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NEMATICIDAL POTENTIAL OF *TERMINALIA CATAPPA* L. AGAINST *MELOIDOGYNE JAVANICA* (TREUB) CHITWOOD

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ABSTRACT

Terminalia catappa L. (wild almond) is a medicinal plant, distributed throughout the tropics in coastal environments. In the present study with aqueous extracts of leaves, old bark and seeds of *T. catappa* was tested against root-knot-nematode *Meloidogyne javanica* (Treub) Chitwood, *in vitro* and under greenhouse conditions on growth of cowpea (*Vigna unguiculata* L. (Walp)) plants. *In vitro* studies with aqueous extracts of leaves, old barks and seeds at 0, 25, 50, 100% concentrations showed differential nematicidal activity on egg hatching and mortality of second stage juveniles (J_2) of root-knot nematode *Meloidogyne javanica.* Minimum hatching and maximum larval mortality percentage was achieved in leaf extracts. Soil organic amendment with wild almond (at 0.3%) enhanced seed germination of cowpea and other growth parameters including shoot and root length, root weight and number of leaves. While at 0.5% of leaves amendment showed maximum root length, root weight, shoot weight and number of leaves. Whereas, reduced number of knots were obtained in 0.5% of bark and 0.3% of seeds. Number of eggs and root-knot index (RKI) was found lowest in 0.5% leaves and 0.3% bark amendments.

KEYWORDS: *Terminalia catappa*, Root-knot nematode, *Meloidogyne javanica*, Cowpea.

INTRODUCTION

Plant diseases cause severe losses and reduce yields of various crop plants and can adversely affect the agricultural economy of a country (Hafeez, 1986). In Pakistan, root-knot nematodes cause more severe losses and have a wide host range. *Meloidogyne* infection affects water and nutrient uptake and upward translocation in the root system. By disrupting the host plant physiology, root-knot nematodes may not only reduce crop yield but also product quality (Alam, 1990).

Medicinal plants are known to produce natural compounds like phenols, flavonoids, quinons, tannins, alkaloids, saponins, sterols, and volatile essential oils which have been reported to show, antimicrobial, insecticidal properties (Akhtar *et al*., 2008). Several plant products have shown environmentally safe, selective and effective nematicidal potential (Javed *et al*., 2006; Dawar *et al*., 2008; Ntalli *et al*., 2009; Ntalli and Caboni, 2012). Organic amendments have an inhibitory effect on plant parasitic nematodes and fungi (Muller and Gooch, 1982).

Genus *Terminalia* belongs to family Combretaceae. About 250 species of *Terminalia* are distributed in the tropical regions. Many pharmacological studies have reported that the extract of *Terminalia catappa* plant parts exhibit antioxidant, antiviral, antidiabetic, anti-inflammatory, anticancer, antimicrobial, anti-HIV reverse transcriptase, anti-inflammatory, anti-diabetic and hepatoprotective activities (Tan *et al*., 1991; Fan *et al*., 2004; Kloucek *et al*., 2005; Chen *et al.*, 2000; Chen and Li, 2006; Chyau *et al*., 2006; Kinoshita *et al*., 2007;Nair and Chanda, 2008; Shinde *et al*., 2009; Naitik *et al*., 2012; Anand *et al*., 2015). In the present study nematicidal potential of *Terminalia catappa* L. is investigated *in vitro* and under greenhouse conditions.

MATERIALS AND MEHTODS

Collection of plant sample: *Terminalia catappa* L. fresh leaves, old bark and almond (seeds) samples were obtained from a tree near Institute of Marine Sciences, Karachi University campus. Samples were washed under tap water and shade dried for 20-25 days then dried samples were powdered in an electrical blender before use and kept at room temperature in brown bottles.

Preparation of plant extracts and dilutions: 10g dried powder of different samples (leaves, old bark and seeds) were soaked separately in 100 mL distilled water for 24 h., then filtered through filter paper (Whatman No.1). It was considered as stock solution (100%). Further dilutions (25, 50) were prepared by the addition of required distilled water in stock solution.

Egg hatching test: Eggs suspension of *M. javanica* was prepared in water as described by Hussey and Barker (1973). One mL of eggs suspension (30-40 eggs) and one mL of plant extract were transferred in cavity block and kept at room

temperature. Each treatment had three replicates and treatment without extract served as control. The number of hatched eggs was recorded at 0, 24, 48 and 72 h, under a low power (4x) of compound microscope.

