



DIVERSITY AND POPULATION OF NON-PARASITIC NEMATODES IN VARIOUS ACCESSIONS OF PEPPER (*Piper nigrum* L.) BANGKA ISLAND

Lesta^{1*}, Riski Meliya Ningsih¹, Badriyah¹, Nita Anggi Felisha¹

¹Pertanian Presisi, Rekayasa Elektro dan Industri Pertanian, Politeknik Manufaktur Negeri Bangka Belitung

*Correspondence : Lesta@polman-babel.ac.id

Received April 11, 2025; Revised May 15, 2025; Accepted May 19, 2025

ABSTRACT

Nematodes are soil organisms that play an essential role in agricultural ecosystems, both as plant pathogens and soil health indicators. Non-parasitic nematodes or free-living nematodes play an essential role in soil food webs. This study aims to analyze the diversity and population of non-parasitic nematodes in *Piper nigrum* L. from various pepper accessions on Bangka Island. soil samples taken from seven locations and extracting nematodes using centrifugation flotation techniques. Nematodes were identified based on morphological characteristics by using a stereo and binocular microscopes and validated to the nematode identification keybook. The population of identified nematodes was calculated using the absolute population calculation formula. The results showed variations of non-parasitic nematodes in each location, which were influenced by environmental factors and types of pepper accessions. The most dominant population of identified nematodes was classified as Rhabditidae with 92.42%, Mononchidae, and *Aphelenchus avenae* (<92.42%). The nematode population in Lampung Daun Lebar accession was 41 individuals, Lampung Daun Kecil (17), Nyelungkup (9), and Merapin (healthy control) 65 individuals. Non-parasitic nematodes have roles as bacterivores (soil bacteria eaters), fungivores (soil fungus eaters), predators of parasitic nematodes, and decomposers in the soil.

Key words : Accession of pepper, Bangka island, Diversity, Free-living nematodes, Population.

INTRODUCTION

The demand for pepper continues to increase in Indonesia, the world's third-largest producer and exporter (The Center for Agricultural Data and Information System Agricultural Statistic, 2022). *Piper nigrum* L. is a spice plant contributing to the Indonesia's foreign exchange (Suryanti *et al.*, 2015). Moreover, national pepper productivity has decreased. The decline of pepper production in Indonesia during 2022-2023 was 6.58 thousand tons from 89.28 to 83.70 thousand tons.

The decline in pepper plant productivity occurred in several pepper plantation centers in Indonesia, including the Bangka Belitung Islands. Various factors have caused the decline in pepper plant productivity, one of which is the attack by Plant Pest Organisms (PPO), yellow

disease, which caused a decrease in the harvest area by 1.81 ha in Indonesia (Ministry of Agriculture of The Republic of Indonesia, 2020) and the health of the soil environment which was relatively low with poor nutrients. Organic content, structure, and microbiome in the soil, influences nematode populations (Gives, 2022).

Lesta, (2023) reported that the incidence of disease due to yellow disease in Bangka Regency reached 25% in the Lampung Daun Lebar (LDL) accession. Several pepper accessions in Bangka include Lampung Daun Lebar, Lampung Daun Kecil, Chunuk, Jambi Variety, Nyelungkup, Bogor, Peremis, Keriwil, Merapin Daun Kecil (Prayoga *et al.*, 2020). The Lampung Daun Lebar (LDL) accession is a pepper accession that farmers in

Bangka widely plant because it has a high productivity number but is susceptible to yellow disease (Hamid & Rahayuningsih, 1990). The diversity of nematodes and the population of each group of nematodes in an agroecosystem can be used as a benchmark for soil quality conditions that refer to soil health (Widowati *et al.*, 2014). Kimenju *et al.*, (2009) added that nematodes can interact with soil organisms in the food web or the complexity of the food web.

Non-parasitic plant nematodes can live freely, play an important role in the decomposition of organic matter, and recycle nutrients in the soil. Most non-parasitic nematodes are eaters of soil bacteria (bacterivores) and soil fungi (fungivores). Non-parasitic nematodes do not eat organic soil matter directly, but

rather bacteria and fungi that act as decomposers. According to Widowati *et al.* (2014), these non-parasitic nematodes play an important role in accelerating the decomposition and mineralization process by recycling minerals and nutrients from bacteria, fungi, and other substrates so that they can be returned to the soil in a more straightforward form and easily absorbed by plant soil (McSorley, 2009).

