Original Research Article

Bionematicidal Potential of Some Incorporating Plants on Meloidogyne javanica Control on Tomato

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ABSTRACT

Experiments carried out to estimate the potential of incorporating garlic (Allium sativum), onion (Allium cepa) and Lemon grass (Cymbopogon citratus) on root-knot nematode Meloidogyne javanica affecting tomato under glasshouse conditions and concentration at 1, 2 and 3% of each plant used. Plants un-inoculated by nematode keep as control in four replicates. Results elucidate that nematode population and tomato plant growth parameters affected by using the different plants. Observation of root system revealed that number of egg-masses, galls and females/root system affected markedly. The garlic at 3% give the highest reduction in nematodes parameters followed by onion 3% while the least one was Lemon grass at 1% comparing to untreated infected plants. Fresh root weight and shoot fresh and dry weights raise compared to control plants. Therefore, using these plants may provide a prime and importance method in the integrated nematode management (INM) practices as environmental safety and economical as anew alternative to originally chemical nematicides.

Keywords
Tomato, Meloidogyne javanica, Root-knot nematodes, Control

Introduction

In Egypt, Tomato (Lycopersicon esculentum L.) is a vegetable crop member of the family Solanaceae grown in wide areas in open fields or greenhouses all over the year according to climate conditions. Different soil biotic pathogens attacks tomato with different severity according to the cultivar, soil type, environmental condition in the present of the causal organism. Plant parasitic nematodes (PPN) put themselves as aninfamous destructive member of tomato plant pathogens worldwide. The estimated annual losses in tomato due to PPN in Egypt reached 12% in 2014 [1]. Root-knot nematodes, Meloidogyne consider one of the limiting factors for profitable tomato plantation [2, 3]. Infected plants appear yellowish, stunt developed to wilting and root galling. Those, uptake of water and nutrient elements affected which refer to decrease quality and productivity of tomato. Damage varying according to the cultivar, nematode species, soil type, level of soil infestation and environment [4]. As a global problem, the solution will be by any method, e.g. physical, chemical or biological control methods. Farmers usually using chemical nematicides as the maximum effective method rather than other approaches in controlling and for limiting the damage of PPN. Unfortunately, Indiscriminate use of this
nematicides sometimes results in different dangerous ecological effects [5]. Awareness of nematologists about the harmful effects of nematicides and environmental consideration try to shift from the conventional use of chemicals to eco-friendly alternative control strategies [6]. Using of bionematicides from plant origin is from the most promising alternatives because they degrade to non-toxic products, less harmful to non-target organisms and environment friendly [7]. Plants from different families reported to showed nematicidal effect on plant parasitic nematodes [8]. Soil amendments with organic amendments using botanical materials not only safe but also improve soil structure, fertility and control of soil borne pathogens [9]. Moreover it reported to be effective in suppress PPN on different crops. Nematicidal effect of garlic, onion and Lemon grass reported against plant parasitic nematode in banana, eggplant and sugar beet [10, 11]. Therefore, the objective of this study was to evaluate the bionematicidal effect of some incorporating botanicals on root-knot *M. javanica* control in tomato plants.

**Materials and Methods**

**Collection and preparation of used plants.**

Garlic (*Allium sativum*), onion (*Allium cepa*) and Lemon grass (*Cymbopogon citratus*) plants collected from the research farm, Faculty of Agriculture, Menoufia University, Egypt.

**Multiplication and preparation of root-knot nematodes inoculum**

*Meloidogyne javanica* were isolated and identification done according to [12] using perineal pattern technique. Single egg masses was used to inoculate black nightshade (*Solanum nigrum*) seedlings transplanting in plastic pots 30 cm in diameter filled with sterilized clay–sand soil (1:2 v/v) and the pots maintained at 25 ± 2°C in the experimental glasshouse, Department of Agricultural Botany, Faculty of Agriculture, Menoufia University, Egypt. Two months after black nightshade infested with *M. incognita*. Plants uprooted and galled infested roots washed carefully with tap water. Roots cutting to short pieces then macerated for two periods of 10 seconds using a blender. Then macerated root placed in a bottle containing 0.5 % of sodium hypochlorite (NaOCl) [13]. Bottle vigorously shaken for 3 minutes to increase lysis gelatin matrix of egg masses consequence eggs releasing from the egg matrix. Solution then poured through sieves to remove the root tissue. Eggs collected on the 20μm sieve then washed several times with tap water to remove residual NaOCl. Eggs then transferred to a beaker containing tap water. Number of eggs/ml was counting under a stereomicroscope.

