African Journal of Agricultural Research Vol. 5(14), pp. 1876-1880, 18 July, 2010
Available online at http://www.academicjournals.org/AJAR
DOI: 10.5897/AJAR09.158
ISSN 1991-637X ©2010 Academic Journals

Full Length Research Paper

Evaluation of the control of citrus nematode (*Tylenchulus semipenetrans*) by leaf extracts of many plants and their effects on plant growth

Kavous Ayazpour[1]*, Hamed Hasanzadeh[2] and Mohammad Sadegh Arabzadegan[2]

[1] Department of Plant Pathology, Islamic Azad University of Jahrom, Jahrom, Iran.
[2]Department of Horticulture, Islamic Azad University of Jahrom, Jahrom, Iran.

Accepted 26 June, 2010

The citrus nematode is an important nematode that is distributed in citrus gardens worldwide and causes decline and growth reduction of citrus plants. A control way of this nematode is chemical control that it is dangerous to environment. In this research, leaf extract of *Allium sativum, Brassica campestris*, *Capsicum frutescents, Glycyrrhiza glabra, Datura innoxia, Chenopodium botrys* and *Foeniculum vulgar*e was used to evaluate them on plant growth and citrus nematode control in Fars province, Iran. In laboratory conditions different plant extracts, concentration of each extract and duration time of exposure extracts was examined in Petri dishes. Results showed that increase in concentration and duration time of exposure extracts caused increased percentage of nematode mortality. In laboratory experiments extracts of *A. sativum* and *C. frutescents, D. innoxia, F. vulgar*e were more effective respectively. In pod experiments, nematode populations decreased in seedlings that were treated with *A. sativum, C. frutescent*s and *F. vulgar*e compared with control sample. In these, pods increased plant growth. These results confirm that it is possible to use local plant extracts to control citrus nematode instead of using chemical nematicides.

Key words: Citrus nematode, plants extract, citrus nematode control, *Tylenchulus semipenetrans*, Fars province, Iran.

INTRODUCTION

Citrus nematode is one of the most important root nematodes of plant trees that have worldwide distribution and cause reduction of crop production and vegetative growth. In addition, this nematode creates slow decline of citrus trees (Bains et al., 1948; van Gandy, 1958). 80 species and varieties of citrus are susceptible to this nematode (Muhammad Shakeel Ahmad et al., 2004). Yield reduction by citrus nematode depending on the citrus nematode infection rate, is 10 to 30% (Verdego-Lucas and McKenry, 2004). Researches in Fars province, Iran, indicated that this nematode is distributed in all citrus orchards. 100% of orchards and 85% of trees that were sampled was infected by citrus nematode in Fars province (Ayazpour and Ghanaatian, 2004).

For several decades, the use of chemical components for controlling plant nematodes, was not only an effective way but also was as an assured way. After several years, many dangerous amounts of these substances were found in springs' water in USA (Agbenin, 2004). Residues of other materials remained in the soil. Some of these pesticides inhibited activities of bacterial nitrification and so they caused accumulation of plant's toxical amounts of nitrite in soil. The consequences of the consumption were that nature lovers groups strongly prohibited using these materials. In recent years, the use of plant materials and animal manures are in the forefront of nematode control researches (Agbenin, 2004).

There are reports that certain parts of some plants or those extracts have nematicides property. Application of these plant's parts or extracts in soils contaminated with nematode, directly affect or stimulate soil microbes to reduce the nematode population (Muhammad et al., 2004).

*Corresponding author. E-mail: Kayazpour@yahoo.com. Tel: 0098-791-4447001, 4447002. Fax: 0098- 791- 3331019.

In this context, the use of plant extracts with the nematicides property is effective, cheaper and healthier, than nematicides.

