# Structure and function role of soil nematode communities in different types of vegetation in Yogyakarta and Central Java, Indonesia

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Abstract. Indarti S, Utami SNH, Putra NS, Maharani R. 2024. Structure and function role of soil nematode communities in different types of vegetation in Yogyakarta and Central Java, Indonesia. Biodiversitas 25: 2765-2772. Soil nematodes are crucial parts of the soil organism community and play a significant role in many ecological soil processes. It is necessary to pay greater attention to the lack of knowledge regarding the composition and role of the soil nematode community in different vegetations. Hence, the nematode structure community from minimal to soil stillages have been observed and compared from seven distinct types of vegetation. This study aimed to determine the soil nematode community structure, which includes the dominance, abundance, richness, diversity, and functional role of nematodes, as well as other chemical soil properties. Vegetation type represents soil management and has an impact on the life of soil organisms such as nematodes. A total of approximately 2328 soil nematodes were found, and 18 genera were identified. The Nematode Indicator Joint Analysis (NINJA) revealed that the vegetation type, land use, and soil chemical properties influence the nematode's abundance, dominance, and structure differently. Through those results, the authors confirmed that soil nematodes in cultivated vegetation are dominated by the trophic groups of plant-parasitic nematodes and bacterivores. Conversely, predator, omnivores, and fungivores nematodes dominate in soil with minimal tillage. Overall, this study highlights the importance of soil nematode analysis in sustainable agricultural soil ecosystems.

Keywords: Bioindicators, diversity, dominance, nematode abundance, soil status

# **INTRODUCTION**

Nematodes community characteristics are an effective bioindicator of soil disturbance (Boutsis et al. 2011) or any other ecosystems (Ridall and Ingel 2021) and provide ecosystem services (Mokuah et al. 2023; Neher and Powers 2023). They are considered an important part of the soil food web. Bioindicators for soil environment disturbance and the state of the soil food web are obtained from a nematode faunal study. Due to their feeding types and life strategies, soil nematodes are an important part of terrestrial biodiversity. Their activities also impact important ecological processes such as maintaining it, including the cycling of carbon, breaking down of organic matter (Kouser et al. 2021), and feeding on other soil microorganism populations (Iqbal and Jones 2017). Nematode community indices, which have been constructed based on the functional guilds, can be used to establish a connection between variations in the composition of nematode communities and ecological processes (Kouser et al. 2021).

The nematode community's structure provides an effective way for evaluating the soil's biological quality and function because for many reasons such as they are found everywhere decomposition occurs, their morphology reflects feeding behavior, their interactions with other soil organism, their food-specificity, short response time, ease of isolation from the substrate, and their genus identification is relatively straightforward (Bongers and Bongers 1998). Since nematodes' trophic position may be used to infer information about their functional role in soils, they are frequently grouped into trophic groups according to the guilds in which they feed (bacterivores, fungivores, herbivores, omnivores, and predators) (Guan et al. 2018; Li et al. 2022). Therefore, the presence and abundance of particular taxa serve as markers for the trophic levels at which those species are found, indicating the complexity of the food web at those levels (Yeates et al. 1993; Ferris et al. 2001). However, broad-scale biogeographical trends in nematode abundance and functional group composition remain poorly understood (van den Hoogen et al. 2019).

Nematode diversity has already been demonstrated to be impacted by changes in land use. Land physical impacts such as crop rotation, cover crops, tillage, and compaction affect the microbial composition, including nematodes (Nisa et al. 2022; Shokoohi 2023). Agricultural practices and fallow land, as well as spring and autumn seasons, also showed distinct nematode communities (Vink et al. 2020). In heavy tillage management, nematode diversity was found to be lower (Freckman and Ettema 1993). Soil organisms and biodiversity decreased or were less diverse in intensive agricultural practices. Intensification also resulted in low functional guilds or soil food web complexity (Tsiafouli et al. 2014). Besides that, soil processes could affect the nitrogen content in the soil directly or indirectly, which could affect the nematodes (Xing et al. 2022). Different climate regions showed different impacts of any practices on nematode diversity and abundance. As reported by Xing et al. (2022), the addition of N impacted nematodes differently in cold and warm regions. Therefore, climate and ecosystem could influence the nematodes by nitrogen and water content availability, as well as the temperature of the soil (Xing et al. 2022). At the same time, the number of dominant nematode genera may be caused by environmental filtering brought on by crop types and water management (Liu et al. 2016). The impacts of ecosystem disruption on soil health may be mediated by soil nematodes, particularly bacterivores that live freely (Shokoohi 2023). Due to various things that could affect the nematodes, the purpose of this study was to investigate whether variation in vegetation, nematode communities, and nematode functionalities varied predictably with soil properties. The accurate investigation of soil nematode communities is essential to understand their ecological function. Bioindicator studies can have great potential to contribute to optimizing agricultural systems, input practices and crop management, as well as creating policies to regulate landscape management (Devi 2020) for the sustainability and synergy of agricultural and natural ecosystems.

