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PHYTOCHEMICAL SCREENING, CHARACTERIZATION OF ESSENTIAL OIL AND ANTIMICROBIAL ACTIVITY OF SCHINUS MOLLE (ANACARDIACEAE) COLLECTED FROM EASTERN HARARGHE, ETHIOPIA

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Abstract

In the present study, antibacterial activity of leaf, stem bark and root bark extracts of Schinus molle (Anacardiaceae) was evaluated against two bacterial (Xanthomonas and Ranstonia) and two fungal (Aspergillusniger and Fusariumvert) species using agar diffusion method. The stem and root ethanol extracts of Schinusmolle exhibited relatively higher zone of inhibition (11.3mm) against Xanthomonas campestris, pv. Campestris. Higher zone of inhibition (10mm) was also observed by the ethanolic extracts of the root of Schinus molle against Fusarium verticillioides followed by the chloroform extracts of the root (8mm) against Aspergillus niger. Phytochemical analysis of leaf, stem bark and root bark extracts of Schinus molle revealed the presence of flavonoids, alkaloids, saponins, steroids, terpenoids, phenols, quinines, carbohydrates and proteins. The occurrence of these biologically active chemicals in the plants parts may justify their wide usage in traditional medicine. From GC-MS analysis, three terpenoids namely monoterpene (66.02%) as a major components, sesquiterpene hydrocarbons (13.63%) and oxygenated sesquterpenes (11.07%) were identified from the roots. The essential oils obtained from leaf have been also rich in oxygenated sesquterpenes (68.28%), followed by sesquiterpene hydrocarbons (18.33%), and alcohols (5.17%). The finding indicated that essential oils from different parts of Schinus molle have a promising potential on inhibiting activity of pathogenic microbes.

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Introduction

For thousands of years, human beings used plants to treat various ailments and still many peoples from developing countries rely on traditional doctors and the collections of medicinal plants to treat health problems (Mehani and Segni, 2013). The medicinal plants are the plants whose parts (leaf, seed, fruits, stems, roots, barks), extracts, infusions, decoctions, powders are used in the treatment of different diseases of humans, plants and animals (Samy, 2005). The medicinal value of plants lies in some chemical substances that produce a definite physiological action on the human or animal body part of the plant. Extracts of many plants are highly efficient against parasitic as well as microbial infections (Bharti *et al*., 2013; Jameela *et al*, 2016; Parekh and Chanda, 2006). Unlike synthetic pesticides and fungicides, antimicrobials of plant origin are not associated with many side effects and are environmental friendly.

The essential oils are volatile aromatic substance extracted from the plant. They are produced in aromatic and medicinal plants as secondary metabolites and are obtained from various plant parts (flower, seed, leaves, bark, herbs, wood, fruits and roots) and stored in secretory cells, cavities, vessels, or epidermal cells called glandular trichomes (**Pedro, 2012).** They have many therapeutic properties. In herbal medicine, they are used for their antiseptic properties against infectious diseases of fungal and bacterial origin (Mehani and Segni, 2013). The inherent activity of an oil extract may be related to its chemical composition, the proportions of the components and the interactions between them. The chemical composition of the essential oil consists mainly of monoterpene hydrocarbons (e.g., α-pinene, β-pinene, sabinene, terpinen-4-o), and some sesquiterpenes such as (+) spathulenol and germacrene-D (Lisin et al., 1999). Essential oils act against microorganisms often causing instability in the plasma membrane leading to cell lysis. Although the antimicrobial activity can be triggered by a single chemical compound, it usually appears to result from a synergy between several chemicals in the oil (Bhavanani and Balow, 1992; Elhayouni *et al*., 2008).

Schinus molle L. (Anacardiaceae) is one of very important medicinal plant native to Argentina, Bolivia and Peru but has since been introduced to Ethiopia and known with local name as "Kundo Berbere" (Amharic) (Basil and Zakaria, 2014). This plant plays an important role in pharmacology and pharmaceutical chemistry because of its high essential oil content in its different parts (Belhamel *et al,* 2008). In Ethiopia, especially in Eastern part of Ethiopia, the fresh leave was traditionally used to repel insects. The insecticide effect of this plant might be combination of bio active intergradients produce a physiologic action on the insects.