Since today water extract of *Azadirachta india* (Agbenin, 2004; Muhammad et al., 2004), *Acacia ciliata*, *Borelia* sp., *Ocimum gratissimum*, *Acalypha ciliata*, *Tamarindus indica*, *Cassia siamea* and garlic were tested for activity. It was shown that many plants, including 57 families have nematicides property. *In vitro* and *in vivo* experiments have shown that extracts of *Calendula officinalis*, *Enhydra fluctans* and *Solanum khasiamum* inhibit the eggs hatch and the gall formation of root knot nematode.

The nematicides property of ginger (*Zingiber officinale*), pepper (*Capsicum annum*), garlic (*Alium sativum*) (Agbenin, 2004), *Glyricida maculate* (Jasy and Koshy, 1992), some plants from family Solanaceae (Haseeb and Butool, 1996), 9 commercial varieties of genus *Brassica* (Walker, 1997). *Calotropis procera, Datura alba* (Muhammad et al., 2004), *Glycyrrhiza glabra*, *Withania somnifera*, *Cassia anguistifolia*, *Asparagus racemosus, Andrographis paniculata* (Goel et al., 2005), *Allium sativum*, *Brassica campestris*, *Capsicum frutescens* and oil of *Brassica campestris* (Neves et al., 2005) has proven.

MATERIALS AND METHODS

Preparation of extraction

First *Allium sativum*, *Brassica campestris*, *Capsicum frutescens*, *Glycyrrhiza glabra*, *Datura innoxia*, *Chenopodium botrys* and *Foeniculum vulgare* were washed in tap water and then cut to small pieces. After that 25 g of crushed leaves of each plant sepa-rately was put in an electrical blender and 100 ml of distilled water was added and grinded well. After 24 h the samples by filter paper No. 1 were filtered. This extract was considered as a stan-dard conc-entration (S) and dilution of S/2 and S/4 were prepared by adding the necessary amount of distilled water.

Determine the effect of extracts on nematode larvae mortality in laboratory conditions

For determining the effect of different water extracts on citrus nematode larvae mortality, 1 ml of nematode suspension containing 100 larvae were put in a Petri dish and was added 5 ml of the extract. This method was used for all extracts. Petri dishes containing distilled water were kept as controls. Each treatment had four replications. Petri dishes were maintained at room temperature between 25-30°C. Dead and survival nematodes were counted after 12, 24, 48 and 72 h. The percentage of larval mortality was calculated with the following formula:

mortality %= (number of dead larvae in treatment- number of dead larvae in control)/total number of larvae×100.

Assessment the effect of extracts on nematode and plant in pot conditions

Effect of standard concentration of extracts was evaluated in the pots. For this purpose, single 3-month seedlings of key lime (*Citrus aurantifolia*) were transferred to the pots with sterile soil. One week after transferring of seedlings, each pot with approximately 5000 citrus nematode larvae was infected. 5 days after the contaminating pots were irrigated by 100 ml of extract per pot. Each treatment had 5 replicates. After 3 months the seedlings carefully were brought out of the pots and their roots were washed with tap water. Infor-mation on plant height, fresh and dry weight of foliage and roots, the final nematode population in soil and the number of females per gram of roots were recorded. In both cases a completely rando-mized experimental design was used. Statistical analysis was done by the computer software Mstat-C.

RESULTS AND DISCUSSION

Results of determining the effect of extracts on nematode larvae mortality in laboratory conditions

Mean mortality of different concentrations showed that increasing of concentration of extracts cause more mortality. Also, mortality increases when increase dura-tion of exposure but after 48 h, exposure time can't decrease survival of nematode larvae (Chart 1). This means, plant extracts can kill nematodes very fast. Before that, Haseeb and Butool (1996) had told that exposure time and concentration of extracts can increase the larvae mortality of nematode larvae.

Sativum and *C. frutescens* caused highest mortality and don't have significant differences together. After that, *D. innoxia* and *F. vulgare* and then *C. botrys*, *G. glabra* and *B. campestris* were more effective. All extracts had significant differences to kill the larvae in comparison with control samples (Chart 2). These results significant match with findings (Agbenin, 2004; Neves et al., 2005) about *A. sativum* and *C. frutescens* extracts on nematode lar-vae mortality.

