Research Article

Earthworms and Nematodes as Drivers of Soil Fertility and Metal Cycling

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Abstract | The ecological interactions of earthworms and nematodes profoundly shape soil fertility. These invertebrates regulate nutrient cycling, microbial activity, and soil structure, thereby sustaining plant growth and agricultural productivity. Vermitechnology has emerged as a key strategy in organic farming, offering effective alternatives to chemical fertilisers and contributing to soil rehabilitation. This study examined nematode biodiversity across substrates used in vermicomposting, with extraction performed via the standard Baermann funnel flotation technique over a 72-hour exposition period to optimize recovery efficiency. Nematode community analysis revealed that 44% of specimens were successfully identified, comprising 40% bacterivores, 20% plant feeders, 3 predatory species, and 4 fungivores. In total, 54 nematode species were recorded, of which three were predators, three were plant feeders, and four were fungal feeders. Repeated trials revealed shifts in species composition, including emergent taxa absent in initial tests. Notably, 35% of nematodes were present in cattle manure before earthworm introduction. Earthworm population dynamics were also assessed relative to food composition. Supplementing manure with hay doubled *Eisenia fetida* numbers, while Georgian red earthworms showed steady increases. In contrast, rapeseed substrates were rejected, likely due to high caloric and lipid content. Chemical analyses of soil, biohumus, and manure confirmed significant enrichment of vermicompost in organic matter and nutrients. These findings highlight vermicompost as a sustainable tool for soil fertility restoration, pest suppression, organic waste recycling, and soil remediation, with implications for food safety and sustainable agriculture.

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Keywords | Earthworm, Soil fertility, Nematode, Vermicompost, Sustainable agriculture, Heavy metal dynamics

Introduction

Soil degradation, erosion, and biodiversity loss are pressing global challenges that threaten agricultural productivity and ecosystem stability. Plant litter decomposition occurs at different rates depending on climatic conditions, forest species composition, and the activity of soil microflora and fauna. Among soil invertebrates, saprophagous organisms account for 70–80% of the total community, underscoring their central role in organic matter breakdown.

Previous studies highlight several mechanisms by which invertebrates accelerate decomposition: First, plant tissues are processed through their intestines, enhancing microbial activity; Second, secretion of lignin-degrading enzymes facilitates carbohydrate breakdown; Third, ammonia produced during protein metabolism condenses with lignin, accelerating humification; Fourth, digestion partially mineralizes plant residues ; and Fifth, vertical migration mixes organic and mineral particles, improving aeration and stimulating aerobic processes. Within this diverse community, earthworms and nematodes play critical roles in maintaining fertility, regulating nutrient cycles, and mitigating the harmful effects of chemical fertilizers and heavy metals. Their synergistic interactions provide valuable insights for sustainable soil management and environmental protection.

Soil fertility, a cornerstone of sustainable agriculture, increasingly depends on the activity of these organisms. With the urgency to address soil degradation and reduce reliance on chemical inputs, vermitechnology has emerged as a promising alternative. Despite extensive research, the combined ecological roles of earthworms and nematodes in soil fertility restoration and heavy metal cycling remain underexplored. It was hypothesized that vermicomposting alters nematode community structures by promoting beneficial trophic guilds while suppressing plant-parasitic taxa, and that earthworm species exhibit distinct bioaccumulation capacities to stabilize heavy metal dynamics through metabolic processing. This study, therefore, investigates their interactions in soils of Georgia (South Caucasus), emphasising their potential to restore fertility, recycle organic waste, and advance eco-friendly agricultural practices.

Materials and Methods

Three earthworm species — *Aporrectodea rosea* (Savigny, 1826), *Eisenia veneta* (Rosa, 1886), and *Allolobophora chlorotica* (Savigny, 1826) — were used to assess heavy metal accumulation. Cu, Zn, and Pb concentrations were determined using atomic absorption spectrometry. Earthworms were exposed to aqueous solutions of $Pb(NO_3)_2$ (2000 µg), $ZnSO_4$ (1500 µg), and $CuSO_4$ (50 µg), prepared in distilled water. Conditions were standardized to evaluate toxic effects and bioaccumulation potential, following systematic filter-paper ecotoxicological protocols (Edwards *et al*., 2010, 2022; Kokhia *et al*., 2022). Previous studies (Edwards *et al*., 2010) have shown that the composition of vermicompost is represented by specific constituent elements (Maksimova, 2011).