Larval mortality test: Eggs or eggs masses of *M. javanica* were placed in water and incubated at 28 ± 2°C. After hatching, the juveniles were collected and a suspension of juveniles in water was prepared. One mL of larval suspension (30-40 larvae) and one mL of plant extract were transferred in cavity block and kept at room temperature. Each treatment had three replicates and treatment without extract served as control. The number of dead larvae was counted at 0, 24, 48 and 72 hours, under a low power (4x) of compound microscope.

Greenhouse experiment: Sandy loam soil was collected from the experimental field of Karachi University. Dry powder of *T. catappa* fresh leaves, old bark and seed sample mixed with soil @ 0.3% and 0.5% w/w, transferred into 8 cm diameter plastic pots. Non-amended soil served as control. Five seeds per pot of cowpea were sown and after germination two seedlings were kept in each pot. When plant achieved two leaves stage plants were inoculated with 4 to 10 egg masses of *M. javanica* per pot. The plants were uprooted after 45- 50 days of nematode inoculation. Data was collected on growth parameters and root knot index.

Statistical analysis: Data were subjected to Analysis of variance (ANOVA) and Least Significant Difference (LSD) ($p<0.05$) was determined on Cost' at.

RESULTS

Effect on egg hatching (%): Different treatments of *Terminalia catappa* (leaf, old bark and seeds) in relation to time have shown nematicidal activities at significance level ($p<0.05$) against *Meloidogyne javanica*. Maximum inhibitory effect on hatching was recorded in leaf extract in all concentrations, compared to bark and seed amendments. Greater hatching was recorded in control and bark extract at all concentration i.e. 0, 25, 50 and 100% (Fig. 1).

Effect on larval mortality (%): Different treatments of *Terminalia catappa* leaf, old bark and seeds in relation to time have shown nematicidal activities at significance level ($p<0.05$) against *Meloidogyne javanica*. Maximum percentage of mortality was recorded in leaf extract in all concentration i.e., 0, 25, 50 and 100% (Fig. 2).

GREEN HOUSE EXPERIMENTS

Growth parameters

Shoot length: Different treatments of *Terminalia catappa* leaf, bark and seeds (almond) were used as amendment to cowpea against the pathogenicity of *Meloidogyne javanica*. Maximum percentage of shoot length was recorded in bark amendment at 0.5% concentrations and seed at 0.3% as compared to other soil amendments of pot experiments. The control has shown least growth in comparison to other treatments i.e. leaves, seed and bark (Fig. 3).

Root length: Maximum percentage of root length was recorded in amendment with leaves at 0.5% and amendment with seeds at 0.3% as compared to other pot treatments. Bark amendment showed lesser growth of plant root (Fig. 4).

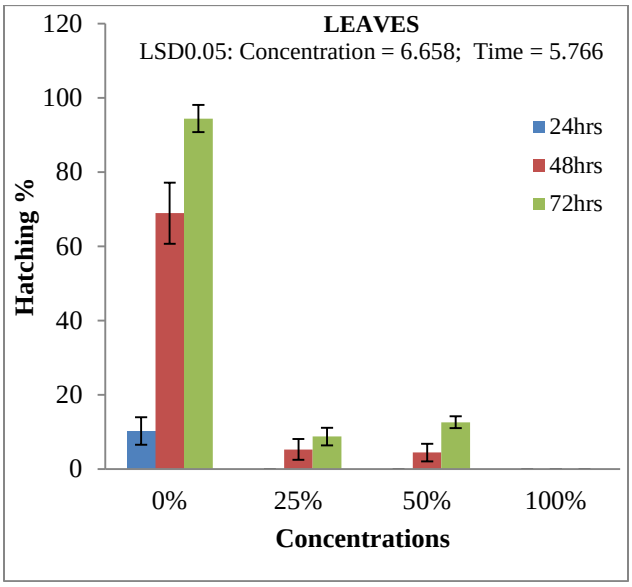
Shoot weight: Maximum shoot weight was recorded in pots amended with leaves (0.5%). Bark amendment have shown lesser growth of host plant root in comparison with other amendments (Fig. 5).

