ISSN: 2347-4688, Vol. 7, No.(1) 2019, pg. 62-67

Current Agriculture Research Journal

www.agriculturejournal.org

# Assessment of the Potential of Eco-Friendly Nematicides in the Management of Root- Knot Nematodes on French Beans in Kenya

**E.O. OGUMO,* W.M. MUIRU, J.W. KIMENJU and D.M. MUKUNYA**

Department of Plant Science and Crop Protection, University of Nairobi, Nairobi- 00100, Kenya.

**Abstract**

Root-knot nematodes (RKN) (*Meloidogyne spp*) are a serious pest causing heavy economic losses in a wide range of agricultural crops. A trial was carried out to evaluate the effectiveness of various eco-friendly nematicides in the management of RKN affecting French bean. The field trial was carried out in two seasons with the following treatments; Rigel-G (*salicylic acid*), Phyto Protect (Sesame oil extract), Mytech (*Paecilomyces lilacinus*), Neemraj 0.3% (Azadirachtin), Vydate® (*Oxamyl*) as a positive control and an untreated control. Various rates; Rigel –G (2.5 ml/l), Phyto Protect (10 l/ha), Mytech (125 g/ha) Neemraj 0.3% (3L/ha) and Vydate® (6 l/ha) of treatments were administered and damage on plants was assessed based on galling indices, crop biomass and yield whereas nematode reproductive potential was assessed based on the J2 counts. There was no significant difference ($P \geq 0.05$) in the nematode population densities and galling indices observed among the eco-friendly nematicides and the conventional nematicide (Vydate®). Eco-friendly nematicides had a significant ($P \leq 0.05$) reduction of RKN J2 population densities compared to the negative control. The negative control had the highest mean of root-knot nematode densities (240 RKN/200 cc soil) and a galling index of 3.77 while Vydate and Neemraj had the lowest mean density (40 RKN/200 cc soil) in the first season. Similar results were observed in the second season with control having the highest RKN J2 population densities (285 RKN/200 cc soil) and a galling index of 3.89 and Vydate had the lowest (23 RKN/200 cc soil). The results of this study clearly indicate that eco-friendly nematicides can be fully adopted to suppress RKN in French beans as alternatives to conventional nematicides.

**Article History**

Received: 01 February 2018
Accepted: 02 March 2019

**Keywords**:

Biomass;
Galling Index;
Plant Extracts;
Root Knot Nematodes.

**CONTACT** E.O. Ogumo wmmuiru27@yahoo.com Department of Plant Science and Crop Protection, University of Nairobi, Nairobi- 00100, Kenya.


Doi: http://dx.doi.org/10.12944/CARJ.7.1.08

## Introduction

Horticulture is a very important in Kenya with French beans being one of the key crops cultivated for export. French bean production is labor intensive and hence it employs millions of people who obtain their livelihood from the crop.[20] Of the biotic challenges facing French bean production in Kenya, root knot nematode is a serious problem due to the hidden nature of the problem and significant yield losses are experienced.[14,20] The root- knot nematode species, *Meloidogyne incognita*, is the most widespread and the most serious plant-parasitic nematode affecting wide range of legume plants including French beans in the tropics and subtropical regions.[19] French beans plants become stunted then wilt and eventually die. Bean yield can be reduced by 26 to 63% depending on the bean cultivar grown. There is increasing interest in the development and adoption of environmentally friendly strategies for managing nematodes all over the world as synthetic fumigants and other chemical nematicides have harmful effects on the environment.[17] Different fungal species such as *Paecilomyces lilacinus* have demonstrated potential in managing nematodes by parasitizing eggs and females of root-knot nematode.[16] This fungi has been used successfully against *M. incognita* on tobacco and tomato.[2]

Natural plant extracts such as sesame oil, neem extracts, *salicylic acid* have also showed nematicidal properties against root- knot nematodes.3 Sesame oil is derived from extracts of specific cultivars of hybrid sesame plants.[5] Neem (*Azadirachta indica*) releases pre-formed nematicidal constituents into the soil and has been found effective for the control of several nematode species. Neem constituents, such as nimbin, salanin, thionemone, *Azadirachtin* and various flavonoids, have nematicidal properties.[1]