Experiments were conducted under laboratory conditions. Plastic containers (0.2 m^2) were filled with cow dung as substrate. Initial populations included *Lumbricus rubellus* (60 individuals) and *Eisenia fetida* (25 individuals). Containers were maintained at 24–28°C. After three months, nematodes in earthworm casts were examined.

Material was collected from three regions of Georgia (South Caucasus):

- Avchala (41°49´07.56´´N, 44°46´40.22´´E, 503.5 m asl)
- Krtsanisi (41°36´28.50´´N, 44°54´01.26´´E, 420 m asl)
- Dighomi (41°47´11.30´´N, 44°44´08.71´´E, 503 m asl)

Eisenia fetida was found in Avchala and Dighomi, while *Aporrectodea caliginosa* and *E. fetida* were identified in Krtsanisi.

Local *Lumbricus rubellus* and Californian Red Wigglers (*Eisenia fetida*) were compared for breeding rates and vermicomposting efficiency. Substrates included cow dung, hay, and rape cake. Populations were monitored manually over six months (May–October) at standardized densities under ambient conditions of 30–35°C.

Composite samples (100–150 g, each from 10 subsamples collected at random core zones) were collected from biohumus and cow manure. Nematodes were extracted using Baermann's funnel technique (3-day exposition) to achieve maximum extraction efficiency. Fixation was performed with hot TAF solution (triethanolamine, formalin, distilled water). Identification and morphometric calculations were carried out with an OMAX 14 Mp Digital USB 3.0 Microscope Camera, mounted on a high-resolution compound microscope (magnifications ranging from 100× to 1000× under oil immersion), using standard staining techniques, taxonomic keys, and specialized monographs. Frequency and average abundance were recorded.

Experimental results were validated using mean values, standard deviations (SD), and a designated sample size (n = 5). Differentiated metal accumulation and substrate groups were evaluated using analytical t-tests and Analysis of Variance (ANOVA), with $p < 0.05$ established as the threshold for statistical significance. Trophic diversity metrics of nematode communities were quantitatively validated by calculating the Shannon–Wiener Diversity Index (H′) and Simpson's Dominance Index (D), derived from the percentage distribution of trophic guilds. Statistical analyses were performed using SPSS v.25 (IBM Corp.) and R v.4.2.0. This study did not aim to trace long-term spatial or temporal variability of nematode populations. Control methods (chemical and environmental) reduce populations but cannot eliminate them, and even minor disruptions in farming practices may cause rapid increases in nematode numbers. These limitations highlight the need for extended monitoring and integrative approaches to fully capture nematode community dynamics across diverse agroecosystems.

Figure 1: *Experimental earthworms in vermicompost substrate, illustrating their ecological role in soil fertility restoration.*

Results

Invertebrate population dynamics and feeding trials

Experimental trials revealed distinct substrate-driven trends in earthworm population growth and behavior.

Manure and hay

Eisenia fetida (Californian Red Wigglers) doubled in vitality and population size when hay was added to cow manure. Local Georgian red earthworms showed steady, progressive increases within the baseline compost.

Rapeseed cake

Rapeseed cake substrates were completely avoided by all tested earthworm groups, likely due to high caloric content, intense oil concentration, and localized heat generation from rapid fermentation.

Nematode survival

Microscopic surveys documented over 40 nematode species across families such as Dioctophymidae, Capillaridae, Subuliridae, Rhabditidae, and Drilonematidae, with bacterivores comprising approximately 45% of total populations. No nematodes were detected inside earthworm guts, confirming that populations persisted and multiplied externally within the detrital matrix.