Based on the explanation regarding the importance of the occurrence of yellow disease and the role of non-parasitic nematodes in the soil in various pepper accessions in Bangka, further studies are needed regarding the diversity and population of non-parasitic nematodes that can be used as an effort to improve the soil environment for pepper plantations.

MATERIALS AND METHODS

Sampling Sites

Soil samples were taken from three pepper plants with symptoms of yellow disease in each garden (gardens were selected based on the presence of diseased plants). One plant was taken at three sampling points and repeated three times (three gardens). The soil taken was 500 grams of pepper planting soil to a depth of 10-15 cm using a soil drill. The soil samples were then placed in labeled sample plastic and stored in a storage box to prevent damage due to temperature and humidity fluctuations during the trip (Plant Quarantine Agency, 2010).

Extraction and Isolation of Non-Parasitic Nematodes from Soil Samples

Soil nematode extraction used the modified centrifugation flotation method from Caveness & Jensen, (1955). Soil samples from each point were composited and then ground until homogeneous. 100 ml of soil was mixed with 800 ml of water, stirred evenly, and waited until there was no sediment. The suspension was sedimented for ± 40 seconds. Then, the suspension was poured onto a graduated sieve at 50, 100

and 400 mesh density. The suspension was poured into a 15 ml centrifuge tube and centrifuged for five minutes at 1500 rpm. The water in the tube was discarded, and the sediment was added to a 40% sugar solution and homogenized. The suspension was centrifuged again at 1700 rpm for one minute. The supernatant was poured into a 50 ml bottle containing 35 ml of distilled water. This aims to dilute the sugar solution so that the nematodes do not die due to the difference in osmotic pressure that is too high. The supernatant was immediately filtered with a 400 mesh sieve and rinsed with running water to lose the sugar solution. A 25 ml nematode suspension was poured into a collection bottle and then identified based on morphological characteristics or stored in $\pm 1\%$ formalin (Hasanah *et al.*, 2016).

Observation of Non-Parasitic Plant Nematodes

Observation of the morphology of non-parasitic nematodes with preparation of preparations using stereo and binocular microscopes. Observation activities include observing non-parasitic nematodes' morphological characteristics and calculating the number or population of

nematodes. Morphological characters are validated to the identification key book from Tarjan *et al.*, (1977) the identification key based on the ecology of non-parasitic nematodes, and the identification key based on morphological characters. According to Yeates *et al.*, (2003) the grouping of nematodes based on feeding groups is divided into eight indicators, namely plant parasites, soil fungus eaters, soil bacteria eaters, organic substrate eaters, animal eaters, unicellular eukaryotic microbe eaters, specific phase as a plant parasite, and omnivore. In addition, Bongers, (1990) and Ferris *et al.*, (2001) added that the grouping of nematodes can also be based on c-p (colonizer-persisters) with a scale of cp-1 to cp-5.

Data analysis

The absolute population of non-parasitic plant nematodes was calculated using the nematode suspension for each sample in each replication using the Norton & Norton, (1978) formula. The calculation of the absolute population of nematodes uses the following formula.

$$PA = \frac{\sum_{i=1}^n \frac{pxV}{v}}{n}$$

p is the population density of the nematode species observed in the counting dish of the i-th is replication, V is the volume of the nematode suspension extracted from the i-th replication, v is the volume of the nematode suspension when in the counting dish of the i-th replication and n is the number of replications used in the observation.

RESULTS AND DISCUSSION

Sampling locations on pepper plants of various accessions

Pepper plant samples were taken from eight points in the Bangka region (Figure 1), namely Paku, Pangkal Buluh, and Nadung Villages, Payung District (South Bangka Regency), Terentang III and

Nibung Villages, Koba District (Central Bangka Regency), Dendang, Tugang, and Air Bulin Villages, Kelapa District (West Bangka Regency). Soil samples were taken from plants over one year and from pepper plantations with different accessions with environmental factors presented in Table 1.



