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Isolation and Identification of Phytonematodes of Leafy Vegetable Crops of Telangana, India

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Abstract: Phytonematodes are known to cause significant economic losses across a wide range of crops, including leafy vegetables. These crops are often severely infected by various phytonematode species. However, limited studies have been conducted on the diversity of phytonematodes affecting commonly cultivated leafy vegetables. Isolation and identification of these nematodes are essential steps toward developing targeted and effective pest management strategies. The present study was undertaken in two selected villages of Telangana, Godumakunta (Keesara Mandal) and Peerzadiguda (Uppal Mandal). Soil samples were collected from the rhizosphere of commonly grown leafy vegetables such as Spinach, Mentha, Amaranthus, and Coriander. The samples were processed using the Baermann funnel method to isolate nematodes, which were subsequently examined under a light microscope and identified using a nematology manual. The identified phytonematode species included *Psilenchus dunensis*, *Tylenchorhynchus brevilineatus*, *Belonolaimus longicaudatus*, *Hoplolaimus indicus*, *Peltamigratus indicus*, and *Aorolaimus perscitus*. These species were found in association with all the sampled leafy vegetables. This study provides valuable insights into the diversity of phytonematodes infecting major leafy vegetable crops in the region and serves as a foundational step toward the development of location-specific nematode management strategies.

Keywords: Phytonematodes, Leafy vegetable crops, Amaranthus, Mentha, Pest management, Diversity

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Introduction

Phytonematodes are an extremely important limiting factor in vegetable production, therefore crop protection is an integral part of food production and must be considered within the context of modern agriculture and sustainable development. Effective crop protection is essential both to combat the threat of wide spread diseases and to provide the effective pest management

programmes. The majority of the plant species, which account for the major world's food supply, is susceptible to attack from phytonematodes which are capable of causing sustainable economic losses in the quantity and quality of the crops (Jain *et al*., 2007; Berry *et al*., 2008). The crop losses caused by phytonematodes in economic terms estimated about $ 157 billion annually to the world agriculture (Abad *et al*., 2008). Yield losses due to root-knot nematodes (*Meloidogyn*e spp.) range from 35.0 to 39.7% (Reddy, 1985; Jonathan *et al*., 2001). In India the losses of agriculture by phytonematodes estimated at about Rs. 210 crore annually (Jain *et al*., 2007). Root-knot nematode (*Meloidogyne spp.)* is a serious pest of vegetables especially chilli (*Capsicum annuum*), which is being grown everywhere. Root-knot nematode, *Meloidogyne incognita* is one of the major limiting factors affecting the production of chilli in India (Nagnathan, 1984; Jain, 1992). Various plant parasitic nematodes like lesion nematode (*Pratylenchus spp.*), reniform nematode (*Rotylenchulus reniformis*), burrowing nematode (*Radopholus similis*), citrus nematode (*Tylenchulus semipenetrans*), rice root nematodes (*Hirschmanniella* spp.), cyst nematodes (*Heterodera* spp.) and root-knot nematodes (*Meloidogyne* spp.), attack a wide variety of major food crops (Khan *et al*., 2010). Based on various studies, it is evident that several species of phytonematodes can cause significant damage to a wide range of crops. Effective pest management begins with the accurate identification of the specific phytonematode species infecting the crops, followed by the implementation of a targeted control strategy. However, there have been limited studies focused on identifying phytonematodes affecting leafy vegetables commonly cultivated in different regions of Telangana, India. In this context, the present study aims to investigate the diversity of phyto-nematodes infecting leafy vegetables grown in Godumakunta village (Keesara Mandal) and Peerzadiguda village (Uppal Mandal), located near the Moosi River in Telangana, India.

Materials and Methods

To identify different species of phytonematodes, soil samples were collected from Godumakunta village (Keesara Mandal) and Peerzadiguda village (Uppal Mandal), and were processed. For this study, four different leafy vegetable crops were selected, viz. Amaranthus, Spinach, Coriander and Mentha. The soil sample is mixed thoroughly, but gently when tumbling, to homogenize the nematodes within the soil. The method which was followed was a modified Baermann tray or funnel. In this case, the required volume of soil is rinsed through a 864 µm sieve and over a 38 µm sieve. The captured material is rinsed into a coffee filter placed within a plastic bowl or funnel and supported by a screen. The water level is brought up to at least 1.0 cm above the coffee filter and allowed to incubate for 24 h. Following incubation, the filter and screen were removed from the bowl and the water left in the bowl or funnel base was poured over a 25 µm sieve. The material contains only live, mobile nematodes and can be observed under a light microscope.

