a changing environment. *J. Exp. Biol.* **2015**, *218*, 1856–1866. [[CrossRef](#)]
58. Payne, R.J.; Thompson, A.M.; Standen, V.; Field, C.D.; Caporn, S.J.M. Impact of simulated nitrogen pollution on heathland micro-fauna, mesofauna and plants. *Eur. J. Soil Biol.* **2012**, *49*, 73–79. [[CrossRef](#)]
59. Wang, S.; Chen, H.Y.H.; Tan, Y.; Fan, H.; Ruan, H. Fertilizer regime impacts on abundance and diversity of soil fauna across a poplar plantation chronosequence in coastal Eastern China. *Sci. Rep.* **2016**, *6*, 20816. [[CrossRef](#)] [[PubMed](#)]
60. Kou, X.; Tao, Y.; Wang, S.; Wu, Z.; Wu, H. Soil mesofauna community composition predicts ecosystem multifunctionality along a coastal-inland gradient of the Bohai Bay. *Land Degrad. Dev.* **2021**, *32*, 4574–4582. [[CrossRef](#)]
61. de Oliveira Filho, L.C.I.; Zeppelini, D.; Sousa, J.P.; Baretta, D.; Klauberger-Filho, O. Collembola community structure under different land management in subtropical Brazil. *Ann. Appl. Biol.* **2020**, *177*, 294–307. [[CrossRef](#)]

Disclaimer/Publisher's Note: The statements, opinions and data contained in all publications are solely those of the individual author(s) and contributor(s) and not of MDPI and/or the editor(s). MDPI and/or the editor(s) disclaim responsibility for any injury to people or property resulting from any ideas, methods, instructions or products referred to in the content.