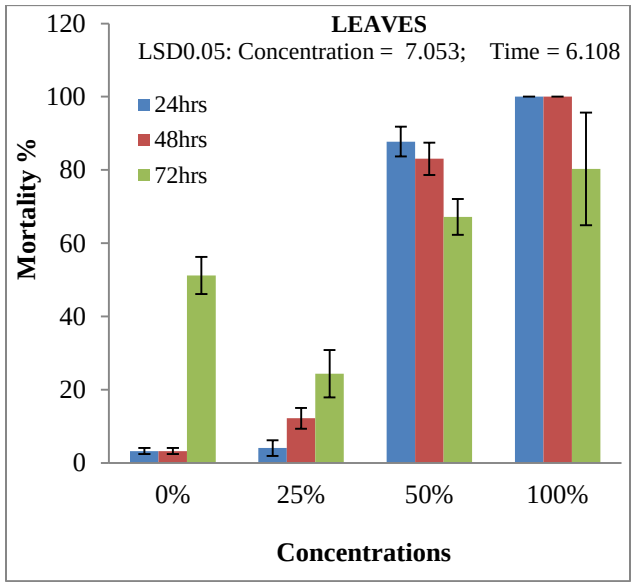
Root weight: Maximum root weight was recorded in pots amended with seeds powder 0.3% followed by 0.5% leaves amendment. Pots with bark amendment showed lesser growth of cowpea plant root (Fig. 6).

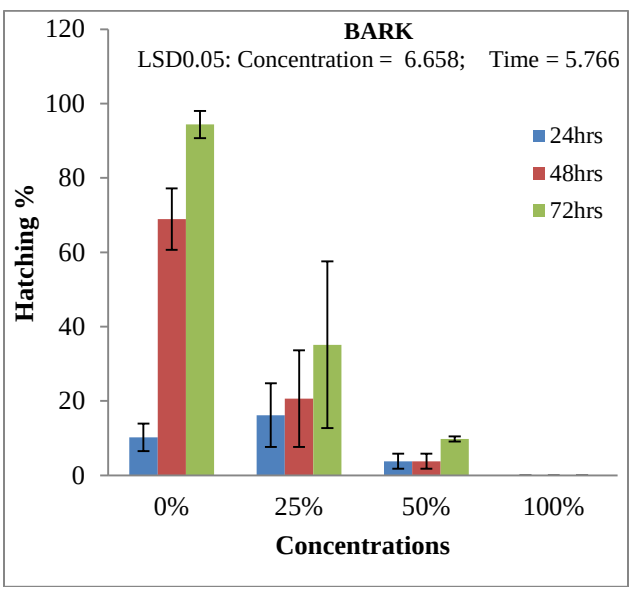
Number of leaves: Maximum number of leaves of cowpea plants was found in pots amended with leaves @ 0.5%. It was followed by the number of leaves in pots amended with seed powder @0.3 and 0.5% (Fig. 7).

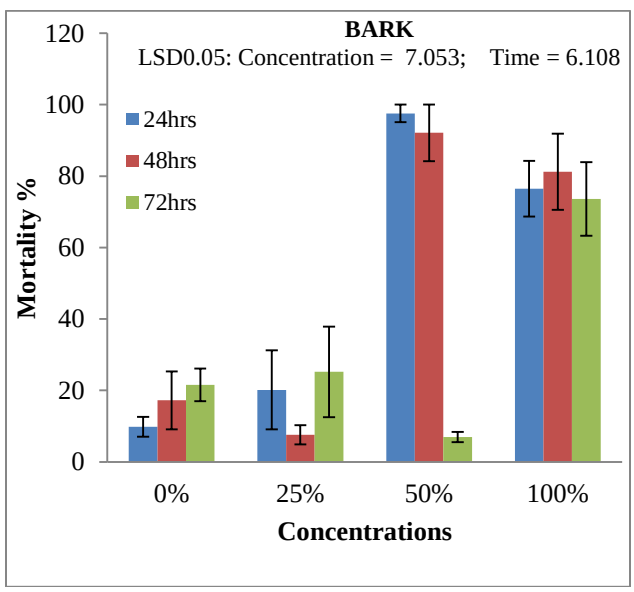
Number of knots: Maximum number of knots was recorded in roots of control secondly in leaves amended pots at all concentrations as compare to other treatments. Minimum number of knots was recorded in 0.5% bark and 0.3% seed amendments (Fig. 8).