*salicylic acid* (SA) is a phenolic phytohormone found in plants and it plays a role in plant growth and development. It has been found to limit the degree of J2 infestation by inhibiting the effect on the nematode reproduction index.[13] The role of *salicylic acid* in defense response of many crops such as tomato to RKN is conferred by the gene Mi-1 which is associated with a localized hypersensitive response (HR) by the cells at the site of infection.[18] This study was carried out to investigate the potential of eco-friendly nematicides in the management of root- knot nematodes on French beans.

## Materials and Methods

### Experimental Design

The study was carried out at Finlays Horticulture Kenya Limited, Kingfisher Farm in Naivasha. The soil type in the study area is sandy loam/loamy, sand to loam soil. The topsoil is predominantly sandy with poor water holding capacity (WHC) and nutrient retention capacity. Due to semi-arid weather conditions of the area, proper irrigation is therefore necessary. The experiment consisted of six treatments namely; Rigel-G (*salicylic acid*), Phyto Protect (Sesame oil extract), Mytech (*Paecilomyces lilacinus*), Neemraj 0.3% (*Azadirachtin*), Vydate® (*Oxamyl*) as a positive control and an untreated control. The treatments were arranged in a randomized complete block design (RCBD) in blocks measuring 3 m by 4 m replicated five times for two seasons. Watering was done on a daily basis in line with the normal agronomic practices for French beans as used in the farm. Rigel-G (*salicylic acid*) was applied at a rate of 2.5 ml/l, Phyto Protect (Sesame oil extract) at a rate of 10 l/ha at planting, repeated at the rate of 6 l/ha in week 2, 6 l/ha in week 4, 5 l/ha in week 6 and finally 5 l/ha in week 8. Mytech (*Paecilomyces lilacinus*) was applied at the rate of 125 g/ha 14 days before planting, repeated 4 and 6 weeks after germination at the same rate while Neemraj 0.3% (*Azadirachtin*) was applied at the rate of 3L/ha applied at planting time and Vydate® (*Oxamyl*) was applied at the rate of 6 l/ha soon after germination. Harvesting started after the crop attained 9 weeks and this was done for two weeks. At 12 weeks, the experiment was terminated and the crop uprooted for assessment of galling index, nematode density and dry biomass.

### Assessment of Nematode Infestation

Assessment for various parameters was done 12 weeks after sowing. Data collected included root-knot nematode (*Meloidogyne* spp) densities, galling indices and dry biomass. Five plants from each block were randomly selected and the roots washed free of soil for determination of galling index. The plants were then dried in an oven to constant dry weights. A composite soil sample which consisted of five cores was obtained from each treatment from which nematodes were extracted from 200 $cm^3$ of soil using

the modified Baermann technique described by Hooper.[8] The The galling index was determined by scoring the galls using a scale of 0-10 adopted from Bridge[4] where; 0 = No galls on roots, 1 = Few small galls difficult to find, 2 = Small galls clearly visible, main root clean, 3 = Some larger galls visible, main roots clean, 4 = Larger galls predominate but main root clean, 5 = 50% of roots infested, galling on parts of main roots, reduced root system, 6 = Galling on main roots, 7 = Majority of main roots galled, 8 = All main roots galled, few clean roots visible, 9 = All roots severely galled, plant usually dying and 10 = All roots severely galled, no root system, plant already dead.

**Nematode Analysis**

The roots and soil samples were assessed for galling as follows; a composite soil sample of 200 $cm^3$ was taken from each plot in the field from a depth of 5-10 cm at the beginning and end of the season. Root-knot nematodes were extracted from each of the 200 cm3 of soil samples at Dudutech Laboratory using the modified Baermann method and enumerated in a 5 ml aliquot of nematode suspension. The results were expressed in nematode population densities per 200 $cm^3$ of soil.

