Evaluation of Plant Materials as Root-Knot Nematode (*Meloidogyne incognita*) Suppressant in Okra (*Abelmuscous esculentus*)

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Summary

Plant parasitic nematodes are one of the most important pests on agricultural fields; they cause serious damage to crops. Synthetic pesticides have been used lately to combat the menace of nematodes. Pesticide residues have however been detected in fruits and vegetables due to the residual effect of nematicides in the environment. To minimize the negative effect of synthetic nematicides, it is important to search for alternative plant protection methods. In this study, plant materials (*Euphorbia heterophylla, Hyptis suaveolens, Eucalyptus officinalis, Ocimum gratismum* and *Crotolaria juncea*) were investigated as soil amendments for possible reduction of root-knot nematode (*Meloidogyne incognita*) populations on okra (*Abelmuscous esculentus*). Results revealed that the highest rate of amendment with *E. officinalis* reduced significantly ($p<0.05$) the number of root galls, egg masses and juveniles, with a simultaneous increase in okra yield. Thus soil amendments with *E. officinalis* could be an alternative to synthetic nematicide in *M. incognita* management.

Key words

euphorbia heterophylla, hyptis suaveolens, eucalyptus officinalis, ocimum gratismum, crotolaria juncea

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INTRODUCTION

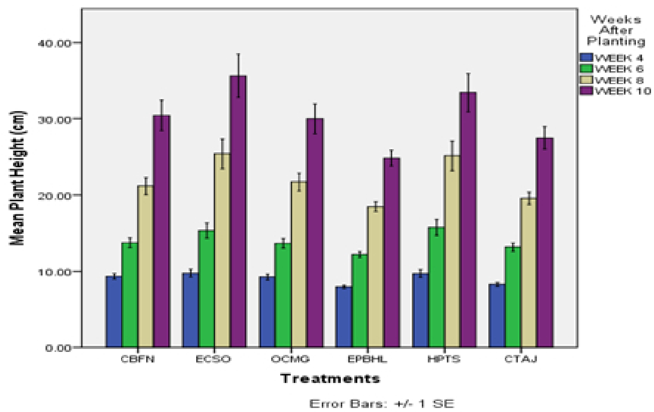
Enormous losses of crops to diseases occur globally. The loss occurs in the field and/or in storage. Among the pests responsible for crop losses are plant parasitic nematodes (PPN). Serious losses on a wide range of crops have been attributed to PPN (Afolami and Orisajo, 2003; Javed et al., 2007). *Meloidogyne* spp. have been reported to be the most important of the PPN; it has been indicated to be responsible for about 12.3% yield loss of crops worldwide (Goodey et al., 1965; Sasser and Carter, 1985; Cristobal-Alejo et al., 2006). In Nigeria *Meloidogyne incognita* is the most damaging and abundant (Adegbite and Adesiyan, 2005; Fabiyi et al., 2018a). It has a very high rate of reproduction; a female *M. incognita* could produce about a thousand eggs making control rather difficult (Williamson and Hussey, 1996; Natarajan et al., 2006). A characteristic nutrient deficiency, stunting and root galling are symptoms of infection. Okra, an important vegetable which is a good source of calcium and potassium, is one of the crops that is highly susceptible to *M. incognita*, leading to reduced yields of the highly relished fruits of this crop (Adekunle, 2009; Hussain et al., 2014). Basically, synthetic nematicides have been used in the control of *M. incognita* on okra, but excessive and indiscriminate use has led to the development of resistance by the organisms and more importantly, pesticide residues abound in vegetables in the market, most often in developing countries (Yudelman et al., 1998; Fabiyi, 2014; Fabiyi et al., 2018b). This necessitated research into the use of organic soil amendments, which in some cases has brought about encouraging results. Plant materials such as *Tagetes erecta, Azadirachta indica, Cymbopogon citratus, Lawsonia inermis* and *Chromolaena odorata* have been reported to be nematicidal (Saravanapriya and Sivakumar, 2005; Fabiyi and Atolani 2011; Hayat et al., 2012; Onyeke and Akueshi, 2012). The aim of this study is to investigate the prospect of using *Euphorbia heterophylla, Hyptis suaveolens, Eucalyptus officinalis, Ocimum gratismum* and *Crotolaria juncea* as soil amendments in the control of *M. incognita* on okra.

