Distribution of plant-parasitic nematode genera in relation to host vegetable crops and ecological regions in Benin, West Africa

Elodie N.D. Coffi[1*], Antoine Affokpon[1], Djibril Aboubakar Souna[2,3], Antoine de Troij[2,4], Alphonsine S.S. Adanmaho[1], and Safiou Sero[1]

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Diversity, food security, *Meloidogyne* spp., nematode survey, Sub-Saharan Africa, urban and peri-urban agriculture

Abstract

Plant-parasitic nematodes (PPNs) are a serious threat to vegetable production. A survey was conducted from April to December 2023 across 49 villages/towns in Benin for a comprehensive understanding of their diversity and spatial distribution. Twenty PPN genera were morphologically identified, with African eggplant and pepper fields exhibiting the highest nematode diversity (16 genera). Genus richness, Shannon-Wiener, and evenness indexes were significantly influenced by ecological regions. The Niger valley was the most diverse ecological region with 15 PPN genera. *Meloidogyne* was the most prevalent. The highest *Meloidogyne* densities were recorded from pepper fields, with 2,151 nematodes per g root in the Oueme valley and 691 nematodes per 100 cm³ soil in the Niger valley. Significant associations were observed between nematode genera, vegetable crops, and ecological regions. This study highlights the relationship between PPN diversity, vegetable crops, and ecological regions and provides valuable insights for developing effective PPN management strategies.

Introduction

Vegetable production is essential for ensuring global food and nutritional security and for reducing poverty. A recent comprehensive review of the available literature on the importance of vegetable consumption for human health concluded that a high intake of vegetables reduces the risk of chronic diseases, improves gastrointestinal health, and strengthens the immune system (Solin et al. 2024). In Sub-Saharan Africa (SSA), vegetable value chains not only provide an affordable source of nutrition to populations but also create employment for women, youth, and vulnerable groups, providing a vital source of income for the stakeholders. Despite its importance, developing the vegetable sector remains a challenge in most SSA countries. Farmer yields and product quality remain far below the potential production due to several factors, including soil type and nutrient status, crop variety, agronomic practices, weather conditions, and pests and diseases (Affokpon 2011).

Among the pests affecting vegetable production, plant-parasitic nematodes (PPNs) are a serious concern, particularly in tropical regions (De Waele and Elsen 2007; Hallmann and Meressa 2018). PPNs cause direct damage to vegetables by disrupting root systems, which affects nutrient and water uptake, leading to reduced root systems, root malformations, stunted growth, leaf yellowing, premature wilting, and ultimately significant reductions in yield and quality of harvested products (Desaeger et al. 2023; Phani et al. 2021). Indirectly, nematode infections weaken the plant's resistance to pests and diseases (Coyne et al. 2018), exacerbate pressures from pathogens by damaging roots (Greco and Di Vito 2009), and serve as entry points for other soil-borne microorganisms (Hallmann and Meressa 2018; Desaeger et al. 2023). Globally,

[1]Nematology Unit, Faculty of Agronomic Sciences, University of Abomey-Calavi, 01 BP 526 Cotonou, Benin,
[2]CIRAD, UR Hortsys, Université de Montpellier, Montpellier, France
[3]Institut de l'Environnement et de Recherches Agricoles (INERA), 01 BP 476 Ouagadougou 01, Burkina Faso
[4]World Vegetable Center, West and Central Africa-Coastal and Humid Regions, IITA Campus, 08 BP 0932 Cotonou, Benin
* Corresponding author email: nevrilec@gmail.com

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PPNs are responsible for approximately 8 - 15% of crop losses worldwide, with an estimated cost of at least US$80 billion (Kiontke and Fitch 2013).

Numerous PPNs have been reported to be associated with vegetable crops worldwide, with root-knot nematodes (*Meloidogyne* spp.) considered the most damaging and widespread group. Other genera also cause significant economic damages, although their impact is often underestimated due to the indistinct symptoms exhibited by infested plants (Hallmann and Meressa 2018). The diversity of soil nematodes is influenced by a number of factors, including ecological, edaphic, and vegetation-related parameters (Nisa et al. 2021). Previous studies have reported that PPN diversity and richness are positively correlated with host plant species diversity and altitude (Shanmugam et al. 2021). Additionally, soil physicochemical properties also play an important role in PPN communities (Baimey et al. 2009; Mokrini et al. 2019).

In Benin, vegetable crops widely cultivated and consumed include tomato (*Solanum lycopersicum* L.), pepper (*Capsicum* spp. L.), African eggplant (*Solanum macrocarpon* L.), onion (*Allium cepa* L.), and okra (*Abelmoschus esculentus* M.) (Houessou et al. 2021). These crops are cultivated across the country in various ecological regions (coastal plains, lowlands, plateaus, and valleys) (ACDD 2019) and climatic zones (Sudanian, Sudano-Guinean, and Guineo-Congolian) (DGEC 2022). Vegetable production sites are characterised by the simultaneous occurrence of various nematode genera rather than the dominance of a single species (Baimey et al. 2009; Coyne et al. 2018). This situation, which is common to many West African countries, complicates sustainable PPN management (Coyne et al. 2018). Knowledge gaps persist regarding PPN distribution patterns in vegetable growing areas at the country and subregional levels. Understanding PPN diversity and spatial distribution based on local key vegetable crops and ecological regions is useful information for developing sustainable and agro-ecologically oriented management strategies.

The objectives of this study were (i) to determine the current status of PPN diversity in vegetable production systems in Benin and (ii) to investigate the influence of ecological regions and crops on their spatial distribution.

Materials and methods

Field selection for surveys and sample collection

Field surveys were carried out between April and December 2023 in 49 villages/towns across 14 districts that represent the agro-ecological areas suitable for the development of vegetable production in Benin (Houessou et al. 2021), and where PPNs had been reported as a major constraint (Affokpon et al. 2015) (Figure 1). A total of 153 vegetable-growing fields were surveyed, of which 45 were located in the coastal areas, 12 in plateaus, 21 in the southern lowland areas, 7 in the northern lowland areas, 24 in the Oueme valley, and 44 in the Niger valley (Table 1). At each field, a composite soil sample and a composite root sample were collected.

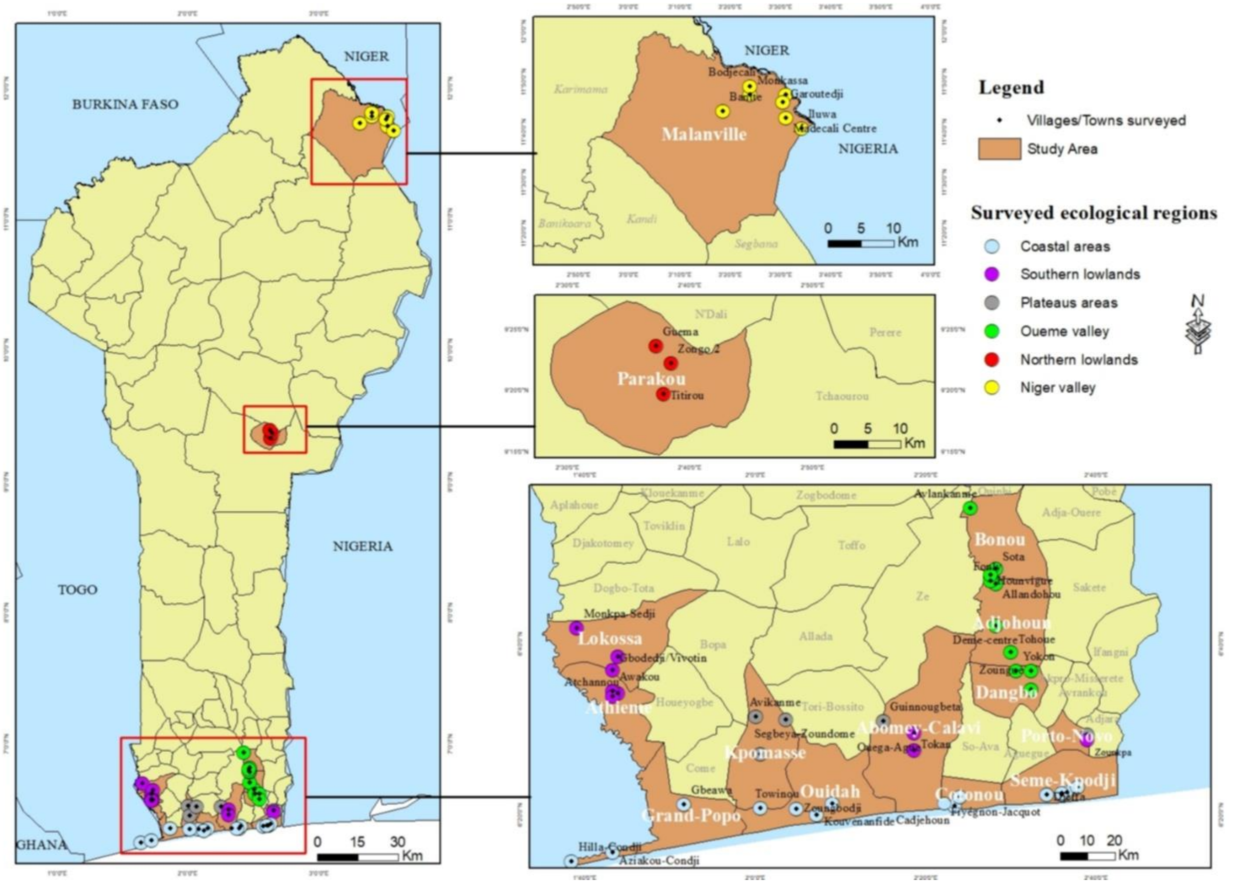


Figure 1: Map of the surveyed villages/towns

Target crops were the commonly produced and consumed fruit (tomato, pepper and okra), leafy (African eggplant) and root/bulb (onion) vegetables, including the top three priority vegetables (tomato, pepper, onion) for investments in Benin (Houessou et al. 2021). Of the 153 fields sampled, 49 were planted with pepper, 37 with tomato, 36 with African eggplant, 21 with onion, and 10 with okra.