**Data Analysis**

All data were subjected to Analysis of Variance (ANOVA) using the generalized linear model (GLM) procedure of Statistical Analysis Systems (SAS) for means to check for any differences in treatments. The means obtained were separated using Student-Newman-Kuel's (S-N-K) test at the 95% confidence level.

**Table 1: Effect of eco-friendly nematicides on root-knot nematode (rkn) Densities and galling indices in french beans under field condition**

| Nematicide | Mean RKN in 200 cc soil | | | Mean Galling index | | |
|---|---|---|---|---|---|---|
| | Season 1 | Season 2 | % Reduction in RKN | Season 1 | Season 2 | % Reduction in Galling index |
| Salicylic acid | 50 [a] | 28 [a] | 63.0 | 2.51 [a] | 2.00 [a] | 68.5 |
| Sesame oil | 65 [a] | 33 [a] | 51.9 | 2.91 [ab] | 2.30 [a] | 62.9 |
| *P. lilacinus* | 42 [a] | 24 [a] | 68.9 | 2.52 [a] | 2.44 [a] | 73.0 |
| *Azadirachtin*. | 40 [a] | 24 [a] | 70.4 | 2.88 [ab] | 2.61 [a] | 73.0 |
| +ve Control Oxamyl | 40 [a] | 23 [a] | 70.4 | 2.09 [a] | 2.04 [a] | 74.2 |
| Untreated Control | 240 [b] | 285 [b] | | 3.77 [b] | 3.89 [b] | |

**Table 2: Effect of eco-friendly manures on crop biomass and its percentage increase under field conditions**

| Nematicide | Season 1 Crop Biomass | | Season 2 Crop Biomass | |
|---|---|---|---|---|
| | Mean | % Increase | Mean | % Increase |
| Salicylic acid | 181.97a | 17.44 | 169.43 a | 4.21 |
| Sesame oil | 174.26a | 13.78 | 165.88 a | 2.16 |
| P. lilacinus | 205.15a | 26.77 | 185.29 a | 12.41 |
| Azadirachtin | 184.25a | 18.46 | 176.60 a | 8.1 |
| Oxamyl | 265.07a | 43.32 | 231.93 a | 30.02 |
| -Ve Control | 150.24a | | 162.30 a | |

## Results

### Effect of Eco-Friendly Nematicides and Bio-Control Agents in Suppressing Root-Knot Nematodes (Rkn)

In both the first and the second season, eco-friendly nematicides had a significant ($P \leq 0.05$) reduction of RKN J2 population densities compared to the untreated control (Table 1). The untreated control had the highest mean RKN densities (240 RKN/200 cc soil) and a galling index of 3.77 while Vydate and Neemraj had the least mean RKN density (40 RKN/200 cc soil) in the first season. Similar results were observed in the second season with control having the highest RKN J2 population densities (285 RKN/200 cc soil) and a galling index of 3.89 and Vydate had the lowest (23 RKN/200 cc soil) (Table 1).

The highest galling index among the eco-friendly nematicides was observed in Phytoprotect treatments with a score of 2.91 in season one and Neemraj with a score of 2.61 in season 2. Rigel G and Mytech had no significant differences in galling indices in season one. Subsequently, treatment with Vydate® resulted in the highest percent reduction in root galling (74.16%) while the least reduction was observed following treatment with Phytoprotect (Table 1).

The resulting crop biomass was highest in the plots treated with Vydate® in both seasons (265.07 and 231.93 grams, respectively) these represented the highest increases in crop biomass for the two seasons (43.32% and 30.02%, respectively). The lowest crop biomass was recorded in the untreated control plots in the two seasons (150.24g and 162.30g). However, there were no significant differences ($P \geq 0.05$) in crop biomass between the eco-friendly nematicides and the untreated control (Table 2).

## Discussion

All eco-friendly nematicides tested in this study reduced the second stage juveniles of RKN. This indicates the importance of the eco-friendly nematicides in management of RKN in French bean production. The reduction was comparable between Vydate and eco-friendly nematicides. Lopez-Perez *et al.*[12] reported reduced galling following treatment with Vydate® which also reduced nematode reproduction as evidenced by the RKN densities. This confirms that use of eco-friendly nematicides is effective in managing RKN.

