

CHEMICAL CHARACTERIZATION OF *Hyptis suaveolens* AND *Bridelia ferruginea* AND THEIR ASSESSMENT FOR NEMATICIDAL PROPERTIES AGAINST *Meloidogyne incognita* INFECTING SOYBEAN (*Glycine max*)

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ABSTRACT

Soybean production is constrained by root-knot nematodes (RKNs) that attack the root system leading to reduced productivity in Nigeria. Thus, these field and greenhouse experiments investigated the use of powdered leaf extracts of *Hyptis suaveolens* and *Bridelia ferruginea* as plant-based nematicides in the control of RKNs infecting two varieties of soybean. The studies were 2 x 3 factorial experiments fitted into a randomized complete block design. Each pot in the greenhouse was inoculated with 1000 eggs at planting, whereas the field was naturally infested. *Hyptis suaveolens* and *Bridelia ferruginea* were applied at 200 g per stand, twice, at a four-week interval, first at one week after planting. The untreated pots and plots served as the controls for the experiments. Phytochemical screening was conducted on samples of the plant materials to determine the presence of organic compounds. Data collected on growth parameters, pod production, and nematode population were subjected to analysis of variance and means were separated using the Fisher's Least Significance Difference Test at $P=0.5$. The results revealed that the treated plants recorded significantly higher ($P=0.5$) growth and yield than the controls. The untreated TGXP1448-2F plants recorded the highest final nematode populations. In spite of the susceptibility of both varieties to *M. incognita*, powdered form of *Hyptis suaveolens* had significantly higher performance than that of *Bridelia ferruginea*, especially on the LOCAL variety. Phytochemical screening of the botanicals revealed the presence of certain organic compounds including phenol and phytol. The prospects of combined powdered extracts of *Bridelia ferruginea* and *Hyptis suaveolens* as efficacious integrated nematode management strategy in soybean production is intriguing.

Keywords: Soybean, *Meloidogyne incognita*, organic nematicides, *Bridelia ferruginea*, *Hyptis suaveolens*, Phytochemical screening

INTRODUCTION

Soybean (*Glycine max* (L.) Merr.) is an industrial crop supplying approximately half of the world demand for vegetable oil (Nieto and Lorenzo, 2022). It has the highest protein quality among plant proteins and the protein quality is comparable to some animal proteins. Given the risks associated with excessive intake of animal proteins like red meat, soybean can serve as substitute for animal products because, it contains essential amino acids and less fat content unlike other plant proteins hence no major dietary adjustment is needed (Ahnen *et al.*, 2019). Besides its use in livestock feed and industrial purposes, it lowers blood pressure, improves blood vessels, improves bone health, protects against breast cancer, improves cognitive function and visual memory (Rizzo *et al.*, 2018). For these reasons, its global consumption and production is increasing.

However, there is a huge gap in the yield of the crop in Nigeria (1,049,000 T) compared to Brazil (122 MT), USA (113 MT), etc. (FAOSTAT, 2019), yet Nigeria is the second-largest producer of the crop in Africa. This is among other factors, due to the crop's susceptibility to pests and diseases including the cosmopolitan nematodes especially the root-knot nematode that parasitizes its root systems. Chemical nematicides are not only expensive and out of reach for the rural-dwelling farmers, but also poses several serious health and environmental concerns. Thus, the need for cost effective and environment-friendly alternative control measures.

Many plants have been tested and confirmed with varying efficacies, as intriguing phytochemical compounds either as plant extracts or soil amendments, (Prakash and Rao, 2018).

Botanical pesticides are biodegradable, renewable, remain active across various agro-climatic zones, seasons and crops; maximize crop yields, protect the environment, biodiversity and human health; and exhibit selective toxicity to target pests (Neeraj et al., 2017). The pesticidal abilities of extracts from several plant parts including roots, tree barks, leaves, seeds and fruits have been tested on various economically-important plant-parasitic nematodes (PPNs) in crop production (Mokrini et al., 2018).

