

Biofumigation Potentials of Some Brassica Crops to Control Root-knot Nematodes *Meloidogyne* spp., on Tomato Under Field Conditions

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ABSTRACT

The experiments were conducted to study the efficacy of fodder radish (*Raphanus sativus* var. Terranovah) and rocket salad (*Eruca sativa* cv. Baladi) as biofumigation crops for controlling root-knot nematodes *Meloidogyne* spp., on tomato plants during two successive seasons 2017 and 2018 under field conditions. Three months after cultivation of fodder radish, and rocket salad (the full blooming stage) all parts were incorporated with soil and covered with a transparent polyethylene film. After 4 weeks, the plastic sheets were removed and soil was left for two weeks later, before transplanting tomato seedlings. Results in both seasons, indicated significant reduction ($p \leq 0.05$) of nematode parameters on tomato plants. Results revealed that effect of *R. sativus* var. Terranovah as biofumigation crop as indicated by the percentage reduction in number of galls, egg-masses/root system, and a number of second-stage juvenile (j_2)/250 g soil (84, 90, and 84%), and (90, 87, and 88%) in seasons 2017 & 2018 respectively. On the other hand, nematicide Vydate (oxamyl) 24% L recorded the percentage reduction in the number of galls, egg-mass/ root system as well as number of second-stage juveniles in soils j_2 (90, 87, and 87%), and (95, 93, and 90%) in season 2017 & 2018 respectively. Results revealed that all plant growth criteria on tomato plants increased significantly ($p \leq 0.05$) by using the tested treatments. Results indicated that the effect of *R. sativus* as biofumigation crop recorded the average highest percentages (57, and 92%), and (64, and 102%) in seasons 2017 & 2018 respectively. At the same time, nematicide Vydate (oxamyl) 24% L was the most effective in the average highest percentages. i.e., the plant growth vigor, increase, and fruit yield per plant (64, and 98), (73, and 107) in two successive seasons 2017 & 2018 respectively.

Gas liquid chromatography-mass spectrometry (GLC-MS) analysis of dichloromethane extract of fodder radish indicated the presence of four glucosinolates, which were identified through their volatile autolysis products. Gluconapin, the major compound was identified by 3- butenyl isothiocyanate, while glucoerucin was identified by 4-(methylthio) butyl isothiocyanate, which is commonly known as erucin. Sulphoraphane was released from 4-(methylsulfinyl) butyl glucosinolate (glucoraphanin) while 4-(methylsulphony) butyl isothiocyanate commonly known as erysolin was liberated from glucoerysolin. Furthermore, five GLS were also identified in rocket salad. Gluconapin was detected and its presence was identified through its epithionitrile; 4,5-epithiopentanenitrile. The isomers progoitrin and epiprogoitrin were detected by the two hydrolysis products diastereomers threo and erythro 1-cyano-2-hydroxy-3,4-epithiobutane respectively. The aromatic glucosinolate; gluconasturtiin could be identified through its liberated nitrile named as 1-benzenepropane nitrile. Sativin, the major identified compound is 4-mercaptobutyl isothiocyanate.

Keywords: *Meloidogyne* spp; tomato (*Solanum lycopersicum* L.); biofumigation; *Raphanus sativus*; *Eruca sativa*; Isothiocyanates; Glucosinolates.

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INTRODUCTION

Tomato (*Solanum lycopersicum* L.) is the second most important vegetable crops in the world next to potato with approximately 182.3 million tons of tomato fruits produced on 4.85 million ha each year, Europe, America, and Africa produced 13.5, 13.4, and 11.8% of the total tomato yield, respectively [1]. Egypt is the fifth country in the world in terms of the production of tomatoes. It's grown all year round. The cultivated area reached 475.514 thousand feddan produced approximately 7.94 million tons [2].

Among the several pests and diseases that affect tomato plants, the plant-parasitic nematodes pose a major threat, resulting in an estimated annual monitoring loss of USD 80 billion [3]. The most prevalent and destructive among the phytonematodes of tomato are *Meloidogyne* spp., which causes major economic losses on vegetables, especially in tropical and subtropical areas [4-6]. In Egypt, tomatoes are known to be highly susceptible to root-knot nematodes *Meloidogyne* spp., caused annual losses of 12% [7].

