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First report of root-knot nematode, *Meloidogyne incognita* infesting king chilli (U-morok) from Manipur, India

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Abstract

An extensive survey was carried out in all the blocks of Churachandpur district of Manipur in 2021 for root-knot nematodes, *Meloidogyne* spp. infesting high valuable crops of Manipur. During the survey, it was observed for the first time that the king chilli (U-Morok) were infested by *Meloidogyne* spp. showing the general symptom of patchiness, yellowing of leaves and wilting of infested plant as well as the typical symptom of gall formation with the presence of egg masses attached to it. Morphological studies in laboratory reveal that, the heavily infested king chilli plant sample collected from farmer's polyhouse of Sagang village, Churachandpur district, Manipur was by the root-knot nematode, *M. incognita*.

Keywords: *Meloidogyne incognita*, root-knot nematode, king chilli, Manipur

Introduction

King chilli locally known as U-morok in Manipur is a high valuable cash crop in the state belonging to the genus *Capsicum* of Solanaceae family. It is an interspecific hybrid chilli pepper of *Capsicum chinense* and *Capsicum frutescens* (Bosland, and Baral, 2007) [5] cultivated in Northeast India. It is also known as ghost pepper, ghost chilli, naga jolokia and bhoot jolokia ranking among the one of the hottest chilli in the world. In Manipur, it is consumed in the form of both fresh or dried and used for its distinct flavour and aroma in various culinary preparations. Due to its distinct taste and pungency, people of Manipur commonly use this chilli for making pickles as well as adding hotness to non-vegetarian food stuff. In recent years, this crop is gaining importance because of its high content of capsaicin (3-5%) as compared to other Indian chilli. King chilli or U-morok is used as spice, food and medicine. It can be used as a relief for asthma patients at low quantities. Regular consumption in small quantities has been known to treat gastro-intestinal abnormalities. The capsaicin, the active principle which accounts for the pharmaceutical properties of chillies is useful as a counter-irritant, anti-arthritis, analgesic and anti-cancer agent. Capsaicin has also received increased attention for its anti-obesity effect. 600 ha of area under King chilli has been fully certified under standards laid down and administered in India by APEDA. The local varieties grown are highly pungent with a unique flavour and aroma and are also used in hot sauces. This chilli variety is available from the month of June till November in Manipur (Anonymous, 2022) [1].

Plant parasitic nematodes belonging to the family Meloidogynidae (Order: Tylenchida) are obligate endoparasite widely distributed in both tropical and temperate climate. They feed on roots of plants and form galls on the roots. The infective second stage juvenile penetrate into the roots at the zone of elongation and establish feeding sites in the phloem causing disruption, hypertrophy and hyperplasia of cells resulting in formation of giant cells and swellings at the sites of establishment due to which the nutrient uptake of the root is hampered; crop become stunted in growth. Berkeley (1855) [3] first reported that root-knot nematode, *Meloidogyne* species was responsible for causing injury in cucumber plant grown inside the glasshouse in England. In India, Barber (1901) [2] first reported root-knot nematode, *Meloidogyne* spp. on tea from Kerela, which was also the first ever report on occurrence of a plant parasitic nematodes in India. In India, yield losses due to *Meloidogyne* spp. in vegetable crops such as okra, tomato and brinjal were estimated to be 91 percent, 42 to 54 percent and 18 percent, respectively (Bhatti and Jain, 1977; Subramanian *et al.*, 1990) [4, 11].

Heavily infested king chilli by root-knot nematodes has been observed in the farmer's polyhouse of Sagang village under Churachandpur district, Manipur.

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The above ground symptoms shows that of abiotic stress which is similar to general symptom infested by nematode like yellowing of leaves, shedding of leaves, patches, stunted plant growth, wilting and poor fruit size. The infested root shows the typical symptom of galling when uprooted and upon washing shows the egg masses attached to the galls developed on the roots leading to poor root system (Fig1).