Hyptis suaveolens, like some other local plants have been reported to exhibit pesticidal activity in certain crops infected by nematodes. Okechalu et al. (2020) reported its nemastatic ability in *Meloidogyne incognita*-infected onions. There are various reports on the chemical composition of *Hyptis suaveolens* (Stevenson et al., 2017; Oscar et al. 2020) and how these components could possibly contribute to its antimicrobial functions. Few studies have thus incorporated *Hyptis suaveolens* in the preparation of environment-friendly nematicides with different results. Anifowose et al. (2023) tested and confirmed that incorporating leaves of *Hyptis suaveolens* in the preparation of effective microorganisms (EM); microbial inoculants that modifies treated soils to become disease suppressive resulted in a EM mix with interesting nematicidal activity. Such microbiota were made possible by compounds present in the EM mix (antioxidants), which are toxic to eggs and/or juveniles of nematodes (Kankam et al. 2015). Furthermore, combining the EM mix with compost manure provided even more tolerance to *Meloidogyne incognita* in sweet potatoes and tomato plants treated with the EM and compost manure mix (Anifowose et al. 2023; Sossou et al. 2022).

The study hence determined response of two varieties of soybean infested with root knot nematode to two plant based extracts.

MATERIALS AND METHODS

Experimental Sites

The field experiment was conducted at the Teaching and Research Farm, University of Ilorin, (8°29' N and 4°35' E) situated within the Southern Guinea Savannah ecological zone, while the pot experiment was carried out in the

Screenhouse of the Department of Crop Protection, University of Ilorin between July and October 2018.

Source of seed and inoculum

Seeds of two varieties of soybean: TGX1448-2F and LOCAL were obtained from a certified agro-allied company in the Ilorin metropolis while the galled roots of *Meloidogyne incognita* infested *Celosia argentea* plants were obtained from the Oyun area, within the Ilorin metropolis, Kwara State.

Preparation of treatments

Leaves of *Hyptis suaveolens* and *Bridelia ferruginea* were obtained from around the University of Ilorin. The leaves were shredded, air dried and and blended into powdered form.

Phytochemical Screening

Samples of the powdered leaves of *Hyptis suaveolens* and *Bridelia ferruginea* were subjected to gas chromatography-mass spectrometry analysis at the Department of Chemical Engineering Laboratory, Faculty of Engineering, University of Ilorin, to determine the bioactive compounds and essential oils present in *Bridelia ferruginea* and *Hyptis suaveolens*

Flushing and running of the samples was carefully done to ensure effective separation of the different constituents of the samples, and the identification of the various bioactive compounds:

Flushing

The extraction solvent, in this case, methanol was used to flush the instrument to remove dirt and achieve uniform results. Three (3) wash bottles were placed in the GC column, two of which contained methanol while the last one was left empty. The syringe took solvent from the first and the second washed bottles three times each into the inner part of the GC and the injector, flushed the solvent and dispose the whole content into the empty bottle provided. This flushing was done repeatedly for 30 minutes (Sarker and Nahar, 2012).

Running of the samples

The running of the sample took place after flushing. The rule is to not use more than 1µml of

the extract so that the toxicity will not affect the machine's effectiveness.

Vial bottles containing the 1µl of the extracts were placed in the GC column (The GC used could run 15 samples at a time). The auto injector drew the extract up from the vial bottle and inject into the GC over a set temperature range for the machine (ion source temperature, 250°C, interface temperature, 300°C). Running of the samples took at least forty-five minutes. Ms Solution software provided by supplier was used to control the system and to acquire the data: Identification of the bioactive compounds and essential oils was carried out by comparing the mass spectra obtained with those of the standard mass spectra obtained from National Institute of Standards and Technology, NIST standard database library software (Mallard and Linstrom, 2008).

Field experiment:

Experimental design

The experimental field was ploughed, harrowed and ridged. It was designed as a 2x3 factorial experiment fitted into a randomized complete block design (RCBD) replicated four time. The first factor was Soybean variety at two levels: TGX1448-2F and LOCAL, and the second factor was the Plant extract source (*B. ferruginea*, *H. suaveolens* and Control).