The continuous use of chemical nematicides to control root-knot nematode had a considerable environmental impact and has resulted in the onset of resistance phenomena within some populations of nematode pests, this situation has led to an increased demand for environment-friendly products in order to reduce the effects of widespread nematicides utilization in crop protection [8].

Several studies reported the ability of certain plants to suppress nematodes through the nematicidal activity of the secondary metabolites [9,10]. Several researches on biofumigation had focused on using brassicaceous crops [11]. The suppressive effect of brassicaceous, biofumigants on Phyto-nematodes were explained in numerous studies under *in vitro*, *in vivo*, and field conditions [12-14]. The mechanism responsible for the biocidal effect of decomposing brassica crops was thought to be based on a chain of chemical reactions ultimately resulting in the formation of biologically active products [15]. Brassicaceae plants contain glucosinolate compounds, which are β -D thioglucosides, sulphur containing stable and non-toxic compounds located in the cell vacuoles distinguished from one another by differences in their organic side chains (R groups) and classified as aliphatic, aromatic or indole forms, occur in all parts of the plant and degrade via enzymatic hydrolysis [16]. The fumigant action of these volatile compounds that are released, suppresses plant pathogens soil-borne pathogens [17,18].

The aim of this study was to determine the biofumigation effects of fodder radish, and rocket salad on the root-knot nematodes, *Meloidogyne* spp., and tomato yield under field conditions in addition to identification of bioactive compounds of the two plants under investigation using Gas liquid chromatography-mass spectrometry (GLC-MS) and correlate between the glucosinolate autolysis products and nematicidal activity.

MATERIALS AND METHODS

Source of plant seeds

Two brassica plants, i.e., fodder radish (*Raphanus sativus* var. Terranovah-4-169/0300), was obtained from Joordens Zaden Company, Netherlands. Rocket salad (*Eruca sativa* cv. Baladi) was obtained from Horticultural Research Institute, Agriculture Research Center, Egypt, Giza.

Field experiments

The experiments were conducted at naturally infested with root-knot nematodes *Meloidogyne* spp., in the experimental farm of Faculty of Environmental Agricultural Sciences, Arish University during two successive seasons of (2017, and 2018) using the same field design plots of applied treatments. The experiments were divided into 16 plots, each plot (2.5 × 3.5 m²), all treatments were added in the two seasons as follows:-

1. Biofumigation crops of fodder radish (*Raphanus sativus* var. Terranovah).
2. Biofumigation crops rocket salad (*Eruca sativa* cv. Baladi).
3. Nematicide treatment Vydate (oxamyl) 24% L.
4. Control treatment (without any treatment).

Preparation of soil

Sixteen individual plots were arranged 2.5 × 3.5² with the 1m interval between cross plots. Plots were plowed to 30 cm depth by the garden tiller. Seeds of (fodder radish, and rocket salad) were sterilized before using and sowed in arranging plots as a recommended amount (2.4kg/Fed) (15 December 2017). Vydate (oxamyl) 24% L as a nematicide was used at 4L/Fed (recommended dose) was used for comparison. Control treatment kept free without any treatment. The experiments were in a completely randomized block design with four replicates.

Incorporation of brassica plants and evaluation of their biofumigation effects on tomato plants

After 3 months (15 March 2018) at the full blooming stage (3 months after cultivation), 4 plants were randomly selected from each replicate. Shoots and roots were dried at 70°C till a constant weight to determine both roots, shoot and whole plant dry weights (g/plant). After that, the dried plant materials were subject to the determination of C/N ratio of the whole plant.

Another 4 plants were used to determine nematicidal phytochemical profiles using GIC-MS. The brassica plants were incorporated into the top 25-30 cm of the soil by a garden tractor with a rotary tiller. The plots were irrigated to field capacity and covered with a transparent polyethylene film (50-micron thickness) in order to prevent any evolved gasses from escaping to the atmosphere, as well as, to increase the temperature to accelerate the decomposition process of brassica as biofumigation crops. Irrigation was continued at 3 day-interval during the decomposition period (four weeks). After that, plastic sheets were removed, and the soil was left for two weeks before tomato transplanting. Tomato seedlings were planted, and all soil partitions were planted at the same time with a tomato seedling (*Solanum lycopersicum*) cv. "Elisa"35 days old with 4-5 true leaves". Seedlings were transplanted in each plot at a distance of 40 cm in the row and 40 cm between rows. Tomato plants were grown by drip irrigation.