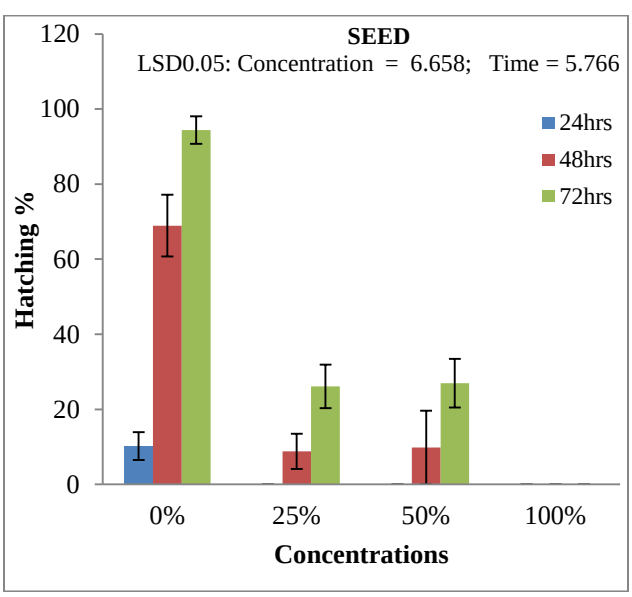
Number of nodules: Maximum number of nodules was recorded in pots amended with leaves powder at all concentrations as compared to other treatments i.e. control, bark and seed. Minimum number of nodules was recorded in roots of control plants followed by in 0.5% bark and 0.3% of seed amended pots (Fig. 9).











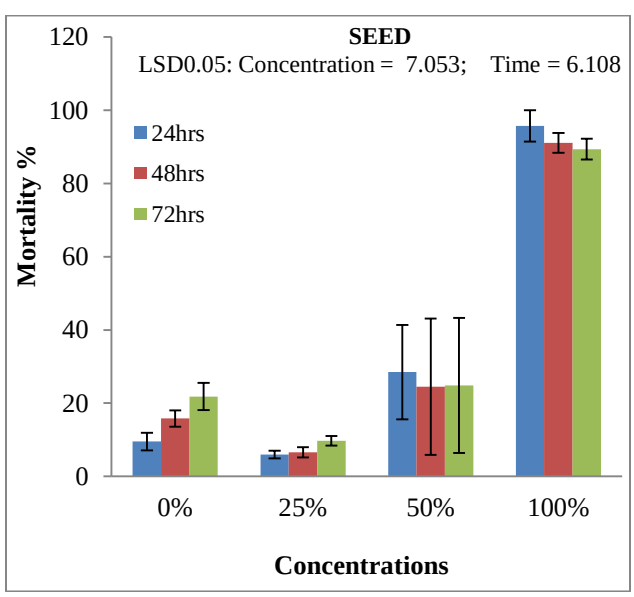


Fig. 1. Effect of aqueous extract of *Terminalia catappa* on egg hatching of root-knot nematodes.

Fig. 2. Effect of aqueous extract of *Terminalia catappa* on J2 mortality of root-knot nematodes.

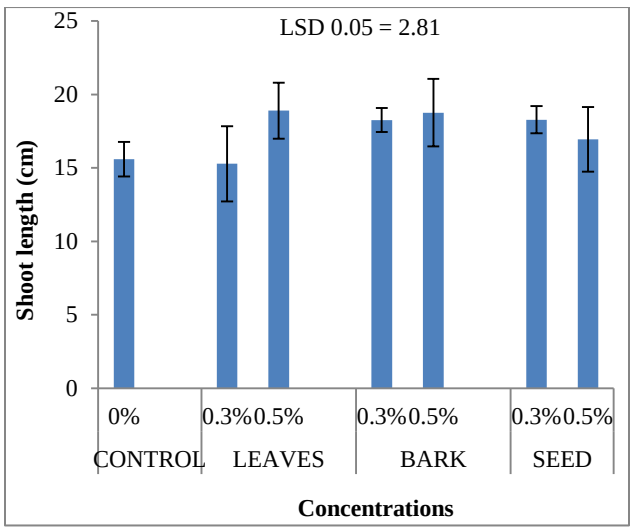


Fig. 3. Effect of soil amendment of different parts of *Terminalia catappa* on shoot length.

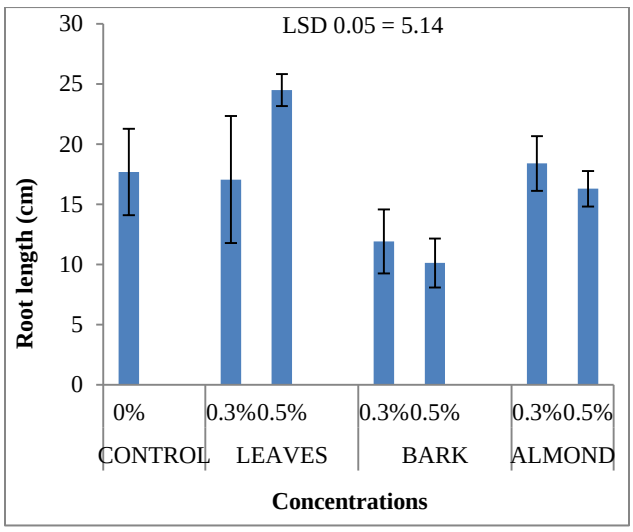


Fig. 4. Effect of soil amendment of different parts of *Terminalia catappa* on root length.

