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Population Dynamics and Eco-friendly Management of Root Grub and Root-knot Nematode using Entomopathogenic Nematode and Fungi in Small Cardamom

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Abstract: The present investigation was undertaken to study the seasonal fluctuations and eco-friendly management of root grub, *Basilepta fulvicorne* (Jacoby) and plant-parasitic nematodes (PPN), *including Meloidogyne javanica* (Treub) in small cardamom from January 2022 to December 2023. This study documented that the *B. fulvicorne* incidence was negligible during November to January, with no grubs recorded from February to April 2022 and March to April 2023. Populations increased thereafter and peaked during September 2023. Plant-parasitic nematodes (PPN) populations followed a similar pattern, with minimum densities in February 2022 and January 2023, reaching a maximum during August 2023. Pearson correlation analysis showed a strong positive association between *B. fulvicorne* and PPN populations and significant correlations with rainfall, rainy days and relative humidity. The combined application of *Heterorhabditis indica* (ICRI EPN-18), *Metarhizium anisopliae* (ICRI MA RG-3), and *Purpureocillium lilacinum* resulted in complete suppression of *B. fulvicorne* and reduced root-knot nematode galling by more than 85 per cent, offering an effective eco-friendly management strategy for small cardamom.

Keywords: Root-knot nematode, Cardamom, PPN, Entomopathogenic nematode root grub

Small cardamom, *Elettaria cardamomum* (L.) Maton belongs to the family Zingiberaceae and popularly known as the "Queen of spices" is grown under forest ecosystem. In India, it is cultivated over an area of about 73,795 hectares, primarily confined to the Western Ghats regions of Kerala, Karnataka, and Tamil Nadu (Narayana et al., 2017). Soil pests on small cardamom include cardamom root grub, *Basilepta fulvicorne* (Jacoby) and root knot nematodes (*Meloidogyne* spp.), which are considered as seasonal pests (Varadarasan et al., 2011). Among the root-knot nematodes (RKN), *Meloidogyne incognita* and *M. javanica* are of major importance, causing significant damage to the crop. Additionally, *Radopholus* sp. and *Pratylenchus* sp. are also recorded when cardamom is grown as an intercrop with arecanut, coffee, or banana (Praveena et al., 2013). In recent years, the excessive use of pesticides to manage *B. fulvicorne* and *Meloidogyne* spp. has led to the accumulation of chemical residues in cardamom soils, degrading soil health and leaving harmful residues on capsules, ultimately compromising the export quality of cardamom. The use of entomopathogenic fungi (EPF) such as *Metarhizium* and *Purpureocillium,* in combination with entomopathogenic nematodes (EPN) belonging to the genera *Steinernema* and *Heterorhabditis*, offers a promising alternative to chemical pesticides. The forest-based agroecosystem in which cardamom is cultivated provides a favourable environment for the survival and persistence of EPN populations (Poinar, 1990, Grewal and Georgis 1998). Among EPN species, *Heterorhabditis indica* (Josephrajkumar et al., 2005, Varadarasan et al., 2011), and *Oscheius* spp. (Pervez et al., 2016) has been reported to be effective biocontrol agents against *B. fulvicorne*. India has a great potential to exploit these beneficial nematodes, EPNs for the suppression of insect pests, however, the technology has yet to be developed and adopted at the farmer level. The seasonal incidence of *B. fulvicorne* and plant-parasitic nematodes (PPNs) in small cardamom remains poorly documented, yet is critical for understanding pest dynamics and designing effective management strategies. As cardamom is an export-oriented crop, minimizing pesticide residues in capsules is essential. Therefore, this study investigates the population dynamics of key soil pests and evaluates the combined efficacy of EPNs and EPFs for their eco-friendly management.

MATERIAL AND METHODS

Study area: The study was conducted at the research farm, Indian Cardamom Research Institute (ICRI), Spices Board India, Myladumpara, Idukki Dt., Kerala (longitude of 9^{0}8'E and latitude of 77^{0}2'N at an altitude 1050 MSL) from January 2022 to December 2023 in small cardamom ecosystem. Field experiments were conducted during June - November for two consecutive years 2022-23 and 2023-24 in naturally infested soil.