# MATERIALS AND METHODS

#### Samples collection and nematode extraction

A randomized sampling method was conducted, and samples were gathered from the soil at 0-20 cm (in-depth) below the ground of 7 different vegetations, including Spruce (*Picea* sp.), teak tree (*Tectona grandis* L.f.), malapari (*Pongamia pinnata* (L.) Pierre), banana (*Musa* sp.), potato (*Solanum tuberosum* L.), beetroot (*Beta vulgaris* L.), tobacco (*Nicotiana tabacum* L.), representing different soil management in Yogyakarta and Central Java, Indonesia. Each location's soil samples were composited from 5-10 cores (depending on the land-wide area). The nematodes were extracted from the soil using a modified version of the Whitehead-Tray extraction method (Coyne et al. 2014), while the cyst nematodes were collected using the Baunacke method (van Bezooijen 2006).

# Nematode identification

Soil nematodes were identified to the level of the genus. Morphological characteristics such as body characteristics, lip region, stylet type, head shape, pharyngeal overlap, tail type, caudal alae, spicule, and vulva position were used (Heyns 1971; Mai and Lyon 1975). The morphology of nematodes was observed by using a light microscope (Olympus CX-31, Tokyo, Japan) with an optical camera (OptiLab Miconos, Yogyakarta, Indonesia). Nematodes were picked and put on glass slides with 20  $\mu$ l of sterilized water. The genus identification was finalized using a light microscope with up to 400x magnification. The number of viable cysts was determined using a counting dish under a stereoscopic microscope (Leiva et al. 2020).

#### Nematode analysis and data visualization

Soil nematodes were categorized into a 1-5 colonizerpersister (c-p) class and plant-parasite (p-p) class, which vary from extreme R- to extreme K-strategists according to Bongers (1990) and Bongers and Bongers (1998). Depending upon the abundance of functional guilds of nematodes, various indices were calculated to analyze the functional role of nematodes, ecological indices, and function indices using NINJA (Nematode Indicator Joint Analysis) (https://shiny.wur.nl/ninja/). NINJA is a tool to perform calculations for nematode-based monitoring of soil quality (Sieriebriennikov et al. 2014). All identified nematodes were classified into five main trophic habits, i.e., bacterivores, fungivores, herbivores/Plant-Parasitic Nematodes (PPNs), omnivores, and predators (Yeates et al. 1993). Each nematode taxon in the investigated areas was analyzed to calculate the nematode biodiversity. The diversity of the soil nematode in various vegetations was analyzed using the Shannon-Weiner index (H') (Ifo et al. 2016).

ID= H'= 
$$-\sum$$
Pi ln P<sub>i</sub> where Pi  $=\frac{ni}{N}$ 

Where:

ID = Index diversity

- H' = Shannon-Weiner diversity index
- ni = individual number of each genus of nematodes
- N = total number of all genera of nematodes
- Pi = abundance index

Chemical properties of soil, such as the percentage of C and organic matter, were analyzed using the Walkley and Black method (Walkley and Black 1934). N total was analyzed using the Kjedahl method (FAO 2021). The C/N ratio was calculated by comparing C and N values. Principal Component Analysis (PCA) correlation biplots were constructed between nematode parameters and soil properties with Minitab 17.

# **RESULTS AND DISCUSSION**

### Nematode analysis

In this survey, 18 nematode genera were found, with a total of 2328 nematodes in all 7 vegetations. These genera were grouped based on the nematodes' feeding type (Table 1) and are classified as c-p1, c-p2, c-p4, p-p2, and p-p3. The abundance of nematodes in each vegetation varies (Table 2). In contrast, vegetation with a total nematode abundance above 500 nematodes/100 mL of soil was found in potato (*S. tuberosum*) and beetroot (*B. vulgaris*) plants. Potato and beetroot vegetation in this research is classified as having high soil management due to being cultivated by farmers with crop management.