Some studies have concluded that the Essential oils as a whole have much greater antibacterial activity than a mixture of the major components, which suggests that minor components are critical for this activity and may have a synergistic effect or potentiating influence (Burt, 2004; Ljalem and Unnithan, 2014; Marongiu *et al*, 2004). However, in Ethiopia, there is no sufficient work on antimicrobial effect of leaf, stem bark, fruit and root bark extract of *S. molle* on growth and activity of different fungal and bacterial strains. Therefore, this study was aimed to screen phytochemicals, investigated the chemical constituent of essential oils and evaluates antimicrobial activities of leaf; stem and root bark extracts of S. *molle* against selected plant pathogens (*Fusariumverticillioides, Aspergillusniger*, *Xanthomonascampestris*, pv. Campestris and *Ralstoniasolanacearum*).

Material and Methods

Plant Material Collection

The aerial parts (stem bark and leaf) and root bark of *Schinus molle* plant were collected from Eastern Hararghe, Ethiopia. Fresh plant materials were thoroughly washed under tap water, dried with blotting paper and made ready for extraction. The plant species deposited at the National Herbarium of the Addis Ababa University, Ethiopia and a voucher specimen (No. TT-12) has been given.

Preparation of Plant Extracts

Fresh plant parts (stem and root barks and leaf of *Schinus molle*) were collected and thoroughly washed under tap water and shade-dried at room temperature or three weeks. The dried plant parts were crushed to fine powder using an electric grinder. Exposure to direct sunlight was avoided to prevent the loss of active components. The powder of collected plant

materials (50 grams of each plant parts) were extracted successively with 600 mL of n-hexane, chloroform and ethanol for 6hours using Soxhlet extraction. Each solvent was removed by rotary flash evaporator at lower temperature under reduced pressure and the crude extracts were stored in dark vials at 4°C for future uses.

Hydro-distillation and purification of essential oil

The fresh leaf and stem bark of *Schinusmolle* (600mg*)* was soaked in water at a ratio of 1:2 (w/v) and subjected to hydro-distillationfor 6hours using a modified Clevenger type glass apparatus. The essential oil was separated from the aqueous layer by separatory funnel, using the non-polar chloroform as a solvent. The residual oil was dried over anhydrous Na_2SO_4 and kept in a refrigerator (5°C) for subsequent experiments. Analysis of essential oil was carried out by Clarus 600/600 P Gas Chromatograph/Mass Spectrometer (Perkin Elmer, USA) system.

Microbial Culture preparation

Bacterial species (Xanthomonascampestris, pv. Campestris and *Ralstoniasolanacearum*.) were used for the antibacterial screening in this study. These organisms were maintained on agar slope at 4^{o}C and sub-cultured for 24hr before use. Similarly, the fungal plant pathogens *Fusarium verticillioides* and *Aspergillus niger* were used for the antimicrobial test of the extract.

Microbial Susceptibility Testing

Standardized inoculums with 0.5 McFarland standards were introduced onto the surface of sterile agar plates, and a sterile glass spreader was used for even distribution of inoculums. McFarland is a barium sulphate standard against which the turbidity of the test and controlled inoculum was compared. McFarland was prepared by mixing two, solution "A" is 1 % v/v solution of sulphuric acid (H_2SO_4) and solution "B" is 1 % w/v solution of barium chloride ($BaCl_2$). To get 0.5 McFarland standard, concentration equivalents to cell density of about 10^7- 10^8 $CFUg^{-1}$, the amount of 0.5 ml $BaCl_2$ of 1 % solution "A" was mixed with 99.5 ml H_2SO_4 of 1 % solution "B" (Bauer *et al.,* 1966). A sterile paper disc previously soaked in known concentration of extracts will be carefully placed at the centre of the seeded labeled agar. The plates were incubated aerobically at 37^{o}C and examined for zone of inhibition after 24 hrs. Each zone of inhibition was measured with a ruler and compared with the control (Bauer *et al.,* 1966).