The experimental field was divided into four blocks which comprised of 6 plots each, each plot measured 10 m x 3 m with alley of one metre. The field experimental plots were naturally infested, but the initial nematode population per 1kg of soil was 80 juveniles

Screenhouse experiments

The experimental design for the screenhouse trial was also a 2 x 3 factorial experiment fitted into a randomized complete block design, and there were five replicates. A total of thirty buckets perforated at the bottom were filled with sterilized soil and placed on elevated platform to avoid reinstestation. The first factor was Soybean variety at two levels: TGX1448-2F and LOCAL, and the second factor was the Plant extract source (*B. ferruginea*, *H. suaveolens* and Control).

Extraction of eggs and juveniles from galled roots

Galled roots of *Celosia argentea* plants was the source of inoculum. Nematode extraction was carried out at the Department of Crop Protection Laboratory and confirmed at the International Institute of Tropical Agriculture (IITA), Ibadan. 10 grams of galled roots with *M. incognita* of *Celosia argentea* plants were washed and chopped into small pieces. The eggs were extracted from the excised galls as described by Hussey and Baker (1973), whereas the Baermann tray extraction tray extraction method was employed to extract the juveniles. Overall, 1000 eggs were obtained from 10 grams of the galled roots and each pot in the screenhouse was inoculated with 10 grams of galled roots.

Treatment preparation

Powdered forms of *Bridelia ferruginea* and *Hyptis suaveolens* were applied at 200g per stand twice: first at one week after planting and finally at four weeks after the first treatment application

Cultural practices

In addition to weeding was maintained on the field and in the screenhouse.

Scarecrow was another cultural practice maintained on the field to ward off birds

Data collection

Data were collected on initial and post harvest nematode counts, plant height, fruit (unshelled) and pod weight (shelled). All numerical data were subjected to analysis of variance and significant difference in means were separated using the Fisher's Least Significance Difference Test at P=0.5.

RESULTS AND DISCUSSION

The effect of variety was not significantly different for all parameters except fruit weight and final nematode populations (Tables 1, 2, 7, 8, 13 and 14). Variety 2 (LOCAL) had significantly lower final nematode population than variety 1 (Tables 13 and 14) and significantly higher pod, fruit, root, and shoot weight than variety 1 (Tables 7 and 8). Plants treated with powdery forms of *Bridelia ferruginea* and *Hyptis suaveolens* had

significantly higher ($p = 0.5$) values for plant height (Tables 3 and 4), fruit weight and pod weight (Tables 9 and 10) than the control plants. Meanwhile, the treatment effect was not significantly different for plant height between plants treated with powdery forms of either of the plant extracts (Tables 3 and 4). And treatment 2 recorded significantly lower final nematode populations than treatment 1 and control.

The effect of interaction between the treatment and variety was significant for all parameters (Tables 5, 6, 11, 12, 17, and 18). Treatment 2 with variety 2 performed significantly higher than with variety 1 and control plants with respect to plant height (Tables 5 and 6) and yield parameters (Tables 11 and 12), and recorded significantly lower final nematode populations (Tables 17 and 18).

The significantly better results recorded in plants of the LOCAL variety of soybean treated with powdered extract of *Hyptis suaveolens* may be because the powdered botanical, *Hyptis suaveolens* used possibly interrupted the life cycle of *Meloidogyne incognita* halting the increase in their population and enhanced the growth of the plants. Izuogu *et al.* (2015) observed that there was 100% inhibition of egg-hatching and 100% juvenile mortality of root-knot nematode when tested with some botanicals. And in 2016, Izuogu *et al.* where in aqueous leaf extract of *Hyptis suaveolens* improved the vegetative growth of cowpea and possibly improved their photosynthetic ability. This is in agreement with the findings of Okechalu *et al.* (2020), who reported that aqueous extract of *Hyptis suaveolens* improved the growth and yield of treated onions compared to their untreated controls.

Poor growth parameters and reduction in the yield of control plants may be attributed to the higher level of soil nematode population in these plants, probably, because they were not treated. Thus, this shows that the two varieties of soybean are susceptible or potential hosts to *Meloidogyne incognita*, and control measures should be directed at them.

The plants treated with *H. suaveolens* and *B. ferruginea* had lower final nematode population than the control counterpart (Tables 9 and 10). This is in agreement with the observation of

Izuogu *et al.* (2016), who observed that *H. suaveolens* caused suppression and reduction of population density of the root-knot nematodes infecting cowpea due to their bioactive ingredients.