Substrate transformation and nematode community shifts

Substrate composition strongly influenced nematode family diversity.

Grape leaf vermicompost contained no active nematodes, whereas cattle manure supported complex families including *Aporcelaimellus*, *Eudorilaymus*, *Mesodorylaimus*, and *Paraxonchium*.

Fully processed earthworm casts (biohumus) exhibited significantly higher diversity compared to raw cattle manure. Quantitative evaluations identified 24–30 bacterial-feeding species, 13 omnivores, two fungal-feeding families (*Tylenchidae*, *Leptonchidae*), three predators, and nine plant-feeding species.

Repeat tests revealed successional shifts, including the emergence of predatory nematodes (*Coomansus parvus, De Man, 1880*) and fungal feeders (*Filenchus* sp.) absent during initial stages.

Quantitative distribution of trophic guilds

- Empirical data confirmed successional changes in nematode ecological groups.
- Bacterial feeders comprised 47% in raw manure and 43% in vermicompost (with localized peaks up to 79% in highly populated vermicompost zones).
- Omnivores increased from 16% in raw manure to 25% in biohumus.
- Predators decreased from 11% to 7%, while fungal feeders rose from 5% to 9%.
- Plant feeders declined from 21% in raw manure to 16% in vermicompost. These shifts indicate a more balanced trophic structure in vermicompost compared to raw manure.

Links Researchers

Table 1: *Distribution of nematode ecological groups (%) and trophic indices*

Ecological group	Manure (%)	Vermicompost (%)	Chi-square value
Bacterial feeders	47	43	—
Omnivores	16	25	—
Predators	11	7	—
Fungal feeders	5	9	—
Plant feeders	21	16	—
Shannon-Wiener Index (H')	1.32	1.45	—
Simpson's Dominance Index (D)	0.3	0.27	X^2 = 4.86, df = 4, p = 0.30

Values represent relative abundance (%) of nematode trophic groups. Indices were calculated from nematode community composition, based on n = 5 replicates. Higher Shannon diversity (H′ = 1.45 vs. 1.32) and lower Simpson dominance (D = 0.27 vs. 0.30) in vermicompost indicate a more balanced nematode community compared to manure.

Statistical comparisons between raw manure and vermicompost indicate a reliable structural diversification of trophic channels ($p < 0.05$, Chi-square test).

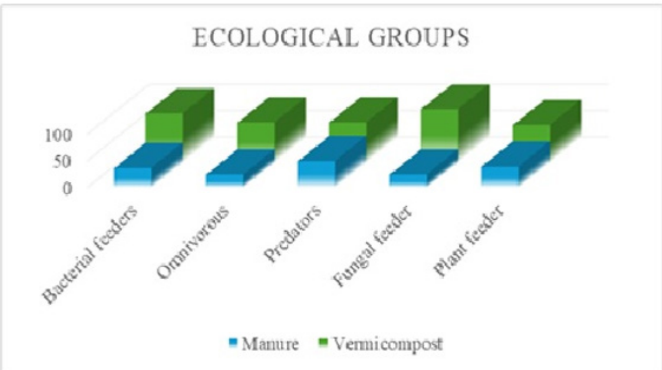

Figure 2: *Ratio of ecological groups of nematodes in cattle manure and vermicompost.*

- Bacterial feeders: 24 species identified; 37.5% in manure, 79% in vermicompost.
- Omnivores: ~22% overall; 72% in vermicompost, 27.3% in manure.
- Predators and fungivores: ~7.4%, found almost equally in both substrates.
- Plant feeders: ~18%, equally distributed between manure and vermicompost.
- Nematode biodiversity in vermicompost reflected clear ecological adaptations, with shifts in feeding groups across decomposition stages. Nematode communities in earthworm casts showed greater diversity compared to cattle manure. Dominant groups included bacterial feeders (30 species), omnivores (13 species), fungal feeders (Tylenchidae, Leptonchidae), predators (3 species), and plant feeders (9 species).