Three months after transplanting tomato seedlings, harvest was done and 10 tomato plants from each plot were randomly selected and the growth parameters were: shoot and root length (cm), fresh shoot and root weights (g) as well as dry weight (g). Also, fruit yield weights (g) were recorded. Increasing percentages fresh weight of the whole plant, and fruit yield (g) % = Treatment - control/control × 100.

The roots of these plants were carefully removed and recorded numbers of galls, gall index, number of egg masses/ root system, as well as, number of juveniles in each plot [19]. Root galling was estimated according to [20], whereas: 0= no galls or egg-mass 1= 1-2 galls or egg-mass 2= 3-10 galls or egg-mass 3= 11-30 galls or egg mass 4= 31-100 galls or egg-mass 5= more than 100 galls or egg-mass. Egg-masses were stained prior to counting by dipping the infected roots in phloxine-B solution (0.015%) for 20 minutes as described by Daykin et al. [21]. Reduction percentages of root knot nematode galls, egg-masses numbers/ root system, and number of the second-stage juveniles (J_2) in 250 g of soil were calculated in comparison with a control treatment.

Preparation of glucosinolates hydrolysis products by natural autolysis

Each plant under investigation (20 g fresh weight) was homogenized with distilled water (200 mL) and left for natural autolysis overnight (15 hours) at room temperature ($\approx 25^\circ\text{C}$). Each mixture was shaken with dichloromethane (50 mL) for 20 min and centrifuged for 5 min at 3500 rpm. Anhydrous sodium sulfate was used to dry the separated organic layer which then concentrated under nitrogen to about 0.5 mL and kept (in a tightly closed vial) in deep freezer until analysis [22].

Gas liquid chromatography-mass spectrometry (GLC-MS)

1 μL of dichloromethane extract of each plant autolysate was injected

into an Agilent 6890 gas chromatography (USA) equipped with PAS-5MS capillary column (30 m \times 0.32 mm; 0.25 μm film thickness), attached to an Agilent 5973 quadrupole mass spectrometer. The injector temperature was 250°C and the temperature program started at 45°C isothermal for 3 min and raised to final temperature 280°C at $5^\circ\text{C}/\text{min}$, 10 min isothermal. Helium was the carrier gas (1 ml/min). The mass spectrophotometry detector was operated in an electron impact ionization mode and ionizing energy of 70 eV scanning from m/z 40 to 500. The temperature of ion source was 230°C . The percentage composition was computed from the total ion chromatogram peak areas (Table 1).

Statistical analysis

All experiments were performed twice in a completely randomized design with three replicates in each treatment. Data were subjected to analysis of variance (ANOVA) using MSTAT-C program version 2.10 [23]. Means were compared by Duncan's multiple range test at $P = 0.05$ probability [24].

RESULTS AND DISCUSSION

Effect on nematode population

Results obtained in both seasons 2017 and 2018 were almost identical Table 2 indicated that significant ($p \leq 0.05$) reduced nematode parameters on tomato plants, under field conditions. Average percentages of nematode population reduction were

Table 1: Chemical analysis of two studied brassica species and its dry weight (g/plant).

Brassica species	Root (g)	Shoot (g)	Whole plant (g)	Root/Shoot ratio	Organic matter, %	Total Nitrogen, %	Organic Carbon, %	C/N ratio	Protein %
<i>R. sativus</i> var. Terranovah	9.32	39.22	48.54	0.24	29.1	3.10	33.3	10.1	19.4
<i>E. sativa</i> cv. Baladi	2.74	18.96	21.70	0.14	20.8	3.20	29.5	9.20	20.0

Table 2: Effect of Brassica as biofumigation crops (*R. sativus*, and *E. sativa*) on root galling and nematode reproduction of *Meloidogyne* spp., under field conditions during successive seasons 2017 & 2018.