Figure 1. Locations for pepper soil sampling in the Bangka region: Paku, Pangkal Buluh, and Nadung Villages, South Bangka Regency, Terentang III and Nibung Villages, Central Bangka Regency, Dendang, Tugang, and Air Bulin Villages, West Bangka Regency.

Table 1. Pepper plant samples from various accessions and vegetation environments							
Accession	P	H	Wc	St	V	S	Pt
LDL	6,4	35,4	22,13	SCL	Oil palm	Dead	Ic
	6,2	39,6	27,36	SLL	Grass, Oil palm	Dead	Ic
	6,1	38,4	25,50	SLL	Grass, Oil palm	Dead	Ic
LDK	5,3	39,6	18,16	LS	Grass, Oil palm	Dead	Ic
	5,7	46,0	12,40	SL	Grass, Citrus	Dead	Ic
	5,1	36,2	18,11	LS	Grass, Oil palm	Dead	Ic
NYP	5,0	38,0	6,730	LS	Oil palm	Dead	Ic
	6,1	34,4	35,67	SLL	Grass, Oil palm	Dead	Ic
	6,2	39,6	32,28	SCL	Grass, Oil palm	Dead	Ic

Description: LDL (Lampung Daun Lebar), LDK (Lampung Daun Kecil), Nyp (Nyelungkup), P (pH), H (Humidity), Wc (Water content), St (Soil texture: SCL (sandy clay loam), SLL (silty clay loam), LS (loamy sand), SL (sandy loam), V (Vegetation), S (Stands used), and Pt (Planting pattern).

Absolute Population and Diversity of Non-Parasitic Nematodes

The study results showed that there were non-parasitic nematodes in the pepper plant soil with a population of 132 individuals per ± 500 grams of soil. The nematode population in the Lampung Daun Lebar accession was 41 individuals,

Lampung Daun Kecil 17 individuals, Nyelungkup 9 individuals, and Merapin 65 individuals. Non-plant parasitic Nematodes (NNPT) found in pepper soil were Rhabditidae, which had the highest dominance with a dominance value of 92.42% (Figure 2).

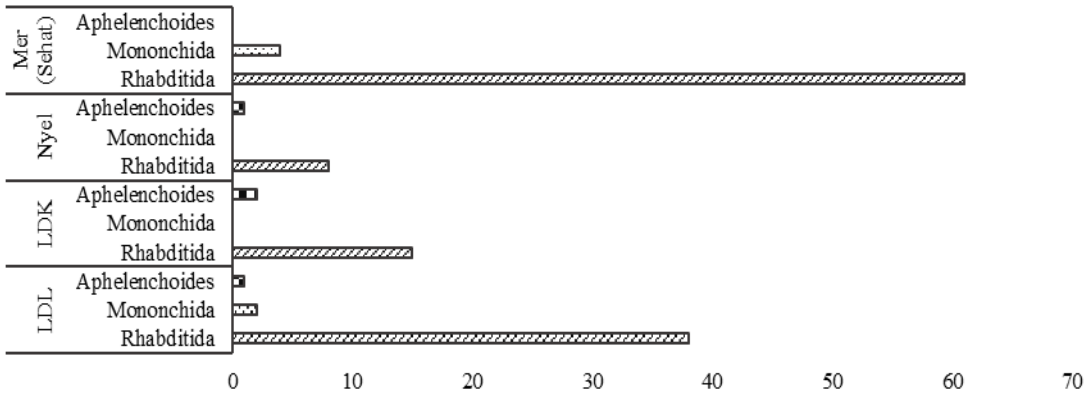


Figure 2. Population of non-parasitic nematodes in pepper plantations

Non-parasitic nematodes found in pepper plantations in the Bangka region include the Rhabditida, Mononchida and Aphelenchus aveane groups. Rhabditida has morphological characteristics of a short and slightly rounded body, crown-like lips, and a cavity (cylindrical). The tail is tapered and elongated (Figure 3a-c). The morphological characteristics of Mononchida are a long body with moderate annulation, crown-like lips, and a cavity (subglobular) with teeth on the inside. The metacarpus is oval, with spicules at the tip of the tail (Figure 3d-f). *A. aveane* has an elongated body and flat lips. The metacarpus is oval and the tip of the tail is slightly rounded (Figure 3g-i).