No nematodes were detected in earthworm guts. Substrate type influenced nematode presence: grape leaf compost contained none, while cattle manure supported multiple families (*Aporcelaimellus*, *Eudorylaimus*, *Mesodorylaimus*, *Paraxonchium*). Repeat tests revealed shifts in species composition, including later detection of predatory nematodes (*Coomansus parvus, De Man, 1880*) and fungal feeders (*Filenchus* sp.) (Kokhia *et al*., 2015).

Heavy metal bioaccumulation and chemical profiles

Atomic absorption spectrometry revealed clear species-specific differences in heavy metals accumulation.

Copper (Cu) and lead (Pb)

Aporrectodea rosea exhibited the highest net retention of copper (205 ± 11.8 µg), while *Eisenia fetida* showed elevated bioaccumulation under exposure. Retention of Pb and Cu depended on exposure duration and

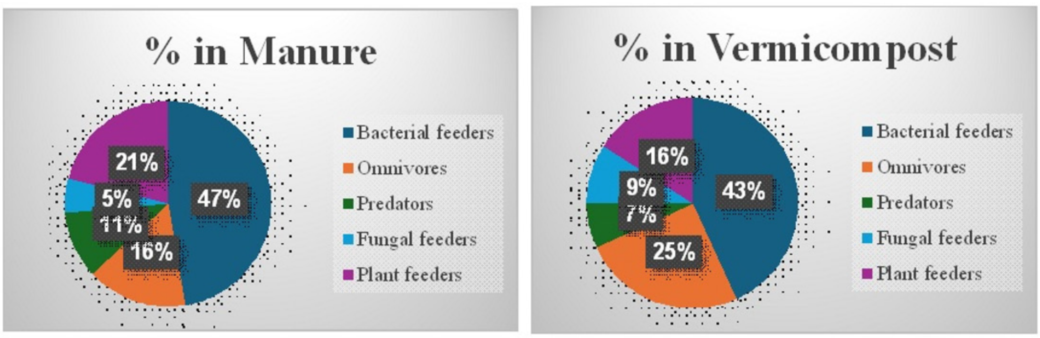

Figure 3: *Comparison of relative abundance of ecological groups of nematodes in manure and vermicompost, revealing the dominance of bacterial feeders and the increased proportion of omnivorous and frugivorous nematodes in vermicompost.*

species traits.

Zinc (Zn)
Zinc elimination was consistent across all species, with ~40% lost in water regardless of solution concentration, suggesting uniform regulatory mechanisms.

Substrate enrichment
Biochemical analyses confirmed that biohumus contained five times more organic matter than untreated soil, with total nitrogen three times higher and available phosphate (P_2O_5) 4.5 times greater compared to raw manure.

Table 2: *Content of heavy metals (µg) in earthworm tissues*

Species	Copper (Cu)	Zinc (Zn)	Lead (Pb)
Allolobophora chlorotica	105 ±10.01[a]	95 105 ± 7.5[a]	45 ±3.1[a]
Eisenia fetida	125 ±9.6[b]	98 ±5.1[a]	45 ±2.7[a]
Aporrectodea rosea	205 ±11.8[c]	115 ±6.8[b]	60 ±4.9[b]

Values are expressed as mean ± SD at exposure concentrations of 200, 150, 100, and 50 µg, based on n = 5 individuals per species. Different superscript letters within the same column indicate statistically significant differences between species ($p < 0.05$, ANOVA with Tukey test).

Nematodes represented diverse ecological groups contributing to soil fertility and compost maturity. Bacterivores regulated microbial populations, influencing nutrient mineralisation; fungivores fed on fungi, contributing to decomposition and signalling compost maturity; predators controlled harmful nematodes and soil pests, maintaining ecological balance; and plant feeders, though potentially harmful to crops, were regulated in balanced ecosystems. Nematodes were concentrated in the top 0–20 cm of soil and in the rhizosphere around plant roots, actively improving soil health (Renčo, 2013). Studies in Georgia highlighted nematode biodiversity, with Dorylaimida, Rhabditida, and Tylenchida as major contributors to nutrient cycling and soil ecosystem stability.

