MANAGEMENT OF ROOT-KNOT NEMATODE *MELOIDOGYNE INCognITA* OF TOMATO PLANT WITH SOME MEDICINAL PLANT EXTRACTS

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ABSTRACT
Controlling of root-knot nematode *Meloidogyne incognita* (Kofoid and White) Chitwood is a growing concern for vegetable producers, because of the cost and demerits of the chemical nematicides. An alternative way to chemical control of *Meloidogyne incognita* on tomato plant was evaluated by using four plant extracts including leaf extracts of *Hibiscus mutabilis*, *Syzygium aromaticum*, seeds extract of *Albizia lebbeck* and extract of fruit of *Terminalia chebula*. *Albizia lebbeck* and *Hibiscus mutabilis* extracts applied by soil drench, reduced nematode infestation and promoted plant growth. These two extracts have significantly reduced *Meloidogyne incognita* infestation. The extracts have decreased root galling, nematode population, root protein content and consequently increasing plant biomass. Further, *Syzygium aromaticum* was found to significantly reduce root weight, nematode population in soil and root protein content, whereas *Terminalia chebula* was found to have no effect in controlling root-knot nematode.

Keywords: Biopesticide, *Meloidogyne incognita*, plant extracts, Tomato plant

INTRODUCTION
Plant parasitic nematodes are serious threats to agricultural food and fiber crops, causing an estimated loss of US $125 billion per year worldwide (Chitwood, 2003). Among all the nematodes, the root-knot nematodes, *Meloidogyne* spp., are the most damaging as they not only cause significant loss in yield but also deteriorate the product quality and fetch less value. The impact of this nematode genus is enhanced by its wide host range of more than 5,000 plant species (Ntalli et al., 2010). Among all *Meloidogyne* spp., *Meloidogyne incognita* (Kofoid and White) Chitwood is the most common species of root-knot nematodes and infect almost all cultivated plants, which makes it perhaps the most damaging of all pathogens (Sasser and Freckman, 1987). It has been estimated that global losses of $78 billion is due to *Meloidogyne incognita* infection (Chen et al., 2004). Davis and May (2005) reported that the yield loss of cotton production caused by *Meloidogyne incognita* in 2002 was estimated to be between 18.0 to 47.3%. Nematodes are difficult to control because of their wide host range and high rate of reproduction, with female capable of producing up to thousand eggs (Natarajan et al., 2006). The management tactics used against root-knot nematodes includes soil fumigation, soil pasteurization, rotation with non-host crops and use of resistant cultivars. Extensive survey of vegetable fields of tomato and okra in western Uttar Pradesh revealed that over and indiscriminate use of nematicides for control of root-knot nematodes caused damage to soil, plant and human health (Goswami et al., 2012). As general awareness of the harmful effects of chemical pesticides increases and so also public attitudes towards environmental pollution, chemical nematicides are losing their popularity among farmers (Elbadri et al., 2008). However, due to environmental concerns about chemical nematicides and the limited number of available resistant cultivars, alternative methods, such as biopesticides

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obtained mainly of plant origin, are being exploited (Echeverrigaray et al., 2010; Harish et al., 2008). Biopesticides have shown great promise in the sense that they are very often non-phytotoxic, non pollutant, easily biodegradable and do not leave toxic residue in the edible plant parts. Nematicidal phytochemicals are generally safe for the environment (Chitwood, 2002). Many plant species have been reported to have nematicidal properties but the active principles have been identified in only a few. They belong to different families of herbs, shrubs and trees. The effective parts are roots, barks, leaves, fruits or seeds. A review by Sukul (1994) on nematicidal plants included a list of 176 of such plants. The potential of using plant extracts in controlling plant parasitic nematodes has been documented by several authors (Dos et al., 2003; Pavaraj et al., 2012). But the effects of many plants are yet to be investigated for their nematicidal properties. In my earlier studies, I have observed strong nematicidal potential of plant extracts of some medicinal plant (Nandi, 2016; Nandi, 2017). Hence, the present investigation has been conducted to evaluate the in vivo controlling activity of some of these plant extracts on root-knot nematode in tomato plant system.

