

Influence of seasonal variations on population dynamics of phyto-parasitic nematodes in soil and roots of bell pepper (*Capsicum annuum* L.) in Otari, Rivers State, Nigeria.

Abstract

Weather instability affects nematode proliferation in soil, affecting plant growth. Identifying endemic species and favourable seasons can guide farmers in cultivating crops during specific seasons, minimizing infections and improving yield. A survey to evaluate the influence of seasonal disparity on plant parasitic nematodes population in soil and roots of bell pepper (*Capsicum annuum* L.) was carried out during dry and rainy seasons in Otari, Abua/Odual Local Government Area of Rivers State, Nigeria. Five bell pepper cultivated fields were randomly selected and a total of 300 soil samples were collected and assayed for soil nematodes. The soil samples were collected using a soil auger at 0-15 cm depth, and roots were collected by the use of sterilized kitchen knife simultaneously for the isolation of nematode. The modified sieve plate method was employed for the extraction of nematodes. Nematode identification was carried out using a pictorial key. A total of 2,220 nematodes from 11 genera were reported in this study, among which 1,290 (58.1%) were recovered during the rainy season while the dry season showed 930 (41.9%) nematodes. Nematode population vary significantly between seasons in soil ($P = .13$) and roots of bell pepper ($P = 3.90$), with *Heterodera* species having higher density (1.44 and 1.03) during the rainy and dry seasons respectively in the soil. High density of *Meloidogyne* (0.97) was recorded during the rainy season and *Radopholus* spp (1.40) during the dry season in the root of bell pepper plant. The study showed that seasonal variations impacted on nematode population abundance in fields, establishing that nematodes are responsive to every unsteady condition of the soil environment resulting from seasonal disparity, and only species who adapt rapidly survive. Farmers should increase crop cultivation during the rainy season to boost yield, as nematodes population decline as rainfall increases.

Keywords: Bell pepper, Nematodes, population dynamics, Seasonal disparity, Soil.

1.0. Introduction

Generally, nematodes have been implicated as crop pests in every cultivated field in Nigeria. Nematodes are versatile [1], [2], inflicting plants with a variety of injuries [3], [4]; yet understanding its community compositions with regards to season is not clear. However, the rate of survival for certain nematode species may be predicted depending on the stability of soil factors which are directly or partially regulated by seasonal disparity. Cedergreen *et al.* [5] reported that the proliferation of nematodes in the soil can be influenced by weather instability. This is because changes in weather may propel a change in soil water content as well as temperature and pH which could inhibit nematode development or incite propagation.

Since nematode survival chances are high in plant tissue, understanding seasonal variations and nematode abundance in soil may aid suitable control measures and guide farmers on the best time to plant to be able to mitigate severe infections and increase yield. Reports have shown that nematodes display host preference [6-7], [4], therefore, limited vegetation in certain fields propelled by change in season may result to low populations of nematodes and affect composition at a given time and season. This is because not every plant will be available all season for active parasitism with species of specificity. [8] and [9] opined that fluctuations in season impact on soil factors and influence soil organisms including phyto-parasitic nematodes. Hassan *et al.* [10] stated that nematodes manifest high populations in soil between June and August (rainy season) and showcase a steady decline from October (dry season). Renco *et al.* [8] reported gradual increase of nematodes in continuous cropping system and more stable seasonal dynamics of nematode species.

Bell pepper, a vegetable crop widely used in Nigeria, thrives well in Otari, yet, quality yield is impaired by the activities of plant parasitic nematodes [2]. Studies on seasonal adaptation of nematodes and severity of infection in relation to bell pepper yield is lacking in Otari and constitute a major set-back in the control of pests. Therefore, a good understanding on nematode propagation and affluence in soil with regards to seasonal disparity will be beneficial to the rural farmers. Also since nematodes are host specific, identification of endemic species and the most favourable season of abundance can help with the most appropriate information regarding the most suitable crop for cultivation against specific season of the year to minimise infections and ameliorate yield. Therefore, this survey is aimed at evaluating the influence of seasonal variations on the population abundance of soil nematodes in rhizosphere and root tissues of bell pepper.