MATERIAL AND METHODS

The leaves of *Euphorbia heterophylla* (EPBHL), *Hyptis suaveolens* (HPTS), *Eucalyptus officinalis* (ECSO), *Ocimum gratismum* (OCMG) and *Crotolaria juncea* (CTAJ) were collected from the University of Ilorin campus. They were air dried in the laboratory for a month and then pulverized using a Steelman diesel engine milling machine with a 7- horsepower capacity; model R175A and then stored away in non-transparent light brown paper bags at room temperature until use. Sandy loam soil was heated at 70 °C for three hours. It was air dried and left to stabilize for four weeks after which 15 kg of substrate was distributed into 40 cm diameter experimental pots. Pure culture of *M. incognita* eggs was extracted from 55-day old *Celosia argentea* cv TLV8 roots infected with *M. incognita* race2 (identified with the perineal pattern based on Eisenback et al., 1981) following the Husssey and Baker (1973) method. The desinfected soil was mixed thoroughly with the different plant materials (EPBHL, HPTS, ECSO, OCMG, CTAJ) at 50 g, 100 g and 150 g per pot with 15 kg of soil. Okra cv local seeds were planted and inoculated with approximately 1000 eggs of *M. incognita* two weeks after seedling emergence in a hole made at the base of each plant in the experimental pots (Fabiyi, 2019). The experiment was a randomized complete block design with six treatments (*Euphorbia heterophylla, Hyptis suaveolens, Eucalyptus officinalis, Ocimum gratismum, Crotolaria juncea* and Carbofuran) coded EPBHL, HPTS, ECSO, OCMG, CTAJ and CBFN respectively. Each treatment had four rates of application and three replicates each. The inoculated but untreated pots served as the negative control which is the zero (0) rate of application. Carbofuran was used as a positive check for the plant materials. Data taken in the course of the experiment include plant height, number of fruits per plant and fruit weight per plant. Number of egg masses, nematode population in 10 g of plant root and 250 g soil were ascertained in the laboratory after harvest. Root gall per root was rated using the rating scale of Bridge and Page (1980), where 0=root knots absent, 1=very small invisible knots, 2=small but very visible with main roots clean, 3=knots largely visible with main roots clean, 4=large root knots abound with main roots clean, 5=50% of roots affected, with knots on main root, 6=main root knotted, 7=a handful of main root knotted, 8=all parts of main root knotted, few clean roots visible, 9=severe root knots on root system, 10=severe root knots all over, plant may be dead. Data obtained were analysed using GenStat 5.32. Comparisons were made among different treatments means with Duncan's multiple range test at alpha level <0.05.

RESULTS

The effects of different plant materials on the vegetative growth of okra plants are depicted in Fig. 1a to Fig. 6b. Plant heights were significantly (p=0.05) higher in okra plants treated with ECSO in the 10th week after planting than plants from treatments EPBHL and CTAJ. This was closely followed by plants treated with HPTS (Fig.1a), while significantly lower plant height was observed in EPBHL treated plants. Higher number of fruits and fruit weights was observed in plants treated with ECSO and HPTS (Fig. 2a and Fig. 3a). The untreated (control) plants produced significantly (p=0.05) lower number of fruits and fruit weights compared to treated plants. Nematode population in the root and soil was lower in CBFN, ECSO and HPTS treated plants, while higher nematode population was recorded in EPBHL treated plants (Fig. 4a). Significantly fewer egg masses were seen on the roots of plants treated with ECSO, HPTS and CBFN (Fig. 5a). The untreated control plants had higher number of egg masses, higher nematode populations in their soil and roots, with a corresponding significantly higher root gall rating (Fig. 4b, 5b and 6b). There were significant (p=0.05) differences amongst the rate of application of plant materials. Vegetative growth was significantly higher in plants treated with the highest rate of application (150 g), while reduced growth was observed in the untreated control plants (0 g). Nematode population in soil and roots was also significantly reduced in plants treated with the highest rate as opposed to the lowest rate (Fig. 4b).