Table 1: Distribution of surveyed fields by ecological regions, climates and vegetable crops

Ecological regions	Dominant soil characteristic	Climates	No of fields surveyed	Vegetable crops	Sampling season
Coastal areas	Sandy	Guinean-Congolian zone	45	Tomato, pepper, African eggplant, okra, onion	Rainy season (August)
Plateau areas	Ferralitic	Guineo-Congolian zone	12	Tomato, pepper, African eggplant, okra	Rainy season (August)

Table 1 *continued*

Ecological regions	Dominant soil characteristic	Climates	No of fields surveyed	Vegetable crops	Sampling season
Southern lowlands	Hydromorphic	Guineo-Congolian zone	21	Tomato, pepper, African eggplant, okra, onion	Rainy season (August)
Oueme valley	Hydromorphic	Guineo-Gongolian zone	24	Pepper, African eggplant, okra	Pre-flooding period (April)
Northern lowlands	Hydromorphic	Sudano-Guinean transition zone	7	Pepper, African eggplant, okra, onion	Dry season (December)
Niger valley	Hydromorphic	Sudanian zone	44	Tomato, pepper, okra, onion	Dry season (December)

Source: ACDD (2019); DGEC (2022)

Depending on the sites, field sizes varied, but sampling intensity and pattern were kept constant for all sites (Affokpon et al. 2015). For smaller fields (e.g., in urban areas), the soil and root samples were collected from four sampling points, whereas for larger fields (e.g., in peri-urban areas), six to nine sampling points were selected. At each sampling point, soil samples were collected at depth of 5 - 30 cm using an auger at 10 cm distance from each selected plant (Affokpon et al. 2011). For root samples, whole plants were uprooted using a spade, and the entire root system was removed. Soil or root sub-samples collected from the same field were combined, and a composite representative sample of 1 kg of soil or approximately 20 g of roots was removed and kept in labelled polyethylene bags. Soil samples were kept at ambient temperature while root samples were stored in insulated boxes with ice packs. All samples were transported to the laboratory within 3 days after collection, stored at 4°C and processed within 5 days. Subsequently, 153 soil and 153 root samples were collected for nematological analyses.

Nematode extraction, identification, and quantification

The extraction of nematodes from soil and roots was performed in the laboratory of the Nematology Unit (UNema) at University of Abomey-Calavi, Benin. For each composite sample, nematodes were extracted from 250 cm^3 soil and 15 g roots, using the centrifugation technique (Affokpon et al. 2011).

Nematodes were then identified at the genus level based on the morphological characteristics described by Hunt et al. (2018) and counted from 2 x 3 mL aliquots taken from a 30 mL suspension under an optical microscope (Euromex iScope IS.1153-PLi) at 40x magnification. The microscope was equipped with a camera (Euromex HD-Ultra VC. 3036), which was used to photograph the identified nematode genera.

Assessment of PPN diversity

The diversity of the PPN community associated with the vegetable crops and the ecological regions was determined using the genus richness (G), Shannon-Wiener diversity index (H´), and evenness index (J´) (Ferris 2012).

The genus richness (G) sums up the total number of genera present in a sample reflecting its biodiversity. The Shannon-Wiener diversity index (H′) quantifies the heterogeneity of diversity within a community by considering the relative abundance of the different genera. The Shannon-Wiener diversity index ranges from 0 to ln(G). Values closer to ln(G) correspond to a heterogeneous community.

$$\mathbf{H' = - \Sigma\ (P_i \times \ln P_i)}$$

where P_i is the relative abundance of the i^{th} genus

The evenness index (J') indicates how uniformly individuals are distributed among the different genera. Evenness values range from 0 to 1, with values closer to 1 indicating a more uniform distribution.

$$\mathbf{J' = H' / \ln (G)}$$

Assessment of PPN incidence

The incidence of the PPN community was assessed by determining the frequency of occurrence, densities and abundance of each genus, using the following formulas (Boag 1993):

Frequency of occurrence = (Number of samples with a specific nematode genus × 100)/Total number of samples collected

Population density = (Number of nematodes counted × Volume of suspension obtained)/Volume of suspension counted

Maximum density = Maximum number of a particular nematode genus recovered from a sample

Abundance = Total number of individuals of a specific genus in positive samples/Total number of positive samples

A nematode genera distribution diagram was then generated for soil and roots based on the computed abundance and frequency of each genus (Sawadogo et al. 2009). Abundance was expressed per 1000 cm³ soil and gram roots. According to Fortuner and Merny (1973), a nematode is considered abundant if its abundance value is equal to or higher than 1.3 (equivalent to 20 nematodes per g roots) or 2.3 (equivalent to 200 nematodes per 1000 cm³ soil). A nematode was considered frequent in soil or roots if it was observed in at least 30% of the samples.