2.0. Materials and Methods

2.1. Study Area

This study was carried out in Otari. Otari is made up of 7 communities in Abua/Odual Local Government Area of Rivers State and lies between latitude $4^{\circ}50'13''N$ and longitude $6^{\circ}39'24''E$. The average rain fall in Otari is about 2000-2500mm and temperature range of $25^{\circ}C$ – $32^{\circ}C$. The vegetation is typical of tropical rain forest. The indigenes are commercial farmers that concentrate mostly on cassava and vegetable crops. The area experiences two seasons, dry season (November to April) and rainy season (May to October).

2.2. Experimental Design

The random sampling design was employed for the study. Five bell pepper monoculture farms were randomly selected and sampled during the dry and rainy seasons. Soil and root sampling was done between December, 2020 to February, 2021 for dry season while rainy season sampling was done between June – August, 2021. The farms were designated F₁, F₂, F₃, F₄ and F₅

2.3. Sampling

2.3.1. Collection of soil and roots

Soil samples were collected randomly from the rhizosphere of ten stands of bell pepper at 0-15 cm depth per farm. These samples were collected once per month between December 2020– February, 2021 and June – August, 2021. The soil samples were collected using a soil auger and a total of fifty soil samples were collected in each month (ten samples in each farm) of sampling per season making a total of one hundred and fifty samples with a **Grand total** soil samples of three hundred. The soil samples were packed into properly labelled white waterproof bags and subsequently transported to the laboratory for nematode extraction.

At each time of soil sampling, ten randomly selected bell pepper stands were uprooted from each farm, and the roots taken at the same time as for soil with the aid of sterilized kitchen knife. The samples, placed into properly labelled polythene bags, were transported to the laboratory for nematodes extraction.

2.3.2. Nematode Extraction Procedure

Nematodes were extracted using the modified sieve plate technique as described in [1]. The soil samples in each water proof sampling bag were examined and detected debris removed. The soil samples from each water proof bag were spread evenly on tissue paper supported on a plastic sieve placed on plastic plates. Water was added to the side of the plates until the soil become wet

but not immerse. After 48 hours, the soil samples were removed and the nematode suspensions emptied into a clean specimen bottles fixed with 5% for viewing using the microscope. The bell pepper roots collected were rinsed with Eva water to remove soil particles. The roots were chopped using a kitchen knife and macerated in an electric blender (BLG 450) for 15 seconds at low speed. Thirty (30) sub-samples were derived from the standardized samples and set up for extraction at each sampling time. The macerated root suspensions were spread evenly on tissue paper supported on a plastic sieve placed on a plastic plate. Water was added to the side of the plates until the root suspensions became wet but not immersed. The extraction set-up was left undisturbed for 48 hours at room temperature. The plate sieve containing the macerate roots was discarded appropriately. The nematode suspensions in the plastic plates were poured into clean specimen bottles and were subsequently fixed with 5% formalin and stored.

2.4. Identification of nematodes

Aliquot of 0.1ml of the nematode suspension extracted from soil and roots samples were taken with pipette and then dispensed on slides. The slides with the nematode suspension were observed using x4 and x10 objective of light microscope for identification and counting of nematodes. Identification of nematodes was done using pictorial keys according to [11] and [12].

2.5. Data analysis

The analysis of data among farms within season to ascertain nematode frequency and density was done using simple percentage ($n \times 100/N$) while **independent t-test** was used to test the significant influence of season on nematodes population abundance.

3.0. Results

3.1. Nematodes population during dry and rainy seasons in soil grown with bell pepper.

Examination of soil during the dry and rainy seasons revealed a total of 1,652 nematodes belonging to 12 genera. Among the 1,652 nematodes recovered in soil cultivated with bell pepper, 1079 (65.3%) were extracted from soil samples collected during the rainy season while the dry season yielded a total of 573 (34.7%) nematodes.