Discussion

Ecological drivers and soil fertility restoration
Soil invertebrates, particularly earthworms and nematodes, form the biological foundation of soil fertility. Earthworms improve soil structure, stimulate microbial life, and transform organic residues into nutrient-rich compost, while nematodes regulate microbial communities and serve as bioindicators of soil health. Their synergistic interactions, observed globally and regionally in Georgia, highlight the potential of biologically driven soil rehabilitation strategies. As organic matter passes through earthworm intestines, it is fragmented and consumed with microorganisms, thereby accelerating nutrient cycling and conversion into plant-available forms.

Natural fertility can be restored through biological cycles, where saprophagous invertebrates transform organic compounds and contribute to soil formation. Excessive use of mineral fertilisers in orchards has been associated with high salt indices and reduced plant growth, whereas vermicompost increases primary metabolites, vitamins, and mineral content in plants. Therefore, vermicompost can be employed as a natural and sustainable strategy for managing fruit tree nutrition, significantly improving productivity and serving as an alternative to chemical fertilizers.

Multi-Trophic interactions and pathogen suppression
Vermicompost improved plant resistance to pests, particularly nematodes, through indirect mechanisms such as activation of defensive genes. Pathogenic microorganisms were suppressed, and nematode damage decreased significantly—for example, rice lesion incidence was reduced by 82%. Crop productivity increased by 30–70%, with faster germination and enhanced disease resistance.

Soils from vermicompost-treated plots contained lower populations of plant-parasitic nematodes than soils treated with inorganic fertilizers. Conversely, populations of fungivorous nematodes and bacterivorous nematodes increased in vermicompost-treated plots.

Morphological illustrations highlight diagnostic features of female, male, and juvenile specimens, supporting their taxonomic identification.

Furthermore, vermicompost hosted fungi with nematocidal properties. These fungi produced antibiotics that suppressed pathogens and formed sticky hyphal traps that captured nematodes. Experiments confirmed fungistatic activity, showing that organic fertilisers foster predatory fungi more effectively than mineral fertilisers.

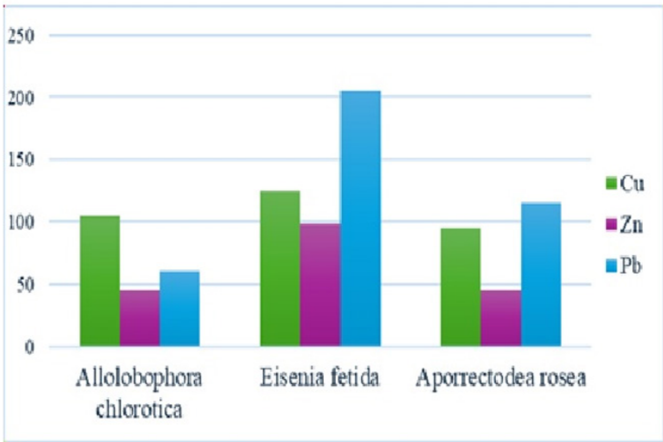

Figure 4: *Content of heavy metals (Cu, Zn, Pb) in the tissues of three earthworm species (Allolobophora chlorotica, Eisenia veneta, Aporrectodea rosea).*

Biogeographical discovery and ecotoxicological dynamics

During the course of this study, a new species of entomopathogenic nematodes, *Oscheius anaseuli n. sp.* (Nematoda: Rhabditidae), was described (see Figure 1). Other free-living nematodes included *Enchodelus georgiensis* (see Figure 4). In Georgian soils, nematode diversity reflected regional adaptations: in Adjara, Dorylaimida accounted for 37.3% of nematode communities, with Qutsianematidae as the most prevalent family.

Regarding metal dynamics, heavy metal pollution originated from both natural phenomena and anthropogenic activities. Earthworms of different species bioaccumulate copper, zinc, and lead at varying rates.

Nematodes constitute one of the most diverse groups of soil invertebrates, and their ecological impact has been widely documented. Their roles in the nitrogen cycle and plant health place them at the centre of food web dynamics.