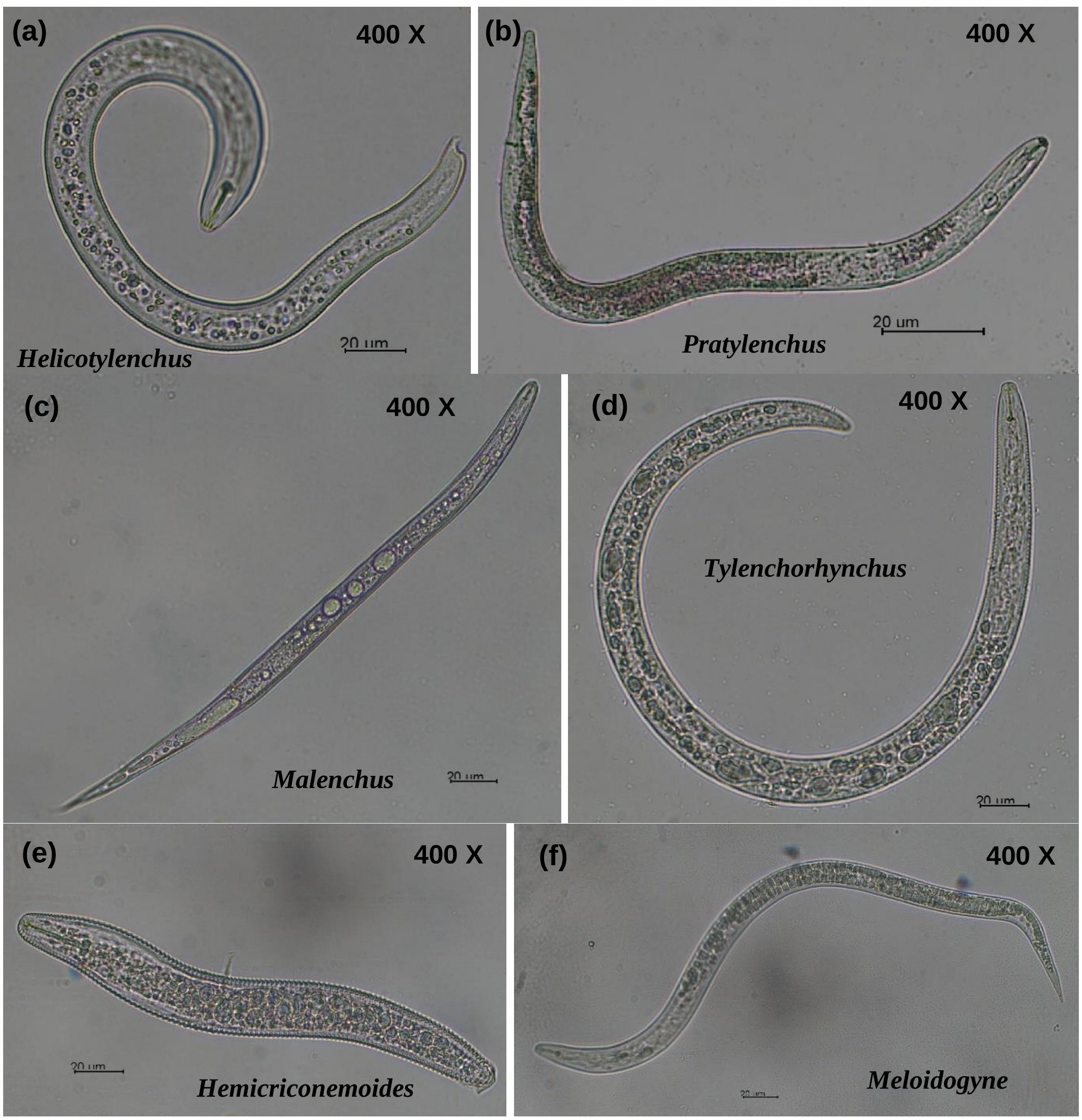
Data analysis

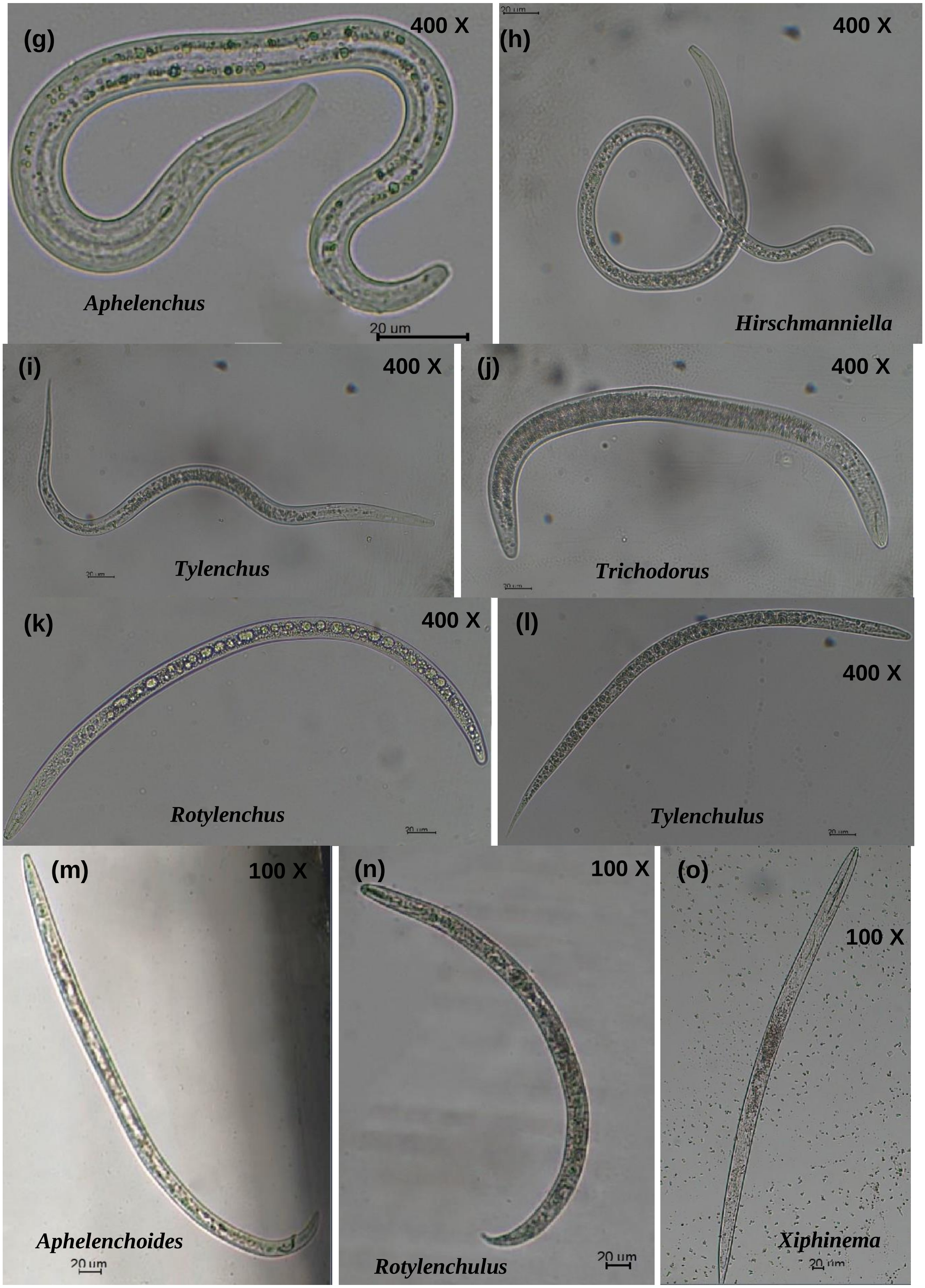
All statistical analyses were performed using R software version 4.4.2 (R Core Team 2024). Diversity indices were computed with the diversity function of the vegan R-package (Oksanen et al. 2024). One-way ANOVA was performed to assess statistical differences between diversity parameters, and means were separated by Tukey’s honestly significant difference (HSD) test using the HSD test function of the agricolae R-package (de Mendiburu 2023). Distribution diagrams were generated using the ggplot2 R-package (Wickham 2016). Before generating the graphs, the abundance variables were transformed using $\log_{10}(x + 1)$. To describe the variation in PPN genera identified across vegetable crops and ecological regions, a contingency table based on nematode genera occurrence was produced and subjected to a Pearson's chi-squared test followed by correspondence analysis using the FactoMineR R-package (Lê et al. 2008).

Results

Diversity and distribution of PPNs in relation to host plants and ecological regions

PPNs were recorded in 98.0% of the surveyed fields; 20 PPN genera viz: *Aphelenchoides*, *Aphelenchus*, *Ditylenchus*, *Geocenamus*, *Helicotylenchus*, *Hemicriconemoides*, *Hirschmanniella*, *Hoplolaimus*, *Malenchus*, *Meloidogyne*, *Pratylenchus*, *Quinisulcius*, *Rotylenchulus*, *Rotylenchus*, *Trichodorus*, *Trichotylenchus*, *Tylenchulus*, *Tylenchorhynchus*, *Tylenchus* and *Xiphinema* were found to be associated with African eggplant, okra, onion, pepper and tomato (Figure 2). Of these genera, 19 were detected in the soil (*Hirschmanniella* being the only absent) while only *Meloidogyne*, *Pratylenchus* and *Hirschmanniella* were recovered from the roots.




(g)
400 X
Aphelenchus
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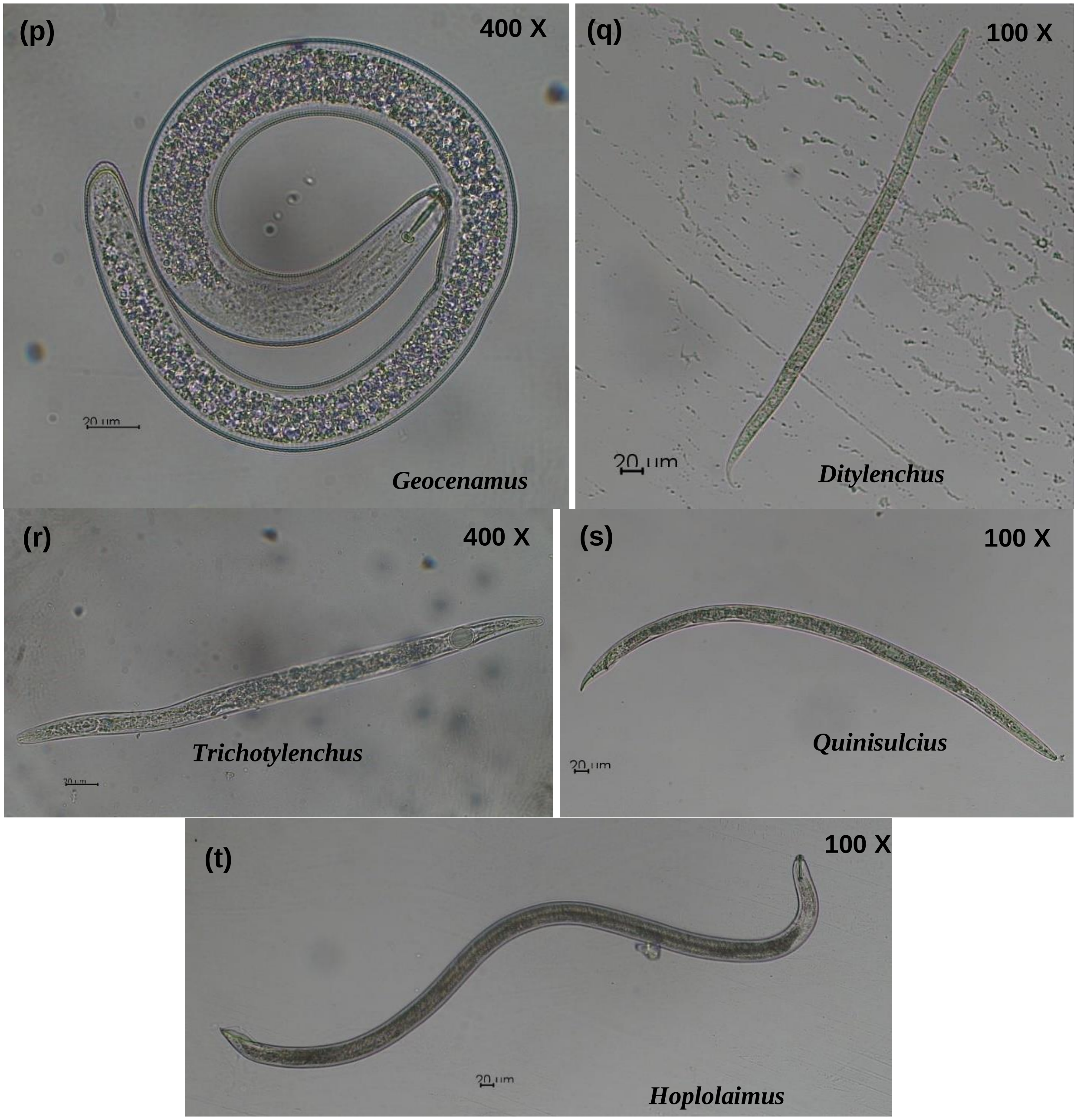

Figure 2: Microscopic illustrations of plant-parasitic nematode genera associated with vegetable crops in Benin (Scale bar: 20 µm)

Dimensions: (a) 596 × 468; (b) 764 × 468; (c) 859 × 654; (d) 705 × 618; (e) 781 × 373; (f) 694 × 390; (g) 726 × 511; (h) 652 × 511; (i) 662 × 372; (j) 678 × 381; (k) 732 × 372; (l) 625 × 372; (m) 565 × 673; (n) 544 × 616; (o) 373 × 654; (p) 1083 × 609; (q) 766 × 771; (r) 679 × 382; (s) 763 × 408; (t) 1072 × 482

Among vegetable crops, African eggplant and pepper fields had the highest number of PPN genera (16 genera) and okra fields the lowest (10 genera) (Table 2). Nine genera (*Meloidogyne*, *Pratylenchus*, *Tylenchus*, *Helicotylenchus*, *Tylenchulus*, *Aphelenchoides*, *Aphelenchus*, *Hemicriconemoides* and *Tylenchorhynchus*) were common to all crops. In contrast, some genera were specific to some crops, namely *Hirschmanniella* and *Trichotylenchus* were detected exclusively in soils grown to pepper, *Xiphinema* in onion, *Quinisulcius* and *Malenchus* in African eggplant. However, the genus richness, Shannon-Wiener diversity index and evenness values were not significantly different between crops (Table 3).

Table 2: Occurrence of plant-parasitic nematodes genera across vegetable crops

Nematode genera	African eggplant	Okra	Onion	Pepper	Tomato
Meloidogyne	+	+	+	+	+
Pratylenchus	+	+	+	+	+
Hirschmanniella	-	-	-	+	-
Tylenchus	+	+	+	+	+
Rotylenchus	+	-	-	+	-
Helicotylenchus	+	+	+	+	+
Tylenchulus	+	+	+	+	+
Aphelenchoides	+	+	+	+	+
Aphelenchus	+	+	+	+	+
Hemicriconemoides	+	+	+	+	+
Rotylenchulus	+	-	-	-	+
Tylenchorhynchus	+	+	+	+	+
Xiphinema	-	-	+	-	-
Hoplolaimus	-	+	+	+	-
Malenchus	+	-	-	-	-
Ditylenchus	+	-	+	+	+
Geocenamus	+	-	+	+	+
Quinisulcius	+	-	-	-	-
Trichotylenchus	-	-	-	+	-
Trichodorus	+	-	+	+	+
Total	**16**	**10**	**14**	**16**	**13**

- Not detected in soil or root samples; **+** Present in soil and/or root samples

Table 3: Diversity parameters of plant-parasitic nematodes community across vegetables crops

Vegetable crops	Genus richness		Shannon-Wiener index		Evenness	
	Soil	Roots	Soil	Roots	Soil	Roots
African eggplant	$2.67^a \pm 0.32$	$1.31^a \pm 0.11$	$0.55^a \pm 0.09$	$0.18^a \pm 0.04$	$0.69^a \pm 0.07$	$0.50^a \pm 0.10$
Okra	$3.40^a \pm 0.60$	$1.10^a \pm 0.21$	$0.72^a \pm 0.17$	$0.07^a \pm 0.08$	$0.61^a \pm 0.10$	$0.34^a \pm 0.24$
Onion	$3.57^a \pm 0.42$	$1.05^a \pm 0.15$	$0.91^a \pm 0.12$	$0.18^a \pm 0.06$	$0.78^a \pm 0.08$	$0.43^a \pm 0.12$
Pepper	$3.06^a \pm 0.27$	$1.29^a \pm 0.10$	$0.68^a \pm 0.08$	$0.16^a \pm 0.04$	$0.61^a \pm 0.05$	$0.48^a \pm 0.08$
Tomato	$2.49^a \pm 0.31$	$1.41^a \pm 0.11$	$0.56^a \pm 0.09$	$0.22^a \pm 0.04$	$0.58^a \pm 0.06$	$0.56^a \pm 0.09$
P value	***0.215***	***0.336***	***0.125***	***0.595***	***0.283***	***0.847***

Values in columns are mean ± standard error. Means followed by different letters are significantly different ($P \leq 0.05$) based on Tukey's Honestly Significant Difference (HSD) test.