Determination of Total Tannin and Phenol Content

Tannin

Condensed tannin was analyzed by vanillin-HCl method of (Price *et al.,* 1980) using the modified Vanillin-HCl methanol method. The Vanillin-HCl reagent was prepared by mixing equal volume of 8% concentrated HCl in methanol and 1% Vanillin in methanol. The solutions of the reagent were mixed just prior to use. About 0.2 g of the ground sample was placed in small conical flask. Then 10 mL of 1% concentrated HCl in methanol was added. The conical flask was capped and continuously shaken for 20 min and the content then is centrifuged at 2500 rpm for 5 minutes. About 1mL of the supernatant was pipette into a test tube containing 5 mL of Vanillin-HCl reagent. Absorbance at 450 nm was read on spectrophotometer after 20 minutes incubation at 30^0C. A blank sample was also analyzed and its absorbance was subtracted from sample absorbance. A standard curve also prepared from catechin (1 mg/mL). Tannins content was expressed as catechin equivalent as follows:

$$\text{Tannin}(\%) = \frac{C \times 10 \times 100}{200}$$

Where: C is concentration corresponding to the optical density, 10 is volume of the extract (mL), 200 is sample weight (mg).

Phenolic

Total phenolic content was determined according to Folin-Ciocalteu method (Sharma and Gupta, 2010). Sample 200 mg was extracted with 4mL of acidified methanol (HCL/Methanol/water, 1:80:10 v/v/v) at room temperature (25^{O}C) for 2 hrs. Aliquot of extract (200 µL) was added to 1.5 mL freshly diluted (10 fold) Folin-Ciocalteu reagent. The mixture was allowed to equilibrate for 5 min and then mixed

with 1.5 mL of sodium carbonate solution (60 g/l). After incubation at room temperature (25^{O}C) for 90 min, the absorbance of mixture was read at 725 nm (6505 uv/vis spectrophotometer, Model 6505, U.K, GENWAY). Acidified methanol was used as a blank. Form the stock solution of standard gallic acid, a series of six standard solutions (0, 5, 10, 15, 20, 25, 30 and 35 ppm) was prepared. The amount of total phenolic was estimated from the calibrated curve as gallic acid equivalent (GAE) in milligram per kilogram of sample. Total phenolic content was calculated by the following formula:

$$\text{Total phenolic (ppm)} = \frac{(\mu g/mL)xDfx100}{\text{Sample mass (db)}}$$

Where: µg/mL is the absorbance reading concentration, Df is dilution factor.

Procedure of Phytochemical Tests

The different qualitative chemical tests were performed for establishing the profile of given extracts to detect various phyto-constituents present in them. The phytochemical were analyzed according to standard screening tests using conventional procedures.

Test for Flavonoids

Lead acetate Test: -Extracts were treated with few drops of lead acetate solution. Formation of yellow colour precipitate indicates the presence of flavonoids.

Test for Alkaloids:

Wagner's Test: -Filtrates were treated with Wagner's reagent (Iodine in Potassium Iodide). Formation of brown/reddish precipitate indicated the presence of alkaloids.

Hager's Test: -Filtrates were treated with Hager's reagent (saturated picric acid solution). Presence of alkaloids was confirmed by the formation of yellow coloured precipitate.

Detection of Saponins

Foam Test: 0.5 gm of extract was shaken with 2 mL of water. If foam produced persists for ten minutes it indicates the presence of saponins.

Test for Steroid

5 drops of concentrated H_2SO_4 was added to 1cm^3 of the extract. A reddish brown colour indicates the presence of steroids.

Test for Terpenoids

Salkowski test; - 5 mL of various solvent extract was mixed in 2 mL of chloroform followed by the careful addition of 3 mL concentrated (H_2SO_4). A layer of the reddish brown coloration was formed at the interface thus indicating a positive result for the presence of terpenoids.

Detection of Phenols

Ferric Chloride Test: Extracts were treated with 3-4 drops of ferric chloride solution. Formation of bluish black colour indicates the presence of phenols.

Test for Quinones

One ml of each of the various extracts was treated separately with ferric chloride solution. Quinines give coloration ranging from red to blue.