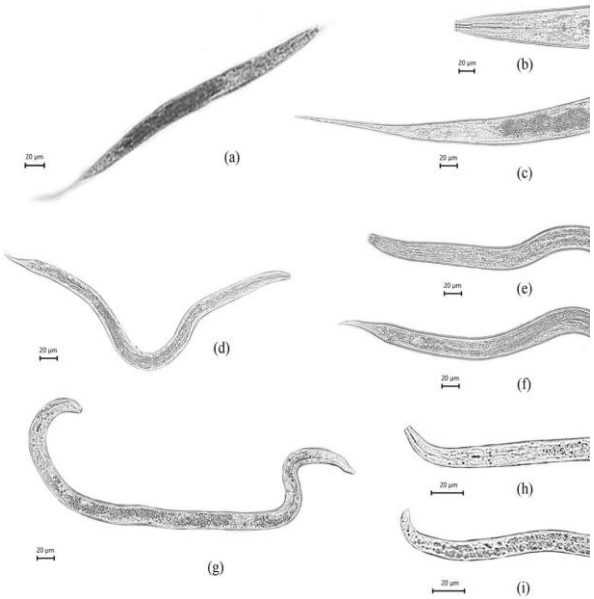


Figure 3. Non-parasitic nematodes on pepper plants in Bangka: (a) Rhabditida, (b) anterior part of Rhabditida, (c) posterior part of Rhabditida, (d) Mononchida, (e) anterior part of Mononchida, (f) posterior part of Mononchida, (g) *Aphelenchus avenae*, (h) anterior part of *A. Avenae*, and (i) posterior part of *A. avenae*. Scale a, d, g 50 μm and b, c, e, f, h, i 20 μm .

Non-parasitic or free-living nematodes are important in the soil's food web and bioindicators of soil conditions (Ferris *et al.*, 2001). According to Widiowati *et al.*, (2014; Gives, 2022), free-living non-parasitic nematodes are very important and beneficial in the soil. Rhabditida do not have setae or are sometimes unclear, the stoma wall is tubular to eat bacteria. These nematodes do not have stylets or teeth; the esophagus is well developed, and the metacarpus part is enlarged (Widowati *et al.*, 2014; McSorley, 2009; Panahandeh *et al.*, 2018). Mirsam *et al.*, (2020) added that the order Rhabditida has lips that tend to be flat, stoma with a cylindrical type (funnel-like channel) consisting of cheilostomes, gymnostomes, and stegostomes. The metacarpus is enlarged and the tail is smaller (tends to be tapered and blunt towards the tip).

Rhabditida acts as bacterivores and are found in large numbers in all soil types. In addition, Rhabditida is also helpful in the decomposition of organic matter by eating decomposing bacteria (Yeates, 2003). Mononchida has distinctive morphological characteristics. These nematodes have flat and slightly wavy lips, have a subglobular stoma (the open funnel), and consist of dorsal teeth. The tail is relatively long cylindrical (Mirsam *et al.*, 2020).

According to Tahsen and Rajan, (2009), the female Mononchida order is medium-sized, the lips are fused with the body, and the dorsal teeth are in 25-30% of the anterior cavity. The tail of Mononchida is long cylindrical, while the male nematode is not. Mononchida acts as a predator

(omnivore) that eats other nematodes with comparable body size (McSorley, 2009). Mirsam *et al.*, (2020) added that predatory nematodes play an important role in regulating the population of parasitic nematodes. *Aphelenchus* has a style that functions as a tool to inject enzymes into fungi, hydrolyze hyphae, and suck hyphal contents (Mulyadi, 2009).

McSorley (2020) stated that *Aphelenchus* is a fungivorous nematode (fungus eater) that is beneficial in the decomposition of organic matter by eating decomposing fungi. Healthy soil has a higher population of non-parasitic plant nematodes than soil infected with parasitic nematodes (Widowati *et al.*, 2014). Non-parasitic nematodes are beneficial in microhabitats that contain many food sources to increase the population of these microbes. Non-parasitic nematodes can also excrete and provide up to 19% dissolved N (Wang & McSorley, 2007).

