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Abundance of Tylenchid Nematodes in Banana (*Musa* sp.) fields with soil abiotic factors in Vasai, Palghar (M.S), India

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ABSTRACT

Bananas are a major commercial crop that the Vasai region majority of farmers cultivate. One of the biggest challenges to the cultivation of bananas is nematodes. Tylenchids have long been recognized as a major challenge to banana production, leading to significant yield losses. As a result, this study aims to examine how the abundance of tylenchid nematodes changes in response to various abiotic soil factors. According to the results of the current study, the tylenchid nematodes population were high throughout the rainy, winter and remained comparatively lower in the summer season.

Key words: *Tylenchids, Soil abiotic factors, Banana, Vasai.*

Introduction

In India, bananas are cultivated in 20% of agriculture.Maharashtra is the second-largest state in terms of land and the most productive state in India. The Jalgaon district has more than 45,000 hectares of this crop grown (Economics Survey of Maharashtra, 2019-20). The Thane district of Konkan, with a total area of 911 hectares, predominantly in the Vasai and "Palghar" tehsils, is where the banana is produced commercially. The marketing system for bananas acts as an intermediary between a widely dispersed and independent producer and a broadly dispersed and independent consumer. A successful marketing strategy makes sure that the product is offered to customers at a fair price, enabling the producer to be paid (Babhulkar, 1998; Kshirsagar and Chavan, 2019). As a result, the current study was conducted in the Vasai, Palghar district with a particular objective in mind. On a global and regional basis, there are several estimates of the economic significance of nematodes in agricultural production, but precise estimates of losses are missing. Nematodes play a significant role in crops because the environment is essential for the growth of nematode populations. Nematodes are important as disease-causing organisms, although this has frequently been forgotten. Nematode infestation symptoms are vague and often confused with signs of nutritional insufficiency. Nematode density in the soil affects how much harm nematodes do to crops. Nematode damage does not become apparent until populations reach a certain level of economic harm (Lavhe *et al*., 2019).

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Plant-parasitic nematodes are tiny worms that live mainly in soil and roots. In the case of banana plants, the most damaging species spend most of their life cycle in root and corm tissues. The plant parasitic nematodes as pests of different horticultural and field crops are responsible for an annual monetary loss of Rs. 300 cores/annum in Tamilnadu alone. Namely, 50 species of plant parasitic nematodes have been reported in association with banana root systems (Parvatha Reddy, 1993) and are mainly responsible for controlling banana production to a greaterextent (Koshy*et al*., 1978, Rajendran *et al.*, 1979; Rajendran *et al.*, 2002 and Sundararaju, 2005). Infestation by Plant Parasitic Nematodes is also a serious worldwide concern for banana cultivation. Globally, more than 151 nematode species of 51 genera are known to be associated with *Musa* sp. (Gowen and Quenehenve, 1990; Koshy and Gulsar Banu, 2000) causing an estimated 19.7% yield loss which approximately amounts to US $ 17.8 million (Sasser and Freckman, 1987). In India, 71 species of Plant-parasitic nematodes are known to be associated with bananas (Krishnappa and Reddy, 1995; Koshy and Sosamma, 2001). They identified 21 species of 14 Plant-parasitic nematodegenera from the banana rhizosphere in West Bengal. The most common genera of Plant-parasitic nematodes associated with bananas are *Pratylenchus, Meloidogyne, Helicotylenchus, Tylenchorhynchus, Hoplolaimus, Rotylenchus, Hirschmanniella, and Criconemoides* (Khan and Mukhopadhyay, 2004). The majority of the farmers in the area gave and planted the saplings for Vasai's first harvest of the Velshi or elaichi kind of bananas. Given the unsettling nature of this pest, the current investigation was made to look at the diversity of plant parasitic nematodes connected to the widely cultivated bananas in Vasai.

Materials and Methods

The methodology is based on existing guidelines and has the following distinct features:

Site selection for taxonomic and ecological studies: After a detailed survey of some Banana fields was selected for the collection of soil samples for the taxonomy and ecology of nematodes. The selected fields are from different villages Nirmal, Girij, Gass and Bhuigaon B.K of Vasai Tahsil in Palghar district Maharashtra, India. The sampling method is very simple i.e., pooled two soil samples with more or less regular monthly. Periodicity is collected from each collection site and about 250 gm of soil from each Banana field from March, 2021- April, 2023.