Results and Discussion

After successful processing of the two samples, which were collected from Godumakunta village (Keesara Mandal) and Peerzadiguda village (Uppal Mandal) extraction of phytonematodes, they were stained and identified accordingly.

Nematodes found in selected leafy vegetable plant soils

In this work, we have identified several nematodes in the soil of leafy vegetable crops like Spinach, Mentha, Amaranthus and Coriander. Phytonematodes, such as *Psilenchus dunensis*, *Tylenchorhynchus brevilineatus*, *Belonolaimus longicaudatus, Hoplolaimus indicus, Peltamigratus indicus, and Aorolaimus perscitus* were found in all leafy vegetables grown soil. The details of these phytonematodes are presented in Figures 1-6.

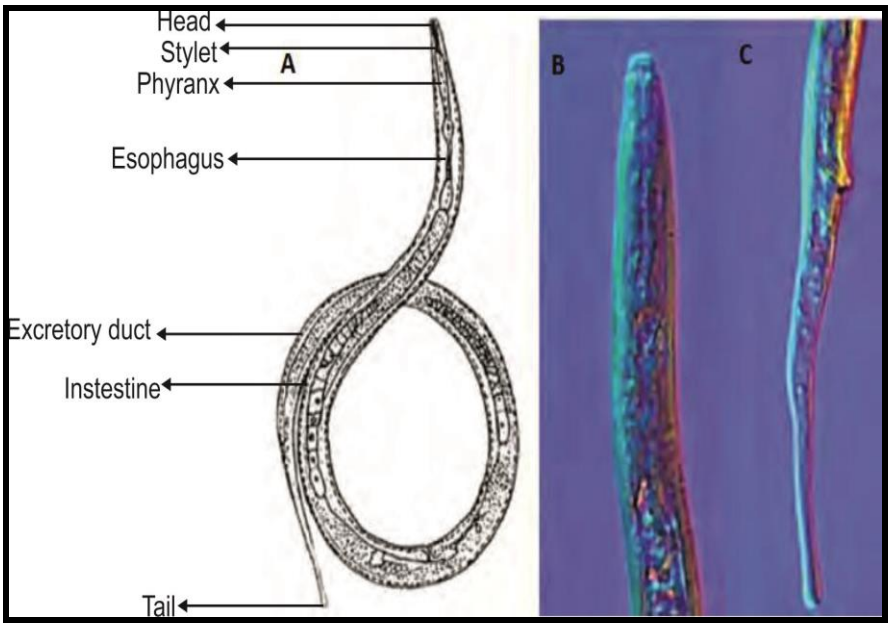

Fig. 1: *Psilenchus dunensis* whole body (A), head region (B), and male tail (C). 400x. (Kingdom –Animalia; Phylum-- Nematoda; Class-- Secernentea; Order-- Tylenchida; Family--Tylenchidae; Genus--*Psilenchus*; *Species*--*dunensis* de Man, 1921).