The means comparisons of interaction between con-centration and treatments on larvae mortality by Duncan test at 1% level showed that the concentration S of *A. sativum* and *C. frutescens* had most effective and then the concentration S of *D. innoxia*, that verifies results of Chart 2. The concentration S/4 of *G. glabra* had the low-est effect. All treatments showed statistical differences with control at 1% level (Chart 3).

Results of Chart 4 show that some extracts such as extract of *Datura*, is more effective when increase the exposure time and will become equaled to most effective extract. From this chart, we conclude that *A. sativum*, *C. frutescens*, *F. vulgare* and *D. innoxia* after 48 and 72 h have a good effect on larvae mortality.

Results of greenhouse experiments

In the study of plant growth, it was found that treatments had effect on seedlings and cause more growth. After the control treatment that did not have nematode, seedlings that was treatmented with extracts of *A. sativum, C. frutescens* and *F. vulgare* had most plant length, foliage, wet weight, root wet weight, foliage dry weight and root

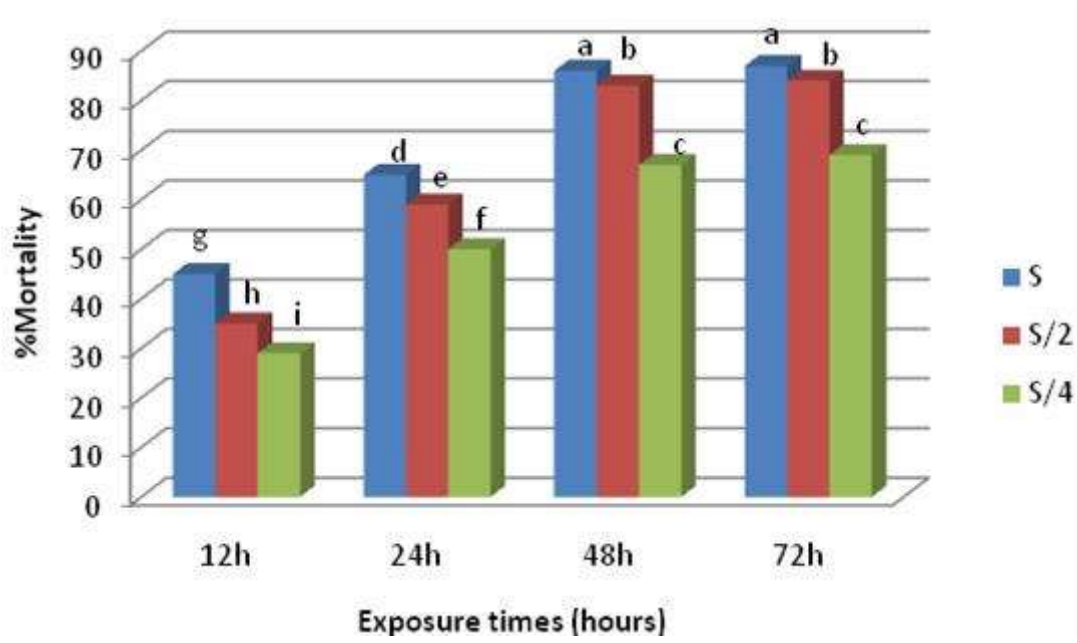

Chart 1. Comparison of average mortality in interaction between concentration and exposure time with Duncan test. (Means with at least a common letter, in the Duncan test at 1% level, do not have significant difference).