Treatment of experimental plots with Phytoprotect (Sesame oil extract) resulted in reduction of RKN population densities. This reduction was comparable to the standard control (Vydate®) since this essential oil has nemastatic effects on RKN. In a study by Oka *et al.*[15], essential and fixed oils from 25 species and aromatic plants were evaluated for their nematicidal effect on M. *javanica*. Twelve out of the 27 oils evaluated immobilized more than 80% of M. javanica juveniles at a concentration of 1ml/ litre$^{-1}$ in vitro and inhibits egg hatching. Fixed oils have been found to immobilize the juveniles and inhibit eggs hatching at ≥125µl$^{-1}$ *in vitro* and reduced root galling of cucumber seedlings[15,21] agreeing with the findings of this study that plant extracts are effective in reducing impacts of RKN infection.

Mytech (*Paecilomyces lilacinus*) performed much better than both Rigel and Phytoprotect in reducing RKN populations. Kerry[11] found that *P. lilacinus* gave variable results in a range of conditions and that it required relatively high soil temperatures to be effective. Other eco-friendly chemicals like Rigel G and Neemraj also gave results which were significantly different from the untreated control, however, they varied in their suppression of RKN. Many factors may be attributed to these variation for instance physiological characteristics such as the permeability of nematode cuticles which may favor the penetration of certain compounds.[22] Bio-chemical differences between different nematode species may also have affected the degradation or detoxification of the compounds, therefore reducing activity of the compounds.[11]

Mytech (*P. lilacinus*) and Vydate® had no significant difference in suppression of RKN and in crop biomass but both showed increased crop biomass compared to the other treatments. The increase in crop biomass can be attributed to better control of the RKN especially in early stages of crop growth and therefore allowing the French bean crop to grow vigorously. These findings agree with those of Kahn *et al.*[9] who reported efficacy of *P. lilacinus* in the management of root- knot nematodes as evidenced by reduction of the number of galls and egg masses.

Neemraj (*Azadirachtin spp.*) and Phytoprotect (Sesame oil extract) gave variable results with respect to crop biomass and performed poorly than the positive control (Vydate®) in both seasons. Although Neemraj and Phytoprotect are expected to have boosted crop biomass, this was not the case and it seems their effect was overwhelmed by the significantly high RKN populations. Studies by Khalil *et al.*[10] showed that *P. lilacinus* and biopesticide agents such as neem reduced root galls, juvenile populations and remarkably increased in tomato growth parameters. This study demonstrated that *P. lilacinus* and *Azadirachtin* plant extracts and biocontrol agents from fungal species can be used to manage RKN problems in crops and this agrees with reports of other workers who have also reported successful management of plant-parasitic nematodes using fungi and plant extracts.[7,20]

**Acknowledgements**
The author appreciates the help and support from Dudutech Kenya Ltd and Orion Future Technologies for their support during this study.