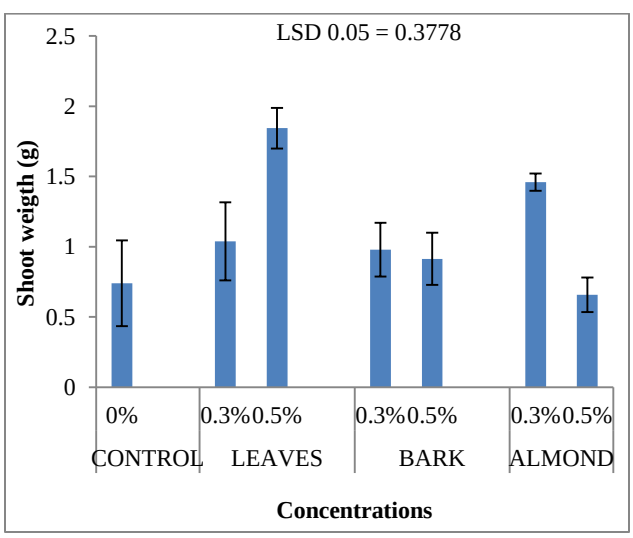


Fig. 5. Effect of soil amendment of different parts of *Terminalia catappa* on shoot weight.

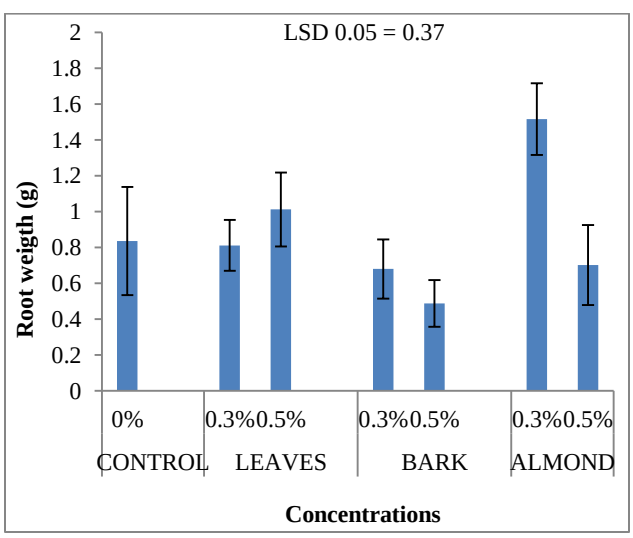


Fig. 6. Effect of soil amendment of different parts of *Terminalia catappa* on root weight.

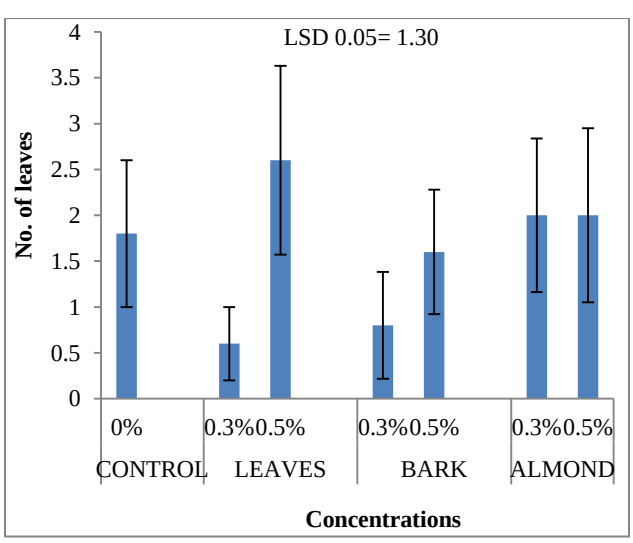


Fig. 7. Effect of soil amendment of different parts of *Terminalia catappa* on Number of leaves.