A well-balanced ecosystem with high biodiversity above and below the soil surface, including its soil microbial diversity, is essential for soil health (Wright et al. 2015). Criteria of bioindicator use are described by Ridall and Ingels (2021), such as that biological indicators need to be representative of all ecosystem components, abundant, common, and present in an undisturbed region, well studied, cheap, and easy to learn. Nematodes as soil organisms can be bioindicators to determine soil health and soil quality because they can be found in all environments, in all types of soil, are simple to collect, and are neatly categorized into functional feeding types (Bileva et al. 2014; Ghanem et al. 2024). This study showed that from 18 nematode genera, only one genus omnivore was found in almost every vegetation type, even at low levels. Research by Laasli et al. (2022) showed that omnivores are at low levels at a depth of 15 cm due to the possible influence of other nematode groups. Omnivorous nematodes are rather large and more sensitive to environmental changes (Zhao and Neher 2013). Laasli et al. (2022) also showed that omnivorous nematodes were normally more associated with silty soils with high-weighted abundance. Except for omnivores, the total nitrogen content increased in all nematode trophic groups (Kouser et al. 2022). However, cultivation and synthetic chemical fertilizers have been reported to reduce some nematode genera, mostly bacterialfeeding nematodes (Zhao and Neher 2013).

 Table 1. Main nematode taxa identified from various soil vegetations in Yogyakarta and Central Java, Indonesia (c-p: colonizer-persister; p-p: plant-parasite)

Family	Genus	Feeding type	c-p Class	p-p Class
Criconematidae	Criconemoides	Herbivores - ectoparasites	0	3
Hoplolaimidae	Helicotylenchus	Herbivores - semi-endoparasites	0	3
Hoplolaimidae	Hoplolaimus	Herbivores - semi-endoparasites	0	3
Hoplolaimidae	Rotylenchulus	Herbivores - sedentary parasites	0	3
Heteroderidae	Globodera	Herbivores - sedentary parasites	0	3
Heteroderidae	Meloidogyne	Herbivores - sedentary parasites	0	3
Pratylenchidae	Pratylenchus	Herbivores - migratory endoparasites	0	3
Tylenchidae	Tylenchus	Herbivores - epidermal/root hair feeders	0	2
Aphelenchoididae	Aphelenchoides	Fungivores	2	0
Aphelenchidae	Aphelenchus	Fungivores	2	0
Cephalobidae	Acrobeles	Bacterivores	2	0
Cephalobidae	Cephalobus	Bacterivores	2	0
Cephalobidae	Eucephalobus	Bacterivores	2	0
Rhabditidae	Mesorhabditis	Bacterivores	1	0
Rhabditidae	Rhabditis	Bacterivores	1	0
Steinernematidae	Steinernema	Bacterivores	1	0
Dorylaimidae	Dorylaimus	Omnivores	4	0
Mononchidae	Mononchus	Predators	4	0

Table 2. Mean abundance of nematode genera associated with various soil vegetations in Yogyakarta and Central Java, Indonesia (nematodes/100 mL soil)

Locat	tions	1	2	3	4	5	6	7
Vegetations		Spruce ( <i>Picea</i> sp.)	Teak (Tectona grandis)	Malapari (Pongamia pinnata)	Banana (Musa sp.)	Potato (Solanum tuberosum)	Beetroot (Beta vulgaris)	Tobacco (Nicotiana tabacum)
Soil management		No	Less	Less	Less	High	High	High
Meloidogyne		0.00	0.00	0.00	48.00	5.33	23.33	13.33
Hoplolaimus		1.33	0.00	0.00	0.00	0.00	30.00	0.00
Helicotylenchus		0.00	0.00	0.00	80.00	0.00	233.33	1.67
Criconemoides		0.00	24.00	5.33	0.00	0.00	0.00	0.00
Globodera		0.00	0.00	0.00	0.00	58.67	0.00	0.00
Pratylenchus		0.00	0.00	0.00	176.00	0.00	0.00	0.00
Rotylenchulus		0.00	0.00	0.00	0.00	0.00	26.67	0.00
Tylenchus		0.00	10.67	5.33	0.00	0.00	16.67	6.67
Dorylaimus		0.00	13.33	24.00	58.60	48.00	90.00	23.33
Mononchus		1.33	2.67	18.67	5.33	10.66	0.00	0.00
Rhabditis		4.00	0.00	0.00	0.00	474.67	130.00	90.00
Steinernema		9.33	37.33	26.67	0.00	26.67	0.00	0.00
Mesorhabditis		0.00	0.00	0.00	10.67	240.00	6.67	5.00
Eucephalobus		0.00	0.00	0.00	21.33	42.66	30.00	6.67
Cephalobus		0.00	0.00	0.00	0.00	0.00	30.00	5.00
Acrobeles		0.00	0.00	0.00	0.00	0.00	3.33	1.67
Aphelenchoides		0.00	0.00	0.00	0.00	37.3	0.00	0.00
Aphelenchus		0.00	0.00	0.00	0.00	0	0.00	26.67
Total nematodes in each vegeta Total nematodes	ation	15.99	88.00	80.00	399.93 2327.89	943.96	620.00	180.01