CBFN - carbofuran, ECSO - *Eucalyptus officinalis*, OCMG - *Ocimum gratismum*, EPBHL - *Euphorbia heterophylla*, HPTS - *Hyptis suaveolens*, CTAJ - *Crotolaria juncea*

Figure 1a. Effects of Treatment Materials on Plant height of Okra

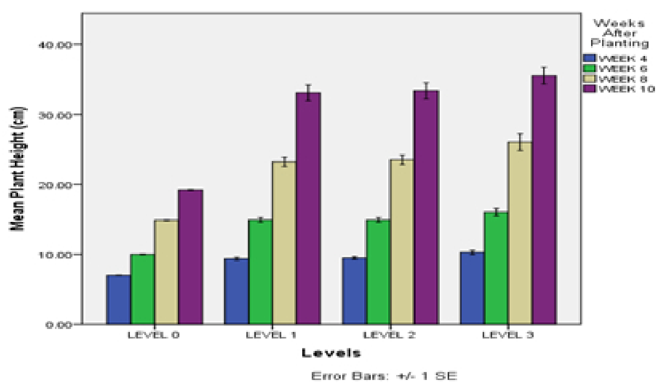


Figure 1b. Effects of Levels of Application of Treatment Materials on Plant height of Okra. Each level represents dosages (0 g, 50 g, 100 g and 150 g)

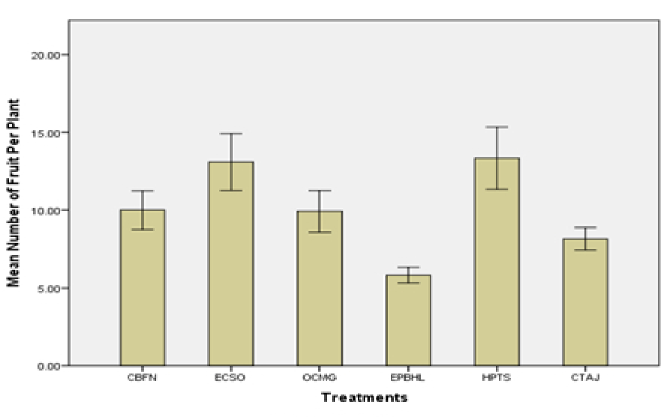


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Figure 2a. Effects of Treatment Materials on Number of Fruits of Okra

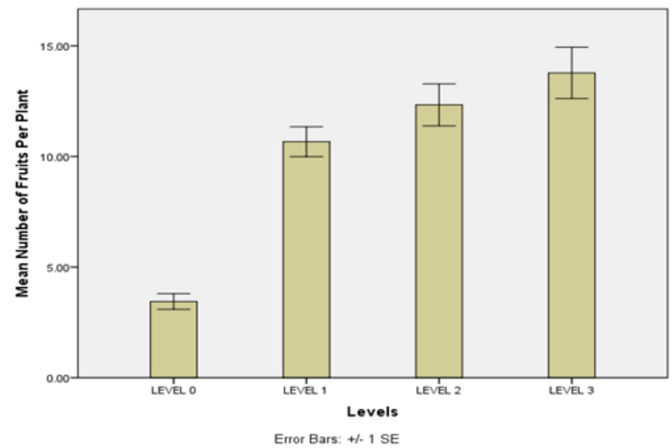

Figure 2b. Effects of Levels of Application of Treatment Materials on Number of Fruits of Okra. Each level represents dosages (0 g, 50 g, 100 g and 150 g)

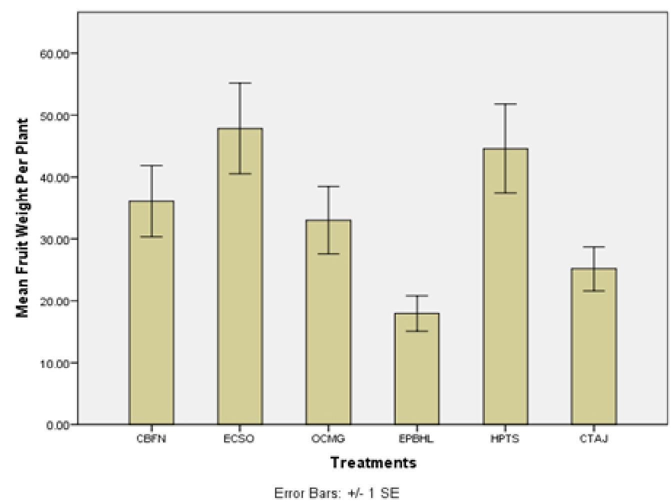

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Figure 3a. Effects of Treatment Materials on Fruit Weights of Okra