The effectiveness of *Hyptis suaveolens* to reduce nematode population as a result of reduced activities of nematode was due to nematicidal substances present in *Hyptis suaveolens*. The major compounds found from phytochemical screening of *Hyptis suaveolens* (Figure 1) were 3,7,11,15-tetramethyl-2-hexadecen-1-ol (12.05%), Phenol, 2,4-bis (1-phenylethyl) (6.14%), 1,2,4-oxidiazol-5(4H)-one, 4-(2-chlorophenyl)-3-(3 pyridyl) (7.6%) and Squalene (22.25%) (Table 11). Whereas n-Hexadecanoic acid (6.99%), Hexadecanoic acid, Ethyl ester (33.54%), Phytol (27.10%), 9,12,15-Octadecatrienoic acid, 2,3-dihydroxypropyl ester, (ZZZ) (21.45%) and Octadecanoic acid, ethyl ester (5.44%) were the major compounds found from phytochemical screening of *Bridelia ferruginea* (Figure 1; Table 12). In vitro studies by Denilson *et al.* (2019), proved that phenol compounds have both direct and indirect nematicidal activity on *Meloidogyne incognita*. Fujimoto *et al.* (2021) reported that exogenous application of Phytol inhibited the penetration of root-knot nematodes into arabidopsis plant although the Phytol did not exhibit any nematicidal activity. Finally, in 2020, Chun-Mei *et al.* reported that a novel strain of *Pseudomonas aureofaciens* produced nematicidal organic volatiles of which octanoic acid ethyl ester demonstrated the strongest nematicidal activity. These reports support the present study and implies that the nematicidal substances in *Hyptis suaveolens* including phenol and those of *Bradelia ferruginea*, Phytol and ethyl ester are responsible for the reduced nematode population density in treated plants.

Consequently, besides the powdery forms of *Hyptis suaveolens* that convincingly suppressed the nematodes, powdery forms of *Bradelia ferruginea* also gave impressive control of the nematodes. The combination of several plants with related compounds whose synergy is effective against nematodes may have an exciting nematicidal effect on nematodes. The high weight of pod and fruits in spite of RKN

infection indicates the tolerance of the crop to nematodes aided by the presence of these treatments.

CONCLUSIONS AND RECOMMENDATIONS

Soybean production, especially, in Nigeria, is constrained by *Meloidogyne incognita*. Powdered leaves of *Hyptis suaveolens* and *Bridelia ferruginea* are excellent nematocidal substances. Thus, the use of *Hyptis suaveolens* and *Bridelia ferruginea* as well as other plant materials could provide effective, eco-friendly and cheap method of controlling root-knot nematode infecting soybean. These plant materials have a wide range of mode of action that would suit possibly, every parasitic nematodes. The materials are readily available as unwanted plants, but could also be deliberately planted on non-arable lands to avoid competition with food crops for space.

Due to the observed increase in yield, reduction in nematode population density and the abundance of *H. suaveolens* as a weed, research should be focused on commercialization of *H. suaveolens*-based nematicides. The challenges impeding their formulation and commercialization should be urgently addressed to ease their adoption and utilization for sustainable plant-parasitic nematodes management. Additionally, the active ingredients in this weed plant could be synthesised to fasten the process and the formulation be made to suit every agro-ecological zone. Further, farmers should be enlightened to combine the powdery forms of *H. suaveolens* with other plant pesticides with similar synergistic effect as an integrated pest management strategy.

Data Availability Statement:

The data recorded on the growth and yield parameters, nematode population, and phytochemical screening are available if need be.

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Table 1: The effect of variety on the mean plant height of two soybean varieties on a nematode infested field

Varieties	1WAP	2WAP	3WAP	4WAP	5WAP	6WAP	7WAP	8WAP
TGXP1448	5.07a	7.03b	9.84b	14.05a	24.76a	35.01a	42.63a	54.30a
LOCAL	5.16a	7.98a	11.13a	15.39a	26.21a	34.81a	43.66a	55.19a
SEM	0.18	0.24	0.31	0.59	0.67	0.74	0.48	0.48

Means with the same letter(s) are not significantly different according to fishers least significant difference at 5% level of significance. KEY: WAP= Week After Planting; *Bridelia*= *Bridelia ferruginea*; *Hyptis* = *Hyptis suaveolens*; SEM= Standard Error of Means, V1= TGXP1448; V2= LOCAL; M.I= *Meloidogyne incognita*; Pract= *pratylenchus*; Pop = Population.