Season, 2017									
Treatment	No. galls/ root system	Reduction (%)	Root gall index	No. egg masses/ root system	Reduction (%)	Egg masses index	No. J_2 /250g of soil	Reduction (%)	
<i>R. sativus</i> var. Terranovah	13.3c	84	3	4c	83	2	78c	84	
<i>E. sativa</i> cv. Baladi	21.3b	75	3	10.3b	55	2	115b	77	
Vydate (oxamyl) 24% L	8c	90	2	3c	87	2	61.6c	87	
Control (without any treatment)	85.3a	-	4	23a	-	3	491.3a	-	
Season, 2018									
<i>R. sativus</i> var. Terranovah	12c	90	3	3.6c	87	2	66 c	88	
<i>E. sativa</i> cv. Baladi	21b	82	3	7.6b	73	2	130.6b	75	
Vydate (oxamyl) 24% L	5.6c	95	2	2c	93	1	52c	90	
Control (without any treatment)	115.3a	-	5	28a	-	3	528a	-	

Data are average of 4 replicates.

*Different letter (s) indicate significant differences among treatments within the same column according to Duncan's multiple range test ($P \leq 0.05$). * Root gall index (RGI) or egg-masses index (EI) was determined according to Taylor and Sasser (1978) where G.I and E.I were determined as follows: 0 = no galls, 1= 1-2, 2 = 3-10, 3 = 11-30, 4 = 31-100, 5= more than 100 galls or egg- masses per root system.

calculated compared with control treatment. In general the effect of *Raphanus sativus* var. Terranovah, as biofumigation crop recorded the average of the percentage reduction in number of galls, egg-masses/root system, and a number of second-stage juvenile (j_2)/250 g of soil (84, 90, and 84%) respectively in season 2017. While, were (90,87, and 88%) in season 2018 respectively. On the other hand, *ErUCA sativa* cv. Baladi as biofumigation crop recorded (75, and 82%) of the percentage reduction in number of galls, (55% and 73%) egg-masses/root system, and a number of second-stage juvenile (j_2)/250 g of soil (77% and 75%) in seasons 2017 & 2018 respectively. The ability of certain plants to suppress nematodes through the nematicidal activity of the secondary metabolites has been reported [25]. Research has furthermore proved that many Brassicaceous species show nematicidal efficacy of plant-parasitic nematodes [26,27]. The plant species that generally are considered for biofumigation are found mostly in family Brassicaceae and include *ErUCA sativa* (rocket salad), and *Raphanus sativus* (radish), [28,29]. The present results are agreement with those obtained by Aydınli et al. [30] reported that used radish (*Raphanus sativus*) and rocket salad (*ErUCA sativa*) as biofumigation crops decreased gall index and egg masses of root-knot nematode *M. arenaria* on tomato plants under the greenhouse conditions. The same trend was noticed with Youssef [31], who reported that used two mashed leaves of plants belonging to the genus brassica, cabbage (*Brassica oleracea*) and kohlrabi (*Brassica caulorapa*) were used as pre-and at sowing biofumigants for managing root-knot nematode, *Meloidogyne incognita* on cowpea plants, under greenhouse conditions. They found that the addition of the plant's residue cabbage and kohlrabi 10 days before sowing gave the highest percentages of final nematode reduction (70.4, and 82.1%) respectively. Also, when the C/N ratio of the amendment is less than 20:1, more effect on nematodes is evident [32,33]. On this basis, (*R. sativus* and *E. sativa*) residue used in this study, have a C/N ratio (10.1, and 9.20) so more effect of its toxic biofumigants on the nematode population.

Effect on tomato growth and yield

As shown in Table 3 all plant growth parameters on tomato plants significant increased ($p \leq 0.05$) by using the tested treatments. The obtained results indicated that the effect of *R. sativus* as biofumigation crop recorded the average highest percentages. i.e., the plant growth vigor, increase, and fruit yield per plant compared with control treatment. The increase in fresh weight of whole tomato plant, and fruit yield were (57, and 92%), and (64, and 102%) in seasons 2017 & 2018 respectively, this may be attributed to the relatively high biomass accumulation by fodder radish (48.54g/plant) Table 1. The same trend occurred for *E. sativa* as biofumigation crop increased in fresh weight of whole tomato plant, and fruit yield were (57, and 79%), and (46, and 93%) in seasons 2017 & 2018 respectively. The present results are agreement with those obtained by Anita [6] who, reported that ethanol extracts of cabbage, cauliflower, radish and chinese cabbage leaves reduced population of *M. hapla* and improved celery plant growth criteria, of which, radish leaf residue increase 41.9% in celery green leaves and stalk yield.