Regarding the ecological regions, fields in the Niger valley exhibited the greatest diversity of nematode genera (15 genera), whereas fields in the plateau areas showed the lowest diversity (7 genera) (Table 4). Five genera *Meloidogyne*, *Pratylenchus*, *Helicotylenchus*, *Tylenchulus*, and *Aphelenchus* were recovered from all five ecological regions, while *Rotylenchus*, *Xiphinema*, *Malenchus* and *Quinisulcius* were detected exclusively in the Oueme valley, coastal areas, southern lowlands and northern lowlands, respectively. Ecological regions significantly influenced the genus richness, Shannon-Wiener and evenness indexes of PPN communities in soil samples (Table 5). The northern lowlands had the highest genus richness (4.86) while the Oueme valley exhibited the highest evenness (0.79). Coastal areas had the lowest soil nematode diversity (1.62) and evenness (0.50). Lowlands and valleys areas had significantly higher Shannon-Wiener index values, compared to plateau and coastal areas. For root samples, only the genus richness index varied significantly ($P = 0.009$) among ecological regions; Shannon-Wiener and evenness indexes were similar between surveyed areas (Table 5).

Table 4: Occurrence of plant-parasitic nematodes genera according to ecological regions

Nematode genera	Coastal areas	Plateau areas	Southern lowlands	Oueme valley	Northern lowlands	Niger valley
Meloidogyne	+	+	+	+	+	+
Pratylenchus	+	+	+	+	+	+
Hirschmanniella	-	-	-	-	-	+
Tylenchus	+	-	+	+	+	+
Rotylenchus	-	-	-	+	-	-
Helicotylenchus	+	+	+	+	+	+
Tylenchulus	+	+	+	+	+	+
Aphelenchoides	-	-	+	+	+	+
Aphelenchus	+	+	+	+	+	+
Hemicriconemoides	+	+	+	-	-	+
Rotylenchulus	-	+	+	-	-	-
Tylenchorhynchus	-	-	+	-	+	+
Xiphinema	+	-	-	-	-	-
Hoplolaimus	+	-	+	-	-	+
Malenchus	-	-	+	-	-	-
Ditylenchus	-	-	-	-	+	+
Geocenamus	-	-	-	-	+	+
Quinisulcius	-	-	-	-	+	-
Trichotylenchus	-	-	-	-	-	+
Trichodorus	-	-	-	-	+	+
Total number of genera	**9**	**7**	**12**	**8**	**12**	**15**

- Not detected in soil or root samples; + Present in soil and/or root samples.

Table 5. Diversity parameters of plant-parasitic nematodes community according to ecological regions

Surveyed ecological regions	Genus richness		Shannon-Wiener index		Evenness	
	Soil	Roots	Soil	Roots	Soil	Roots
Coastal areas	$1.62^{d} \pm 0.24$	$1.18^{ab} \pm 0.10$	$0.32^{b} \pm 0.07$	$0.16^{a} \pm 0.04$	$0.50^{b} \pm 0.06$	$0.44^{a} \pm 0.08$
Plateau areas	$1.92^{bc} \pm 0.47$	$1.67^{a} \pm 0.19$	$0.28^{b} \pm 0.14$	$0.33^{a} \pm 0.08$	$0.55^{ab} \pm 0.12$	$0.72^{a} \pm 0.14$
Southern lowlands	$3.43^{abc} \pm 0.36$	$1.57^{a} \pm 0.14$	$0.73^{a} \pm 0.10$	$0.24^{a} \pm 0.06$	$0.60^{ab} \pm 0.07$	$0.60^{a} \pm 0.12$
Oueme valley	$2.83^{bc} \pm 0.33$	$0.96^{b} \pm 0.13$	$0.76^{a} \pm 0.10$	$0.07^{a} \pm 0.05$	$0.79^{a} \pm 0.07$	$0.32^{a} \pm 0.15$
Northern lowlands	$4.86^{a} \pm 0.62$	$1.14^{ab} \pm 0.24$	$1.03^{a} \pm 0.18$	$0.13^{a} \pm 0.10$	$0.65^{ab} \pm 0.12$	$0.45^{a} \pm 0.24$
Niger valley	$4.02^{ab} \pm 0.25$	$1.32^{ab} \pm 0.10$	$0.94^{a} \pm 0.07$	$0.19^{a} \pm 0.04$	$0.71^{ab} \pm 0.05$	$0.47^{a} \pm 0.08$
P value	***< 0.001***	***0.009***	***< 0.001***	***0.072***	***0.025***	***0.406***

Values in columns are mean ± standard error. Means followed by different letters are significantly different ($P \leq 0.05$) based on Tukey's Honestly Significant Difference (HSD) test.

Prevalence, dominance and densities of PPNs associated with vegetable crops in grown under different ecological regions

The top four prevalent genera were *Meloidogyne* (81.0%), *Pratylenchus* (57.5%), *Helicotylenchus* (51.0%) and *Aphelenchus* (41.8%) (Figure 3A). Based on their frequency (F) and abundance (A) in soil, *Meloidogyne* and *Helicotylenchus* were dominant (F > 50%; A > 218 nematodes/dm^3 soil) and *Aphelenchus* were considered frequent (F > 40%; A = 50 nematodes/dm^3 soil). However, more than 84% of the genera recovered from soil were recorded as rare (F < 30%; A < 145 nematodes/dm^3 soil) including *Pratylenchus* (Figure 3B). Among the three genera extracted from the roots, *Meloidogyne* was found dominant (F > 70%; A = 70 nematodes/g root), *Pratylenchus* frequent (F > 50%; A = 7 nematodes/g root) and *Hirschmanniella* rare (F < 5%; A = 7 nematodes/g root) (Figure 3C).

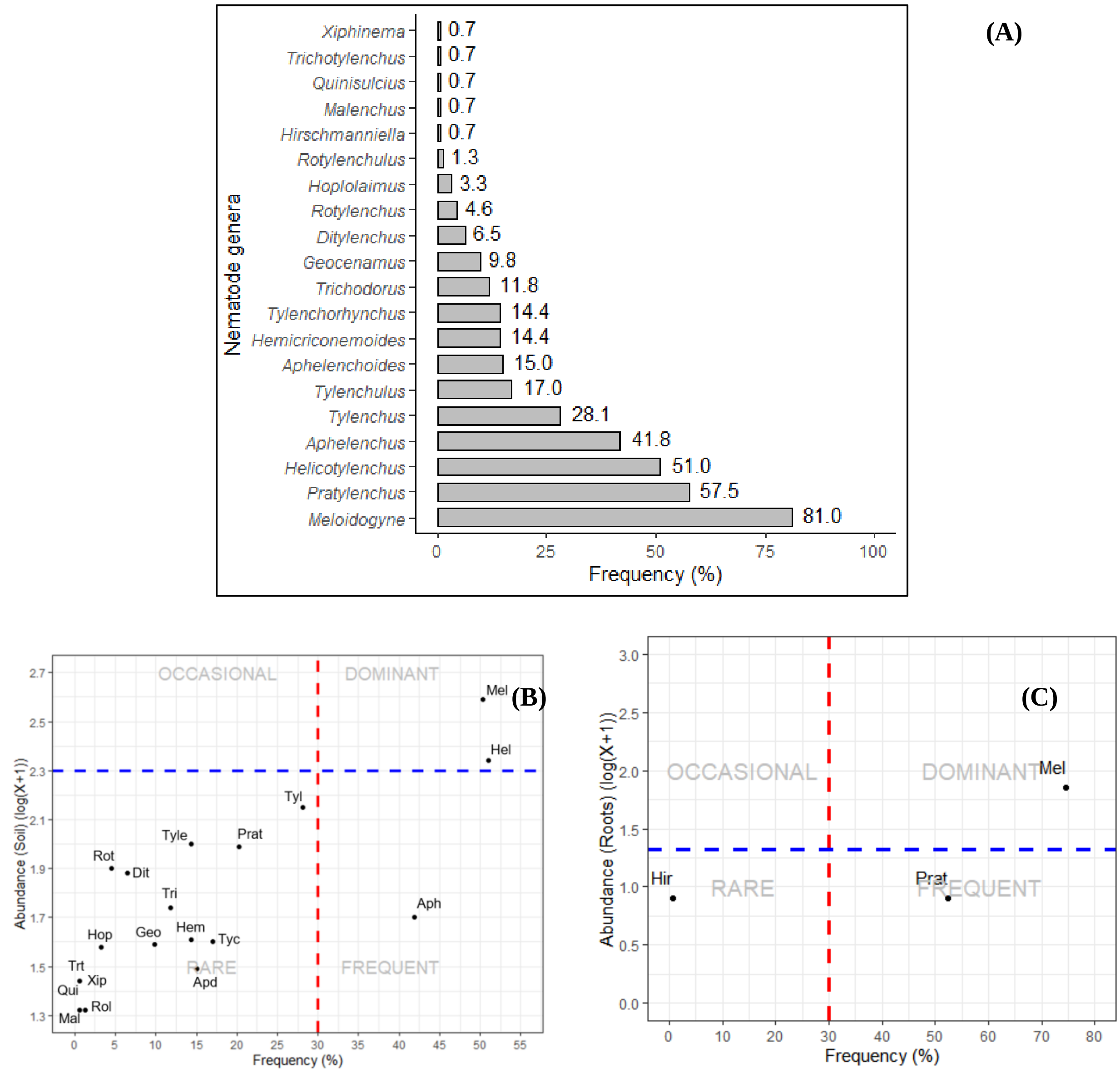

Figure 3: A: Frequencies of occurrence of plant-parasitic nematodes associated with vegetable crops B: Distribution diagram (frequency – abundance in soil) C: Distribution diagram (frequency – abundance in roots).