**Glasshouse experiment**

Complete fresh whole plants of garlic and onion and leaves of lemon grass mashed for small pieces and add at 1, 2 and 3% to 30cm dim pots filled with sterilized mixture of sand and clay soil (2:1, v/v). Three days later, one seedling tomato cv. Beto 86 at 30 days old transplanted in the middle of the pots. Treated pots inoculated with 3000 *M. javanica* eggs/pot made by pouring the aqueous solution into holes made around the seedlings in each pot. Non-inoculated pots served as control. Treatments replicated four times and coordinated in completely randomized block design under glass house conditions. Agricultural processing was done as needed. Two months after nematode inoculation, plant roots gently uprooted and washed using tap water. Fresh root and shoot weights recorded. Number of galls and egg masses /root system counted. Egg-masses counted after dipping in Phloxine-B stain solution at 0.015% for 20
Number of females/root system was counted using a stereomicroscope [15]. Number of second stage juveniles (J2S)/250g soil evaluated using the counting slide under a stereomicroscope according to Goody [16] using serial sieves and modified Baermann technique. Reduction Percentage in nematode parameters calculated according the following equation:

\[
\text{Reduction Percentage} = \left(\frac{\text{Control} - \text{Treatment}}{\text{Control}}\right) \times 100
\]

**Statistical analysis**

Data Statistical analysis using Duncan's Multiple Range test (P=0.05) using costat 6.3 version program.

**Results and Discussion**

Obtained data in table 1 clarified that garlic, onion and Lemon grass at the tested rates significantly (p≤0.05) reduced nematode parameters. However, there was variation among the treatments in reducing nematode parameters. The highest percentage of reduction in number of second stage juveniles in 250 gm soil (69.90%) was obtained at the higher concentration of garlic followed by onion (63.52 %) compared with nematode alone. Results showed also that those treatments significantly (p≤0.05) decreased the number of galls/tomato root system. The treatment Garlic at 3%recorded the highest reduction in number of galls/root system by 75.7%followed by 67.8% in onion compared with inoculated untreated plants. Results showed also that those treatments significantly (p≤0.05) decreased the number of egg-masses and females/root system by 75.5 and 79.4 % of reduction respectively. The lowest reduction in nematode parameters recorded with the treatment by lemon grass at 1% as it recorded 25.1, 49.4, 49.9 and 58.4 % in number of J2S in soil, galls, egg-masses and females respectively compared with plants inoculated by nematode only (Fig.1 and 2).

Significant differences found between all treatments and nematode inoculated plants in all nematode related parameters but not found between all treatments each other. In general, the percentage of reductions positively correlated with used concentration, by increasing the treatment concentration the percentage of reduction increased.

The effect of treatments on tomato plant growth parameters i.e., plant height, root length, fresh root weight; fresh shoot weight and dry shoot weight after inoculation of plants with eggs of *M. javanica* is illustrated in table 2. Results showed that the treatments enhanced plant growth parameters compared with untreated inoculated plants.

The treatments did not result in significant differences in all the parameters. The highest increasing in plant height recorded with plants treated with garlic 3% followed by onion 3% and lemongrass 3% even there is no significance different between the each other while the significance different found with untreated inoculated plants and control. Similar trend recorded in root length, fresh root weight; fresh shoot weight. The lowest plant growth parameters observed in plants with nematode alone.

Applications of botanicals at all tested concentrations were significantly (p<0.05) inhibited nematode related parameters and effective in controlling infestation of nematodes into tomato roots compared to untreated infected plants. In general, the mode of action of plant parasitic nematodes control by botanicals could be one or more of juvenile toxicant, anti-feedant, ovicidal properties, inducers of host resistance, growth disruption or repellent [17]. Earlier studies revealed that phytochemicals change nematode physiology by affecting: ion uptake, membrane permeability, enzymatic activity, cell division and electron transport [18].
Table 1 Effect of garlic, onion and lemon grass on mean number of galls, egg masses and females on tomato plants root system

| Parameters Treatments | Number of J2S / 250gm soil | Nematode parameters /root system |
|-----------------------|-----------------------------|----------------------------------|
|                       | Number of Galls | Number of Egg-masses | Number of Females |
| Garlic 1 %            | 100 d           | 78 b                     | 74.75 b            | 66.5 bc |
| Garlic 2 %            | 74.25 ef        | 63.25 cd                | 59.75 c            | 58.25 d |
| Garlic 3 %            | 54.25 g         | 40.25 f                 | 37.75 d            | 33.75 e |
| Lemon grass 1%        | 135 b           | 83.75 b                 | 77.25 b            | 68 b   |
| Lemon grass 2%        | 115.75 c        | 67.5 c                  | 63.5 c             | 63.5 bcd |
| Lemon grass 3%        | 77.5 ef         | 56 de                   | 44.25 d            | 37.5 e |
| Onion 1%              | 117.75 c        | 80.75 b                 | 76 b               | 67.25 b |
| Onion 2%              | 83.75 e         | 65.5 cd                 | 61 c               | 61 cd  |
| Onion 3%              | 65.75 fg        | 53.25 e                 | 41 d               | 35.75 e |
| Nematode alone        | 180.25 a        | 165.5 a                 | 154.25 a           | 163.5 a |
| Control               | -               | -                       | -                  | -      |

Columns followed by different letters are significantly different according to Duncan's Multiple Test (P≤0.05).