Procedure of sampling of soil: For taxonomic purposes, moist soils around the tender roots of the Banana was collected with the help of a Trowel from a depth of 0-20 cm from the surface. Two sub-samples of about 250 g each were taken from four different locations of a particular Banana field. These soil samples were collected in polythene bags and their open ends were closed with rubber bands to bring them to the laboratory. All relevant taxonomic data like host plants, details of locality, date of collection, name of collector etc. were noted at the time of collection and the labels were kept safely in the folds of the polybags bag of the respective samples. Samples were collected randomly with more or less regular periodicity monthly from March. 2021- April. 2023.

For ecological studies, soil samples were collected for ten consecutive months from March. 2021- April. 2023 from four selected Banana fields. These were collectedon the second day of every week of the month from a distance of about 2 feet from the trunk the of Banana. Two samples of 250 g each, composed of two cores of soil sampled to a depth of 0-20 cms were collected randomly from each Banana field. A portion of soil samples apart from 250 g were kept separate for the estimation of soil abiotic factors.

Processing of soil samples and extraction of nematodes: The Baermann funnel method (Plate No. 4) is an excellent system for separating specimens in soil and condensing them for examination. The apparatus consists of a regular funnel with a short piece of rubber tubing attached to the stem and closed by a clamp or pinch cock. The funnel is supported in an upright position and filled with water. The procedure of this method is very simple described below.

i) Place about 250 g of soil in a glass beaker pack with muslin cloth by rubber band or thread.
ii) Place the beaker on top of a Baermann funnel apparatus and attach it to a stand.
iii) Fill the funnel up to the rim of the water. The nematodes move through a muslin cloth into the water in the funnel. They settle at the bottom of the funnel by gravity, where they are collected (10 - 20 ml suspension) after 1 day. The nematodes are ready for counting and identification under a dissecting microscope.

Fixation and Preservation of Nematodes: The nematodes, kept in 3-4 ml of water in the test tube, were killed and fixed instantly in their characteristic body posture by Steinhorst's method (Steinhorst, 1966) in hot (90-100 ºC) FA (formalin- acetic acid 4:1) solution, the composition of which was as follows:

Formalin (40% formaldehyde): 40 ml
Glacial acetic acid: 01 ml
Distilled water: 89 ml

The fixed nematodes were preserved in the same solution in specimen tubes or vials with appropriate labels.

Picking and dehydration of nematodes: The fixed nematode specimens, obtained from a particular soil sample, were taken in a glass cavity block and placed under a low-power stereoscopic binocular. The nematodes were picked up one by one from the fixative with the help of a micropipette and sometimes with the help of a single soft bristle of camel-hair brush. The picked nematodes were transferred to vials containing 3-4 ml of glycerin-alcohol solution, the composition of which was as follows:

Anhydrous glycerin: 5 parts
30% alcohol (alcohol-water 30:70): 95 parts

Now, the vials containing the nematodes in glycerin-alcohol solution were placed in an airtight glass desiccator with fused calcium chloride (CaCl2) in its lower chamber for slow dehydration of nematodes at room temperature for 3-6 weeks depending upon the seasonal relative humidity.

Counting of nematode population for ecological studies: The live specimens were taken for population counting. After extraction, the nematodes of a particular sample with 3-4 ml of water were taken. A micropipette took 10 ml of this water, containing nematodes, in a counting dish. The counting of nematodes in the counting dish was done under a low-power binocular microscope. The same process of counting was repeated thrice for everytwo samples per Banana field separately. The meanof the nematode population belonging to different genera from three counts of each sample was recorded.

In this way, the individual means the of nematode population for each of the four samples was calculated. After that, the mean to the al population of each genus of two samples for a particular month was finally calculated and recorded for statistical analysis.

Soil abiotic factors of the Four Banana field: A portion of soil samples, collected the from Four Banana field selected for ecological studies, was kept separate to estimate the soil abiotic factors like soil moisture, soil pH and soil temperature.The soil samples were thoroughly air dried under shade, grinded into fine particles, and passed through 2mm a pore size sieve. The soils were stored in containers with labels of locality and the month of collection of about to following factors was carried out.

A) Soil Temperature: Soil temperature was recorded every month from March. 2021- April. 2023 from Four Banana field at the time of collecting soil samples with the help of a soil thermometer. The soil thermometer was pushed in the soil at least up to 10 cm and kept there until a constant temperature was reached andthe temperature was reached and the temperature was recorded.

B) Soil moisture: Small amounts of soil were collected and kept in polythene bags to prevent the loss of moisture content. Initially, 10 g of soil with moisture was taken and kept in the oven at a temperature of around 105 °C overnight and re-weighted. The loss of weight of 10 g of soil a in hot air oven is equal to the weight of its moisture content. Then, the amount of soil moisture was calculated in percentage from the difference in this weight.