Gas liquid chromatography-mass spectrometry (GLC-MS) of *R. sativus* and *E. sativa*

Analysis of dichloromethane extract of fodder radish and rocket salad indicated the presence of glucosinolates hydrolysis products including isothiocyanate, nitriles and epithioalkane nitriles. Total ion current chromatogram of the GLC analysis of dichloromethane extract of autolysis products of fodder radish, and rocket salade, were shown in Tables 4 and 5, and Figures 1 and 2. The identification was based upon a comparison of mass fragments and their relative intensities with the available literature [22, 34-36] and Wiley 9th edition NIST11 (W9N11) USA, database libraries.

Bioactive compound names, retention time, relative percentage, and mass spectral data of each compound in each plant extract of glucosinolates hydrolysis products were listed in Tables 4 and 5.

Table 3: Effect of *Brassica* as biofumigation crops (*R. sativus* and *E. sativa*) on the growth of tomato infected with *Meloidogyne* spp., under field conditions during successive seasons 2017 & 2018.

Treatments	Shoot length (cm)	Root length (cm)	Shoot fresh weight (g)	Root fresh weight (g)	Fresh weight of whole plant (g)	Inc%	Shoot dry weight (g)	Root dry weight (g)	Fresh fruit weight/ plant (g)	Inc%
Season , 2017										
<i>R. sativus</i> var. Terranovah	56.7a	33.6a	287.8b	72.6a	360.4b	57	91b	33b	448.6a	92
<i>E. sativa</i> cv. Baladi	49.6b	28 b	264.6c	65.6b	330.2c	57	77.6c	27.3c	418.3b	79
Vydate (oxamyl) 24% L	58.2 a	33a	302a	75a	377a	64	96.3a	36.6a	463a	98
Control (without any treatment)	28.9c	19.2c	186d	44c	230d	-	60.3d	20d	233.3c	-
Season , 2018										
<i>R. sativus</i> var. Terranovah	64.3b	37a	285.6b	77b	362.6b	64	94b	34a	491b	102
<i>E. sativa</i> cv. Baladi	51c	29.6b	258.3c	64c	322.3c	46	80.6c	26.3b	468.3c	93
Vydate (oxamyl) 24% L	67.6a	37a	299.3a	83a	382.3a	73	104a	35a	504a	107
Control (without any treatment)	42.6d	20.3c	182.3d	39d	221.3d	-	34.6d	19.3c	243d	-

Data are average of 4 replicates. *Different letter (s) indicate significant differences among treatments within the same column according to Duncan's multiple range test ($P \leq 0.05$ Inc (%) = treatment - control / control \times 100.

Table 4: Glucosinolates hydrolysis products obtained by natural autolysis of *R. sativus* var. Terranovah (Fodder radish).