Dotted red lines indicate the nematode frequency threshold (30%) while the dotted blue lines mark the abundance thresholds (1.3 for roots and 2.3 for soil) as defined by Fortuner and Merny (1973).

Apd: *Aphelenchoides*, **Aph:** *Aphelenchus*, **Dit:** *Ditylenchus*, **Geo:** *Geocenamus*, **Hel:** *Helicotylenchus*, **Hem:** *Hemicriconemoides*, **Hir:** *Hirschmanniella*, **Hop:** *Hoplolaimus*, **Mal:** *Malenchus*, **Mel:** *Meloidogyne*, **Prat:** *Pratylenchus*, **Qui:** *Quinisulcius*, **Rol:** *Rotylenchulus*, **Rot:** *Rotylenchus*, **Tri:** *Trichodorus*, **Trt:** *Trichotylenchus*, **Tyc:** *Tylenchulus*, **Tyl:** *Tylenchus*, **Tyle:** *Tylenchorhynchus*, **Xip:** *Xiphinema*

PPN abundance and maximal densities are shown according to vegetable crops and ecological regions in Tables 6, 7, 8, and 9. *Meloidogyne* was the most abundant genus in soil grown to pepper and African eggplant, with 64 and 34 nematodes per 100 cm³ soil, respectively, while *Helicotylenchus*, *Tylenchus* and *Ditylenchus* were the most abundant in okra, tomato and onion soil, with 60, 28 and 16 nematodes per 100 cm³ soil (Table 6). When examining nematode population densities in soil, *Meloidogyne* recorded the maximal values in African eggplant, pepper and tomato with 350, 691 and 164 nematodes per 100 cm³ soil, respectively. Conversely, *Helicotylenchus* and *Tylenchus* exhibited the highest densities in okra and onion soil respectively, with 216 and 59 nematodes per 100 cm³ soil. In the roots, *Meloidogyne* remained the most abundant and exhibited the highest density in African eggplant, pepper, okra, and tomato with the respective highest values of 99 and 2,151 nematodes per g root in pepper. In onion *Pratylenchus* was the most prevalent with abundance value of 12 and density of 49 nematodes per g root (Table 7).

Table 6: Abundance (A) and maximal density (MD) values of plant-parasitic nematode genera in soil samples of vegetable crops

Nematode genera	African eggplant		Okra		Onion		Pepper		Tomato	
	A	MD	A	MD	A	MD	A	MD	A	MD
Meloidogyne	34 (19)	350	20 (6)	72	6 (5)	4	64 (29)	691	19 (18)	164
Pratylenchus	10 (9)	56	4 (2)	6	4 (5)	6	4 (6)	6	19 (9)	134
Tylenchus	8 (12)	30	6 (4)	8	12 (6)	59	18 (15)	136	28 (6)	107
Rotylenchus	3 (1)	3	-	-	-	-	9 (6)	16	-	-
Helicotylenchus	21 (20)	83	60 (8)	216	13 (12)	54	20 (19)	211	17 (19)	96
Tylenchulus	5 (6)	11	3 (1)	3	3 (2)	3	6 (11)	14	3 (6)	4
Aphelenchoides	2 (1)	2	4 (2)	4	3 (3)	3	4 (12)	6	4 (5)	4
Aphelenchus	5 (13)	16	4 (5)	8	5 (10)	18	6 (25)	30	6 (11)	14
Hemicriconemoides	3 (3)	3	8 (3)	8	7 (4)	11	3 (3)	3	3 (9)	4
Rotylenchulus	2 (1)	2	-	-	-	-	-	-	2 (1)	2
Tylenchorhynchus	14 (6)	42	24 (1)	24	8 (6)	24	9 (8)	22	3 (1)	3
Xiphinema	-	-	-	-	3 (1)	3	-	-	-	-
Hoplolaimus	-	-	6 (2)	8	3 (2)	3	3 (1)	3	-	-
Malenchus	2 (1)	2	-	-	-	-	-	-	-	-
Ditylenchus	3 (1)	3	-	-	16 (4)	24	3 (4)	3	3 (1)	3
Geocenamus	3 (1)	3	-	-	5 (10)	19	3 (2)	3	4 (2)	4
Quinisulcius	3 (1)	3	-	-	-	-	-	-	-	-
Trichotylenchus	-	-	-	-	-	-	3 (1)	3	-	-
Trichodorus	3 (1)	3	-	-	3 (5)	3	9 (8)	27	4 (4)	8

Abundance = (Total number of individuals of a specific genus in positive samples)/(Total number of positive samples for that genus); values in brackets represent the number of positive samples for each genus per crop. Maximal density values are nematodes/100 cm^3 of soil
- Genus not detected in the samples

Table 7: Abundance (A) and maximal density (MD) values of plant-parasitic nematode genera in root samples of vegetable crops

Nematode genera	African eggplant		Okra		Onion		Pepper		Tomato	
	A	MD	A	MD	A	MD	A	MD	A	MD
Meloidogyne	74 (29)	570	51 (6)	219	7 (9)	34	99 (42)	2151	46 (28)	428
Pratylenchus	7 (18)	24	4 (5)	12	12 (13)	49	7 (20)	72	6 (24)	22
Hirschmanniella	-	-	-	-	-	-	7 (1)	7	-	-

Abundance = (Total number of individuals of a specific genus in positive samples)/(Total number of positive samples for that genus); values in brackets represent the number of positive samples for each genus per crop. Maximal density values are nematodes/g of root
- Genus not detected in the samples

Meloidogyne recorded the highest soil abundance and density in the Niger Valley (77 and 691 nematodes/ 100 cm³ soil), northern lowlands (42 and 112 nematodes/ 100 cm³ soil) and coastal areas (31 and 350 nematodes/ 100 cm³ soil) (Table 8). *Helicotylenchus* had the highest soil abundance and density in plateau areas (46 and 96 nematodes/ 100 cm³ soil) and southern lowlands (36 and 211 nematodes/ 100 cm³ soil). *Tylenchus* was highest in the Oueme Valley (23 and 136 nematodes/ 100 cm³ soil). In the roots, *Meloidogyne* was the most abundant and had the highest maximal density in all ecological regions, with the highest mean abundance and density values of 141 and 2,151 nematodes, respectively, per g root in the Oueme valley (Table 9).

Table 8: Abundance (A) and maximal density (MD) values of plant-parasitic nematode genera in soil samples taken in ecological regions

Nematode genera	Coastal areas		Plateau areas		Southern lowlands		Oueme valley		Northern lowlands		Niger valley	
	A	MD	A	MD	A	MD	A	MD	A	MD	A	MD
Meloidogyne	31 (21)	350	16 (7)	50	14 (10)	68	22 (12)	92	42 (7)	112	77 (20)	691
Pratylenchus	3 (3)	4	2 (1)	2	21 (11)	134	4 (2)	6	4 (3)	16	5 (11)	11
Tylenchus	3 (3)	4	-	-	20 (13)	107	23 (12)	136	16 (2)	30	4 (13)	6
Rotylenchus	-	-	-	-	-	-	8 (7)	16	-	-	-	-
Helicotylenchus	27 (16)	216	46 (6)	96	36 (17)	211	5 (15)	12	31 (6)	83	9 (18)	54
Tylenchulus	4 (4)	4	2 (1)	2	3 (4)	4	7 (9)	14	3 (1)	3	3 (7)	3
Aphelenchoides	-	-	-	-	2 (1)	2	4 (5)	6	3 (2)	3	4 (15)	6
Aphelenchus	4 (16)	18	3 (4)	3	5 (6)	6	9 (6)	18	4 (4)	6	6 (28)	30
Hemicriconemoides	4 (7)	8	3 (3)	3	3 (3)	3	-	-	-	-	6 (9)	11
Rotylenchulus	-	-	2 (1)	2	2 (1)	2	-	-	-	-	-	-
Tylenchorhynchus	-	-	-	-	14 (4)	42	-	-	10 (3)	16	10 (15)	24
Xiphinema	3 (1)	3	-	-	-	-	-	-	-	-	-	-
Hoplolaimus	3 (2)	3	-	-	8 (1)	8	-	-	-	-	3 (2)	3
Malenchus	-	-	-	-	2 (1)	2	-	-	-	-	-	-
Ditylenchus	-	-	-	-	-	-	-	-	3 (1)	3	9 (9)	24
Geocenamus	-	-	-	-	-	-	-	-	3 (3)	3	5 (12)	19
Quinisulcius	-	-	-	-	-	-	-	-	3 (1)	3	-	-
Trichotylenchus	-	-	-	-	-	-	-	-	-	-	3 (1)	3
Trichodorus	-	-	-	-	-	-	-	-	3 (1)	3	6 (17)	27

Abundance = (Total number of individuals of a specific genus in positive samples)/(Total number of positive samples for that genus); values in brackets represent the number of positive samples for each genus per ecological region. Maximal density values are nematodes/100 cm^3 of soil
- Genus not detected in the samples

Table 9: Abundance (A) and maximal density (MD) values of plant-parasitic nematode genera in root samples taken in ecological regions

Nematode genera	Coastal areas		Plateau areas		Southern lowlands		Oueme valley		Northern lowlands		Niger valley	
	A	MD	A	MD	A	MD	A	MD	A	MD	A	MD
Meloidogyne	57 (35)	570	63 (10)	428	16 (16)	92	141 (18)	2151	134 (6)	516	60 (29)	546
Pratylenchus	6 (18)	24	7 (10)	18	9 (17)	37	5 (5)	15	15 (2)	24	8 (28)	72
Hirschmanniella	-	-	-	-	-	-	-	-	-	-	7 (1)	7

Abundance = (Total number of individuals of a specific genus in positive samples)/(Total number of positive samples for that genus); Values in brackets represent the number of positive samples for each genus per ecological region. Maximal density values are nematodes/g of root
- Genus not detected in the samples

Relationships between PPN genera, vegetable crops and ecological regions

Pearson's chi-squared test showed a highly significant association between PPN genera and vegetable crops ($\chi^2 = 119.74$, df = 76, $P < 0.001$). Correspondence analysis, used to describe the distribution of nematode genera across vegetable crops, revealed that the first two dimensions accounted for 45.8 and 27.5% of the variance, respectively (based on eigenvalues). The projection of nematode genera and vegetable crops in the two-axis system indicated that *Xiphinema*, *Geocenamus*, *Ditylenchus*, and *Tylenchorhynchus* were strongly associated with onion fields. *Hirschmanniella*, *Rotylenchus*, *Trichotylenchus*, and *Aphelenchoides* were particularly linked to pepper crops. *Helicotylenchus*, *Pratylenchus*, and *Hemicriconemoides* were associated with tomato and okra, whereas *Meloidogyne*, *Tylenchus*, *Tylenchulus* and *Aphelenchus* tended to be associated with African eggplant (Figure 4).