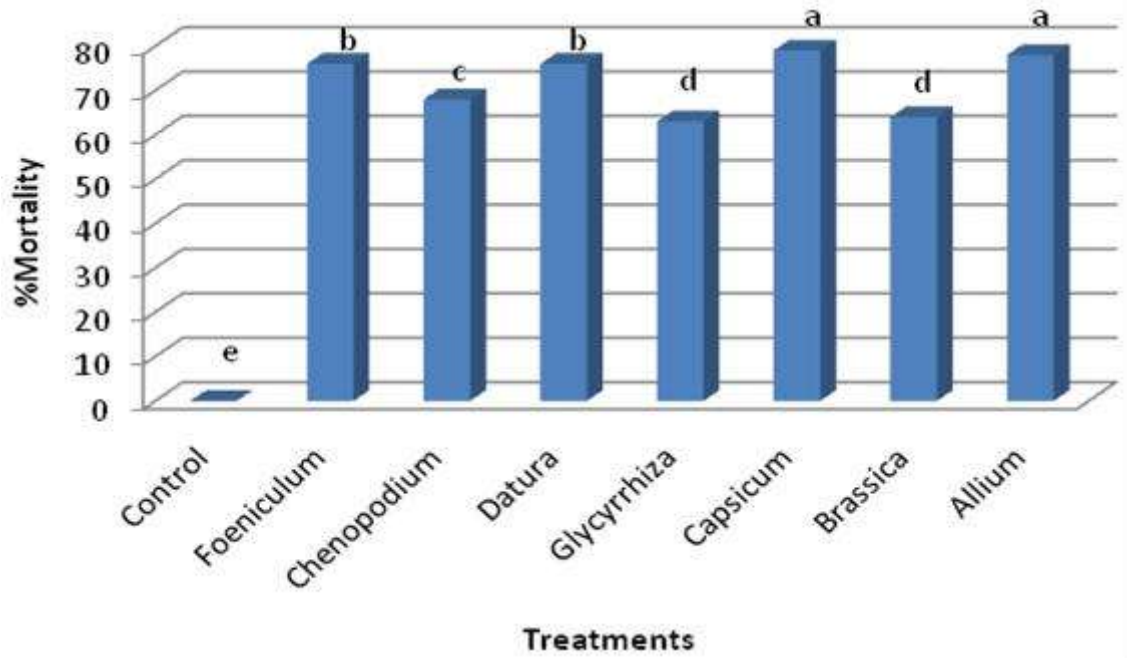

Chart 2. Comparison of average mortality in different treatments with Duncan test. (Means with at least a common letter, in the Duncan test at 1% level, do not have significant difference).

weight (Table 1).

Nematode population in one hundred cubic cm of soil showed that soil treatment with *A. sativum*, *C. frutescens* and *F. vulgare* could reduce nematode population compared with the control (Table 1). As well as nematode population in soil, nematode density on roots were reduced by using *A. sativum*, *C. frutescens*, *Datura innoxia* and *F. vulgare*.