Materials and Method

The egg masses (clearly seen by naked eyes) attached to the infested root system were collected with the help of a forceps and were kept for hatching at room temperature in water for 24 hours for study of morphology in second stage juveniles (J_2) of *Meloidogyne* spp. Further, the entire knotted root system was cut into a small pieces and stained by NaOCl-Acid Fuchsin method (Byrd *et al.*, 1983) [6] for species identification using perineal pattern. Stained female nematode was dissected from the galls of the root with the help of a fine needle and blade. Ten mature females were taken for preparation of perineal patterns. The posterior half of the female body was cut on 45 percent lactic acid on the transparent glass slide and cleared away the body contents. The posterior cuticle was then transferred to a drop of dehydrated glycerol on a glass slide where, it was trimmed to a size slightly greater than the pattern. The cuticular pattern was finally mounted in a drop of anhydrous glycerine on a clean slide with cover slip of 18 mm (No.1) keeping their outer surface uppermost and then, sealed with nail varnishes. The detail studies for identification of the species were made by comparing characteristics observed in the perineal region and size and shape of root-knot nematode with description given by Singh and Gupta in 2011 [10].

After hatching, nematodes were killed, fixed (8 percent formalin) and stored in Mc Cartney bottle with proper label for further processing. Clearing of nematodes (Scienhorst's

slow method, Scienhorst, 1962) followed by mounting and sealing of nematodes for permanent slides to study the morphology of J_2 *Meloidogyne* spp.

Results and Discussions

Morphology of root-knot nematode, *Meloidogyne incognita* collected from King chilli in Manipur

Mature female (n=10): Body pearly white, globular to pear shaped, with projecting neck. Cuticle translucent and glistering. Annulations visible in neck and vulva-anus region. Lip set-off. Stylet slender with distinct rounded knobs. Esophagus well developed with large cylindrical procorpus and rounded metacarpus. Excretory pore just behind the level of stylet knobs. Perineal pattern wavy dorsally. Dorsal arch high and squarish, distinct lateral lines absent. Vulva, anus and phasmid present in perineum (Fig3.). Hence, the sample collected from King chilli (U-morok) was found to be *M. incognita*. Similar finding were also observed by Chitwood (1949) [7] and considered the perineal pattern as the most striking and distinguishing character for identification of root-knot nematode species. Kaur and Attri (2013) [8] also opined that perineal pattern is the most stable characters of root-knot nematodes.

Second stage juvenile, J_2 (n=15): Body slender, lip region continuous with the body contour. Cephalic framework well developed. Stylet slender with distinct round small basal knobs. Metacarpus distinct with basal bulb extension overlapped ventrally, genital primordia developed, rectum and anus clearly visible. Tail with hyaline terminus conoid shaped. L = 0.360-0.440 mm (0.410±0.234), Stylet Length = 10.0-12.0 μ m (10.8±0.7), Lip height = 2.0 μ m, Lip width = 4.0-5.0 μ m (4.2±0.04), a = 26.07-32.53 (30.71±1.61), b = 3.95-5.61 (4.69±0.42), b' = 3.08-3.79 (3.44±0.18), c = 8.29-9.84 (9.28±0.47), c' = 3.75-4.50 (4.10±0.22) and MB = 42.18-53.09 (46.99±4.79) (Fig3.)