Table 2: The effect of variety on the mean plant height of nematode-infected soybean varieties in the greenhouse

Varieties	1WAP	2WAP	3WAP	4WAP	5WAP	6WAP	7WAP
TGXP1448	12.87a	19.24b	30.10b	47.55b	71.69b	90.67b	101.83a
LOCAL	13.46a	21.83a	34.58a	59.76a	89.17a	107.58a	118.38a
SEM	0.27	0.49	1.09	2.71	4.59	4.50	5.61

Table 3: The effect of treatment on the mean plant height (cm) of *Glycine max* naturally infested by *Meloidogyne incognita* on the field

Treatment	1WAP	2WAP	3WAP	4WAP	5WAP	6WAP	7WAP	8WAP
<i>Bridelia</i>	5.06a	7.88a	10.73b	15.83a	26.36a	35.53a	44.13a	56.30a
<i>Hyptis</i>	5.40a	7.94a	11.86a	16.75a	27.85a	37.14a	45.76a	57.28a
Control	4.88a	6.71b	8.88c	11.59b	22.24b	32.06b	39.55b	50.66b
SEM	0.22	0.29	0.38	0.723	0.818	0.91	0.59	0.59

Table 4: The effect of treatment on the mean plant height of *Glycine max* infected with *Meloidogyne incognita* in the greenhouse.

Treatments	1WAP	2WAP	3WAP	4WAP	5WAP	6WAP	7WAP
<i>Bridelia</i>	12.69b	20.99a	34.63a	58.78a	89.28a	110.50a	114.71a
<i>Hyptis</i>	14.71a	22.41a	35.00a	59.69a	88.75a	107.13a	122.94a
Control	12.09b	18.20b	27.00b	42.50b	63.25b	79.75b	92.65b
SEM	0.33	0.61	1.34	3.31	5.62	5.52	6.87

Table 5: The effect of interaction between the treatment and variety on the mean plant height of two soybean varieties on a nematode infested field

Treatment	Varieties	1WAP	2WAP	3WAP	4WAP	5WAP	6WAP	7WAP	8WAP
<i>Bridelia</i>	V1	5.125a	7.40abc	9.93cd	14.93ab	25.20bc	36.03ab	43.75b	55.70a
<i>Hyptis</i>	V2	5.00a	8.35ab	11.53ab	16.73a	27.53ab	35.03abc	44.50ab	56.90a
<i>Bridelia</i>	V1	5.18a	7.30bc	10.75bc	16.10a	26.55ab	36.48a	45.23ab	57.08a
<i>Hyptis</i>	V2	5.63a	8.58a	12.98a	17.40a	29.15a	37.80a	46.30a	57.48a
Control	V1	4.90a	6.40c	8.85d	11.13c	22.52c	32.53bc	38.93c	50.13b
	V2	4.85a	7.03c	8.90d	12.05bc	21.95c	31.60c	40.18c	51.20b
SEM		0.31	0.41	0.53	1.02	1.16	1.29	0.84	0.84

Table 6: The effect of interaction between the treatment and variety on the mean plant height of nematode-infested soybean varieties in the greenhouse

Treatment	Varieties	1WAP	2WAP	3WAP	4WAP	5WAP	6WAP	7WAP
<i>Bridelia</i>	V1	12.50c	19.88bc	32.00ab	49.28bc	76.08bcd	94.25bc	101.40bc
	V2	12.88bc	22.10ab	37.25a	68.28a	102.50a	119.50a	128.03ab
<i>Hyptis</i>	V1	14.10ab	21.00bc	33.55ab	55.88ab	82.75abc	101.50ab	113.63abc
	V2	15.33a	23.83a	37.25a	63.50a	94.75ab	120.00a	132.25a
Control	V1	12.00c	16.85d	24.75c	37.50c	56.25d	76.25c	90.45c
	V2	12.18c	19.55c	29.25bc	47.50bc	70.25cd	83.25bc	94.85c
SEM		0.47	0.86	1.89	4.69	7.95	7.80	9.72