#### Nematode community structure

The greatest variety of nematode genera was found in beetroot vegetation, reaching 11 genera, and the least in spruce (Picea) vegetation, where only 4 genera were found (Table 2). The PPNs with more than 50% fraction percentage of total nematodes were found in organic crop cultivation with beetroot plants. They were found in cultivation with minimal soil treatment, such as banana (Musa sp.) plants. In comparison, the PPNs of each vegetation varied depending on its plant host. Land with less tillage, like teak (T. grandis) and malapari (P. pinnata) vegetation, has a higher percentage of ectoparasitic nematode fractions. Meanwhile, land with intensive tillage, such as land with potato plants, is dominated by only one type of PPN, which is sedentary nematodes, which reach almost 100% of total PPNs. However, the percentage of each feeding group nematode population is more evenly found in land with malapari plants, except for the fungivores group (Figure 1). In this study, the percentage population of non-parasitic nematodes was dominated by bacterivorous nematodes in almost all vegetation, with the highest percentages found in spruce and potato vegetation. It's a bit contrary because the spruce vegetation land represents the land without tillage, while potato vegetation land represents the land with high tillage. Schmidt et al. 2020 explained that minimum tillage land had more bacterivorous nematode than in the plow-based system. Regardless, Griffiths and Bardgett (1997) also explained that the bacterivorous nematode population could increase where PPNs are present due to the greater availability of food distribution in the soil, as can be seen that the PPN population in the potato vegetation was high. Nevertheless, analysis of the diversity and dominance of nematodes in all vegetation in this study showed moderate diversity and low dominance (Table 3). The percentage of herbivore nematodes based on their feeding site (Figure 1.C), their cp and p-p class (Figures 1.D-1.E) varied.

Xiao et al. (2021) mentioned that nematode distribution is affected mostly by organic matter. Organic matter is reflected by the C/N ratio (Garcia et al. 2022). This study showed that organic matter can influence omnivores and herbivores. The C/N ratio and plant vigor are increased by adding organic matter, and these factors have an indirect effect on nematode activity, particularly herbivores or PPNs (Indarti et al. 2023). Research by Garcia et al. (2022) showed that PPN abundances appear to be lower in soil with lower C/N ratios. Soil organic matter could affect nematodes with certain life-strategy, such as the fastgrowing nematodes like bacterivores and fungivores that include r-strategists nematodes have a positive correlation with it (Quist et al. 2019).

Cropping types under control conditions also had a considerable impact on the density of all soil macrofauna (Wang et al. 2018). Soil organic matter can be positively related to soil nematodes due to other microbial presence, such as bacteria and fungi, which can increase the presence of nematodes and influence plant growth and development (Papatheodorou et al. 2004). Besides that, fungivores nematode composition was higher in tobacco plants, which means that decomposition could be slower than that with lower fungivores nematodes due to fungi decomposition that is considered slower compared to the bacterial counterpart (Laasli et al. 2022). Yeates et al. (1983) showed that increasing dominance by Rhabditidae resulted in the reduction of PPNs. Laasli et al. (2022) mentioned that disturbed systems dominated by fungi decomposition. Nematode are community composition within the soil reflects the soil food web's changes, in which smaller ratios of fungivorous and bacterivorous nematodes were associated with faster rates of decomposition and nutrient turnover (Shokoohi 2023).

The c-p triangle was constructed from all different vegetation (Figure 2). The soil from banana and malapari vegetation was close to soil stability, as structure nematodes (c-p3-5) tend to increase by 80%. However, the other soil samples were close to soil enrichment, as basal nematodes (c-p2) decreased alongside the increase in soil enrichment. Based on food web analysis, all locations were shown to have maturing soil in different values, rich in N, low C/N ratio, and regulated soils with bacterial features.

The lowest value of enrichment index is found in the soil of banana vegetation  $(\pm 65)$ , while spruce and teak trees had 100 values (Figure 3). The PCA accounts for 78.3% of the variation, with 48.6% attributed to PC1 and 29.7% to PC2 (Figure 4). Soil chemical variables, nematodes abundance based on their feeding type, and study sites from different vegetations indicated that PC1 was mainly contributed by herbivores or PPNs and omnivores, which are dominant in soil characterized by organic matter and percentage of total C and N. The relative proportions of predators were correlated to the negative side of the plot as opposed to percentages C and N total. Bacterivores and fungivores were correlated to the C/N ratio and predatory nematodes. Bacterivorous and predatory nematodes are known to contribute to nitrogen mineralization, as well as predatory nematodes that feed on microbial grazing nematodes (Yadav et al. 2018). Nevertheless, in this study, they were located close to the center, indicating that they had little impact on the changes.