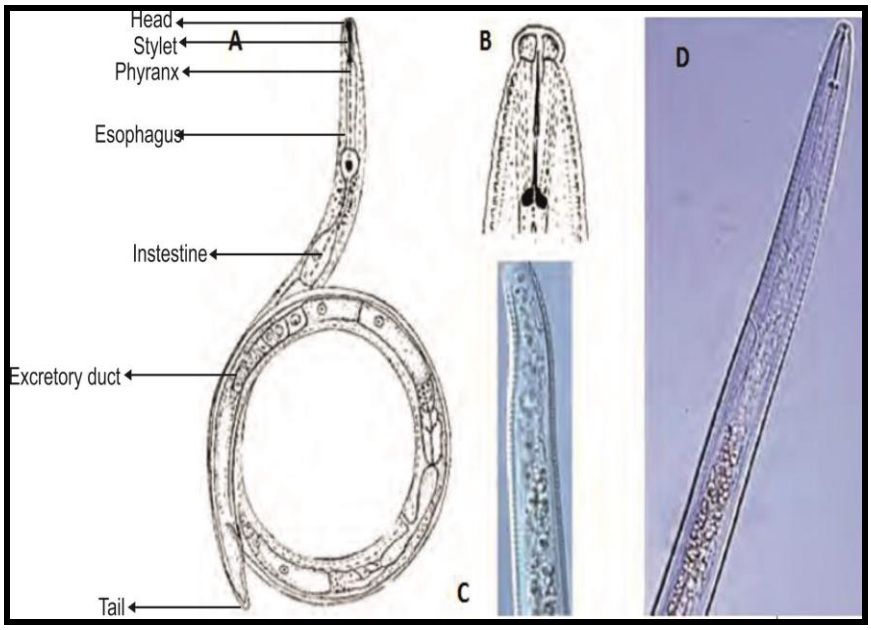

Fig. 2: *Tylenchorhynchus brevilineatus* whole body (A), short stylet (B), round tail tip (C), basal bulb not overlapping intestine (D). 400x. (Kingdom –Animalia; Phylum-- Nematoda; Class-- Secernentea; Order-- Tylenchida; Family-- Belonolaimidae; Genus-- *Tylenchorhynchus*; *Species*--*brevilineatus* Cobb,1913).

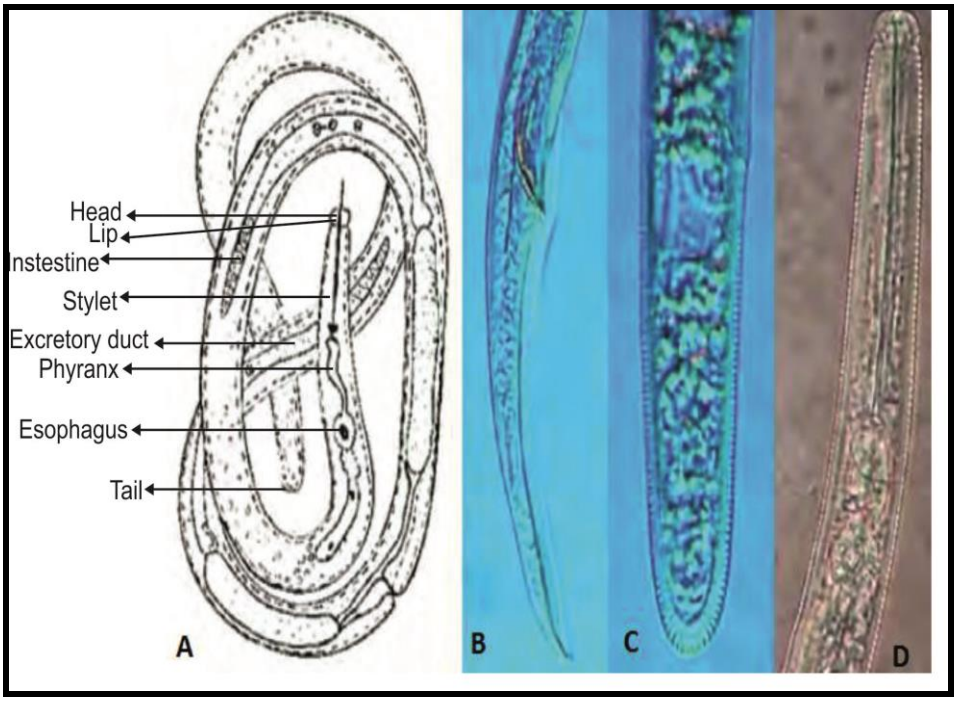

Fig. 3: *Belonolaimus longicaudatus* whole body length (A), male tail region (B), female tail region (C), and head region (D). 400x. (Kingdom –Animalia; Phylum-- Nematoda; Class-- Secernentea; Order-- Tylenchida; Family-- Belonolaimidae; Genus-- *Belonolaimus*; *Species--longicaudatus* Steiner, 1949).