a) General symptom in farmer's polyhouse (Yellowing of leaves, poor fruit size, stunted growth, wilting)



b) Typical symptom of RKN (Galling of roots)



c) Egg masses attached to galls on infested root

Fig 1: Above and below ground symptoms observed under farmer's polyhouse

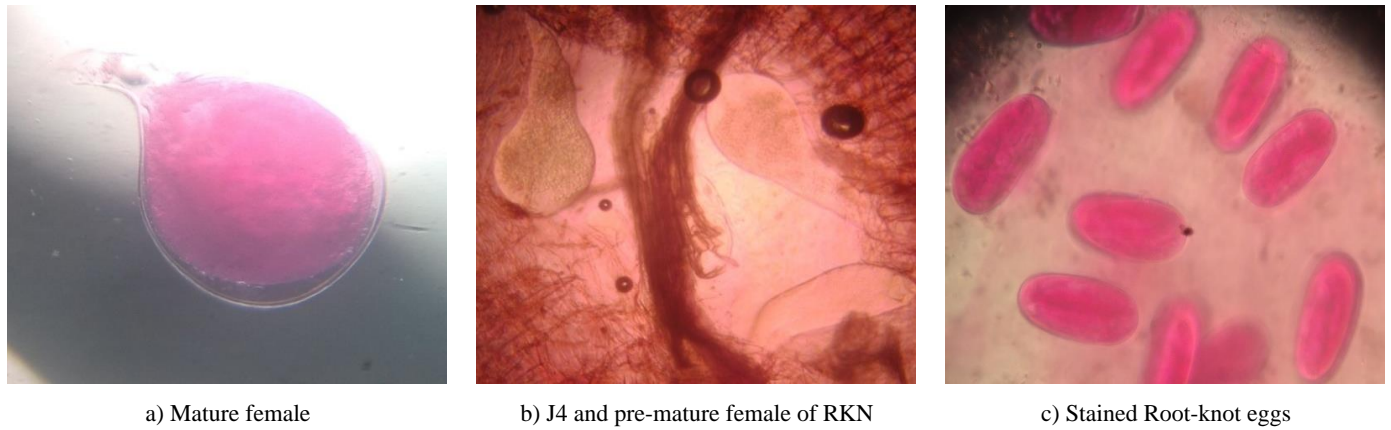


Fig 2: Root-knot nematode observed under microscope

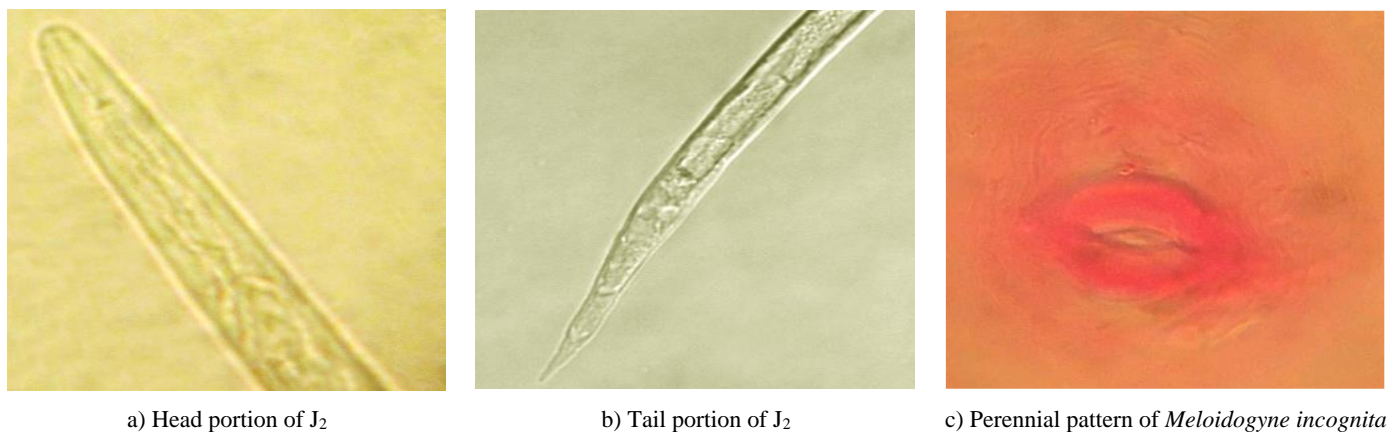


Fig 3: Morphology of root-knot nematode, *Meloidogyne incognita* under microscope

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