**Conflict of Interest**
Authors declare no conflict of interest.

## References

1. Akhtar M., Malik, A. Roles of organic commercial biocontrol product list (online). Soil amendments and soil organisms in the biological control of plant-parasitic nematodes. *Bioresource Technol*, 2000;74: 35-47.
2. Brand D., Soccol C., R., Sabu, A., Roussos, S. Production of fungal biological control agents through solid state fermentation: A case study on *Paecilomyces lilacinus* against root-knot nematodes. *Micol. Aplicada Int.* 2010; 22: 31-48.
3. Brandt. Naturally derived parasitic nematode control- Sesamin EC. Available online: http://www.brandt.co/Portals/0/PDFs/SESAMIN-EC-BROCH-2008.pdf: (2008).
4. Bridge J., Page S., L., J. Estimation of root-knot nematode infestation levels on roots using a rating chart. Tropical Pest Management. 1980; 26: 296–298.
5. Ehlers R., U. Regulation of biological control agents and the EU policy support action REBECA. In: Regulation of Biological Control Agents, R.-U. Ehlers (Ed.), pp. 3-23, Springer Dordrecht, ISBN 978-90-481-3663-6, Heidelberg, Germany. 293 pp. (2011).
6. Export Processing Zone Authority. Horticulture industry in Kenya 2005. Available online: http://epzakenya.com/UserFiles/files/Horticulture.pdf 2005.
7. Hashem M., Abo-Elyousr K., A. Management of the root-knot nematode *Meloidogyne incognita* on tomato with combination of different biocontrol organisms. 2011. Crop Prot. 30: 285-292.
8. Hooper D., J. Extraction and processing of plant and soil nematodes, p. 45-68. In: Luc. M, Sikora, R. A. and Bridge, J. (eds.) Plant Parasitic Nematodes in Sub-tropical and Tropical Agricultural. CAB International. Wallingford, UK: (1990).
9. Kahn A., K,. Williams L., H.K.M. Nevalainen. Infection of plant-parasitic nematodes by *Paecilomyces lilacinus* and Monacrosporium lysipagum. *BioControl*, 2006; 51: 659 - 678.
10. Khalil., M., S. E., Allam, A..F..G., Ahmed, A.S.T. Nematicidal activity of some biopesticide agents and microorganisms against root-knot nematode on tomato plants under greenhouse conditions. 2012; *Journal of Plant Protection Research* 52: 47-52.
11. Kerry B., R. Biological control of nematodes: Prospects and opportunities. FAO Corporate Document Repository. Plant nematode problems and their control in the Near East Region. FAO Plant Production and Protection Paper, 144. URL: http://www.fao.docrep/V9978E00.htm. (1997).
12. López-Pérez J., A., Escuer M., Díez-Rojo M., A., Robertson L., A., Piedra Buena J., López-Cepero B., A. Host range of Meloidogyne arenaria (Neal, 1889) Chitwood, 1949 (Nematoda: Meloidogynidae) in Spain. 2011; Nematropica 41:130-140.
13. Molinari S. Loffredo E. The role of *salicylic acid* in defense response of tomato to root-

knot nematodes. *Physiological and Molecular Plant Pathology* 2006; 68: 69-78.

14. Monda E., O., Munene S., Ndegua A., French bean (Phaseolus vulgaris L.) is an important vegetable export crop in Kenya. *African Crop Science Conference Proceedings*. 2003; 6: 683-687.
15. Oka Y., Koltai, H., Bar-Eyal, M., Mor, M., Sharon, E., Chet, I. Spiegel, Y. New strategies for the control of plant parasitic nematodes. Pest Management Sci. 2000; 56: 983-988.
16. Pau C.G., Leong C.T.S., Wong S.K., Eng L., Jiwan M., Kundat F.R., Aziz Z.F.B.A., Ahmed O.H., Majid N.M. Isolation of indigenous strains of *Paecilomyces lilacinus* with antagonistic activity against *Meloidogyne incognita*. Int. J. Agric. Biol., 2012; 14: 197–203.
17. Schneider S.M., Rosskopf E.N., Leesch J.G., Chellemi D.O., Bull C.T., Mazzola M. Alternatives to methyl bromide pre-plant and post harvest. Pest Manage. Sci. 2003; 59: 814–826.
18. Sergio M., Loffredob E. The role of *salicylic acid* in defense response of tomato to RKN. 2006; Physiol Mol Plant Path, 68: 69–78.
19. Sikora R.A., Greco N., Luc R.A. Sikora & J. Bridge (Eds.), Plant-parasitic nematodes in subtropical and tropical agriculture, pp. 181–235. 1990; CAB International Publishers, Wallingford, UK.
20. Netherlands Development Organisation (SNV) Annual Report. 2012. http://www.snvworld.org/CAR2012
21. Sumbali G., Mehrotra R.S. Evaluation of some fixed oils for the control of certain temperate fruit rot fungi. Indian Phytopath., 1980; 33: 517.
22. Udo I. A., Osai E O., Ukeh D., A. Management of Root-Knot Disease on Tomato with Bioformulated *Paecilomyces lilacinus* and Leaf Extract of Lantana camara. *Brazilian Archives of Biology and Technology*. 2014; 57: 486-492.