Table 7: The effect of variety on the pod, fruit, root and shoot weight of soybean on nematode infested field

Varieties	Pod	Fruit	Root	Shoot
TGXP1448	2283.33a	1825b	182.08b	343.75a
LOCAL	2433.75a	2100a	132.08a	363.75a
SEM	73.9	70.6	12.5	14.6

Table 8: The effect of varieties on the pod, fruit, root and shoot weight of nematode-infested soybean varieties in the greenhouse

Varieties	Pod weight	Fruit weight	Root	Shoot
TGXP1448	2.83b	2.08b	3.02b	5.47b
Local	3.33a	2.70a	2.28a	6.53a
SEM	0.08	0.11	0.10	0.105

Means with the same letter(s) are not significantly different according to fishers least significant difference at 5% level of significance. KEY: WAP= Week After Planting; *Bridelia*= *Bridelia ferruginea*; *Hyptis* = *Hyptis suaveolens*; SEM= Standard Error of Means, V1= TGXP1448; V2= LOCAL; M.I= *Meloidogyne incognita*; Pract= *pratylenchus*; Pop = Population.

Table 9: The effect of treatment on mean weight of pod (grams) and mean weight of fruit (grams) per plant at harvest on the field

Treatments	Pod	Fruit	Root	Shoot
<i>Bridelia</i>	2713.00a	2112.5b	156.875a	461.88a
<i>Hyptis</i>	2750.13a	2412.5a	146.250a	374.38b
Control	1612.50b	1362.5c	168.125b	225.00c
SEM	90.5	86.5	8.84	17.9

Table 10: The effect of treatment on the mean pod, fruit, root and shoot weight of nematode-infected soybean at harvest in the screenhouse

Treatments	Pod	Fruit	Root	Shoot
<i>B. ferruginea</i>	2.91b	2.19b	3.02b	6.28b
<i>H. suaveolens</i>	4.36a	3.70a	2.28a	6.99a
Control	1.98c	1.75c	3.99c	4.74c
SEM	0.10	0.14	0.12	0.12

Table 11: The effect of interaction between the treatment and variety on the total yield for pod, fruit, root and shoot weight per plant at harvest on the field

Treatments	varieties	Pod	Fruit	Root	Shoot
<i>Bridelia</i>	TGXP1448	2475.00b	1975b	185.00b	326.25b
<i>Bridelia</i>	LOCAL	2775.00ab	2250b	128.75a	422.50a
<i>Hyptis</i>	TGXP1448	2725.00ab	2200b	140.00a	458.75a
<i>Hyptis</i>	LOCAL	2951.25a	2625a	127.50a	465.00a
Control	TGXP1448	1650.00c	1300c	196.25b	210.00c
	LOCAL	1575.00c	1425c	165.00b	240.00c
SEM		15.8	12.2	7.22	25.3

Table 12: The effect of interaction between the treatment and variety on the mean pod, fruit, root and shoot weight of nematode-infected soybean at harvest in the screenhouse

Treatments	Varieties	Pod	Fruit	Root	Shoot
<i>B. ferruginea</i>	TGXP1448	2.55c	1.73d	2.63c	5.68c
	Local	3.28b	2.65c	1.80ab	6.88b
<i>H. suaveolens</i>	TGXP1448	4.20a	3.35b	2.13bc	5.68c
	Local	4.53a	4.05a	1.38a	7.78a
Control	TGXP1448	1.75d	1.40d	4.30e	4.95d
	Local	2.20c	1.18d	3.68d	4.53d
SEM		0.14	0.19	0.17	0.17

Table 13: The effect of variety on nematode population for soil and root on the field

Varieties	Soil nematode population		Root nematode population		
	Initial M.I pop.	Final M.I pop	Initial prat. pop	Final pract. pop	Final M.I pop
V1	80.00	255.08b	100.00	7.00b	84.75b
V2	80.00	179.96a	100.00	5.42a	68.42a
SEM	0.00	0.53	0.00	0.396	0.64

Means with the same letter(s) are not significantly different according to fishers least significant difference at 5% level of significance. KEY: WAP= Week After Planting; *Bridelia*= *Bridelia ferruginea*; *Hyptis* = *Hyptis suaveolens*; SEM= Standard Error of Means, V1= TGXP1448; V2= LOCAL; M.I= *Meloidogyne incognita*; Pract= *pratylenchus*; Pop = Population.