Population dynamics of *B. fulvicorne* and PPNs: The five-year-old Njallani Green Gold cultivar plot was selected for the study and population of grubs of *B. fulvicorne* and plant parasitic nematodes (PPN) were counted from randomly selected five plants in an acre which were maintained without pesticide application at monthly intervals throughout study period. For *B. fulvicorne*, the number of grubs per 15 cm^3 of soil was recorded as the mean of five samples (ICRI 2008 and Varadarasan et al., 2011). Similarly, for PPN, the number of nematodes per 200 cc of soil was determined during 2022-23 (Thomas and Goddard 1986, Giné et al., 2014). The population of *B. fulvicorne* and PPN was documented and then correlated with various weather factors, specifically maximum temperature (X_1), minimum temperature (X_2), relative humidity (X_3), rainfall (X_4) and rainy days (X_5). Subsequently, the obtained data was regressed for the prediction of the *B. fulvicorne* and PPN population. Statistical analyses were performed using the “Agricolae” package version 1.4.0 (Mendiburu 2015) and the correlation matrix with scatter plots graphs was prepared using “Performance Analytics” package for (Peterson et al., 2018) in R Studio 2025.09.2+418.

Culturing of EPN and EPF: Infective juveniles (IJs) of native isolate of EPN *Heterorhabditis indica*, a strain ICRI EPN - 18 (Vardarasan et al., 2011) and an isolate ICRI MA RG-3 of *Metarhizium anisopliae* (Metschn.) Sorokin were isolated in the grubs of *B. fulvicorne* from Cardamom Hill Reserve (CHR), Idukki district, Kerala (Fig. 1) were bio-assayed and both are found effective against cardamom *B. fulvicorne*. The EPN strain ICRI EPN -18 and an isolate ICRI MA RG-3 of Entomopathogenic fungi (EPF) were mass multiplied (Sydhic 2005 and Thiyagarajan et al., 2023) at ICRI, Myladumpara and used for field study.

Field experiments: The experiments consisted of eight treatments implemented in a randomized block design with three replications. Five-year-old 'Njallani Green Gold', a popular small cardamom cultivar was used, with each replication consisting of 12 plants. All standard agronomic practices were followed except pesticide applications. Based on preliminary studies, an EPN strain ICRI EPN -18 @ 4 lakhs IJs, native isolate ICRI MA RG - 3 of *M. anisopliae* @ 25g/plant and *Purpureocillium lilacinum*, previously known as *Paecilomyces lilacinus* @ 25g/plant which was collected from Cardamom Research Station, Kerala Agricultural University, Pampadumpara, Kerala were used along with vermicompost @ 2kg/plant. The treatments of bio-control agents consisted of eight treatments including untreated check (Table1). Two applications were given at 90 days intervals, first application during onset of monsoon (June) and second application during September (post monsoon). EPN infected *Galleria* cadavers were implanted in 7.5 cm away from the plant base at a depth of about 5cm. Care was taken to ensure adequate moisture before application of EPN and EPF. The data were recorded at 30 and 90 days after