C) Soil pH: Soil pH was measured by making two preparations – (i) soil in water and (ii) soil with 1(N) KCL solution in a ratio of 1: 2.5ml in both cases. 10

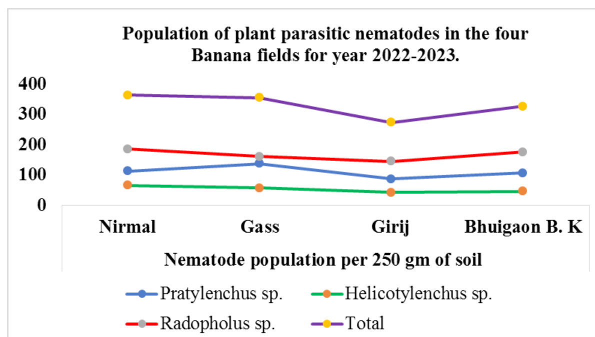
g of soil sample were taken in a beaker and 25 ml of distilled water was added. The content of the beaker was stirred intermittently with a glass rod for half an hour. Now pH meter was adjusted against a standard buffer solution. The electrode of the pH meterwas washed and inserted into the beaker containing the soil solution after stirring the content of the beaker. The pH of the soil solution was recorded by directly readingthe pH meter.

Statistical analysis with the estimated data: Finally, the data obtained from the monthly population count of the genera belonging to the order Tylenchida along with the estimated soil abiotic factors from the respective Banana fields were subjected to appropriate statistical analysise.g., correlation and linear regression analysis, to realise the relation between them by using Kyplot software.

Results and Discussion

Population fluctuation of the nematode order Tylenchida in Banana fields: In Nirmal Village, Vasai Tahsil Banana field, the maximum total population abundance of Tylenchida was noticed in October 2021-22 (51 and 58 / 250 g of soil). Tylenchids were counted minimum in April 2021-22, being 14 and 17. In Gass Village, Vasai Tahsil Banana field, the maximum total population abundance of Tylenchida was noticed in December 2021-22 (58 and 62 / 250 g of soil). Tylenchids were counted minimum in March and May 2021-22, being 17 and 21. In Girij Village, Vasai Tahsil Banana field, the maximum total population abundance of the maximum population of Tylenchida was noticed in November 2021-22 (44 and 48 / 250 g of soil). Tylenchids were counted minimum in June 2021-22, being 12 and 17. In Bhuigaon B.K Village, Vasai Tahsil Banana field, the maximum total population abundance of the maximum of Tylenchida was noticed in November 2021-22 (47 and 45 / 250 gm of soil). Tylenchids were counted minimum in March 2021-22, being 12 and 18 (Table 1, 2 and Graph 1).

In the present observation, the total population of all the nematode orders started coming from September and October for the months under study. It maintained a high population abundance up to December, reaching its highest in September. The population started declining notably from March to May followed by June- July. In the present observation, the total population of all the nematode orders started up from September and October for the months under study and maintained high population abundance up to December, reaching its highest in September. The population started declining notably in March to May followed by June- July. The



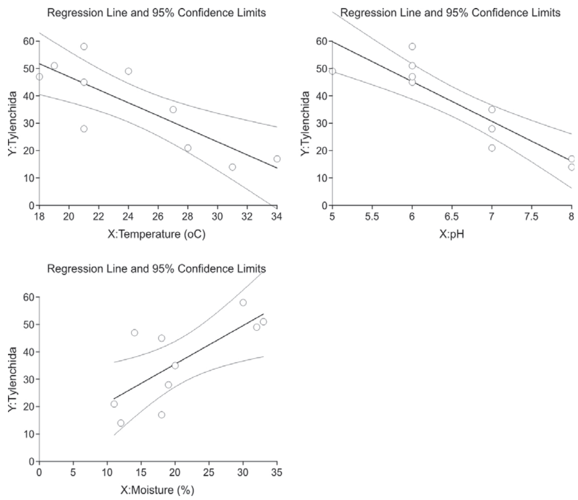
Graph 1. Population of plant parasitic nematodes in the four Banana fields.

Table 1. Soil abiotic factor parameters of soil in Banana fields.