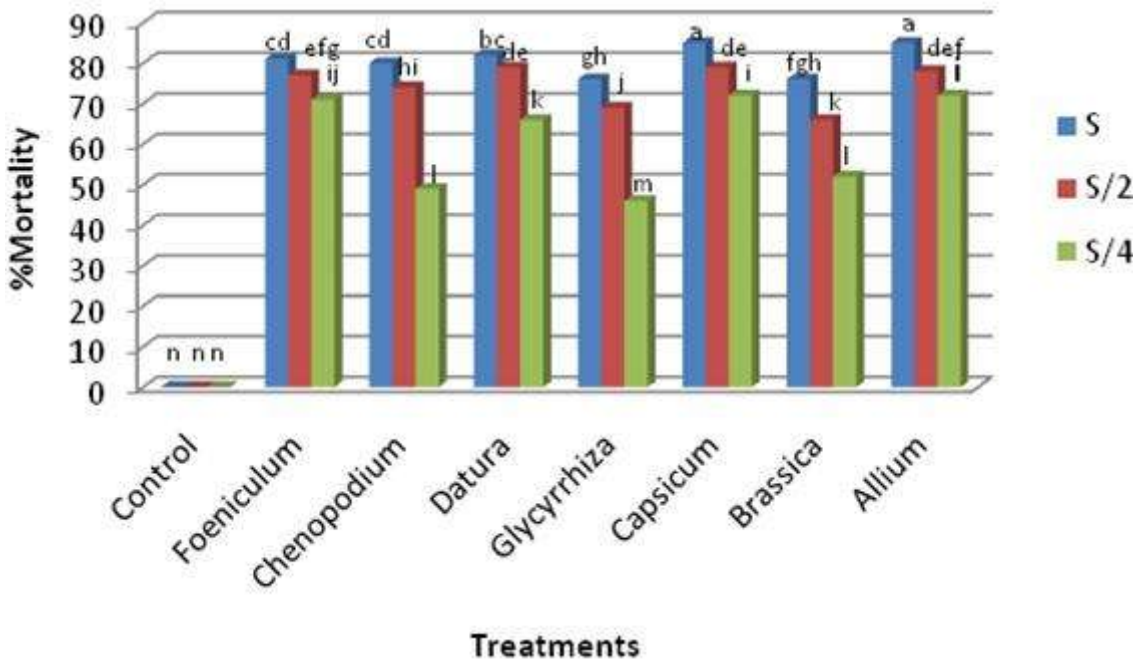

Chart 3. Comparison of average mortality in interaction between concentration and treatments with Duncan test. (Means with at least a common letter, in the Duncan test at 1% level, do not have significant difference).

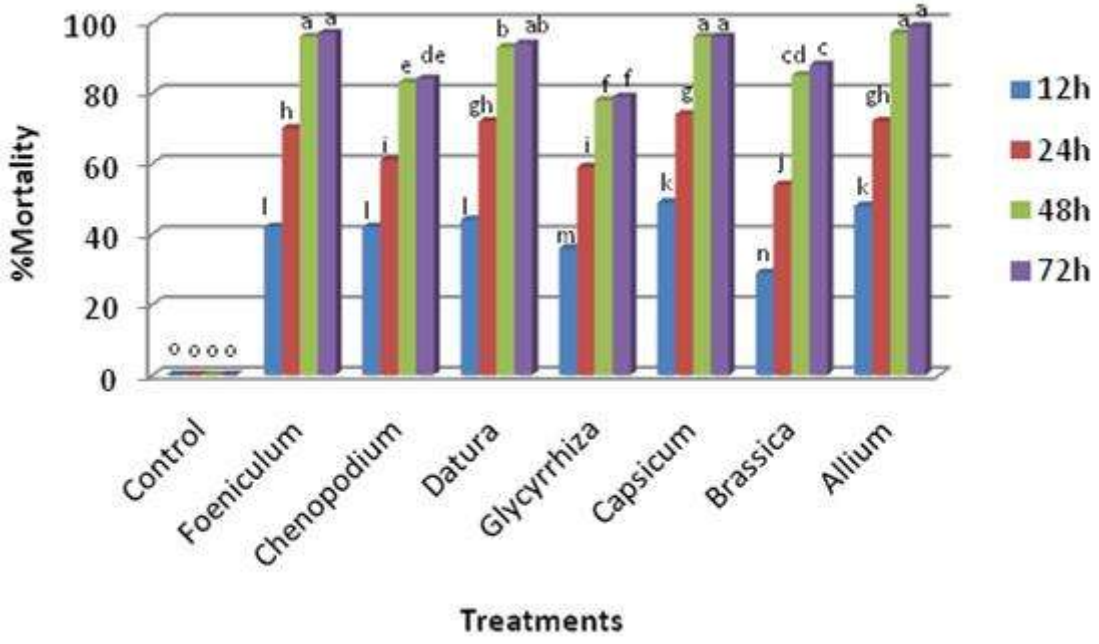

Chart 4. Comparison of average mortality in interaction between exposure times and treatments with Duncan multiple range. (Means with at least a common letter, in the Duncan test at 1% level, do not have significant difference).