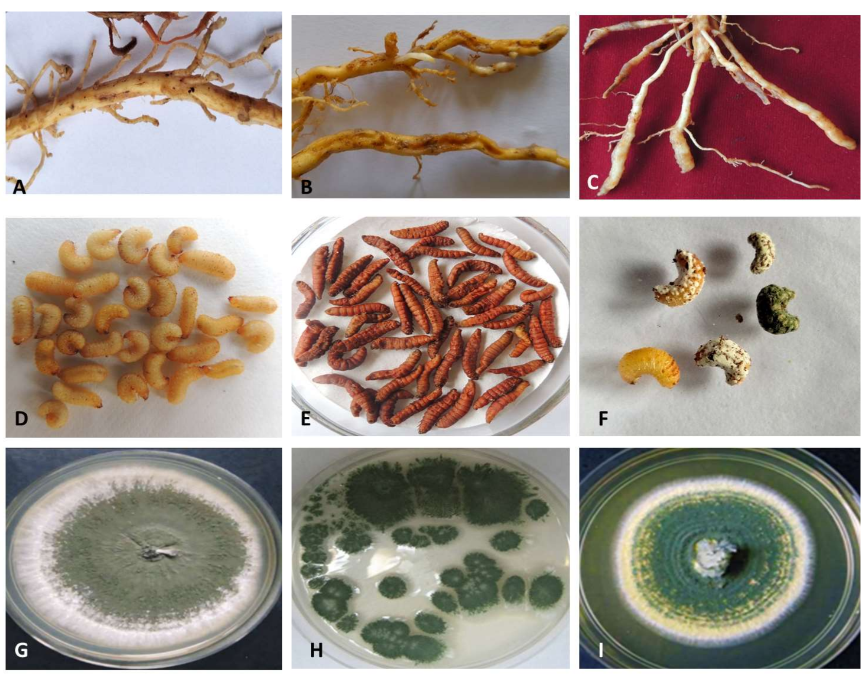

Fig. 1. A. Healthy cardamom root B. Root grub damage C. RKN damage D. Grub of root grub E. ICRI EPN - 18 strain F. Different growth stages of *M. anisopliae* on root grub G. *M. anisopliae* (ICRI MA RG -1) H. *M. anisopliae* (ICRI MA RG -2) I. *M. anisopliae* (ICRI MA RG -3)

treatment application from the randomly selected five plants of each plot. For *B. fulvicorne*, the population of grubs of *B. fulvicorne* counted in the soil in depth of 15cm^3 / plant (Varadarasan et al., 2011). For nematodes, number of galls (RKN) per 5g root were recorded and the initial nematode population per 200cc soil was recorded just before applying the treatments and 30 days and 90 days after treatment (Narayana et al., 2017) (Fig. 1). The percentage reduction (PR) of both root grub and nematodes over the untreated control was calculated as PR [(Control count - Treatment count/Control count) x 100] for each treatment following each application. Data were analysed using randomized block design in SPSS software version 16, and means were separated using Tukey's HSD test at $p \leq 0.05$.

RESULTS AND DISCUSSION

Population dynamics of *B. fulvicorne* and PPNs: Root grub, *B. fulvicorne* incidence remained negligible, with no grubs recorded from February to April 2022 and from March to April 2023 (Fig. 2). The highest population occurred in September 2022 and 2023, with 5.4 and 6.2 grubs, respectively and declining in the subsequent months. PPN populations fluctuated in a similar manner, and this showed that the prevailing weather conditions support their coexistence with the *B. fulvicorne* populations (Fig. 2). The lowest population density of PPN was recorded in February 2022 (12.4/200 cc) and January 2023 (12.6/200 cc). From may onwards, nematode populations increased steadily and peaked during the monsoon months. The highest PPN population was recorded in August 2023 (350.8/200 cc), with similarly high values in September 2022 (328.6) and other monsoon months (Fig. 3). Both *B. fulvicorne* and PPNs are more during rainy months due to adequate moisture in soil. Pearson correlation analysis showed a strong positive correlation between *B. fulvicorne* and PPN populations, exhibiting that both populations respond similarly to changes in environmental conditions. Both *B. fulvicorne* and PPN were also positively correlated with rainfall, the number of rainy days and relative humidity. These findings revealed that rainfall, humidity, and rainy days are the significant environmental factors that promote the populations of *B. fulvicorne* and PPN in the study area (Fig. 4). Similar trend was observed in the studies of Bora et al. (2023) and Dutta and Phani (2023) in large cardamom and other crops, where