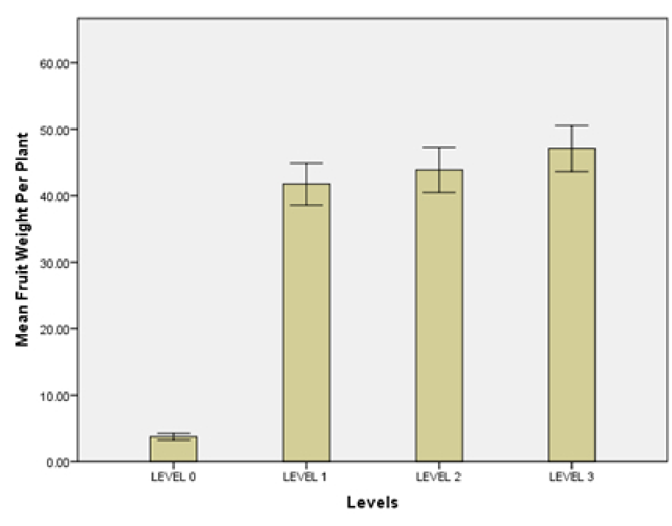

Figure 3b. Effects of Levels of Application of Treatment Materials on Fruit Weights of Okra. Each level represents dosages (0 g, 50 g, 100 g and 150 g)

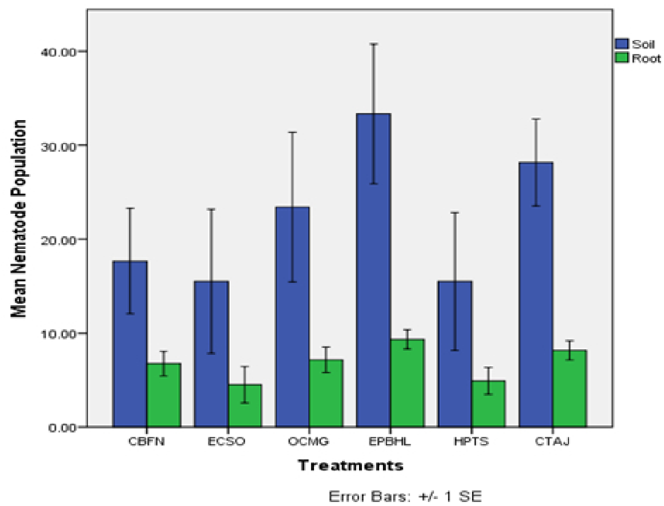

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Figure 4a. Effects of Treatment Materials on Nematode Population in Soil and Roots of Okra

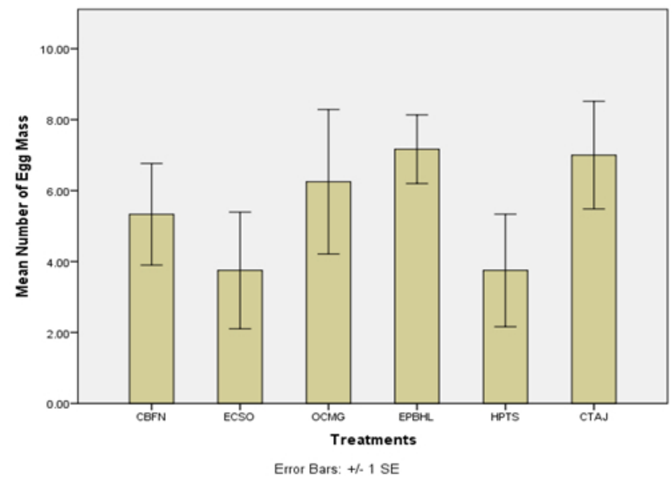

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Figure 5a. Effects of Treatment Materials on Number of Egg Masses