Prominent nematodes recovered during the rainy season were *Gracilachus*, *Helicotylenchus*, *Ditylenchus*, *Heterodera*, *Hoplolaimus*, *Meloidogyne*, *Radopholus*, *Rotylenchus*, *Scutellonema*, and *Tylenchorhynchus* species. Some plant parasitic nematodes were reported in all the five farm sites where samples were collected, while some were missing in certain farms. For instance, *Gracilachus* spp, *Helicotylenchus* spp, *Heterodera* spp, *Meloidogyne* spp, *Scutellonema* spp, were found in all the sampled farms while *Rotylenchus* spp, *Radopholus* spp, *Hoplolaimus* spp and *Ditylenchus* spp were missing in farm F₂, F₃, F₄ and F₅ respectively (Table 1).

The dry season showcased nematodes such as *Heterodera*, *Meloidogyne*, *Pratylenchus*, *Radopholus*, *Rotylenchus*, *Scutellonema* and *Tylenchus*. *Rotylenchus* spp was missing in Farm F₁ and F₂, while *Scutellonema*, *Pratylenchus* and *Tylenchus* were not found in F₁, F₃, F₄ and F₅ respectively. Nematode population showed a significant difference between seasons in soil ($P = .13$) with *Heterodera* species displaying higher density (1.44 and 1.03) during the rainy and dry seasons respectively (Table 1).

Table 1: Nematodes population during dry and rainy seasons in soil grown with bell pepper in Otari, Rivers State, Nigeria.

Season	Nematode genera	Farms grown with bell pepper					Total (%)	Pop. density
		F ₁ (%)	F ₂ (%)	F ₃ (%)	F ₄ (%)	F ₅ (%)		

Rainy season	<i>Gracilachus</i>	37 (14.8)	16 (11.9)	10 (5.4)	19 (6.3)	13 (6.3)	95 (8.8)	0.63
	<i>Helicotylenchus</i>	27 (10.8)	14 (10.4)	30 (16.1)	24 (7.9)	40 (19.3)	135(12.5)	0.90
	<i>Ditylenchus</i>	17 (6.8)	31 (23.1)	22 (17.8)	0	0	70 (6.4)	0.46
	<i>Heterodera</i>	50 (20.0)	27 (20.1)	62 (33.3)	44 (14.5)	33 (15.9)	216(20.0)	1.44
	<i>Hoplolaimus</i>	31 (12.4)	9 (6.7)	13 (7.0)	43 (14.2)	0	96 (8.9)	0.64
	<i>Meloidogyne</i>	33 (13.3)	17 (12.7)	12 (6.5)	37 (12.2)	41 (19.8)	140(12.9)	0.93
	<i>Radopholus</i>	21 (8.4)	9 (6.7)	0	49 (16.2)	30 (14.5)	109(10.1)	0.72
	<i>Rotylenchus</i>	4 (1.6)	0	17 (9.1)	23 (7.6)	9 (4.3)	53 (4.9)	0.35
	<i>Scutellonema</i>	14 (5.6)	5 (3.7)	20 (10.8)	37 (12.2)	17 (8.2)	93 (8.6)	0.62
	<i>Tylenchorhynchus</i>	15 (6.0)	6 (4.5)	0	27 (8.9)	24 (11.6)	72 (6.8)	0.48
Total	249(23.1)	134(27.4)	186(17.2)	303(28.1)	207(19.2)	1,079	7.17	
Dry season	<i>Heterodera</i>	41 (39.4)	32 (27.6)	28 (17.4)	17 (28.8)	37 (27.8)	155(27.1)	1.03
	<i>Meloidogyne</i>	14 (3.5)	24 (20.7)	31 (19.3)	5 (8.5)	12 (9.0)	86 (15.0)	0.57
	<i>Pratylenchus</i>	14 (15.5)	24 (20.7)	31 (19.3)	0	32 (24.0)	96 (16.8)	0.64
	<i>Radopholus</i>	4 (3.8)	17 (14.6)	28 (17.4)	15 (25.4)	23 (17.3)	62 (10.8)	0.41
	<i>Rotylenchus</i>	0	0	27 (16.7)	12 (20.3)	22 (16.5)	91 (15.9)	0.60
	<i>Scutellonema</i>	0	13 (11.2)	16 (9.9)	3 (5.0)	7 (5.3)	39 (6.8)	0.26
	<i>Tylenchus</i>	31 (29.8)	6 (5.2)	0	7 (11.9)	0	44(7.7)	0.29
Total	104(18.2)	116(20.2)	161(28.0)	59 (10.3)	133(23.2)	573	3.8	