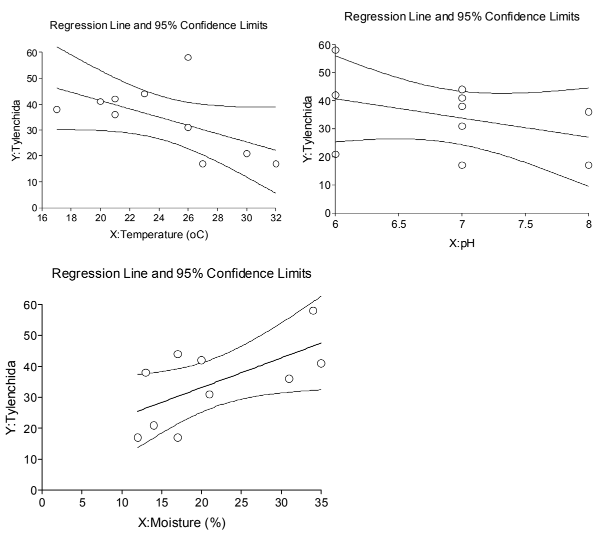
Collection Site	Soil type	Temperature	Moisture	pH
Nirmal	Sandy loam	25.2	20.7	6.6
Gass	Sandy loam	24.2	21.4	6.9
Girij	Sandy loam	21	26.4	6.8
Bhuigaon B. K	Clay loam	21.8	28.2	7

Table 2. Population of plant parasitic nematodes in the four Banana fields.

Nematode Genus	Nematode population per 250 gm of soil			
	Nirmal	Gass	Girij	Bhuigaon B. K
Pratylenchus sp.	113	137	86	106
Helicotylenchus sp.	66	57	42	45
Radopholus sp.	185	161	145	176
Total	364	355	273	327



Graph 2. Linear regression of the Tylenchida population in relation to soil abiotic factors (as mentioned on X-axis) in Nirmal Village Banana field. (Data is shown in Table 3).



Graph 3. Linear regression of the Tylenchida population in relation to soil abiotic factors (as mentioned on X-axis) in Gass Village Banana field. (Data is shown in Table 3).

seasonal variations in the population of plant and soil nematodes were discussed by many scientists (Oostenbrink, 1960; Seinhorst, 1967 and 1968; Wallace, 1969; Brodie, *et al.*, 1970a, b, c and Khan *et al.*, 1971).

Many observers reported seasonal population fluctuation of nematodes concluding differently in regard seasonal population of nematodes concluding differently in regard of their maximum growth from various climatic conditions of tropics and subtropics which are either consistent or inconsistent with the present investigation, yet at least in one point they agree that the soil moisture, rainfall, temperature etc. have definite and significant influence in the growth of nematode population (Marcelino *et al.*, 1978; Harris, 1979; Pinochet *et al.*, 1990; Deshmukh *et al.*, 1990; Srivastava *et al.*, 2000; Rama and Dasgupta, 1987; Rama and Dasgupta, 2000 and Kumar, 2002). Two nematode population peaks - one in October- November and a second peak in July of *Hoplolaimus indicus* around the soil guava in Allahabad, India evidenced in the work of (Dwivedi *et al.*, 1987) resemble close to the present findings.

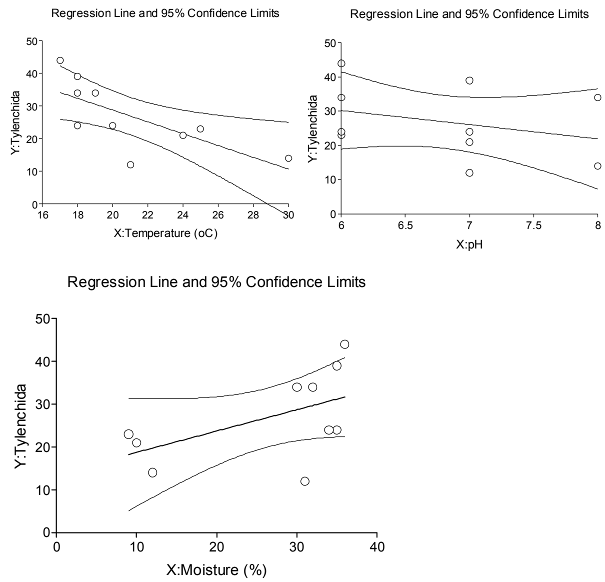
Sundarababu Rajeswari and Muthukrishnan (1995) have similar results on the population prevailed under field conditions in the soils around west Indian cherry trees for a period of one year indicating two peak Periods of increased nematode population, the first being in November to February and the second in April and May. Annamalai and Muzna (1996) also had similar findings for the population of *Pratylenchus jordanensis* in soil did not vary significantly and was high throughout the year. However, the population was lowest in May and August at the first and second sites, respectively.