Based on table 1 regarding the diverse plant vegetation among pepper plants, there are many plant remains or litter which are a source of organic material for non-parasitic nematodes. This aims to return these nutrients to the soil, so plant roots can more easily absorb them (McSorley, 2009). Mirsam *et al.*, (2020) stated that non-parasitic nematodes can contribute to nutrient recycling and decomposition, suppressing pathogenic microorganisms (biological agents), and biodegradation of hazardous compounds. Bacterivores and fungivores do not eat soil organic matter directly, but rather bacteria and fungi that function as decomposers and accelerate the

decomposition and mineralization processes.

According to Khan & Kim, (2007), non-parasitic nematodes that act as predators are divided into four orders, namely Mononchida, Diplogasterida, Dorylaimida, and Aphelenchida. Based on their feeding tools, predatory nematodes are divided into three categories, namely cutter-swallowers (Mononchida), piercing-sucking (Dorylaimida and Aphelenchida), and cutter-sucking (Diplogasterida) (Askary & Martinelli, 2015).

According to Ferris *et al.*, (2001) non-parasitic nematodes are grouped based on feeding groups and c-p (colonizer-persisters). The non-parasitic nematodes are divided into five groups based on these

characteristics. cp-1 (short life cycle, small eggs, high fecundity, bacteria eaters (bacteriovores), can live in nutrient-rich media and can form dauer larvae), cp-2 (longer life cycle and lower fecundity than cp-1, bacteriovores, and fungivores, very tolerant of unfavorable environments and can become cryptobiotic, able to live when food sources are few), cp-3 (longer life cycle, more sensitive to unfavorable environments, bacteriovores, fungivores, and carnivores) cp-4 (longer life cycle, lower fecundity, more sensitive to disturbances, few species are omnivores) and cp-5 (most extended life cycle, largest body size, lowest fecundity, most sensitive to disturbances, primarily carnivores, and omnivores).

CONCLUSION

Non-parasitic nematodes found in pepper plantations of various accessions such as Lampung Daun Lebar, Lampung Daun Kecil, Nyelungkup, and Merapin are the Rhabditida, Mononchida, and Aphelenchus avenae groups. The dominant population is the Rhabditida group with a value of 92.42%.

REFERENCES

- Askary, T.H., & Martinelli, P.R.P. (2015). *Biocontrol Agents of Phytonematodes*. Wallingford: CABI Publishing.
- Bongers, T. (1990). The maturity index: an ecological measure of environmental disturbance based on nematode species composition. *Oecologia*, 83(1), 14-19. <https://doi.org/10.1007/BF00324627>.
- Caveness, F.E., & Jensen, H.J. (1955). Modification of the centrifugal-flotation technique for the isolation and concentration of nematodes and their eggs from soil and plant tissue. *Proc Helminthol Soc Wash*, 25, 87-89.
- Ferris, H., Bongers, T., de Goede, R.G. (2001). A framework for soil food web
- diagnostics: extension of the nematode faunal analysis concept. *Appl Soil Ecol*, 18(1), 13-29. [https://doi.org/10.1016/S0929-1393\(01\)00152-4](https://doi.org/10.1016/S0929-1393(01)00152-4).
- Gives, P.M. (2022). Soil borne nematodes: impact in agriculture and livestock and sustainable strategies of prevention and control with special reference to the use of nematode natural enemies. *Pathogens*, 11(6), 1-24. <https://doi.org/10.3390/pathogens11060640>.
- Hamid, A., & Rahayuningsih, S.T. (1990). Identifikasi dan pengenalan empat varietas utama tanaman lada," in: *Simposium I Hasil Penelitian dan Pengembangan Tanaman Industri*. Bogor: Pusat Penelitian dan Pengembangan Tanaman Industri, pp. 586-590.
- Hasanah, S., Swibawa, I.G., Solikhin, S. (2016). Populasi nematoda *Radopholus* dan *Pratylenchus* pada tanaman kopi robusta berbeda umur di Tanggamus, Lampung. *JAT*, 4(3), 217-221. <https://doi.org/10.23960/jat.v4i3.1855>.
- Khan, Z., & Kim, Y.H. (2007). A review on the role of predatory soil nematodes in the biological control of plant parasitic nematodes. *Appl Soil Ecol*, 3(5), 370-