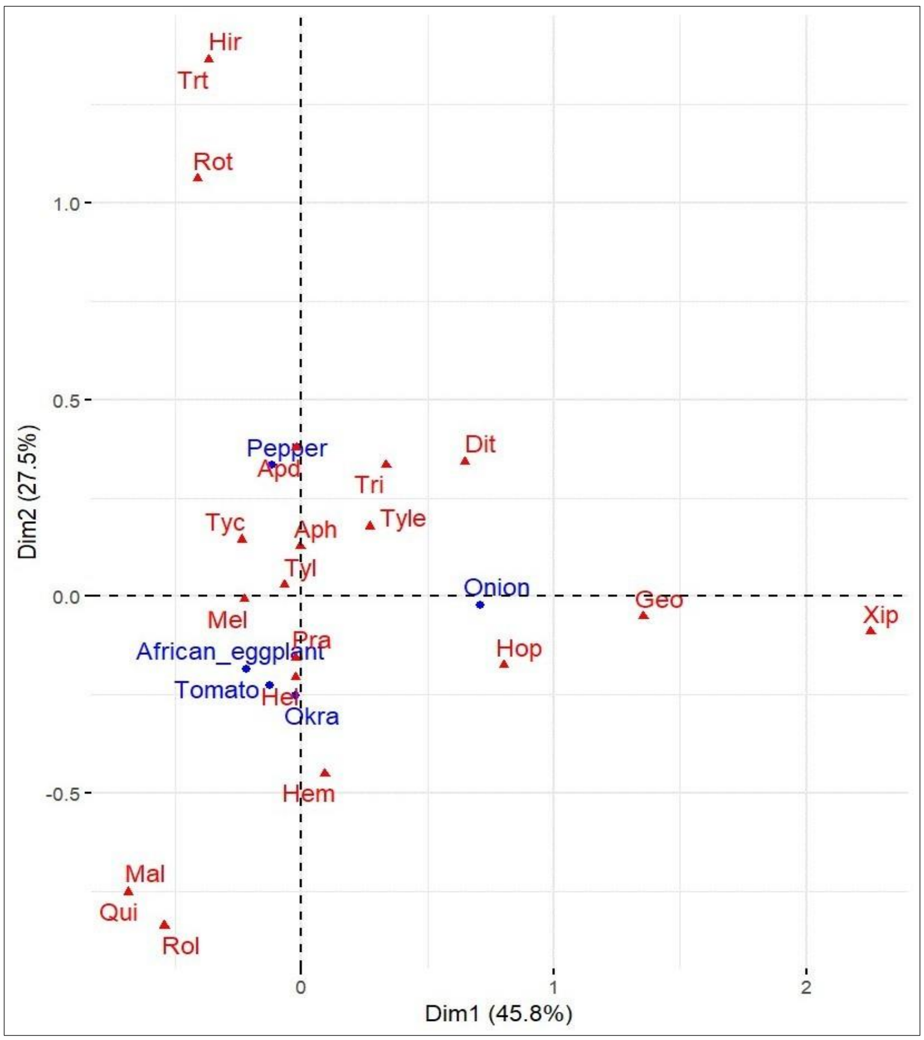

Figure 4: Correspondence analysis of the relationship between plant-parasitic nematodes genera and vegetable crops

A significant relationship was also observed between PPN communities and ecological regions ($\chi^2 = 240.75$, df = 95, $P < 0.001$). Analysis of nematode genera distribution across the ecological regions showed that the total variance explained was 75.4%, with 46.2% attributed to the first dimension and 29.2% to the second dimension (Figure 5). The projection of nematode genera and ecological regions in the two-axis system indicated that *Trichodorus*, *Geocenamus*, *Tylenchorhynchus*, *Ditylenchus* and *Aphelenchoides* were strongly associated with the Niger valley. A strong correlation was observed between *Rotylenchus*, *Tylenchulus* and *Tylenchus* and the Oueme valley fields. Similarly, *Meloidogyne*, *Pratylenchus*, and *Hemicriconemoides* were linked to the coastal areas (Figure 5).

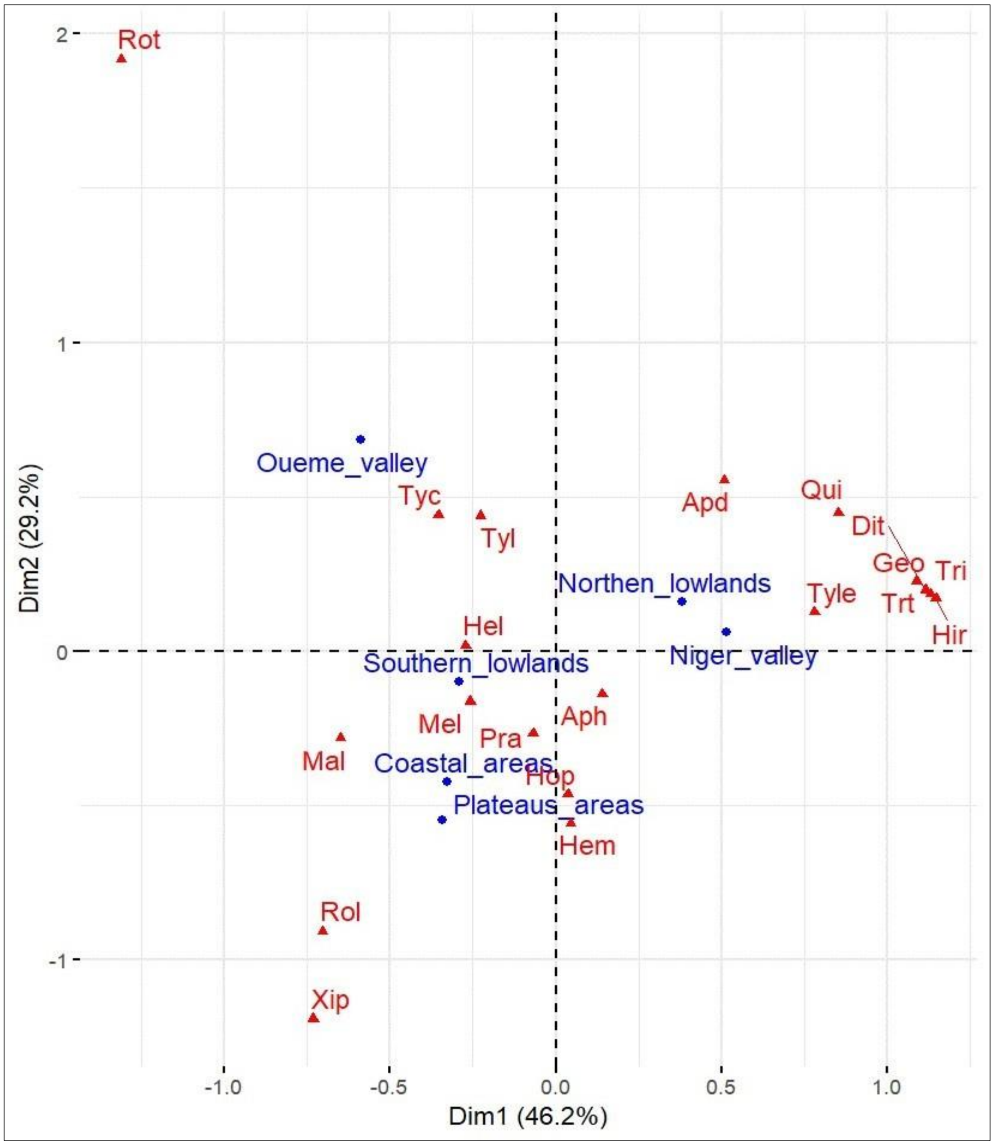

Figure 5: Correspondence analysis of the relationship between plant-parasitic nematodes genera and ecological regions

Correspondence analysis conducted for each individual ecological region showed that the relationship between nematode genera and vegetable crops differed across regions (Figure 6). However, Pearson's chi-squared test revealed no significant association between PPN genera and vegetable crops within specific areas (χ^2 = 9.865, df = 14, P = 0.772 for Oueme valley; χ^2 = 38.077, df = 32, P = 0.212 for coastal areas; χ^2 = 10.10, df = 18, P = 0.928 for plateau areas; χ^2= 25.296, df = 44, P = 0.989 for southern lowlands; χ^2= 23.277, df = 33, P = 0.895 for northern lowlands and χ^2 = 38.330, df = 42, P = 0.633 for Niger valley).