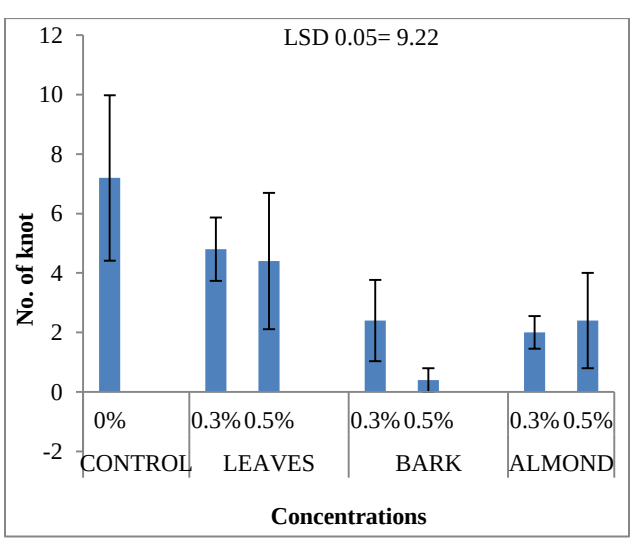


Fig. 8. Effect of soil amendment of different parts of *Terminalia catappa* on number of knots.

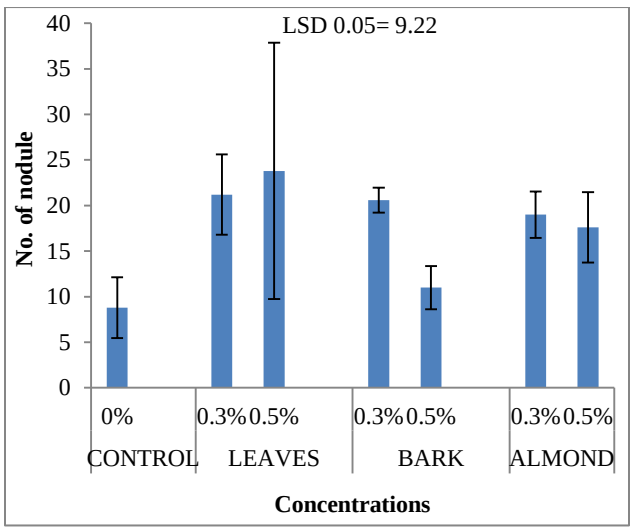


Fig. 9. Effect of soil amendment of different parts of *Terminalia catappa* on number of nodule.

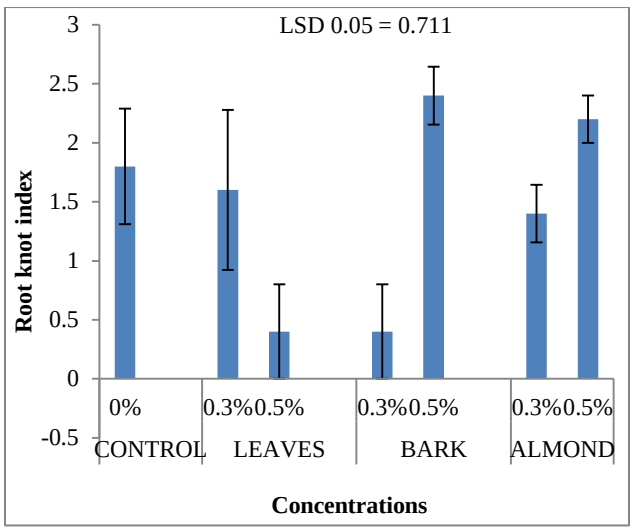


Fig. 10. Effect of soil amendment of different parts of *Terminalia catappa* on root-knot index (RKI).

Root knot index: Root-knot infection has been estimated by the following the scale of Taylor and Sasser (1978). Maximum number of knots was found in *Terminalia* bark and seed amendment at the concentration of 0.5%. Root-knot Index was more or less equal in control plants and plant in pots amended with 0,3% leaf powder. The minimum root knot index was recorded in pots amended with 0.5% leaf and 0.3% bark powder (Fig. 10).

DISCUSSION

Present study showed that hatching of eggs of *M. javanica* decreased and mortality of juveniles increased under the influence of *Terminalia* extracts. Long exposure to the plant extract increased larval mortality rate and percentage of inhibition of egg hatching. The phytochemical analysis of leaf extract by Sofowora (1993) revealed the presence of compounds which are known to exhibit medicinal as well as physiological activities such as phenols, tannins, flavonoids, saponins, glycosides, steroids and alkaloids. Toxic potential of 3, 4-dihydroxybenzoic acid (3, 4-DHBA) from *Terminalia nigrovenulosa* bark has been reported to be active against *Meloidogyne incognita* (Nguyen *et al*., 2013a; 2013b). Methanol extract of *Terminalia arjuna* bark also showed nematicidal activity against *Haemonchus contortus* and ethanol extract of *Terminalia chebula* fruit was toxic to both *Meloidogyne* spp., and *Cephalobus litoralis* (Bachaya *et al.*, 2009; Zia-ul-Haq *et al.*, 2010).