3.2. Population of plant parasitic nematodes of bell pepper root in Otari during rainy and dry seasons.

From the root tissue of bell pepper, 568 nematodes belonging to 7 genera were recovered during the rainy and dry seasons. Among the 568 nematodes recovered, the rainy season yielded 211 (37.1%) while the dry season reported 358 (62.9%) nematodes. Important phyto-parasitic nematodes extracted were *Helicotylenchus* spp, *Pratylenchus* spp, *Radopholus* spp, *Rotylenchus* spp, *Tylenchus* spp, *Scutellonema* spp and *Meloidogyne* species. *Tylenchus* and *Scutellonema* species were peculiar to dry season and were not found during the rainy season. However, during the rainy season, *Rotylenchus* spp was missing in farm F₂ and F₅ while *Helicotylenchus* and *Radopholus* spp were not reported from farm F₁ and F₄ respectively. In the root of bell pepper plant, high density of *Meloidogyne* (0.97) was recorded during the rainy season; while

Radopholus spp showed higher density (1.40) during the dry season. Nematode population showed a significant difference between seasons in roots of bell pepper ($P = 3.90$) (Table 2).

Table 2: Population of plant parasitic nematodes of bell pepper root in Otari

Season	Nematode genera	Farms grown with bell pepper					Total (%)	Pop. density
		F ₁ (%)	F ₂ (%)	F ₃ (%)	F ₄ (%)	F ₅ (%)		
Rainy season	<i>Helicotylenchus</i>	0	4 (14.8)	1 (1.8)	2 (12.5)	6 (9.8)	13 (6.2)	0.14
	<i>Meloidogyne</i>	24(46.2)	11(40.7)	20(36.4)	7 (43.8)	26(42.6)	88 (41.7)	0.97
	<i>Pratylenchus</i>	6 (11.5)	3 (11.1)	9 (16.4)	3 (18.8)	10(16.4)	31 (14.7)	0.34
	<i>Radopholus</i>	12(23.1)	9 (33.3)	17(30.9)	0	19(31.1)	57 (27.0)	0.38
	<i>Rotylenchus</i>	10(19.2)	0	8 (14.5)	4 (25.0)	0	22 (10.4)	0.24
	Total	52(24.6)	27(12.8)	55(26.0)	16 (7.6)	61(28.9)	211(37.1)	2.07
Dry season	<i>Helicotylenchus</i>	16(16.6)	7 (5.8)	5 (7.9)	0	1 (1.2)	29 (8.1)	0.32
	<i>Meloidogyne</i>	21(21.9)	11(13.4)	7 (11.1)	6 (18.8)	24(28.6)	69 (19.3)	0.76
	<i>Pratylenchus</i>	6 (6.3)	12(14.6)	0	13(40.6)	17(20.7)	48 (13.4)	0.53
	<i>Radopholus</i>	32(33.3)	7 (5.8)	41(65.1)	7 (21.9)	39	126(35.3)	1.40
	<i>Rotylenchus</i>	4 (4.2)	20(24.4)	8 (12.7)	0	0	32 (9.0)	0.35
	<i>Scutellonema</i>	0	21(25.6)	2 (3.2)	6 (18.7)	0	29 (8.1)	0.32
	<i>Tylenchus</i>	17(17.7)	4 (4.9)	0	0	3 (3.7)	24 (6.7)	0.26
Total	96(26.9)	82(23.0)	63(17.6)	32 (8.9)	84(23.5)	357(62.9)	3.94	