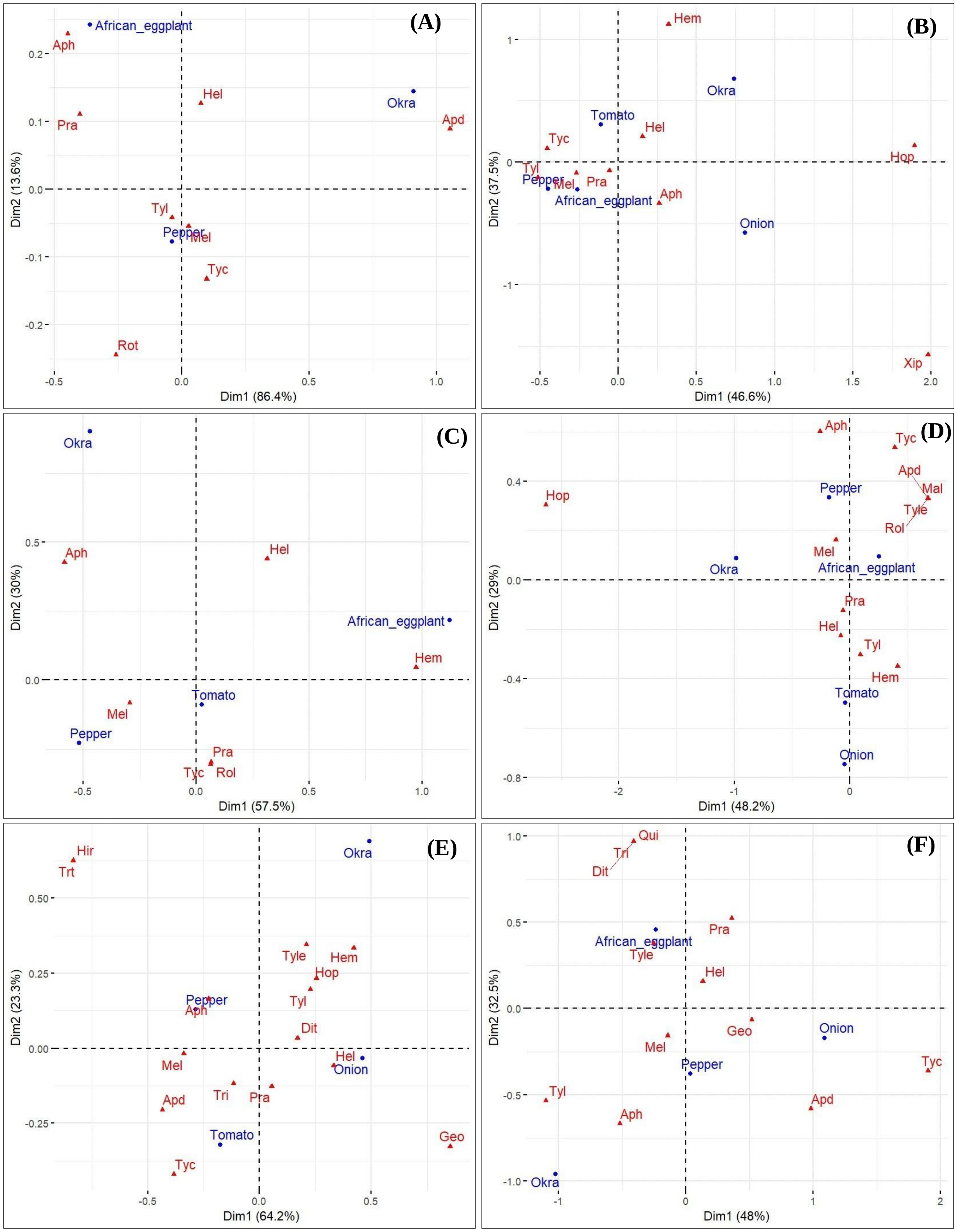

Figure 6: Correspondence analysis of the relationship between plant-parasitic nematodes and vegetable crops in Oueme valley (A), coastal areas (B), plateau areas (C), southern lowlands (D), Niger valley (E) and northern lowlands (F)

Discussion

PPNs are a critical constraint to vegetable production and a threat for food security in SSA (Coyne et al. 2018). Their presence in vegetable-growing areas is likely to reduce crop yields, compromise quality, and increase production costs for farmers, ultimately posing a substantial threat for food security (Coyne et al. 2018; Hallmann and Meressa 2018). Based on the morphological characteristics, 20 PPN genera were identified in the main vegetable growing areas of Benin, with an infestation rate of 98.0%. The study findings underscore the complex nature of PPN communities in Benin's vegetable fields. Although they are often overlooked when *Meloidogyne* is present, the detection in our samples of *Ditylenchus*, *Pratylenchus*, *Rotylenchulus*, *Trichodorus*, *Tylenchorhynchus*, and *Xiphinema* reported as economically important on vegetable crops (Hallmann and Meressa 2018) is an indicator of the potential for PPNs to cause substantial agricultural losses in the country. Compared to previous studies (Baimey et al. 2009; Azandeme-Hounmalon et al. 2023), *Rotylenchus*, *Tylenchorhynchus*, *Geocenamus*, *Trichotylenchus*, *Tylenchus*, *Malenchus*, and *Aphelenchus* were newly recorded. These observations could be due to factors such as sampling sites, agricultural practices, or environmental conditions (Waswa et al. 2020; Cozim-Melges et al. 2025). Among crops, nematode diversity varied with African eggplant and pepper harbouring the highest diversity, probably due to differences in host susceptibility and the suitability of these crops as hosts for PPNs (El-Nuby et al. 2019). These results confirm the nematode host status of African eggplant (Affokpon et al. 2012), the most cultivated and consumed indigenous leafy vegetable in southern Benin, and suggest special attention to its cropping systems to prevent exacerbating nematode problems. In addition to crop-related factors, specific farming practices commonly adopted in vegetable production systems in Benin, including high inputs of chemicals, disrupt soil ecosystems and reduce natural nematode antagonists (Colagiero et al. 2024). Such findings highlight the need to promote better agronomic practices in vegetable cropping systems. Nine PPN genera, including *Meloidogyne*, *Pratylenchus*, *Helicotylenchus*, and *Tylenchorhynchus* were common to the target vegetable crops, whereas others were found in specific crops (e.g., *Hirschmanniella* and *Trichotylenchus* in pepper). These findings indicate the polyphagous nature of some PPN genera and the host specificity of others, as reported earlier by Shanmugam et al. (2021). Consequently, farmer production plans should be considered when designing nematode management strategies. Agroecological management of PPNs could be a suitable option (Sun et al. 2023). Several practices, including cover crops, resistant varieties, crop rotation, intercropping, and biological control, can be made use of and arranged to design sustainable agroecological PPN management strategies (Wezel et al. 2014).

Understanding the distribution of PPNs in relation to ecological regions is essential for locally adapted management practices. In this study, the distribution and diversity of PPNs significantly varied across ecological regions. Many authors have reported the influence of edaphic and ecological factors on nematode communities (Mokrini et al. 2019; Nisa et al. 2021). The distribution of PPNs associated with citrus in Morocco and their interaction with soil patterns were studied by Zoubi et al. (2022), who found that the texture of fine sandy soil positively affects the distribution of PPNs. In this study the Niger valley, characterised by hydromorphic soil and a Sudanian climate, exhibited the highest nematode diversity. These observations can be explained by the variation in soil types and land-use intensity. Five genera, *Meloidogyne*, *Pratylenchus*, *Helicotylenchus*, *Tylenchulus* and *Aphelenchus*, were present in all surveyed ecological regions, indicating their adaptability to a wide range of pedoclimatic conditions. In contrast, some genera were restricted to specific ecological regions, suggesting niche preferences and adaptation to local soil conditions. Similar patterns were reported by Marquez et al. (2021) and Lopez-Nicora et al. (2022), who pointed out the influence of environmental factors and agricultural practices such as soil physicochemical properties, crop types and cultural practices, etc.

Meloidogyne was the most prevalent and dominant genus in both soil and roots, making them the most widespread nematode genus. Their high soil and root densities across most crops and ecological regions confirm their ability to parasitise diverse host plants under diverse environmental conditions. Coyne et al. (2018) and Hallmann and Meressa (2018) noted that *Meloidogyne* are universally associated with vegetable crops due to their broad host range and high adaptability. In addition to *Meloidogyne*, *Pratylenchus* and *Helicotylenchus* were also found to be dominant, as reported earlier by Baimey et al. (2009) and Coyne et al. (2018), requesting further investigation on their potential harmful impact on vegetable crops. In this study, the occurrence of PPNs genera was concomitantly affected by the vegetable crops and ecological regions, proving that PPN occurrence is influenced by the production environment and the cultivated crops. However, it was observed that within individual ecological regions, there was no significant association between PPN genera and vegetable crops, suggesting a random distribution or low specificity of nematodes to crops in given areas.

Conclusion

This study provides a comprehensive understanding of the diversity, geographical distribution, host plants, and ecological region-specificity of PPNs in Benin. The results confirm the ubiquitous nature of *Meloidogyne*, which remains a challenge in sustainable and viable vegetable production. The high prevalence and abundance of other PPNs can be an indicator of their harmful potential and require particular attention when developing an effective management strategy. Further research to understand factors driving nematode distribution and abundance in various ecological and edaphic conditions is crucial for the implementation of sustainable agroecological PPN management strategies in the complex West African vegetable growing conditions.

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Declaration of competing interest

The authors declare no conflict of interest.

References

ACDD (Action - Conseils pour le Developpment Durable). 2019. *Etude d'Identification et de Caractérisation des Flux des Produits Maraîchers Entre le Bénin, le Togo, le Nigéria, le Burkina-Faso et le Niger*. 96p. Ministère de l’Agriculture, de l'Elevage et de la Pêche (MAEP). République du Bénin.

Affokpon, A. 2011. “Potential of Indigenous Biocontrol Agents for Improved Root-knot Nematode Management in Vegetable Production Systems in Benin, West Africa.” PhD Thesis Dissertation, Faculty of Bioscience Engineering, Katholieke Universiteit Leuven, Leuven, Belgium.

Affokpon, A., D.L. Coyne, M. De Proft, and J. Coosemans. 2015. “In Vitro Growth Characterization and Biocontrol Potential of Naturally Occurring Nematophagous

Fungi Recovered from Root-Knot Nematode Infested Vegetable Fields in Benin." *International Journal of Pest Management* **61 (4)**: 273–283. https://doi.org/10.1080/09670874.2015.1043971.

Affokpon, A., D.L. Coyne, C.C. Htay, R. Dossou Agbèdè, L. Lawouin, and J. Coosemans. 2011. "Biocontrol Potential of Native *Trichoderma* Isolates Against Root-Knot Nematodes in West African Vegetable Production Systems." *Soil Biology and Biochemistry* **43 (3)**: 600–608. https://doi.org/10.1016/j.soilbio.2010.11.029.

Azandeme-Hounmalon, Y.G., N.E. Houngbo, M.E. Dossoumou, G.C.A. Mensah, A. Affokpon, O.K. Douro Kpindou, and R. Sikirou. 2023. "*Facteurs de Développment et de Distribution du Flétrissement Bactérien et des Principaux Ravageurs des Solanacées en Maraîchage au Bénin*." 72p. Document Technique et d'Information. Dépôt légal N° 14969 du 17/01/2023, 1er trimestre 2023, Bibliothèque Nationale (BN) du Bénin. ISBN: 978-99982-59-66-9 Ministère de l'Agriculture, de l'Elevage et de la Pêche (MAEP). République du Bénin.

Baimey, H., D. Coyne, G. Dagbenonbakin, and B. James. 2009. "Plant-Parasitic Nematodes Associated with Vegetable Crops in Benin: Relationship with Soil Physico-Chemical Properties." *Nematologia Mediterranea* **37 (2)**: 227–236.