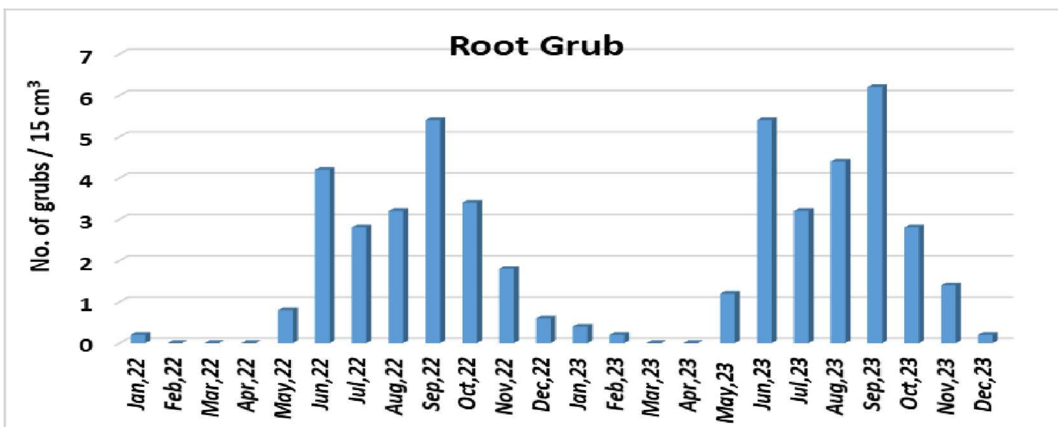

Fig. 2. Monthly variation in the incidence of *B. fulvicorne* on small cardamom during 2022-23

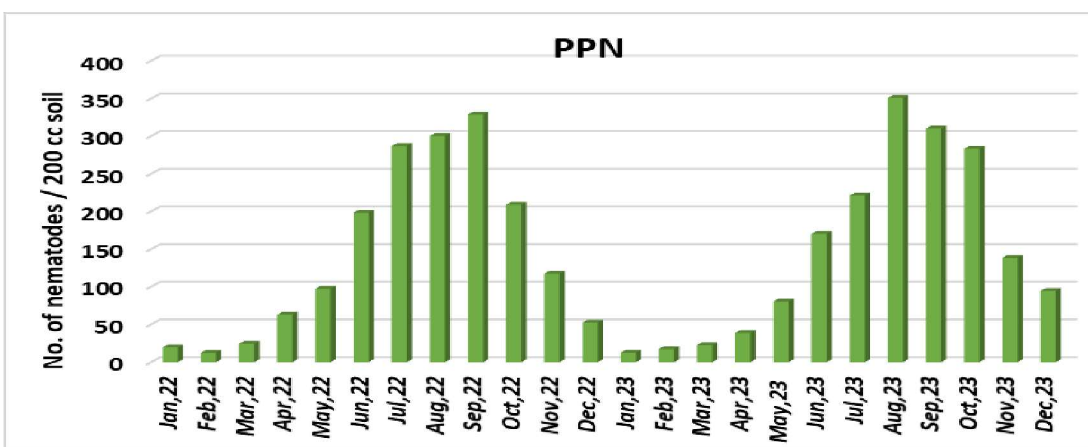

Fig. 3. Monthly variation in the incidence of PPNs on small cardamom during 2022-23

nematode abundance is closely linked to soil moisture, temperature, and rainfall.

The multiple regression analyses provided the linear equation for prediction of *B. fulvicorne* and PPN populations as $Y = -46.596+0.584(X_1)-0.502(X_2)+0.423(X_3)-0.004(X_4)+0.229(X_5)$ and $Y = -1094.874+18.516(X_1)-15.045(X_2)+9.150(X_3)+0.084(X_4)+9.548(X_5)$, respectively. The coefficient of determination (R^2) was recorded to be 47 per cent, and all the weather parameters together had a significant impact on *B. fulvicorne* and PPN populations, respectively. The seasonal fluctuations in *B. fulvicorne* and PPN populations in small cardamom appeared to be largely influenced by weather parameters and edaphic factors, both of which play a significant role in pest management and crop productivity (Varadarasan and Nagarajan 2014, Dutta and Phani 2023).