A large number of research reports have confirmed that addition of a variety of decomposable organic matters to nematode infested soil results in the reduction of population of plant parasitic nematode (Akhtar *et al.*, 2008). 0.3% of almond amendment has shown maximum shoot and root length and weight as well as number of leaves in chickpea plant. Minimum number of knots was obtained in 0.5% of bark and 0.3% of seed treatment. Nodules was lesser in control and 0.5% of bark, number of eggs and RKI was lowest in 0.5% of leaves and 0.3% of bark amended pots. Organic amendment with different parts i.e., leaves, bark and seeds of *Terminalia catappa* showed efficiency in the control of root knot infection. Chemical nematicides are effective in controlling root-knot nematodes but they have environmental concerns, however, there is an increasing interest in discovering nematicidal compounds in the plants (Chitwood, 2002). Plants have the potential to suppress or impair rhizosphere fungal assemblages and the associated nematode populations (Abbasi *et al*., 2008). Maintenance of sufficient high organic matter levels is a prerequisite for sustainable high production levels of crop (Akhtar and Mehmood, 1994).

Organic matter has suppressive effect on plant parasitic nematodes (Alam, 1990). Release of toxic compounds from plant tissues are also reported to reduce plant parasitic nematode infection. Several plant terpenoids and phenolic compounds are known to have nematicidal properties and contain unique biochemical compounds that have biocidal properties (Akhtar *et al.*, 2008; Muller and Goach, 1982).

Terminalia species have nematicidal activity which may vary in degree of efficacy. *T. catappa* appears to have ability of sustaining and protecting cowpea plant from diseases. Further research is needed to explore the bio-chemical properties which may be responsible for biological activity against root-knot nematode.

REFERENCES

Abbasi, W.M., N. Ahmed, M.J. Zaki and S.S. Shaukat. (2008). Effect of *Barleria acanthoides* Vahl. on root-knot nematode infection and growth of infected okra and brinjal plants. *Pak. J. Bot.*, 40(5): 2193-2198.

Akhtar, M. and I. Mahmood. (1994). Potentiality of phytochemicals in nematode control: A review. *Biores. Technol*., 47: 189-201.

Akhtar, Y., R. Yeoung and M.B. Isman. (2008). Comparative bioactivity of selected extracts from Meliaceae and some commercial botanical insecticides against two noctuid caterpillars, *Trichoplusiani* and *Pseudaletia unipuncta*. *Phytochem Rev*., 7: 77e88.

Alam, M. (1990). Neem in nematode control. In: *Nematode biocontrol (Aspects and prospects)*: M.S. Jairajpuri, M.M. Alam and I. Ahmed. BS Publishers and Distributors (Pvt) Ltd. Dehli-110032, India. 51-55pp.

Anand, A. V., N. Divya and P.P. Kotti. (2015). An updated review of *Terminalia catappa*. *Pharmacogn. Rev*., 9: 93-98. doi: 10.4103/0973-7847.162103.

Bachaya, H.A., Z. Iqbal, M.N. Khan, A. Jabbar, A.H. Gilani and I.U. Din. (2009). *In vitro* and *In vivo* anthelmintic activity of *Terminalia arjuna* bark. *Int Agric Biol*., 11: 273-278.

Chen, P. S. and J.H. Li. (2006). Chemopreventive effect of punicalagin, a novel tannin component isolated from *Terminalia catappa* on H-ras-transformed NIH3T3 cells. *Toxicol. Lett*., 163: 44-53.

Chen, P.S., J.H. Li, T.Y. Liu and T.C. Lin. (2000). Folk medicine *Terminalia catappa* and its major tannin component, punicalagin, are effective against bleomycin-induced genotoxicity in Chinese hamster ovary cells. *Cancer Lett*., 52: 115-122.

Chitwood, D.J. (2002). Phytochemical based strategies for nematode control. *Annu. Rev. Phytopathol*., 40: 221e49.

Chyau, C.C., P.T. Ko and J.L. Mau. (2006). Antioxidant properties of aqueous extracts from *Terminallia catappa* leaves. *LWT – Food Sci. Technol*., 39: 1099-1108.