Table 3. Nematode Shanon-Wiener diversity index and soil chemical parameters from various soil vegetations in Yogyakarta and Central Java, Indonesia

Vogetation	H' diversity		C dominance	Soil parameters				
vegetation				C organic (%)		N total (%)	C/N ratio	Organic matter (%)
Spruce	1.07	Moderate	0.42	Low	0.78	1.35	13.91	0.06
Teak Tree	1.36	Moderate	0.29	Low	2.78	4.8	24.35	0.11
Malapari	1.42	Moderate	0.26	Low	2.99	5.16	23.19	0.13
Potato	1.49	Moderate	0.32	Low	13.52	23.32	20.68	0.65
Banana	1.53	Moderate	0.27	Low	8.41	14.5	33.05	0.25
Beetroot	1.85	Moderate	0.22	Low	9.46	16.3	14.78	0.64
Tobacco	1.62	Moderate	0.30	Low	10.17	17.54	16.4	0.62



**Figure 1.** Soil nematode community's fraction from various soil vegetations in Yogyakarta and Central Java, Indonesia. A. Percentages of the total nematode numbers according to their feeding type. B. Percentage of free-living nematodes. C. Percentages of Plant-Parasitic/herbivores Nematodes (PPNs). D. Percentages of free-living nematodes according to c-p (colonizer-persister) values. E. Percentages of PPNs according to p-p (plant-parasitic) values. 1. Banana (*Musa* sp.); 2. Beetroot (*Beta vulgaris*); 3. Malapari (*Pongamia pinnata*); 4. Potato (*Solanum tuberosum*); 5. Spuce (*Picea* sp.); 6. Teak (*Tectona grandis*); 7. Tobacco (*Nicotiana tabacum*)



Figure 2. The c-p (colonizer-persister) triangle depicting soil status from various vegetations in Yogyakarta and Central Java, Indonesia



Figure 3. Food web analysis of nematode communities as bioindicators of soil health from various vegetations in Yogyakarta and Central Java, Indonesia



Figure 4. Principal Component Analysis (PCA) biplot of different feeding habits of nematode abundance and soil chemical parameters from various vegetations in Yogyakarta and Central Java, Indonesia. Data were transformed before analysis to standardize all parameters

Previous studies showed higher nematode abundance was found in higher-input organic farming systems. The lowest tillage treatments had the highest degree of species variety, and all nematodes, especially the free-living ones, prefer organic enhancements as they don't interfere with plant feeders (Freckman and Ettema 1993; Boutsis et al. 2011). Higher nitrogen input could promote the plant root mass and also increase the PPNs abundance (Schmidt et al. 2020). Intensive land use known could decrease nematode diversity. Thev have indicated that herbivores. bacterivores, and omnivores were higher in organic vegetable fields than in conventional fields (Yang et al. 2021; Shokoohi 2023). In this study, beetroot vegetation was cultivated under organic crop management. This also showed a high number of nematodes as herbivores, bacterivores, and omnivores. Each trophic group of nematodes was differently distributed in all localities surveyed. However, there were no significant differences in nematode abundance and diversity among the vegetation. A study by Gutiérrez et al. (2016) showed that heavy metals and contaminants from different land uses did not affect the total number, diversity, and community structure of nematodes. Besides that, genera with k-strategy were negatively affected. Nematode at high soil management tends to shift a little towards land with stressful conditions, but still, all vegetation is grouped into maturing soil with soil enrichment. Similar soil properties might be a good explanation for the lack of notable variations in nematode richness and diversity. Our results showed similar findings to those of Xiao et al. (2021), who found no significant differences in nematode abundance between different ecosystems. The physicochemical characteristics of the soil have a significant impact on the very complex pattern of belowground richness and variety, including soil nematodes (Bardgett and van der Putten 2014; Xiao et al. 2021). Cao et al. (2020) mention that the alterations in soil nutrients, tree species composition, and soil nematode feeding characteristics were the main reasons for the changes in the assembly of the soil microbial community.

Finally, this study showed that soil nematodes in cultivated vegetation are dominated by the trophic groups of plant-parasitic nematodes and bacterivores. Conversely, predator, omnivore, and fungivore nematodes dominate in soil with minimal tillage. Overall, this study highlights the importance of soil nematode analysis in sustainable agricultural soil ecosystems.

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