Field experiments: The pooled results from 2022–23 and 2023–24 showed that root grub populations increased in the untreated control but declined significantly across all treatments after application (Table 1). Among them, T7 consistently produced the highest suppression, *reducing B. fulvicorne* to near-zero levels after the second application, followed by T4. T1 also improved notably after the second application, while T3 alone resulted in only modest reductions. A similar pattern was observed for *M. javanica*. Nematode populations rose in the untreated control but were reduced most effectively by T7 (86.74% at 90 DAT), followed by T5 (Table 2). The ranking for gall reduction showed a clear superiority of T7 over all other treatments (Table 3). The enhanced performance of combined bioagents, particularly T7, indicates strong synergistic effects between EPNs and EPFs. Further, T7 also produced the highest capsule yield (900.90 kg/ha), substantially outperforming the untreated control. Varadarasan et al. (2011) observed 71-93%

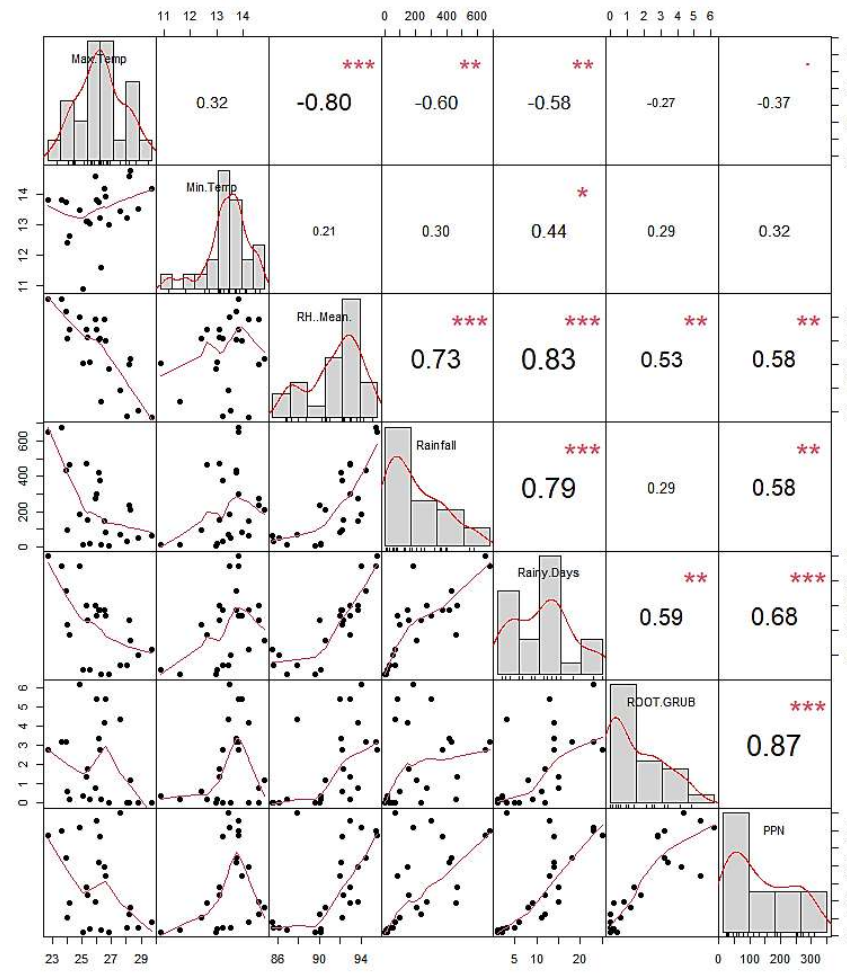

Fig. 4. The population of root grub and PPN in response to weather parameters.
(Graph's diagonal displays the distribution of weather parameters and RG & PPN population. On below, scatter plots with fitted lines. Above, correlation values and significance levels are provided; Each significance level is represented by p-values of 0.001 are denoted as "***", 0.01 as "**", 0.05 as "*" and 0.1 as ".", 1 as " ").

reduction of *B. fulvicorne* using EPN-infected *Galleria* cadavers, and Varadarasan and Nagarajan (2014) demonstrated that EPN-infected cadavers of *H. indica* were more effective than gel formulations in small cardamom. Similar observations by Pervez et al. (2016) highlighted *Heterorhabditis* sp. and *O. gingeri* as promising EPNs for