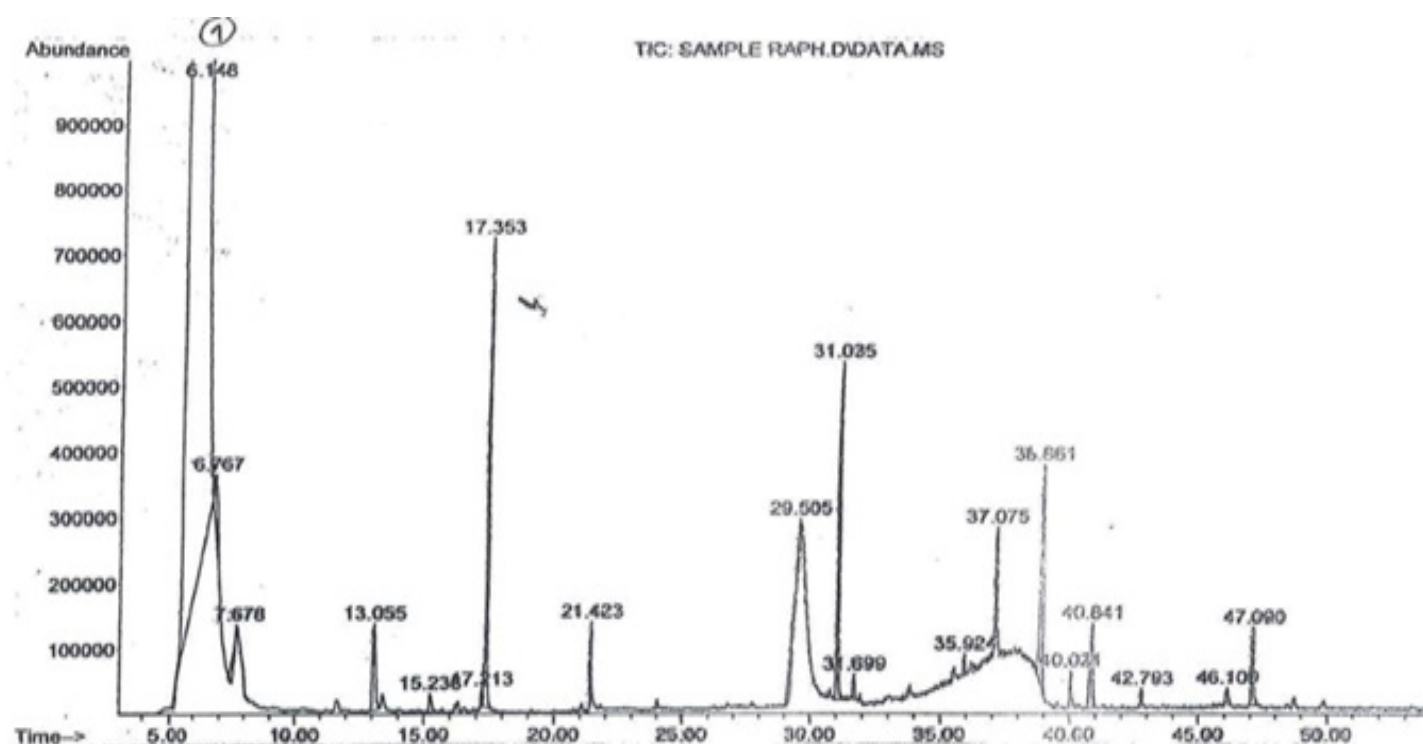
Parent compound Hydrolysis product	Rt (min)	Relative (%) of autolysis products	Major mass fragments*
Gluconapin 3- Butenyl isothiocyanate	17.35	14.15	113 (M ⁺), 72 and 55
Glucoerucin 4- (Methylthio) butyl Isothiocyanate (erusin)	31.033	9.4	161 (M ⁺), 146, 115, 100, 72 and 61
Glucoraphanin 4- (Methylsulfinyl) butyl isothiocyanate (sulforaphane)	38.85	10.37	177 (M ⁺), 160, 114, 72 and 55
Glucoerysolin 4- (Methylsulphony) butyl isothiocyanate (erysolin)	40.02	9.4	193, 135, 114, 86, 72, 55 and 41

*: Base ion in bold

Table 5: Glucosinolates hydrolysis products obtained by natural autolysis of *E. sativa* cv. Baladi (Rocket salad).

Parent compound Hydrolysis product	Rt (min)	Relative (%) of autolysis products	Major mass fragments*
Gluconapin 4,5-Epithiopentanenitrile	22.06	1.2	113 (M ⁺), 86, 80, and 73
Progoitrin (R) 1-cyano-2-hydroxy-3,4-epithiobutane (Threo)	25.35	1.2	129 (M ⁺), 96, 89, 83, 61, 55 and 40
Gluconasturtiin 1-Benzenepropane nitrile	25.63	1.2	131 (M ⁺), 91, 77, 65 and 52
Epiprogoitrin (S) 1-cyano-2-hydroxy-3,4-epithiobutane (Erythro)	25.81	1.2	129 (M ⁺), 111, 96, 89, 83, 61, 59, 55 and 41
Glucosativin 1,3-Thiazepane-2-thione, (4-Mercaptobutyl isothiocyanate) (sativin)	38.21	2.38	175 (M ⁺), 114, 87, 72 and 55

*: Base ion in bold

**Figure 1:** Total ion current chromatogram of GLC analysis of fodder radish.

Results in Figure 1, and Table 4, indicated that four glucosinolates were identified in fodder radish through their volatile autolysis products. Gluconapin, the major compound was identified by 3- butenyl isothiocyanate while glucoerucin was identified by 4-(methylthio) butyl isothiocyanate, which is commonly known as erucin. Sulforaphane was released from 4-(methylsulfinyl) butyl glucosinolate (glucoraphanin) while 4-(methylsulphony) butyl isothiocyanate commonly known as erysolin was liberated from glucoerysolin.