Table 14: The effect of variety on nematode population for soil and root in the screenhouse

Varieties	Soil nematode population		Root nematode population	
	Initial M.I pop.	Mid M.I pop	Final M.I pop (root)	Final M.I pop (soil)
V1	1000a0	551.83b	594.92b	253.54b
V2	1000a	451.88a	434.75a	199.63a
SEM	0.00	0.94	0.58	0.63

Table 15: The effect of treatment on mean initial and final nematode population in soil and root on the field

Treatments	Soil nematode population		Root nematode population			
	Initial M.I pop.	Final M.I pop	Initial prat pop	Final pract pop	Final M.I pop	Final pract pop
<i>Bridelia</i>	80.00a	20.44b	100.00a	3.00b	14.75b	3.00b
<i>Hyptis</i>	80.00a	14.75a	100.00a	0.13a	10.00a	0.13a
Control	80.00a	617.38c	100.00a	15.50c	205.00c	7.75c
SEM	0.00	0.65	0.00	0.49	0.79	0.42

Table 16: The effect of treatment on the initial and final nematode population in soil and root in the screenhouse

Treatments	Soil nematode population		Root nematode population		
	Initial M.I pop	Mid M.I pop	Final M.I pop	Final M.I pop	Final M.I pop
<i>Bridelia</i>	1000.00	155.19b	75.0b	49.56b	
<i>Hyptis</i>	1000.00	125.38a	65.0a	40.313a	
Control	1000.00	1225.00c	1399.5c	589.875c	
SEM	0.00	1.14	0.71	0.77	

Table 17: The effect of interaction between treatment and variety on nematode population for soil and root on the field

Treatments	varieties	Initial M.I pop.	Final M.I pop	Initial prat. pop	Final pract. pop	Final M.I pop	Final pract pop
<i>Bridelia</i>	V1	80.00a	25.75d	100.00a	5.75b	19.75d	5.75b
	V2	80.00a	15.13b	100.00a	0.25a	9.75b	0.25a
<i>Hyptis</i>	V1	80.00a	19.75c	100.00a	0.00a	14.75c	0.00a
	V2	80.00a	9.75a	100.00a	0.25a	5.25a	0.25a
Control	V1	80.00a	719.75f	100.00a	15.25c	219.75f	9.75c
	V2	80.00a	515.00e	100.00a	15.75c	190.25e	5.75b
SEM		0.00	0.93	0.00	0.69	1.11	0.59

Table 18: The effect of interaction between treatment and variety on nematode population for soil and root in the screenhouse

Treatments	Varieties	Soil nematode population		Root nematode population	
		Initial M.I pop	Mid M.I pop	Final M.I pop	Final M.I pop
<i>Bridelia</i>	V1	1000.00a	170.25d	90.00d	59.88d
	V2	1000.00a	140.13c	60.00b	39.25b
<i>Hyptis</i>	V1	1000.00a	134.75b	75.25c	50.75c
	V2	1000.00a	116.00a	44.75a	29.88a
Control	V1	1000.00a	1350.50f	1599.50f	650.00f
	V2	1000.00a	1099.50e	1199.50e	529.75e
SEM		0.00	1.62	1.0	1.09

Means with the same letter(s) are not significantly different according to fishers least significant difference at 5% level of significance. KEY: WAP= Week After Planting; *Bridelia*= *Bridelia ferruginea*; *Hyptis* = *Hyptis suaveolens*; SEM= Standard Error of Means, V1= TGXP1448; V2= LOCAL; M.I= *Meloidogyne incognita*; Pract= *pratylenchus*; Pop = Population.