Table 1. Effect of EPN & EPF on incidence of *B. fulvicorne* in small cardamom

Treatments with Dosage / Plant	PTC	Mean grub population (15 cm^3 / Plant)					
		1 Application			2 Application		
		30 DAT	90 DAT	PR	30 DAT	90 DAT	PR
T_1 - EPN @ 4 lakhs IJs	3.07	2.66^{c}	0.73^{c}	76.08	0.60^{c}	0.13^{b}	92.00
T_2 - *M. anisopliae* @ 25g	3.13	2.73^{d}	1.06^{d}	65.21	0.66^{c}	0.20^{b}	88.00
T_3 - *P. lilacinum* @ 25g	3.20	3.73^{f}	2.26^{e}	26.08	4.80^{d}	1.20^{c}	28.00
T_4 - EPN @ 4 lakhs IJs + *M. anisopliae* @ 25g	3.00	1.80^{b}	0.46^{b}	84.78	0.33^{b}	0.06^{ab}	96.00
T_5 - EPN @ 4 lakhs IJs + *P. lilacinum* @ 25g	3.07	2.46^{e}	0.66^{c}	78.26	0.53b^{c}	0.13^{b}	92.00
T_6 - *M. anisopliae* @ 25g + *P. lilacinum* @ 25g	3.00	2.73^{d}	1.00^{d}	67.39	0.66^{c}	0.20^{b}	88.00
T_7 - EPN @ 4 lakhs IJs + *M. anisopliae* @ 25g + *P. lilacinum* @ 25g	3.20	1.60^{a}	0.26^{a}	91.30	0.06^{a}	0.00^{a}	100.00
T_8 - Untreated control	3.13	4.46^{g}	3.06^{f}	-	6.46^{e}	1.66^{d}	-

PTC: Pre-Treatment Count, DAT: Days After Treatment, PR: Percent Reduction Over Control
In column, means followed by common letters are not significantly different at (P=0.05) by DMRT

Table 2. Effect of EPN & EPF on population of PPNs in small cardamom

Treatments with Dosage / Plant	PTC	Number of nematodes/200g soil					
		1 Application			2 Application		
		30 DAT	90 DAT	PR	30 DAT	90 DAT	PR
T_1	175.30	152.00^{f}	212.30^{f}	30.15	212.30^{f}	75.30^{f}	37.56
T_2	176.00	171.60^{g}	242.60^{g}	20.17	243.60^{g}	87.00^{g}	27.90
T_3	174.60	129.00^{d}	145.60^{d}	52.08	102.00^{d}	34.60^{d}	71.27
T_4	175.00	138.30^{e}	188.30^{e}	38.04	191.60^{e}	72.00^{e}	40.33
T_5	174.30	112.60^{b}	119.60^{b}	60.63	84.00^{b}	28.30^{b}	76.51
T_6	175.60	124.00^{c}	143.00^{c}	52.96	98.60^{c}	31.00^{c}	74.30
T_7	175.00	109.30^{a}	104.30^{a}	65.67	80.00^{a}	16.00^{a}	86.74
T_8	174.00	201.60^{h}	304.00^{h}	-	325.30^{h}	120.60^{h}	-

See Table 1 for details

Table 3. Effect of EPN and EPF on incidence of root galls of *M. javanica* in small cardamom

Treatments with Dosage / Plant	PTC	Number of galls/5g root					
		1 Application			2 Application		
		30 DAT	90 DAT	PR	30 DAT	90 DAT	PR
T_1	14.33	11.33^{e}	17.66^{f}	27.39	20.33^{f}	14.00^{f}	31.14
T_2	14.66	13.66^{f}	19.33^{g}	20.54	23.00^{g}	15.66^{g}	22.95
T_3	14.00	9.33^{d}	11.66^{d}	52.05	10.66^{d}	5.66^{d}	72.13
T_4	14.00	11.33^{e}	16.00^{e}	34.24	19.00^{e}	13.00^{e}	36.06
T_5	14.33	8.00^{b}	10.33^{b}	57.53	8.33^{b}	5.00^{b}	75.40
T_6	14.66	9.00^{c}	11.00^{c}	54.79	8.66^{c}	5.33^{c}	73.77
T_7	14.00	7.66^{a}	8.66^{a}	64.38	7.00^{a}	3.00^{a}	85.24
T_8	14.00	15.00^{g}	24.33^{h}	-	28.66^{h}	20.33^{h}	-