3.3 Actual incidence of nematodes in the soil and roots of bell pepper in relation to month of occurrence in Otari.

Soil sampling recorded high assemblage of nematodes in the month of June (25.3%), July had 15.6 %; while 7.7%, 13.8%, 6.8% and 9.7% of the total population of nematodes occurred in August, December, January and February respectively (Fig. 1). The occurrence in the root tissues of bell pepper was 1.8%, 3.1%, 4.6%, 2.6%, 4.4% and 4.6% for the June, July, August, December, January and February respectively (Fig. 2).

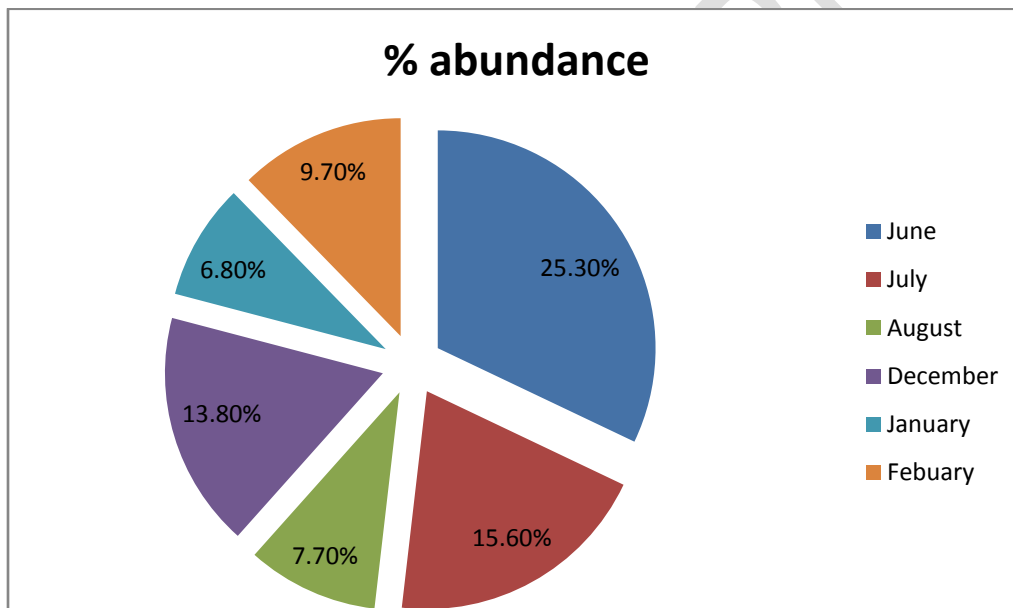


Figure 1: Actual incidence of nematodes in the soil of bell pepper in relation to month of occurrence.