Table 18: The effect of interaction between treatment and variety on nematode population for soil and root in the greenhouse

Soil nematode population			Root nematode population		
Treatments	Varieties	Initial M.I pop	Mid M.I pop	Final M.I pop	Final M.I pop
<i>Bridelia</i>	V1	1000.00a	170.25d	90.00d	59.88d
	V2	1000.00a	140.13c	60.00b	39.25b
<i>Hyptis</i>	V1	1000.00a	134.75b	75.25c	50.75c
	V2	1000.00a	116.00a	44.75a	29.88a
Control	V1	1000.00a	1350.50f	1599.50f	650.00f
	V2	1000.00a	1099.50e	1199.50e	529.75e
SEM		0.00	1.62	1.0	1.09

Table 19: Proposed Retention Time, Compounds and Peak Area (%) of *Hyptis suaveolens* using GC-MS

S. No.	Name of compounds	Retention time	Peak area %
1	Eucalyptol	11.677	1.341
2	Caryophyllene	25.361	2.939
3	Bergamotol, Z -alpha -trans	33.507	0.961
4	Bicyclo [3.1.1] heptane, 2,6,6 -trimethyl -, [1R-(1 α ,2 β ,5 α)]	37.294	3.897
5	3,7,11,15 -tetramethyl -2-hexadecen -1-ol	37.820	1.336
6	n-Hexadecanoic acid	38.770	3.565
7	Hexadecanoic acid, ethyl ester	38.872	1.293
8	Naphthalene, decahydro -1,1,4a -trimethyl -6-methylene -5-(3-methylene) -	38.951	1.008
9	1,3,6,10 -Cyclotradecatetraene, 3,7,11 -timethyl -14-(1-methylethyl) - [S-(E,Z,E,E)] -	39.438	0.894
10	3,7,11,15 -tetramethyl -2-hexadecen -1-ol	39.682	12.052
11	9,12,15 -Octadecatrienoic acid, 2,3 -dihydropropyl ester, (Z,Z,Z) -	39.917	4.436
12	Octadecanoic acid, ethyl ester	40.114	0.798
13	Phytol, acetate	40.225	1.807
14	Cyclotetradecane	40.483	1.233
15	1H-Benzotriazole, 1 -ethenyl -	40.6955	0.748
16	Benzene, 1 -(1-buten-3-yl) -4-pentyl	40.758	5.688
17	10H- Phenoxaphosphine, 2,8 -diethyl -10-hydroxy -, 10-oxide	40.821	12.169
18	1-Phenananthrenemethanol, 1,2,3,4,4a,9,10,10a - octahydro -1, 4a -dimethyl -7-(1-methylethyl) -	41.041	2.889
19	Abietic acid	41.221	3.474
20	Phenol, 2,4 -bis(1 -phenylethyl) -	41.308	6.136
21	1-Heptatriacotanol	41.708	1.239
22	1,2,4-oxidiazol -5(4H) -one, 4 -(2-chlorophenyl) -3-(3-pyridyl) -	42.117	7.670
23	Squalene	44.717	22.427

Means with the same letter(s) are not significantly different according to fishers least significant difference at 5% level of significance. KEY: WAP= Week After Planting; *Bridelia*= *Bridelia ferruginea*; *Hyptis* = *Hyptis suaveolens*; SEM= Standard Error of Means, V1= TGXP1448; V2= LOCAL; M.I= *Meloidogyne incognita*; *Pract*= *pratylenchus*; Pop = Population.

Table 20: Proposed Retention Time, Compounds and Peak Area (%) of *Bridelia ferruginea* using GC-MS

S. No.	Name of compound	Retention time	Peak area %
1	2-Fluoro-4-(trifluoromethyl)acetophenone	24.341	1.024
2	2-6-Pentadecen-1-ol acetate	36.351	1.653
3	n-Hexadecanoic acid	37.696	6.987
4	Hexadecanoic acid, ethyl ester	37.802	33.544
5	Phytol	38.578	27.111
6	Linoleic acid ethyl ester	38.834	2.790
7	9,12,15-Octadecatrienoic acid, 2,3-dihydroxypropyl ester, (Z, Z, Z)-	38.872	21.452
8	Octadecanoic acid, ethyl ester	39.009	5.439