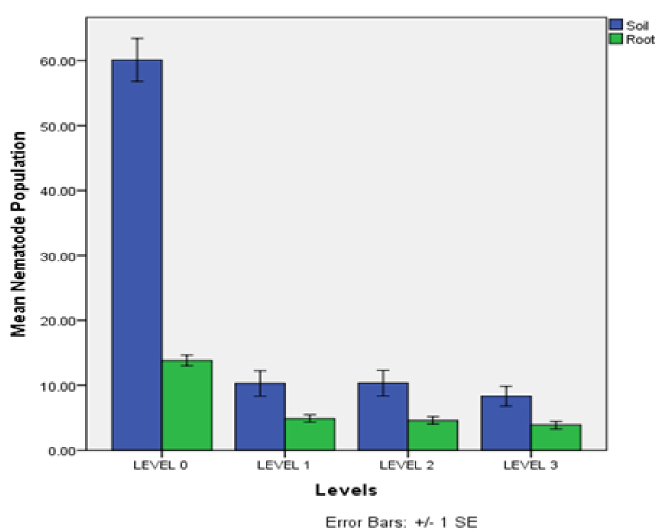

Figure 4b. Effects of Levels of Application of Treatment Materials on Nematode Population in Soil and Root of Okra. Each level represents dosages (0 g, 50 g, 100 g and 150 g)

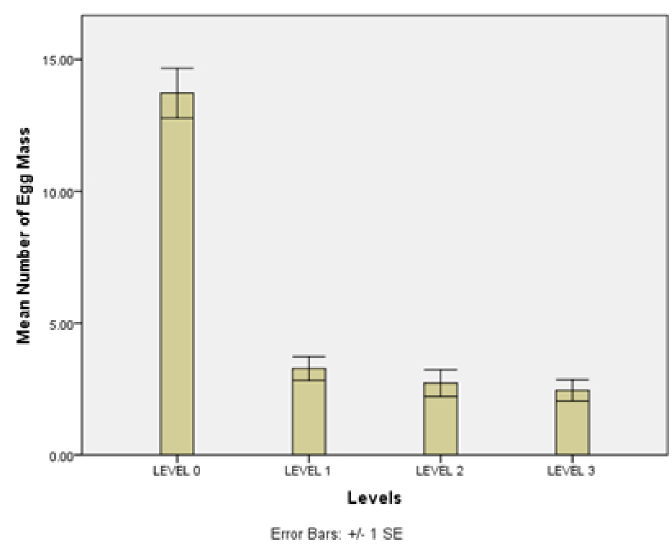

Figure 5b. Effects of Levels of Application of Treatment Materials on Number of Egg Masses. Each level represents dosages (0 g, 50 g, 100 g and 150 g)

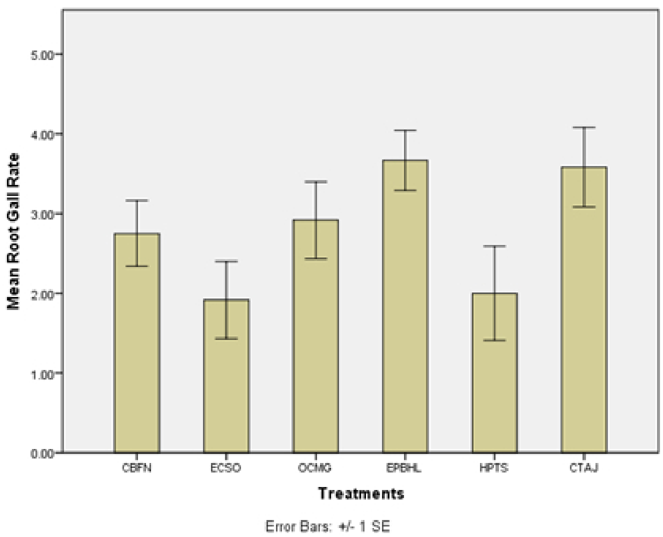

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Figure 6a. Effects of Treatment Materials on Root Gall Rating