See Table 1 for details

managing *B. fulvicorne*, while the pathogenicity of *M. anisopliae* against root grub was earlier confirmed under laboratory and field conditions (Sydhic 2005, Varadarasan et al., 2002). These results corroborate earlier findings of Narayana et al. (2017), where *P. lilacinum* and other bioagents showed high efficacy against root-knot nematodes in cardamom. Studies in other crops further support the present results that PPN suppression by *Steinernema rarum* and *H. bacteriophora*, while increased EPN application rates enhanced PPN and RKN, *M. incognita* suppression in tomato (Perez and Lewis 2002, Caccia et al., 2012, Khan et al., 2016). The present study has shown that two rounds application with combination of bio-control agents, EPN @ 4 lakhs IJs + *M. anisopliae* @ 25g + *P. lilacinum* @ 25g/plant was proved to be most effective against for reducing the population of both *B. fulvicorne* and *M. javanica* in cardamom soil besides encouraging microbial load in soil, which are also earlier reported by workers on small cardamom and other crops, that confirm the current investigation.

CONCLUSIONS

The current findings demonstrate that both root grub, *B. fulvicorne* and PPNs in small cardamom are closely associated with seasonal rainfall patterns, peaking during monsoon periods. Notably, the combination of bio-agents approach utilizing *Heterorhabditis indica* @ 4 lakhs IJs*, Metarhizium anisopliae* @ 25g, and *Purpureocillium lilacinum* @ 25g /plant offers excellent suppression of both pest's population and root gall formation. In light of these results, it is recommended that this eco-friendly pest management strategy be adopted in cardamom cultivation, with further trials conducted under diverse agro-climatic conditions to refine the protocols for maximum efficacy and farmer acceptance.

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AUTHOR'S CONTRIBUTION

All authors contributed significantly to the development of this work. P. Thiyagarajan contributed for population dynamics and supervised the research process. K. A. Saju and Arthra Ancy Joseph involved for characterization and multiplication of multiplication of *Metarhizium*. L. Gopianand performed the analysis the data and interpretation. A. B. Rema Shree edited, and approved the final version of the manuscript.

REFERENCES

Bora SS, Das D, Basumatary B, Ajay D, Deka TN, Bhutia SL and Rema Shree AB 2023. Report on association of plant parasitic nematodes in large cardamom (*Amomum subulatum* Roxb.) at Sikkim Himalaya region of India. *Journal of Spices & Aromatic Crops* **32**(1): 101-105.

Dutta TK and Phani V 2023. The pervasive impact of global climate change on plant-nematode interaction continuum. *Frontiers in plant science* **14**: 01-14.

Caccia M, Lax P and Doucet ME 2012. Effect of entomopathogenic nematodes on the plant-parasitic nematode *Nacobbus aberrans. Biology and Fertility of Soils* **49**: 105-109.

Giné A, López-Gómez M, Vela MD, Ornat C, Talavera M, Verdejo-Lucas S and Sorribas FJ 2014. Thermal requirements and population dynamics of root-knot nematodes on cucumber and yield losses under protected cultivation. *Plant pathology* **63**(6):1446-1453.

Grewal PS and Georgis R 1998. Entomopathogenic nematodes. In: *Methods in Biotechnology, Biopesticides: Use and Delivery*. In: Hall FR and Menn JJ (Eds.), Humana press, Totowa, New Jersey, pp 271-299.

ICRI 2008. *Management of cardamom root grub, Basilepta fulvicorne (Jacoby) with entomopathogenic nematodes*. Final report submitted to Department of Biotechnology, Ministry of Science & Technology, New Delhi, India.