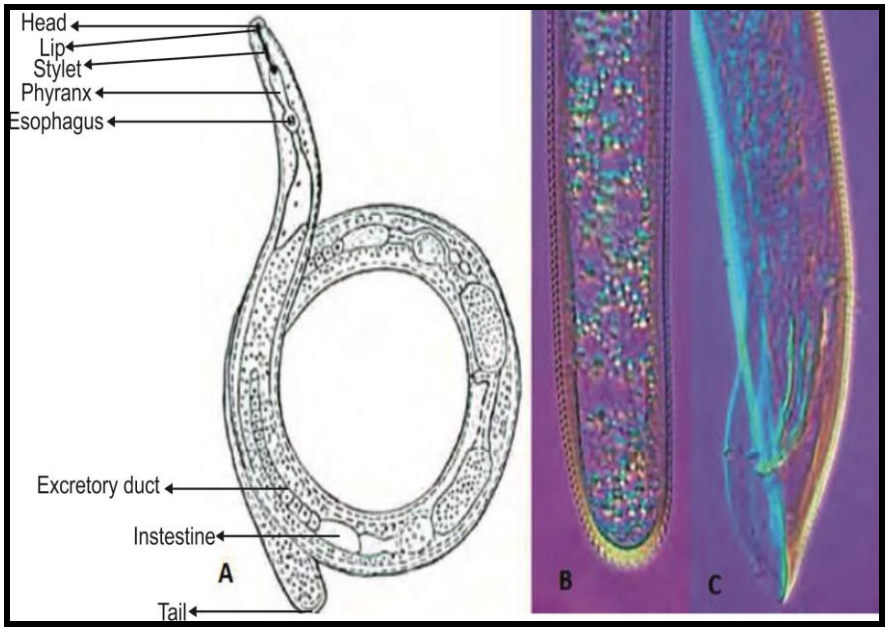

Fig. 4: *Hoplolaimus indicus* whole body (A), female tail (B), and male tail (C). 400x. [Kingdom –Animalia; Phylum-- Nematoda; Class-- Secernentea; Order-- Tylenchida; Family-- Hoplolaimidae; Genus-- *Hoplolaimus*; *Species-- indicus*; Binomial name-- *Hoplolaimusindicus*Sher, (1963)].

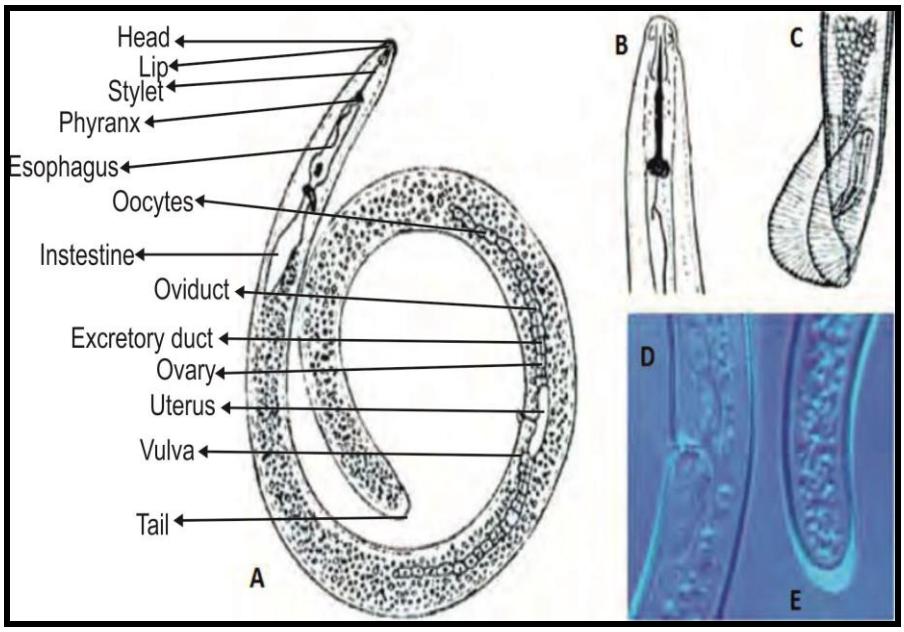

Fig. 5: *Peltamigratus indicus* female whole body with epitygma (A), stylet region (B), male tail (C), epiptygma (D), and female tail region (E). 400x. (Kingdom –Animalia; Phylum-- Nematoda; Class-- Secernentea; Order-- Tylenchida; Family—Hoplolaimidae; Genus-- *Peltamigratus*; *Species--indicus* Sher, 1963).