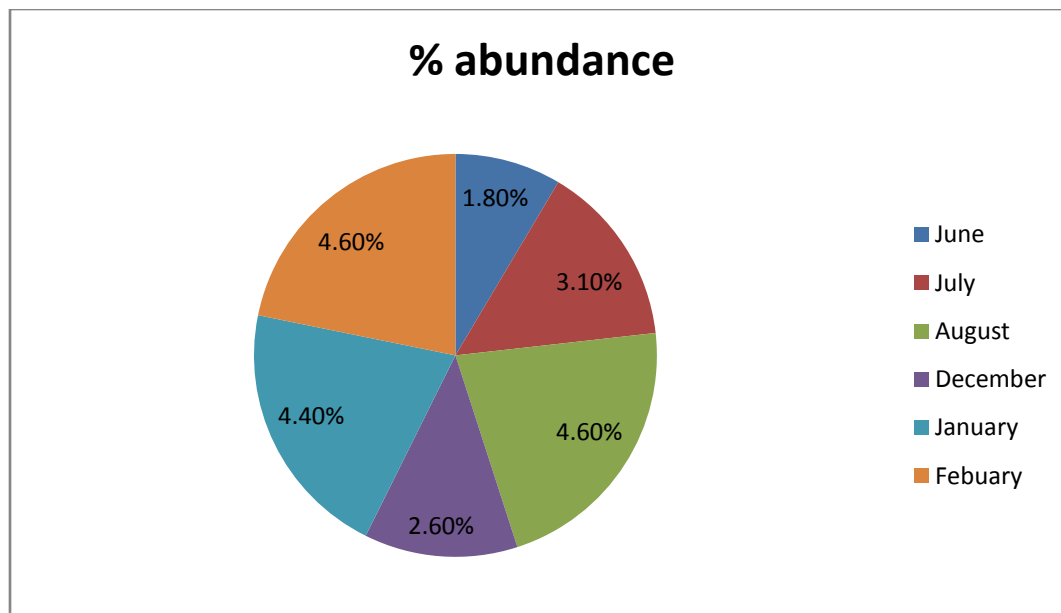


Figure 2: Nematode assemblage in roots of bell pepper and month of occurrence.

3.3. Nematode dynamics in soil and roots of bell pepper during dry and rainy season in Otari, Rivers State.

Total assemblage of phyto-parasitic nematodes during rainy and dry seasons was 2,220. Among the 2,220 nematodes recovered from soil and roots of bell pepper in Otari, 1,290 (58.1%) occurred during rainy season and 930 (41.9%) were extracted at dry season.

There was disparity in the occurrence of nematodes during the sampling seasons. For instance, *Helicotylenchus* spp, *Heterodera* spp, *Meloidogyne* spp, *Scutellonema* spp, *Rotylenchus* spp, *Pratylenchus* spp and *Radopholus* spp occurred during dry and rainy season. However, *Gracilachus* spp, *Hoplolaimus* spp and *Tylenchorhynchus* spp were reported only during rainy season and *Tylenchus* spp was found during dry season only. The abundance of soil nematodes between seasons was statistically significant ($P = .87$). Nematode with most prevalence occurrence during the rainy season was *Meloidogyne* spp (17.8%), closely followed by *Heterodera* spp (16.7%). Significant occurrence was also observed from *Radopholus* spp

(12.9%), *Helicotylenchus* spp (11.5%), *Gracilachus* spp (7.4%), *Hoplolaimus* spp (7.4%) and *Scutellonema* spp (7.2%), with *Pratylenchus* spp (2.4%) showing the least appearance. *Radopholus* spp had the highest frequency of abundance at dry season followed by *Heterodera* spp and *Meloidogyne* species 16.6%. *Pratylenchus* spp was 15.5% while frequency of abundance of *Helicotylenchus* spp was 3.1%. The most prevalent nematode during rainy and dry season was *Meloidogyne* spp (17.3%), closely followed by *Heterodera* spp (16.7%) and *Radopholus* spp (15.9%); while the least occurred nematode was *Tylenchorhynchus* spp (3.2%) (Fig. 3).