The results indicated that rocket salad autolysate had five GLS Figure 2, and Table 5. Gluconapin was detected in rocket salad

also and its presence was identified through its epithionitrile; 4,5-epithiopentanenitrile. The isomers progoitrin and epiprogoitrin were detected by the two hydrolysis products diastereomers threo and erythro 1-cyano-2-hydroxy-3,4-epithiobutane respectively. The aromatic glucosinolate; gluconasturtiin could be identified through its liberated nitrile named as 1-benzenepropane nitrile. Sativin, the major identified compound is 4-mercaptobutyl isothiocyanate liberated from glucosativin according to several researchers [37-39]. Fechner et al., [40] revealed that sativin was proved to be 1,3-thiazepane-2-thione, a tautomer of 4-mercaptobutyl isothiocyanate with cyclic 7-membered ring structure according to NMR data.

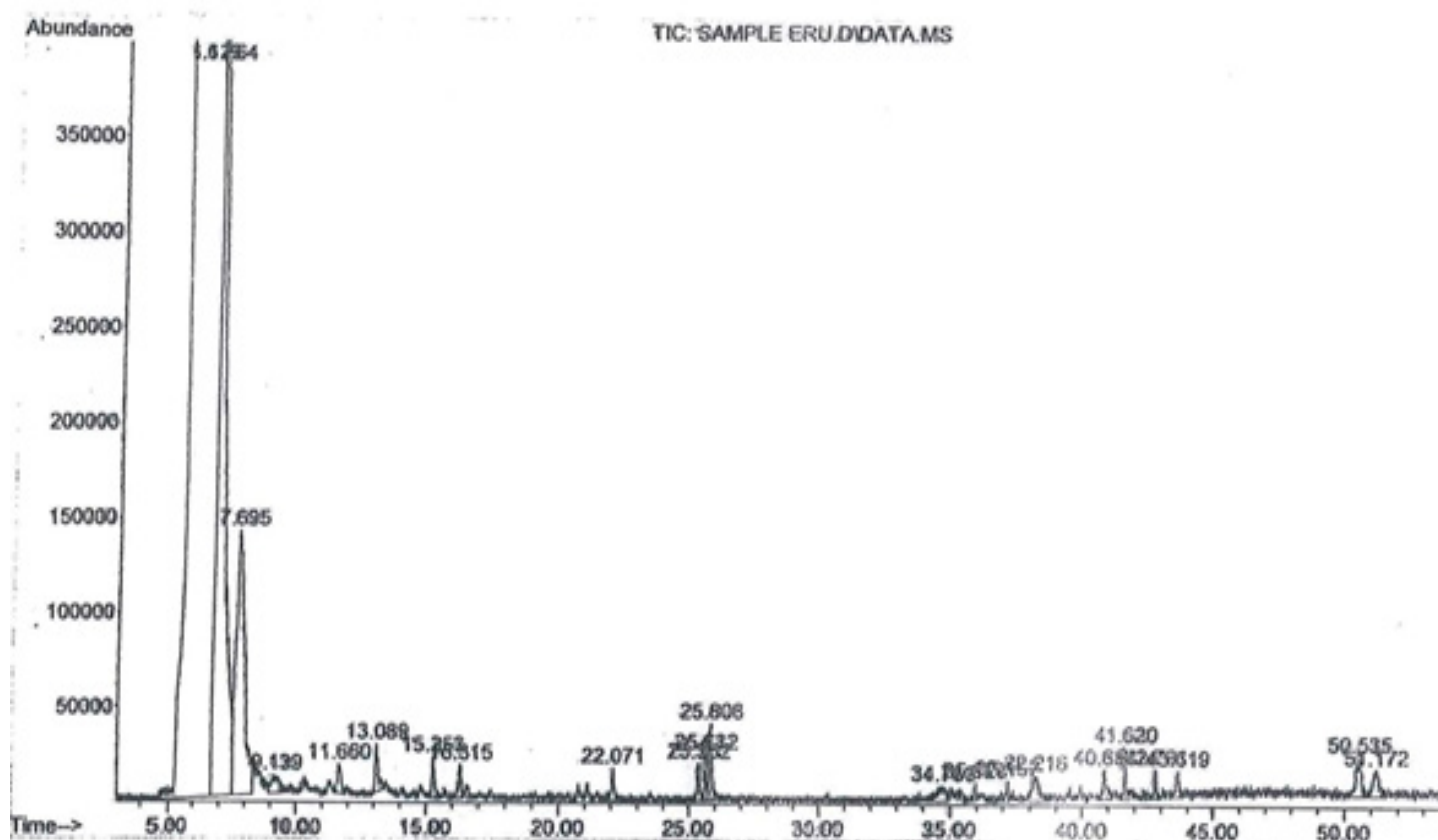


Figure 2: Total ion current chromatogram of GLC analysis of rocket salad.

Gluconapin, glucosinolate (glucoraphanin) 4-(methylsulphony) butyl isothiocyanate. An alternative management strategy that is increasingly receiving interest is biofumigation that are a sustainable approach to manage soil-borne pathogens, nematodes, and weeds. This process was defined as a process that occurred, when volatile compounds with pesticidal properties were released during the decomposition of plant materials or animal products [41]. Some isothiocyanates such as benzyl isothiocyanate, methyl isothiocyanate, and phenyl isothiocyanate have been extracted from brassica species and showed nematicidal effects [42]. Other isothiocyanates also showed some nematicide effects, such as ethyl isothiocyanate benzyl thiocyanate, 1-phenylethyl isothiocyanate, and 2-phenylethyl isothiocyanate. However, acryloyl isothiocyanate and allyl isothiocyanate showed the highest effect against nematodes [43]. Glucosinolates and their derivatives, such as isothiocyanates, isolated from various brassica species differ in their toxicity against nematodes. Species