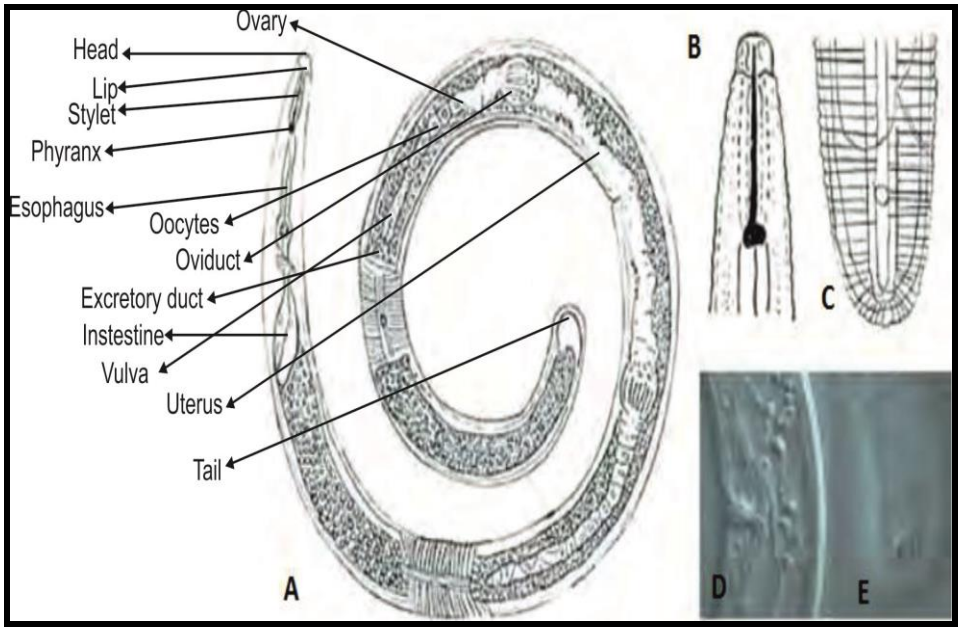

Fig. 6: *Aorolaimus perscitus* female body with phasmid midbody (A), stylet region of *Aorolaimus perscitus* and *Scutellonema* (B), tail region of *Scutellonema* with scutellum (C), vulva without epitygma (D), and scutellum near tail tip of *Scutellonema* (E). 400x. (Kingdom –Animalia; Phylum-- Nematoda; Class-- Secernentea; Order-- Tylenchida; Family-- Hoplolaimidae; Genus-- *Aorolaimus*; *Species—perscitus* Sher, 1963).

In the present study, different soil samples, from two different fields were collected. Vegetable crops, such as Spinach, Mentha, Coriander and Amaranthus were selected as test crops to study phytonematodes in Godumakunta Village and Peerzadiguda village of Telangana state. Among the selected leafy vegetable crops, Spinach grown soil showed high diversity, The results obtained from the current study revealed that the populations of *Psilenchus dunensis, Tylenchorhynchus brevilineatus, Belonolaimus longicaudatus, Hoplolaimus indicus, Peltamigratus indicus, and Aorolaimus perscitus* multiplied freely on Spinach, Mentha, Amaranthus and Coriander in both normal and deep ploughed fields, showing that these crops are a good host for these nematode species. Phytonematode pest management studies have shown that when organic soil amendments were added, they were known to alter the soil environment with decomposable organic matter, which is considered as one of the preferred methods of biological control of plant diseases. The materials used for soil amendment includes dry or green crop residues, agricultural wastes, oil cakes and other organic wastes. These materials were allowed to decompose in the field itself where the target pathogens are supposed to be present. However, the efficacy of an organic amendment against plant parasitic nematodes depends on many different factors including the nematode species present (McSorley and Gallaher, 1996), the chemical composition of the amendment (Mojtahedi *et al*., 1993), including its C:N ratio (Ritzinger and McSorley, 1998), the length of time since application (McSorley and Gallaher, 1996), the other organisms present in the soil environment including those which feed on nematodes (Stirling, 1991) as well as other environmental factors and agricultural practices. However, organic materials with C: N ratios less than 20:1 have higher degradation rates and often nematicidal activities (McSorley and Gallaher, 1995). The compost amendments and the fertilizer applications used, can increase soil organic matter, pH and also soil nutrient levels (McSorley and Gallaher, 1997). These amendments may contribute to the reduction of phytonematode population, thereby contributing to the economic growth.

Ethical Statement

The authors confirm that all ethical issues have been dealt with the research ethics guidelines provided by the Department of Zoology, Nizam College (Affiliated to Osmania University), Opp. LB Stadium, Basheer Bagh Campus, Gunfoundry Hyderabad, Telangana, India.

Author Contributions

Each author has contributed equally to the study's planning, analysis, writing, and editing. All authors have read and agreed to the published version of the manuscript.

Conflict of Interest

The authors declare no conflicts of interest.

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