Conclusion

According to the above results, we can conclude that plant extracts of *A. sativum*, *C. frutescens*, *Datura innoxia* and *F. vulgare* can use as a safe nematicides to environment. These extracts could create better conditions for vegetative growth of seedlings, and could reduce nematode population, and so, they can increase crop yield.

Table 1. Effects of extracts on comparison of mean treatments in correspond to pot experiments with Duncan test.

Adjective	Inoculated control without extract	Control without treatment	*Allium*	*Brassica*	*Capsicum*	*Glycyrrhiza*	*Datura*	*Chenopodium*	*Foeniculum*
Plant length	64 ef	76 a	71.8 abc	65.6 def	73.6 ab	62.4 f	68 cde	70.2 bcd	73.2 ab
Foliage wet weight	49.7 c	59.1 a	55.7 abc	51 bc	57.3 ab	54.5 abc	52.8 abc	54.7 abc	56.7 abc
Root wet weight	29.1 ef	34.5 a	32.6 abc	29.8 def	33.5 ab	28.4 f	31 cde	31.9 bcd	33.3 ab
Foliage dry weight	15.2 ef	18 a	17 abc	15.5 def	17.4 ab	14.8 f	16.1 cde	16.6 bcd	17.2 abc
Root dry weight	7.92 ef	9.4 a	8.9 abc	8.1 def	9.14 ab	7.72 f	8.42 cde	8.68 bcd	9.08 ab
Final nematode population per 500 g soil	3072 a	0 c	1699 b	2860 a	2064 b	3028 a	26.6 a	2970 a	2128 b
Nematode population per 1 g root	516 a	0 c	306 b	479 a	314 b	488a	303 b	474 a	279 b

Means with at least a common letter significant, in the Duncan test at 1% level, do not have significant difference.

REFERENCES

Agbenin ON (2004). Potentials of organic amendments in the control of plant parasitic nematodes. Plant Protect. Sci., 40: 21-25.

Ayazpour K, Ghanaatian A (2004). Determination of root parasite nematodes of citrus in Jahrom (Fars province, Iran). Final Research Project, p.28.

Baines RC, Clarck OF, Bitters WP (1948). Susceptibility of some Citrus species and other plants to the citrus-root nematode, Tylenchulus semipenetrans. (Abs.). Phytopathology, 38: 912.

Goel SR, Madan VK, Verma KK, Nandal SN (2005). Proceeding of national symposium on recent advances and research priorities in Indian nematology. 9-10 December. India. p. 22.

Haseeb A, Butool F (1996). Evaluation of nematicidal properties of some members of the family solanaceae. Biores. Technol., 57: 95-97.

Jasy T, Koshy PK (1992). Effect of certain leaf extracts and leaves of Glyricida maculate steud. As green manure on Radopholus similis. Indian J. Nematol., 22: 117-121.

Muhammad Shakeel Ahmad, Tariq Mukhtar, Riaz Ahmad (2004). Some studies on the control of citrus nematode (Tylenchulus semipenetrans) by leaf Extracts of three plants and their effects on plant growth variables. Asian J. Plant Sci., 3(5): 544-548.

Neves WDS, Freitas LGD, Giaretta RD, Fabry CFS, Coutinho MM, Dhingra OD, Ferraz S, Demuner AJ (2005). Activity of garlic, mustard and chili pepper extracts on Meloidogyne javanica egg hatch. Nematologia Brasileira. 29(2): 273-278.

Van Gandy SD (1958). The life history of the citrus nematode Tylenchulus semipenetrans. Cobb. Nematologica. 3: 283-294.

Verdego-Lucas S, McKenry MV (2004). Managment of the citrus nematode Tylenchulus semipenetrans. J. Nematol., 36(4): 424-432.

Walker GE (1997). Effects of Brassica residues and other organic amendments on abundance and sex ratio of Tylenchulus semipenetrans in soil. Aust. J. Exp. Agric., 37: 693-700.