Table 2 Effect of garlic, onion and lemon grass on tomato plant growth parameters

| Parameters Treatments | Plant height (cm) | Root length (cm) | Fresh root weight (g) | Fresh shoot weight (g) | Dry shoot weight (g) |
|-----------------------|-------------------|------------------|------------------------|------------------------|---------------------|
| Garlic 1 %            | 50.75 bcd         | 15.00 abc        | 4.15 cde               | 20.55 bcd              | 2.97 cd             |
| Garlic 2 %            | 52.75 abcd        | 12.12 c          | 4.55 bcd               | 22.25 abc              | 3.17 bcd            |
| Garlic 3 %            | 57.50 a           | 17.50 a          | 6.32 a                 | 24.30 a                | 4.55 a              |
| lemon grass 1%        | 39.87 e           | 12.87bc          | 3.80 de                | 19.87 cd               | 2.42 de             |
| Lemon grass 2%        | 49.87 cd          | 15.12 abc        | 4.17 cde               | 22.09 abc              | 2.65 de             |
| Lemon grass 3%        | 53.50 abc         | 16.50ab          | 4.90bc                 | 23.70 a                | 3.50 bc             |
| Onion 1%              | 48.25 d           | 13.75 abc        | 4.00 cde               | 20.31 cd               | 2.60 de             |
| Onion 2%              | 51.00 bcd         | 15.75 abc        | 4.27 cde               | 22.15 abc              | 2.95 cd             |
| Onion 3%              | 56.12 ab          | 17.25 a          | 5.17 b                 | 23.00 ab               | 3.90 ab             |
| Nematode alone        | 38.62 e           | 7.62 d           | 2.92 f                 | 10.28 e                | 1.85 e              |
| Control               | 42.00 e           | 12.25 c          | 3.35 ef                | 18.86 d                | 1.87 e              |

Columns followed by different letters are significantly different according to Duncan's Multiple Test (P≤0.05).
Decaying of plant materials in the soil, released compound may be act as nematicides. However, noticing a difference among treatments in their efficacy at different concentrations. This could be due to variance in chemical composition and toxic components concentration. Results showed that lower concentrations were less effective than higher concentration in all tested treatments. Garlic was effectively decreased number of J2S in pots soil, number of galls, egg masses and females /root system and increased fresh root and shoot weight and dry shoot weight of tomato plants. This result is similar to those by [11, 19] when using garlic plant extract or oil. The nematicidal effect of garlic may be due to richness in many component such as garlic oils that mainly
consist of Allicina, diallylsulphide, allyl-
methyltrisulphidea, diallyldisulphidedi-
allytetrasulphide, diallyltrisulphide, and
allypropylidisulphidea which have a
nematicidal activity [20, 21]. A major
members of garlic component is Allicin
which have inhibitory effect against of
M. incognita egg hatching [22]. Garlic may be
have indirect effects on nematode populations
by interrupt their mobility, food absorption
and reproduction [23].

Results cleared that soil amendment with the
leaves of lemon grass affect the nematode
related parameters and enhanced plant growth
parameter of tomato plants. This funding in
agreement with those by previous researcher
[24, 25] who showed nematicidal effect of
lemon grass by reducing number eggs of M.
javanica and M. incognita and root galling of
tomato and African yam bean and inhibit egg
hatching [26]. Lemon grass as a member of
Cymbopogon genus produce essential oils rich
in monoterpenes such as citral, citronellal,
citronellol, elemol, linalool, limonene, 1,8-
cineole, geraniol, β-carophyllene, geranyl
acetate, methyl heptenone and geranylformate
[27]. Cymbopogon citratus contain aldehydecitral, geraniol and monoterpenene
olefins [28].

Allium cepa in the present study showed a
nematicidal effect against root-knot nematode
by reducing J2S in pot soil, number of galls,
egg masses /root system. Similarly Goel and
Gupta [29] reported the nematicidal potential
of onion cv. N- 53 extract against second
stage juveniles of M. javanica and Garvita Joshi et al., [30] reported the effect of onion
in hatching and mortality of M. incognita.

Results confirmed the effectiveness of the
botanicals in enhancing the plant growth
parameters of tomato plants, which may be
due to nutritional effect of the green manure
and reduction in nematode by reducing J2S,
galls and egg masses. Reduction of plant
growth in untreated plants may be due to its
inability to use of water and nutrient from soil
because of root injury, which became unable
to uptake and translate water and nutrients.
Plant growth phytohormons may be also
affect by nematode infection [31]. Although
present study recorded effective potential of
the tested plants, may be this effect vary
according to different factors such as: time of
application, plant age, soil type, collection
time [4, 8]. Further studies must be on focus
on combination of the tested plants as soil
amendment with other different strategy. This
will maximize the nematicidal activity and
develop formulation to improve their efficacy
and stability as an ecofriendly sustainable
method for pant parasitic nematode
management. These could lead to reduce
using of synthetic nematicides and providing
food security.

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