Soybean production and soybean cyst nematode densities were studied about soil pH (Rogovska *et al.*, 2009), and the results showed that relative yield fell with increasing pH compared to resistant growers. The density of soybean cyst nematode populations was more closely connected to soil pH. Although various studies imply that soil pH has no harmful effect on nematodes unless at extremely low or high levels, as reported by (Norton, 1979), soil pH appears to have a minor influence on H. multicinctus and *M. incognita* in this study.

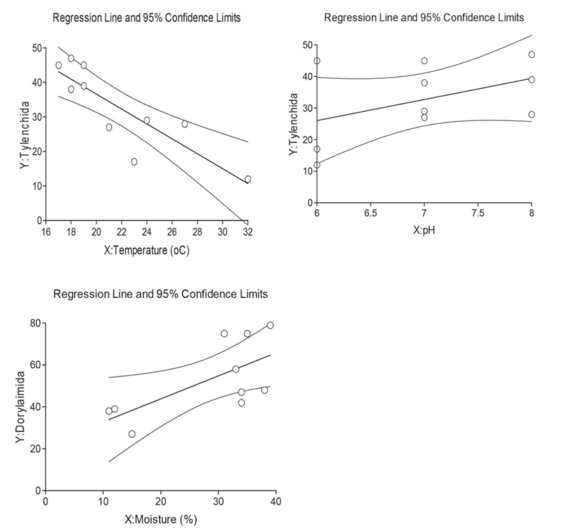
Correlation and regression analyses of the nematode order Tylenchida: Soil abiotic factors like temperature, moisture and pH were also observed during the same period and were recorded and the values of temperature for correlation coefficient (r) show a negative impact on Tylenchid order nematode populations, $P \leq 0.05$. The moisture content of the soil at a significant level (correlation coefficient (r) shows a positive impact on different order nematode populations, $P \leq 0.05$. In contrast, pH of soil also affected the population of all the genera having a negative impact on their population growth with correlation coefficient (r) with orders nematode populations, $P \leq 0.05$. Ingeneral, populations of all orders of nematodes were highest during the rainy and winter seasons (June-December) and remained comparatively lower in summer. The available Tylenchid orders were correlated to three soil abiotic factors mentioned above along with linear regression analyses- these effects of soil abiotic factors on the nematode population. Soil temperature and pH show a negative correlation with the nematode population. Soil moisture was positively correlated ($P \leq 0.005$) with the population of different genera for all four Banana fields (Table 3 and Graph 2,3,4,5).

Annamalai and Muzna (1996) also had similar findings for the population of *Pratylenchus jordanensis* in soil did not vary significantly and was high throughout the year. However, the population was lowest in May and August at the first and second sites, respectively. The correlation between nematode soil populations and soil temperature was not significant ($P > 0.05$). In contrast with soil population, nematode populations in roots fluctuated greatly with a distinct high from July to November at both locations, which coincided with higher soil temperatures. Despite the presence of a high nematode population in the soil, the population in roots was very low from December to June, coinciding with lower soil temperatures. These findings were confirmed by the positive correlations between the populations in roots and the soil temperature (r = 0.433 and 0.324; $P = 0.01$), represented by the linear regression equations, $Y=78.45+29.43x$ and $Y=76.11+3.38x$. The high nematode population in roots from July to November suggested that the penetration and/or reproduction of *P.jordanensis* into roots were favoured by high soil temperature.

In a practical and honest confession, we must accept that the growth/fall of the overall nematode population is the result of the simultaneous interaction of all available biotic components of the soil and nematode population in a naturally existing ecosystem. In light of this, a population-based experiment to establish the impact of abiotic components would be impractical.



Graph 4. Linear regression of Tylenchida population in relation to soil abiotic factors (as mentioned on X-axis) in Girij Village Banana field (Data is shown in Table: 3).



Graph 5. Linear regression of Tylenchida population in relation to soil abiotic factors (as mentioned on X-axis) in Bhuigaon B. K Village Banana field. (Data is shown in Table: 3).

Table 3. Correlation coefficients (r) between order Tylenchida different soil abiotic factors in four Banana fields, Vasai Tahsil, and Palghar district.

Soil abiotic factors	Nirmal	Gass	Girij	Bhuigaon B. K
Temperature	-0.8155*	-0.5731*	-0.7102*	-0.8521*
Moisture	0.7306*	0.6354*	0.5331*	0.4936*
pH	-0.8949*	-0.3828*	-0.3108*	0.4497*

(* Significant correlation ≤ 0.05).

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Conflict of Interest- None

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