containing benzyl or 2-phenylethyl and smaller level allyl isothiocyanate were more effective against *Tylenchulus semipenetrans*, and *M. javanica*, when compared to butyl, ethyl, methyl, phenyl, and 4-methylsulfinyl isothiocyanate [44]. Glucosinolates are one of the most frequently studied groups of defensive secondary metabolites in plants. Upon cellular disruption, e.g. wounding by nematodes, the thioglucoside linkage is hydrolyzed by endogenous enzymes (myrosinases), resulting in the formation of products (e.g. isothiocyanate, thiocyanate, nitrile, epithionitrile, oxazolidine-2-thione) that are active against herbivores and pathogens [45,46]. For instance, glucosinolates purified from Brassicaceae (*Brassica napus*, *B. rapa*, *B. carinata*, *Lepidium sativum*, *Raphanus sativus*, and *Sinapis alba*) were not toxic to J_2 s of the cyst nematode *Heterodera schachtii* in their original form, but enzymatic hydrolysis products of glucosinolates (isothiocyanate, sinigrin, gluconapin, glucotropaeolin, dehydroerucin) were lethal to the nematode [47].

Similarly, 11 glucosinolates and their degradation products did not affect J_2 s of the root knot nematode *M. incognita*, but myrosinase hydrolysis products (gluconasturtiin, glucotropaeolin, glucoerucin, and sinigrin) were highly toxic [48]. Other studies also report that glucosinolates are only lethal to the cyst nematode *Globodera rostochiensis* in the presence of myrosinase [49].

CONCLUSION

From the obtained results, it can be concluded that (fodder radish, and rocket salad) when were utilized as biofumigation crops and compared to nematicide Vydate (oxamyl) 24% L, were effective in reducing population density of root-knot nematode *Meloidogyne* spp., infecting tomato plants and improved plant growth parameters. The highest percentage of the nematode population reduction occurred when of *R. sativus* as a biofumigation crop in two successive seasons 2017 & 2018 respectively.

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