MATERIALS AND METHODS

Preparation of plant extracts: Leaf extracts of Hibiscus mutabilis, Syzygium aromaticum, seeds extract of Albizia lebbeck and fruit extract of Terminalia chebula were tested for their controlling activity of root-knot nematode of tomato plant. Plant materials were collected from different part of Malda District of West Bengal. Plant extract was prepared by following the methodology of Ferris and Zheng (1999). The collected materials were thoroughly washed in tap water, air dried and ground and soaked in distilled water for 24 hours. Standard extract (10% w/v) was prepared by soaking 10 g of powder in 100 ml of distilled water. The extract was passed through a muslin cloth, filtered through Whatman No.1 filter paper. This standard stock solution was diluted 50% by using distilled water and used for pot test experiment.

Planting material: The seeds of tomato (Lycopersicon lycopersicum L. Pusa Ruby) were procured from the local market. The seeds were surface sterilized with 70 % ethanol (1 min.) followed by 0.1 % HgCl₂ (2 min.). Seeds were then washed thoroughly with sterile distilled water for several times and shown into seedling raising pot, filled with autoclaved clay soil and composted manure (2:1 v/v). After four weeks, seedlings were used for pot test.

Pot test: Aseptically germinated seeds of tomato (Lycopersicon lycopersicum L. Pusa Ruby) were sown one seed/pot (32 cm diameter) containing a mixture of clay soil and composted manure (2:1 v/v). The soil filled pots were treated previously with boiling water to dematatised the soil. The pots were divided into seven groups, each of ten pots. The groups were marked as un-inoculated untreated, inoculated untreated, inoculated and treated with Hibiscus mutabilis leaf extract, inoculated and treated with Syzygium aromaticum leaf extract, inoculated and treated with Albizia lebbeck seed extract and inoculated and treated with Terminalia chebula fruit extract. When the plants were at 6-leaf stage, the pots marked as inoculated untreated group and all the inoculated and plant extracts treated groups were inoculated with Meloidogyne incognita juveniles (J₂) at the rate of 2000 ± 100 freshly hatched juveniles/plant. Aqueous plant extracts were applied by soil drench 2 days after inoculation. Treatment was repeated 4 days after the first treatment. The plants were regularly watered and the experiment was conducted outdoor at ambient atmospheric temperature 28 ± 3°C and humidity 60 ± 5 %. All the plants were uprooted 45 days after inoculation and their shoot length, shoot weight, root length and root weight were measured. The root galls on each plant were counted and the nematode population in 2 g root and 200 g soil was estimated by the modified Baerman method (Baermann, 1917). Three samples of roots from
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each group of plants were taken at random and the total protein concentration in each sample was estimated by Lowry’s method (Lowry et al., 1951). Data were analyzed by ANOVA, followed by Duncan’s new multiple range test to compare means (Duncan, 1955).

RESULTS

The results of the pot test are presented in Table I. Albizia lebbeck and Hibiscus mutabilis extracts increased plant growth significantly in terms of shoot weight and root length as compared to those in the inoculated untreated plants. Root weight significantly decreased in Albizia lebbeck, Hibiscus mutabilis and Syzygium aromaticum extracts treated plants. Root gall number and nematode (J₂) population in root and soil significantly reduced in Albizia lebbeck and Hibiscus mutabilis extract treated plants as compared to the inoculated untreated plants. Root protein content significantly decreased in Albizia lebbeck, Hibiscus mutabilis and Syzygium aromaticum extracts treated plants. Syzygium aromaticum reduced J₂ in soil but not effective in reducing root gall number and J₂ in root significantly. Syzygium aromaticum extract is not also effective in increasing plant growth. Terminalia chebula extract is only effective in reducing J₂ in soil significantly but it has no significant effect in increasing plant growth and also reducing nematode infection in terms of root gall number, nematode population in root and root protein content.

DISCUSSION

The nematicidal activity of different medicinal plant and herb extracts against Meloidogyne spp. has been demonstrated by

### Table 1. Increase in growth and decrease of Meloidogyne incognita infestation in tomato following treatment by plant extracts.