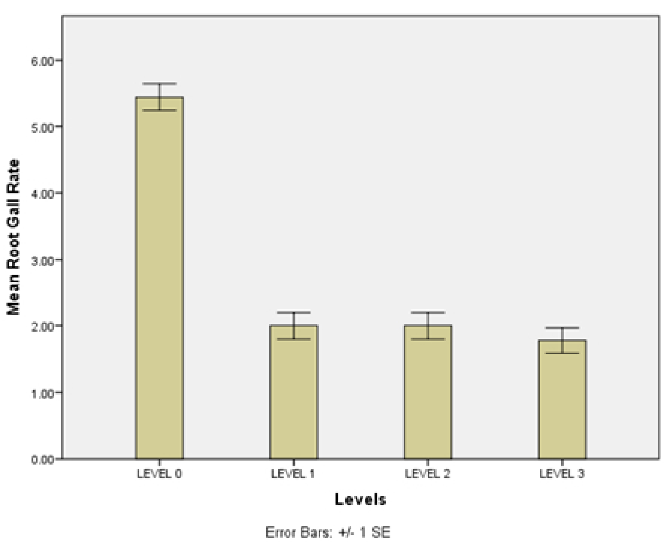

Figure 6b. Effects of Levels of Application of Treatment Materials on Root Gall Rating. Each level represents dosages (0 g, 50 g, 100 g and 150 g)

DISCUSSION

Root knot nematodes are one of the major obstacles in food production (Carter and Sasser, 1982). The use of medicinal plant in the management of PPN is important to reduce the damage caused by these microscopic worms. Among the various plant materials used as soil amendment in this study, *E. officinalis* and *H. suaveolens* (ECSO and HPTS) appear to be the most effective as they exhibited the greatest reduction in nematode population in soil and root, reduced number of egg masses, reduced root galling, with a corresponding higher number of fruits and fruit weights. Moderate nematicidal activity was observed in *O. gratismum* (OCMG) and *C. juncea* (CTAJ), while *E. heterophylla* (EPBHL) was the least effective. In spite of the difference among the various plant materials, they all exhibited a considerable level of effectiveness. The nematicidal activity was higher at the highest level of application. A substantial increase in soil health has been associated with organic soil amendment (Neher, 2001). *O. basilicum* has been indicated in the population reduction of *Xiphinema americanum* infecting date-palm (*Phoenix dactylifera*) Siddiqui et al. (2007). Reports by Hasabo and Noweer (2005) indicated a 61% *M. incognita* mortality with *O. basilicum* with a corresponding 40% increase in fruit weight of eggplant, while Malik et al. (1987) and Sangwan et al. (1990) equally found essential oils from *O. basilicum* to be nematicidal, and moderate nematicidal activity was documented by Mackeen et al. (1997) in their findings on *O. bacilicum*. Fabiyi et al. (2015) reported the toxicity of essential oil, dichloromethane and aqueous extract of *H. suaveolens* to eggs and second stage juveniles of the sugarcane nematode *Heterodera sacchari*. The *in-vitro* anthelmintic properties of *E. heterophylla* on *Ascaris suum* was established by Nalule et al. (2013). Umar and Adamu (2014) stated the nematicidal activity of *E. heterophylla* on egg masses and larvae of *Meloidogyne javanica*, while Anih et al. (2015) reported the nematicidal activity of aqueous and ethanolic extract of *E. heterophylla* on root-knot nematode. Reports by Kushida (2003), substantiated the effectiveness of *C. juncea* on the soybean cyst nematode, *Heterodera glycines*. The lethality of *C. juncea* leaf extract was equally reported by Jasy and Koshy (1994). Studies by Wang (2000) and Wang et al. (2001) also established the toxicity of *C. juncea* leachate on *Rotylenchulus reniformis*. The suppression of *M. incognita* population by *C. juncea*, was also documented by Marahatta et al. (2010). Similarly, the nematicidal properties of fractions from *E. officinalis* were reported by Fabiyi (2016). A 40.70 % juvenile mortality was observed in chromatographic fractions from methanol extract *in-vitro*, while a substantial reduction in *M. incognita* population was achieved in the field.

CONCLUSION

Results obtained in this study demonstrated that *E. heterophylla, H. suaveolens, E. officinalis, O. gratismum* and *C. juncea* are reliable botanicals in the management of PPN. The integration of botanicals offers the development of new strategies in nematode management. It is recommended that field experiments be carried out on a significantly higher number of plants to evaluate the effectiveness of these powdered botanicals.

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