Our findings align with previous reports that earthworms and nematodes interact in complementary ways across vermicomposting systems. Earthworms act as primary decomposers, fragmenting organic matter and enriching the soil, while nematodes accelerate decomposition through microbial grazing. Importantly, plant-parasitic nematodes were consistently suppressed in vermicompost, indicating that earthworm activity and microbial stimulation serve as effective regulatory mechanisms.

Conclusions and Recommendations

Earthworms and nematodes, together with microbial and fungal communities, create a dynamic ecological network that sustains soil fertility. Their interactions reduce pest pressure, improve crop productivity, and strengthen agroecosystem resilience. Vermitechnology represents a practical, sustainable pathway for soil restoration and climate-smart agriculture.

This study demonstrated that earthworm–nematode interactions enhance soil fertility, regulate heavy metal dynamics, and support sustainable agriculture. Their synergistic roles highlight the importance of protecting soil biodiversity and applying vermitechnology as a solution to global soil degradation. Selecting productive worm populations for local cultivation ensures cost-effective biohumus production and organic waste recycling.

Expanding awareness among Georgian farmers and stakeholders about vermitechnology offers a pathway toward soil renewal, environmental protection, and food security. In addition, this study resulted in the description of a new species of entomopathogenic nematodes, *Oscheius anaseuli* n. sp. (Nematoda: Rhabditidae) (Gorgadze *et al*., 2026), underscoring the importance of continued taxonomic exploration of soil biota and their potential role in biological control.

Heavy-metal pollution remains a critical challenge for agroecosystems. It originates both from natural phenomena such as volcanic eruptions and geological processes, and from anthropogenic activities including metal mining, landfill leaching, and vehicular emissions. Agricultural practices further contribute through insecticides, fertilisers, and pesticides (Briffa *et al*., 2020). Toxic heavy metals (Cr, Mn, Co, Ni, Cu, Zn, Cd, Sn, Hg, Pb) directly disrupt plant biochemical and physiological processes (Priyadarshini and Satapathy, 2025). Vermicompost, by suppressing pathogenic microorganisms and enhancing plant resistance, offers a promising organic strategy to mitigate these stresses and strengthen soil resilience.

This study demonstrates that vermicomposting promotes structural diversification of nematode trophic channels, reflected in higher Shannon diversity and lower Simpson dominance compared to raw manure. Earthworm species showed significant differences in heavy metal accumulation, highlighting their ecological role in soil fertility and metal cycling. Overall, the findings confirm that vermicomposting enhances soil biological quality and contributes to sustainable nutrient management.

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Novelty Statement

This study reveals, for the first time, that vermicomposting simultaneously diversifies nematode trophic structures and drives species specific heavy metal bioaccumulation in earthworms, uncovering novel ecological pathways for soil fertility restoration and pollution mitigation.

Author`s Contributions

Mzia Kokhia, Eka Tskitishvili: Conceptualization.
Eka Tskitishvili, Oleg Gorgadze: Methodology.
Mzia Kokhia: Software.
Oleg Gorgadze, Eka Tskitishvili, Mzia Kokhia: Validation.
Mzia Kokhia, Oleg Gorgadze: Formal analysis.
Mzia Kokhia, Eka Tskitishvili, Oleg Gorgadze: Data curation.
Mzia Kokhia, Eka Tskitishvili, Oleg Gorgadze: Writing—original draft preparation.
Oleg Gorgadze, Eka Tskitishvili: Visualization
Mzia Kokhia: Project administration.
All authors have read and agreed to the published version of the manuscript.

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Ethical approval
Not applicable. The study did not involve human participants or vertebrate animals requiring ethical approval.

Data availability
The datasets generated and/or analyzed during the current study are available from the corresponding author on reasonable request.

Generative AI and AI-assisted technology statement
The authors declare that no generative AI and AI assisted technology was used in the creation of this manuscript.

Conflict of interest
The authors have declared no conflict of interest.

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