<table>
<thead>
<tr>
<th>Treatments*</th>
<th>Shoot length (cm)</th>
<th>Shoot weight (gm)</th>
<th>Root length (cm)</th>
<th>Root weight (gm)</th>
<th>Root gall no.</th>
<th>J₂/2gm root</th>
<th>J₂/200gm soil</th>
<th>Root protein (mg/gm)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Uninoculated untreated</td>
<td>65.85 ± 2.48 b</td>
<td>73.14 ± 4.91 b</td>
<td>40.28 ± 4.69 b</td>
<td>21.57 ± 3.18 a</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>3.36 ± 0.47 a</td>
</tr>
<tr>
<td>Inoculated untreated</td>
<td>52.42 ± 2.4 a</td>
<td>59.57 ± 4.58 a</td>
<td>26.71 ± 2.93 a</td>
<td>40.85 ± 4.02 b</td>
<td>337.85 ± 23.08 c</td>
<td>357.33 ± 30.47 c</td>
<td>385.33 ± 10.61 c</td>
<td>5.93 ± 0.58 c</td>
</tr>
<tr>
<td>Inoculated and treated with A. lebbeck extract</td>
<td>60.85 ± 2.74 ab</td>
<td>73.85 ± 5.19 b</td>
<td>37.0 ± 4.77 b</td>
<td>28.0 ± 4.3 a</td>
<td>261.14 ± 21.94 ab</td>
<td>241.33 ± 10.98 a</td>
<td>250.0 ± 12.56 a</td>
<td>4.80 ± 0.21 b</td>
</tr>
<tr>
<td>Inoculated and treated with H. mutabilis extract</td>
<td>59.71 ± 3.12 ab</td>
<td>75.28 ± 4.53 b</td>
<td>38.71 ± 3.5 b</td>
<td>24.42 ± 3.56 a</td>
<td>215.14 ± 16.65 a</td>
<td>233.66 ± 15.68 a</td>
<td>279.0 ± 14.19 a</td>
<td>4.20 ± 0.25 b</td>
</tr>
<tr>
<td>Inoculated and treated with S. aromaticum extract</td>
<td>55.28 ± 3.97 ab</td>
<td>65.85 ± 4.98 ab</td>
<td>33.71 ± 3.58 ab</td>
<td>27.85 ± 3.11 a</td>
<td>285.71 ± 12.81 bc</td>
<td>304.66 ± 22.63 bc</td>
<td>271.33 ± 17.68 a</td>
<td>4.90 ± 0.36 b</td>
</tr>
<tr>
<td>Inoculated and treated with T. chebula extract</td>
<td>53.28 ± 5.57 a</td>
<td>61.0 ± 5.24 ab</td>
<td>31.57 ± 3.07 ab</td>
<td>30.14 ± 4.24 ab</td>
<td>294.85 ± 22.32 bc</td>
<td>328.33 ± 11.36 bc</td>
<td>324.66 ± 19.30 b</td>
<td>5.13 ± 0.35 bc</td>
</tr>
</tbody>
</table>

*Values are given in mean ± SE. Each mean consists of ten replicates. Means in each column followed by the same letters are not significantly different at p = 0.05, according to Duncan’s New Multiple Range Test.
different workers (Wiranto et al., 2009; Ntalli and Caboni, 2012). Extracts of many plants with anti-helminthic and antimicrobial properties have been proven effective in controlling plant parasitic nematodes (Ferris and Zheng, 1999). Many phytochemicals having nematicidal potential have been isolated from various plant parts, usually by aqueous or organic extraction methods (Ferraz and De Freitas, 2004). The suppressive effect of phytochemicals on nematode populations has been well documented in several pathosystems (Chitwood, 2002). Evidently, Albizia lebbeck and Hibiscus mutabilis extracts could significantly ameliorate root-knot disease and improve plant growth in terms of shoot weight and root length. Root protein content in these treated groups also reduced significantly with respect to inoculated untreated group. This result correlates with the observation of Sinha Babu and Sukul (1983). Syzygium aromaticum significantly reduced root weight, nematode population in soil and root protein content. It has no significant effect in plant growth in terms of increasing shoot length, shoot weight and root length. This plant extract is also unable to reduce root-knot nematode infection in terms of root gall numbers and nematode population in root. Terminalia chebula has no effect in controlling root-knot nematode. The inoculated untreated plants recorded the highest weight of root as compared to the treated plants. This could be due to the development of more galls which brought about biochemical and physiological changes in the roots, which caused reduced root efficiency for water and nutrient absorption (Umar et al., 2010). The inoculated untreated plants recorded the highest number of nematodes recovered from root and soil as compared with the treated groups. This was because of more infection and reproduction of nematodes in inoculated untreated plants. Albizia lebbeck and Hibiscus mutabilis extracts treated groups recorded fewer numbers of juveniles in root and soil and this was attributed to the efficacy of these plant extracts which prevented nematodes from attacking tomato plants in these treated groups, thus preventing their reproduction within the roots.

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CONFLICT OF INTEREST
Declared none by the authors.

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