379.
<https://doi.org/10.1016/j.apsoil.2006.07.0007>.
- Kimenju, J.W., Karanja, N.K., Mutua, G.K., Rimberia, B.M., and Wachira, P.M. (2009). Nematode community structure as influenced by land use and intensity of cultivation. *Trop Subtropical agroecosystems*, 11(2), 353-360. https://www.researchgate.net/publication/287492898_Nematode_community_structure_as_influenced_by_land_use_and_intensity_of_cultivation.
- Lesta. (2023). Insidensi Penyakit Kuning, Faktor Lingkungan, dan Populasi Nematoda Parasit Pada Tanaman Lada di Bangka. Thesis. Graduated School. IPB University. Bogor. Indonesia.
- McSorley, R. (2009). *Soil-inhabiting nematodes*," Florida: Featured Creature, pp. 1-3.
- Ministry of Agriculture of the Republic of Indonesia. (2020). Pepper productivity by province in Indonesia 2016-2020," <https://www.pertanian.go.id/home/?show=page&act=view&id=61>.
- Mirsam, H., Muis, A., Nonci, N. (2020). The density and diversity of plant-parasitic nematodes associated with maize rhizosphere in Malakaji Highland, South Sulawesi, Indonesia. *Biodiv J of Biolog Diver*, 21(6), 2654-2661. <https://doi.org/10.13057/biodiv/d210637>.
- Mulyadi. (2009). *Nematologi Pertanian*. Yogyakarta: Gadjah Mada University Press.
- Norton, D.C., & Norton, D.C. (1978). *Ecology of plant parasitic nematodes*," New York: John Wiley and Sons.
- Panahandeh, Y., Abolafia, J., Pourjam, E., Giblin-Davis, R.M., Afshar, F.J., Pedram, M. (2018). Morphological and molecular characterization of *Labrys filiformis* n. sp. (Rhabditida: Tylenchidae) from Iran. *J Nematol*, 50, 343-354.
- <https://doi.org/10.21307/jofnem-2018-031>.
- Plant Quarantine Agency. (2010). Guide to the diagnosis of OPTK nematode group, Jakarta: Plant Quarantine Agency.
- Prayoga, G.I., Ropalia, Aini, S.N., Mustikarini, E.D., Rosalin, Y. (2020). Diversity of black pepper plant (*Piper nigrum*) in Bangka Island (Indonesia) based on agro-morphological characters. *Biodiversitas*, 21(2), 27-32. <https://doi.org/10.13057/biodiv/d21023>.
- Suryanti, S., Hadisutrisno B., Mulyadi, M., Widada, J. (2015). Interaksi *Meloidogyne incognita* dan *Fusarium solani* pada penyakit kuning lada. *JPTI*, 21(2), 127-134. <https://doi.org/10.22146/jpti.29760>.
- Tahsen, Q., & Rajan, P. (2009). Description of *Mononchus intermedius*. *Nematol Mediterr*, 37, 161-167.
- Tarjan, A., Robert, P.E., Shih, L.C. (2007). Interactive diagnostic key to plant parasitic, freeliving and predaceous nematodes. *Journal of the Water Pollution Control Federation*, 49, 2318-2337.
- The Center for Agricultural Data and Information Systems Agricultural Statistic 2022. (2022). Jakarta: Ministry of Agriculture of the Republic of Indonesia.
- Wang, K., & McSorley, R. (2007). Effects of soil ecosystem management on nematode pests, nutrient cycling and plant health.
- Widowati, R., Indarti, S., Rahayu, B. (2014). Distribution of non plant parasitic nematodes genera in *Arabica coffee*. *JPTI*, 18(1), 24-32. <https://doi.org/10.22146/jpti.15572>.
- Yeates, G.W. (2003). Nematodes as soil indicators: functional and biodiversity aspects. *Biol Fertil Soils*, 37(4), 199-210. <https://doi.org/10.1007/s00374-003-0586-5>.