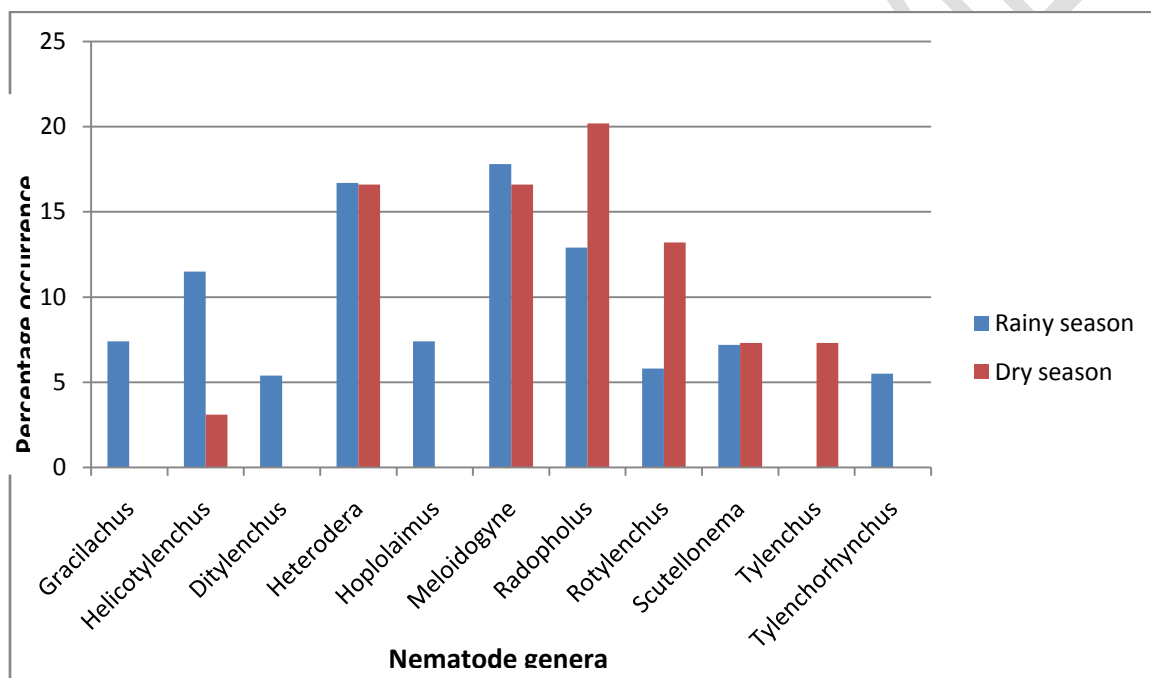


Figure 3: Nematode dynamics in soil and roots of bell pepper during dry and rainy season in Otari.

4.0. Discussion

The result of this study recorded overall occurrence of 2,220 nematodes from 11 genera unlike the result of [13] and [8] which reported 6 and 9 nematode genera in their respective studies on

effects of seasonal fluctuations on nematodes population in the field. However, the result suggests that nematode propagation in Otari occur all through the year not minding season.

The abundance of plant parasitic nematodes in the soil during the dry season (34.7%) was low compare to the recovered population during the rainy season. The low nematode population observed in bell pepper-cultivated soil during the dry season may be attributed to insufficient water in the soil due to the absence of rain as observed in the study area (Otari) during the period under review. The limited water noticed in the soil may have impacted negatively on the propagation and profusion of soil nematodes and have encouraged hypobiosis. However, the high population of nematodes observed during the rainy season could be attributed to nematodes trophic affiliation. This result agrees with [9] who reported low nematode occurrence in their research on occurrence of plant parasitic nematodes and factors that enhance population build up in cereal-based cropping system in Uganda. However, it is in contrast with [10] and [13].

In this study, nematodes were unevenly distributed in soil across the seasons during soil sampling. This scenario is indicative that seasonal fluctuations influence population abundance of plant parasitic nematodes in fields. This result suggests that changes in season directly impact on nematodes abundance in the soil. This is because at each time of switch (from dry season to rainy season), the soil ecosystem is altered and significantly dictate nematode survival chances in the soil [14].