Josephrajkumar A, Devi A, Murugan M and Vasanthakumar K 2005. Entomopathogenic nematodes - Mass production and application in cardamom root grub (Basilepta fulvicorne Jacopy) management. *Indian Journal of Arecanut, Spices & Medicinal Plants* **7**(2): 54-60.

Khan SA, Javed N, Kamran M, Abbas H, SAfdar A and Haq I 2016. Management of *Meloidogyne incognita* Race 1 through the use of entomopathogenic nematodes in tomato. *Pakistan Journal of Zoology* **48**(3): 763-768.

Mendiburu FD 2015. *Agricolae: Statistical Procedures for Agricultural Research*. R Package Version 1: 2–3. https://cran.r-project.org/package=agricolae.

Narayana R, Sheela MS and Thomas S 2017. Management of root-knot nematode *Meloidogyne javanica* infecting cardamom. *Indian Journal of Nematology* **47**(1): 60-64.

Perez EE and Lewis EE 2002. Use of entomopathogenic nematodes to suppress *Meloidogyne incognita* on greenhouse tomatoes. *Journal of Nematology* **34**(2): 171-174.

Pervez R, Eapen SJ, Devasahayam S and Jacob TK 2016. Eco-friendly management of cardamom root grub (*Basilepta fulvicorne* Jacoby) through entomopathogenic nematodes. *Indian Phytopathology* **69**(4s): 496-498.

Pervez R, Eapen SJ, Devasahayam S, Jacop TK, Ansar Ali A and Thiyagarajan P 2016. *Oscheius* spp. an alternative to *Heterorhabditis* spp. for eco-friendly management of cardamom root grub (*Basilepta fulvicorne* Jacoby). *Annals of Plant Protection Sciences* **24**(2): 385-391.

Praveena R, Biju CN and Eapen SJ 2013. Nematod pests affecting cardamom plants. *Spice India* April: 8-11.

Peterson BG, Carl P, Boudt K, Bennett R, Ulrich J, Zivot E and Wuertz D 2018 Package 'performanceanalytics'. *R Team Cooperation* **3**: 13-14.

Poinar GO 1990. Taxonomy and biology of *Steinernematidae* and *Heterorhabditidae*. In: Gaugler R and Kaya HK (Eds.) Entomopathogenic Nematodes in biological control, CRC Press, Boca Raton, Florida, pp 23-60.

Sydhic N 2005. *Studies on pathogenicity/ cross infectivity of biocontrol agents, Verticillium species and Metrhizium anisopliae on major pests of small cardamom (Elettaria cardamomum).* M.Sc. Thesis, Mahatma Gandhi University, Kerala.

Thiyagarajan P, Varna M, Ansar Ali MA and Rema Shree MA 2023. Development of low cost artificial diet for mass production of entomopathogenic nematode, *Heterorhabditis indica* a Strain ICRI EPN-18. *Plant Health Archives* **1**(2): 34-36.

Thomas SH and Goddard C 1986. Population dynamics of selected plant-parasitic nematode species on guayule. *Plant Disease* **70**(6): 579-580.

Varadarasan S, Hafitha NM, Sithara L, Balamurugan R, Chandrasekhar SS, Ansar Ali MA and Thomas J 2011. Entomopathogenic nematodes-science, technology and field outreach for biocontrol of cardamom root grub. *Journal of Plantation Crops* **39**(1): 86-91.

Varadarasan S, Ansar Ali MA, Chandrasekar SS, Suseela Bhai R and Gopakumar B 2002. Evaluation of entomogenous fungus *M. ansisopliae* (Metsch.) Sorokin on cardamom root grub, *B. fulvicorne* Jacoby under field condition. *National seminar on strategies for increasing production and export of spices*, October 24-26, 2002, Calicut, Kerala.

Varadarasan S and Nagarajan K 2014. Studies on suitable formulation of entomopathogenic nematode for the management of cardamom root grub, *Basilepta fulvicorne* (Jacoby). *Journal of Plantation Crops* (India) **42**(2): 262-264.

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