Boag, B. 1993. "Standardisation of Ecological Terms in Nematology." *Fundamental and Applied Nematology* **16 (2)**: 190–191.

Colagiero, M., I. Pentimone, L. Rosso, and A. Ciancio. 2024. "Soil Biodiversity and Microbial Antagonism for Suppression of Plant-Parasitic Nematodes." *CABI Reviews* **19 (1)**. https://doi.org/10.1079/cabireviews.2024.0048.

Coyne, D.L., L. Cortada, J.J. Dalzell, A.O. Claudius-Cole, S. Haukeland, N. Luambano, and H. Talwana. 2018. "Plant-Parasitic Nematodes and Food Security in Sub-Saharan Africa." *Annual Review of Phytopathology* **56**: 381–403. https://doi.org/10.1146/annurev-phyto-080417-045833.

Cozim-Melges, F., R. Ripoll-Bosch, P. Oggiano, H.H.E. van Zanten, W.H. van der Putten, and G.F. Veen. 2025. "The Effect of Alternative Agricultural Practices on Soil Biodiversity of Bacteria, Fungi, Nematodes and Earthworms: A Review." *Agriculture, Ecosystems and Environment* **379**: 109329. https://doi.org/10.1016/j.agee.2024.109329.

de Mendiburu, F. 2023. "Agricolae: Statistical Procedures for Agricultural Research." *R package version 1.3-7*. https://doi.org/10.32614/CRAN.package.agricolae.

De Waele, D., and A. Elsen. 2007. "Challenges in Tropical Plant Nematology." *Annual Review of Phytopathology* **45 (1)**: 457–485. https://doi.org/10.1146/annurev.phyto.45.062806.094438.

Desaeger, J., M.R. Khan, and E.H.C. Silva. 2023. "Nematode Problems in Tomato, Okra, and Other Common Vegetables, and their Sustainable Management." In *Nematode Diseases of Crops and their Sustainable Management*, edited by M. Rahman Khan, and M. Quintanilla, 223–250. New York, USA: Academic Press.

DGEC (Direction Générale de l'Environnement et du Climat). 2022. *Plan National d'Adaptation aux Changements Climatiques du Bénin*. 157p. Ministère du Cadre de Vie et du Développement Durable. République du Bénin.

El-Nuby, A., S. Montasser, and I.A. Khadrawy. 2019. "Diversity of Plant Parasitic Nematodes Associated with Some Vegetables in Sinai Peninsula, Egypt." *Egyptian Journal of Desert Research* **69 (Special Issue)**: 77–100.

Ferris, H. 2012. "Nematodes as Bio-Indicators." In *Practical Plant Nematology*, 1st ed., edited by R.H. Manzanilla-López, and N. Marbán-Mendoza, 677–698. Biblioteca Basica de Agricultura, Montecillo, Mexico.

Fortuner, R., and G. Merny. 1973. "Les Nématodes Parasites des Racines Associés au Riz en Basse-Casamance (Sénégal) et en Gambie." *Cahiers ORSTOM, Série Biologie* **21**: 3–20.

Greco, N., and M. Di Vito. 2009. "Population Dynamics and Damage Levels." In *Root-knot Nematodes*, edited by R.N. Perry, M. Moens, and J.L. Starr, 246–274. Wallingford UK: CABI.

Hallmann, J., and B.H. Meressa. 2018. "Nematode Parasites of Vegetables." In *Plant Parasitic Nematodes in Subtropical and Tropical Agriculture*, edited by R.A. Sikora, D. Coyne, J. Hallman, and P. Timper, 346–410. Wallingford UK: CABI.

Houessou, D., C. Gbedomon, J. van den Broek, K. Gandji, and F. Thoto. 2021. *Roadmap to Strengthen the Vegetables Sector in Benin: Exploring Business Links with the Dutch Private Sector*. 66p. Netherlands Enterprise Agency. The Haque, Netherlands.

Hunt, D.J., J.E. Palomares-Rius, and R.H. Manzanilla-López. 2018. "Identification, Morphology and Biology of Plant Parasitic Nematodes." In *Plant Parasitic Nematodes in Subtropical and Tropical Agriculture*, edited by R.A. Sikora, D. Coyne, J. Hallman, and P. Timper, 20–61. Wallingford UK: CAB International.

Kiontke, K., and D.H.A. Fitch. 2013. "Nematodes." *Current Biology* **23 (19)**: PR862-R864. https://doi.org/10.1016/j.cub.2013.08.009.

Lê, S., J. Josse, and F. Husson. 2008. "FactoMineR: An R Package for Multivariate Analysis." *Journal of Statistical Software* **25 (1)**: 1–18. https://doi.org/10.18637/jss.v025.i01.

Lopez-Nicora, H.D., G.A. Enciso-Maldonado, G.G. Caballero-Mairesse, A.D. Sanabria-Velázquez, A.J. Armandans-Rojas, L.C. Soilán-Duarte, C.J. Grabowski-Ocampos, G.A. Resquin-Romero, A.A. Colmán, L. M. Pedrozo-Fleitas, H.N. Valiente-Raidán, and F.A. Ramiriz-Cardozo. 2022. "Distribution and Abundance of Nematodes in Horticultural Production in Paraguay." *Plant Health Progress* **23 (4)**: 466–475. https://doi.org/10.1094/PHP-01-22-0001-S.

Marquez, J., P.M. Severns, and A. Hajihassani. 2021. "Influence of the Environment and Vegetable Cropping Systems on Plant-Parasitic Nematode Communities in Southern Georgia." *Plant Disease* **105 (10)**: 3181–3191. https://doi.org/10.1094/PDIS-09-20-2019-RE.

Mokrini, F., S-E. Laasli, Y. Karra, A. El Aissami, and A.A. Dababat. 2019. "Diversity and Incidence of Plant-Parasitic Nematodes Associated with Saffron (*Crocus sativus* L.) in Morocco and Their Relationship with Soil Physicochemical Properties." *Nematology* **22 (1)**: 87–102. https://doi.org/10.1163/15685411-00003286.

Nisa, R.U., A.Y. Tantray, N. Kouser, K.A. Allie, S.M. Wani, S.A. Alamri, M.N. Alyemeni, L. Wijaya, and A.A. Shah. 2021. "Influence of Ecological and Edaphic Factors on Biodiversity of Soil Nematodes." *Saudi Journal of Biological Sciences* **28 (5)**: 3049–3059. https://doi.org/10.1016/j.sjbs.2021.02.046.

Oksanen, J., G.L. Simpson, F. Guillaume Blanchet, R. Kindt, P. Legendre, P.R. Minchin, R.B. O'Hara, P. Solymos, M.H.H. Stevens, E. Szoecs, H. Wagner, M. Barbour, M. Bedward, B. Bolker, D. Borcard, G. Carvalho, M. Chirico, M. De Caceres, S. Durand, H.B.A. Evangelista, R. FitzJohn, M. Friendly, B. Furneaux, G. Hannigan, M.O. Hill, L. Lahti, C. Martina, D. McGlinn, M-H. Ouellette, E.R. Cunha, T. Smith, A. Stier, C.J.F. Ter Braak, and J. Weedon. 2024. "Package 'Vegan'." *R package version 2.6-6.1*.

Phani, V., M.R. Khan, and T.K. Dutta. 2021. "Plant-Parasitic Nematodes as a Potential Threat to Protected Agriculture: Current Status and Management Options." *Crop Protection* **144**: 105573. https://doi.org/10.1016/j.cropro.2021.105573.

R Core Team. 2024. *R: A Language and Environment for Statistical Computing 4.4.0.* R Foundation for Statistical Computing. Vienna, Austria.

Sawadogo, A., B. Thio, S. Kiemde, I. Drabo, C. Dabire, J. Ouedraogo, T.R. Mullens, J.D. Ehlers, and P.A. Roberts. 2009. "Distribution and Prevalence of Parasitic Nematodes of Cowpea (*Vigna unguiculata*) in Burkina Faso." *Journal of Nematology* **41 (2)**: 120–127.

Shanmugam, G., A. Mohankumar, P. Sundararaj, and S. Kannan. 2021. "Spatial Distribution of Nematode Genera in Relation to Host Plants of Western Ghats, Tamil Nadu, India." *Acta Ecologica Sinica* **41 (3)**: 189–192. https://doi.org/10.1016/j.chnaes.2020.08.003.

Solin, A., H. Batu Bara, M.I. Auzi, C.A. Tanjung, and B. Amirul. 2024. "The Importance of Consuming Vegetables For Body Health." *Madani: Jurnal Ilmiah Multidisiplin* **2 (5)**: 635–637. https://doi.org/10.5281/zenodo.12210024.

Sun, F., S.F.M. Coulibaly, N. Cheviron, C. Mougin, M. Hedde, P-A. Maron, S. Recous, J. Trap, C. Villenave, and M. Chauvat. 2023. "The Multi-Year Effect of Different Agroecological Practices on Soil Nematodes and Soil Respiration." *Plant and Soil* **490**: 109–124. https://doi.org/10.1007/s11104-023-06062-y.

Waswa, S. J., J. W. Waceke, and S. B. Nchore. 2020. "Diversity of Plant Parasitic Nematodes of Tree Tomato (*Solanum betaceum* Cav.) in Kiambu and Embu Counties, Kenya." *Middle East Journal of Agriculture Research* **9 (3)**: 605–616. https://doi.org/10.36632/mejar/2020.9.3.47.

Wezel, A., M. Casagrande, F. Celette, J-F. Vian, A. Ferrer, and J. Peigné. 2014. "Agroecological Practices for Sustainable Agriculture. A Review." *Agronomy for Sustainable Development* **34 (1)**: 1–20. https://doi.org/10.1007/s13593-013-0180-7.

Wickham, H. 2016. *ggplot2: Elegant Graphics for Data Analysis* xvi+260p. Springer-Verlag, New York, USA.

Zoubi, B., F. Mokrini, A.A. Dababat, M. Amer, C. Ghoulam, R. Lahlali, S-E. Laasli, K. Khfif, M. Imren, O. Akachoud, A. Benkebboura, A.I. Housseini, and A. Qaddoury. 2022. "Occurrence and Geographic Distribution of Plant-Parasitic Nematodes Associated with Citrus in Morocco and their Interaction with Soil Patterns." *Life* **12 (5)**: 637. https://doi.org/10.3390/life12050637.