Dawar, S., A. Sattar and M.J. Zaki. (2008). Seed dressing with biocontrol agents and nematicides for the control of root knot nematode on sunflower and okra. *Pak. J. Bot*., 40: 2683e91.

Fan, Y.M., L.Z. Xu, J. Gao, Y. Wang, X.H. Tang and X.N. Zhao. (2004). Phytochemical and anti-inflammatory studies on *Terminalia catappa. Fitoterapia*, 75: 253-260.

Hafeez, A. (1986). *Plant Diseases*. Pakistan Agricultural Research centre (PARC). Islamabad, pp. 552.

Hussey. R.S. and K.R. Barker. (1973). A comparison of methods for collecting inocula for *Meloidogyne* spp. including a new technique. *Plant Disease Reporter*, 57: 1025-1028.

Javed, N., S.R. Gowen, M. Inam Ul Haq, K. Abdullah and F. Shahina. (2006). Systemic and persistent effect of neem (*Azadirachta indica*) formulations against root-knot nematodes, *Meloidogyne javanica* and their storage life. *Crop Prot*., 26: 911e6.

Kinoshita, S., Y. Inoue, S. Nakama, T. Ichiba and Y. Aniya. (2007). Antioxidant and hepatoprotective actions of medicinal herb, *Terminalia catappa* L. from Okinawa Island and its tannin corilagin. *Phytomedicine,* 14: 755-762.

Kloucek, P., Z. Polesny, B. Svobodova, E. lkova and L. Kokoska. (2005). Antibacterial screening of some *Peruvian* medicinal plants used in Calleria District. *J. Ethnopharmacol*., 99: 309-312.

Muller, R. and P.S. Gooch. (1982). Nematological reviews: Organic amendments in nematode control. An examination of the literature. *Nematropica*, 12(2): 319-326.

Nair, R. and S. Chanda. (2008). Antimicrobial activity of *Terminalia catappa*, *Manilkara zapota* and *Piper betel* leaf extracts. *Indian J. Pharm. Sci*., 70: 390-393.

Naitik, P., T. Prakash, D. Kotresha and N.R. Rao. (2012). Effect of *Terminalia catappa* on lipid profile in transplanted fibrosarcoma in rats. *Indian J. Pharmacol*., 44: 390-392.

Nguyen, D.M.C., D.J. Seo, K.Y., Kim, R.D. Park, D.H. Kim, Y.S. Han, T.H. Kim and W.J. Jung. (2013a). Nematicidal activity of 3, 4- Dihydroxybenzoic acid purified from *Terminalia nigrovenulosa* bark against *Meloidogyne incognita*. *Microb. Pathogenesis*, 59: 52-59.

Nguyen, D.M.C., D.J. Seo, V.N. Nguyen, K.Y. Kim, R.D. Park and W.J. Jung. (2013b). Nematicidal activity of gallic acid purified from *Terminalia nigrovenulosa* bark against the root-knot nematode *Meloidogyne incognita*. *Nematology*, 15(5): 507-518.

Ntalli, N.G. and P. Caboni. (2012). Botanical nematicides: a review. *J. Agric. Food Chem*., 60: 9929e40.

Ntalli, N.G., U. Menkisoglou-Spiroudi, I.O. Giannakou and D.A. Prophetou-Athanasiadou. (2009). Efficacy evaluation of a neem (*Azadirachta indica* A. Juss) formulation against root-knot nematodes *Meloidogyne incognita*. *Crop Prot*., 28: 489e94.

Shinde, S.L., S.B. Junne, S.S. Wadje and M.M. Baig (2009). The diversity of antibacterial compounds of *Terminalia* species (Combretaceae). *Pak. J. Biol. Sci*., 12: 1483-1486.

Sofowora, A. (1993). Recent trends in research into African medicinal plants. *Journal of Ethnopharmacology*, 38(2): 197-208.

Tan, G.T., J.M. Pezzuto, A.D. Kinghorn and S. H. Hughes. (1991). Evaluation of natural products as inhibitors of human immunodeficiency virus type 1 (HIV-reverse transcriptase. *J. Nat. Prod*., 54: 143-154.

Taylor, A.L. and J.N. Sasser. (1978). *Biology, identification and control of root-knot nematodes* (*Meloidogyne* species). Coop. Publ. Dept. Plant. Path. North California state University and USAID, Graphics, Releigh. pp. 111.

Zia-ul-Haq, M., M. Ahmad and M. Akhter. (2010). Nematicidal activity of selected flora of Pakistan. *Pak. J. Bot*., 42: 2119-2123.

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