The abundance of nematodes in relation to the month of sampling showed that during the rainy season, the month of June (25.3%) recorded the highest prevalence of soil nematodes and showed a steady decline in July (15.6%) and August (7.7%). The high population of soil nematodes observed in June could be attributed to activeness of nematodes due to rain. The rain,

during the rainy season in the month of June, impacted on the amount of water present in the soil and may have improved its optimum level, enhancing nematode survival strategies. It also stirred up important soil microbes for active associations in the soil, hence, made more food available for nematodes and improve propagation and abundance. Elsewhere, [15] recorded a peak nematode population for all plant-parasitic nematodes in the summer. The progressive decline in nematodes population observed in July and August could be attributed to increased water in the soil due to frequent downpour experienced in the study area during the study period. This result opined that changes in season, alongside fluctuations in rainfall could result to irregular pattern in the occurrence of nematodes in the soil as suggested by [15]. However, [14], [16-17] reported disparity in nematodes frequency of occurrence with respect to variable water content in soil resulting from different amount of rain fall. Nematodes experience less stress for sustainable amplification at rainy season. This is because moderate rainfall could raise soil water to optimum level and improve nutrient for the sustainability of nematodes span [18]. The hydrological conditions of soil during the rainy season can ease nematode migratory ability [19] and facilitate food search and enhances their opportunity to stay alive.

From the roots of bell pepper, greater population of nematodes were isolated at dry season (62.9%) while the percentage abundance during the rainy season (37.1%) was relatively low compare to the results of [20]. This result implied that dryness of soil due to the absence of rain during the dry season presented unsafe environmental conditions for the survival of plant parasitic nematodes and they borrow into the roots tissues of bell pepper for survival. However, improved soil environment due to rain during the rainy season stir up active associations in the soil and increases nutrient concentration around the bell pepper plant and discourages root borrowing. This result also opined that bell pepper has less rigidity or resistance against plant

parasitic nematodes at dry season. This observation agrees with [21] which reported similar result, but disagree with [22] who extracted higher number of nematode species in the root of eggplant. This disparity could be attributed to study location; crop under review and prevalence nematode genera endemic in the study area.

The nematodes population dynamics seen in this study suggests that seasonal disparity affects nematode propagation and profusion. *Meloidogyne* species had the highest prevalence in the study, followed by *Radopholus* species; while *Tylenchorhynchus* species had the least abundance in the study. This study's observation indicates that *Meloidogyne* was better adapted to seasonal fluctuations than any other species occurring in the study area. This study result also suggests that nematode species are sensitive to unsteady conditions of the soil ecosystem resulting from seasonal disparity and only species that are better adapted to survive. *Meloidogyne*, *Scutellonema*, *Hoplolaimus*, *Pratylenchus*, *Helicotylenchus*, *Rotylenchus* and *Radopholus* were reported at both dry and rainy seasons, while *Tylenchus* was found only during dry season and *Gracilachus*, *Ditylenchus*, *Hoplolaimus* and *Tylenchorhynchus* were peculiar to the rainy season only. This scenario could be attributed to nematode survival strategy and persistence environmental fluctuations in the soil due to seasonal variations. It also signifies that soil nematodes propagate and survive only in environments that they are better supported. This observation agrees with [6] and [23].

5.0 Conclusion

Seasonal variations directly impact on nematodes population abundance in the soil as it impacts on soil factors which also influence nematode chances of survival. Soil nematodes are responsive to unsteady environmental conditions resulting from seasonal disparity and only species who adapt rapidly survive. Due to the fact that nematode populations decline as rainfall increases,

farmers should make the most of the season by increasing crop cultivation throughout the rainy season in order to boost yield. Since Otari's endemic species population declines with frequent rainfall, farmers should try to expose agricultural area cleared for production to flooding by irrigation for at least 21 days before ploughing.

List of abbreviations:

1. F₁ = Farm 1
2. F₂ = Farm 2
3. F₃ = Farm 3
4. F₄ = Farm 4
5. F₅ = Farm 5

Declarations:

Ethical Approval: Not applicable

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