Test for Carbohydrates

Molisch test: Treat extract with few drops of alcoholic alpha-naphthol. Add 0.2mL concentrated sulphuric acid slowly along the sides of test tube, purple to violet colour ring appears at junction was showed the presence of carbohydrates.

Detection of proteins and amino acids

Xanthoproteic Test: -The extracts were treated with few drops of conc. nitric acid. Formation of yellow colour indicates the presence of proteins.

Ninhydrin Test: -To the extract, 0.25% w/v ninhydrin reagent was added and boiled for few minutes. Formation of blue colour indicates the presence of amino acid.

Data Analysis

The data was analyzed by using simple statistical methods**.** Experimental results were expressed as

means ± SD. All measurements were replicated three times.

Results and Discussion

Phytochemical screening of the crude extracts

The different qualitative chemical tests were performed for screening the profile of given extracts to detect various phyto-constituents present in the plant. The phytochemicals were analyzed according to standard screening tests using conventional procedures. The results of constituents of the crude extract of parts of *Schinus molle* were presented in table 1.

Table 1: -Phytochemical screenings results of crude extracts of leaf, stem and root bark *Schinusmolle.*

No.	Phytochemical Components	Leaf			Stem Bark			Root Bark		
		P ether	CF	Eth	P ether	CF	Eth	P ether	CF	Eth
1	Flavonoids	-	+	+	-	+	+	-	+	+
2	Alkaloids	-	+	+	-	+	+	-	+	-
3	Saponins	-	-	-	-	-	-	-	-	+
4	Steroids	+	+	-	+	+	-	-	-	-
5	Terpenoids	+	-	-	+	-	-	++	++	-
6	Phenols	-	+	+	-	+	+	-	-	+
7	Quinones	-	-	+	-	+	++	-	+	++
8	Carbohydrates	+	+	-	++	+	-	++	+	-
9.	Proteins	-	-	+	-	+	+	-	-	+

Note: ++ = strong presence, + = moderate presence, - = absence: P eth= Petroleum ether, CF= Chloroform and Eth= Ethanol.

Phytochemical screenings results (Table 1) of crude extracts of leaf of *Schinus molle* revealed moderate presence of flavonoids, alkaloids, steroids, phenols and carbohydrates. The crude extracts of steam bark showed strong presence of quinones and carbohydrates and moderate presence of flavonoids, alkaloids, steroid and phenols. Phytochemical screenings of crude extracts of root of schinus molle also showed strong presence of terpenoides, quinones and carbohydrates and moderate presence of flavonoids. Flavonoids were present in both ethanolic and chloroform extracts of leaf, stem and root barks of *Schinus molle*. Similarly, alkaloids were found in both chloroform and ethanol extracts of leaf and stem barks of *Schinus molle*. They were also found in chloroform extracts of root barks while absent in ethanol extracts. Terpenoids were found in petroleum ether extracts of all plants parts while steroids were found in both petroleum and chloroform extracts of leaf and stem barks. Quinones were also present strongly in ethanolic extract of the above-mentioned plant parts. On the other hand, carbohydrates were found all petroleum and chloroform extracts of only all plant parts.

Tannin and Phenolic content of the different parts of *Schinus molle*

The result of experiment for total tannin and phenol contents from the leaf, stem and root bark was indicated in table 2.

Table 2: Total tannin and phenolic content.

No	Plant part	Tannin mg/g	Total phenol mg/g
1	Stem bark	13.48	177.27
3	Root bark	13.71	167.27
5	Leaf	5.92	58.86

This result showed that, the methanolic extracts of the leaf, root and stem bark of *Schinus molle* showed high amount of total phenolic content when compared to the total tannin in the three parts of the plant.

Similar finding was reported by Kasmi et al, (2016) there by higher phenolic content was observed on the fruit of *Schinus molle* with aqueous solution extracts.

Our findings showed that *Schinus molle* is rich in phenolic compounds which were mainly responsible for several antimicrobial activities due to the trapping potential of free radicals and activation of antioxidants of the cells. Phenols activate the natural defensive mechanism on the microbes.

Antimicrobial activity of leaf, stem and root bark extract of *Schinus molle*

The result for antimicrobial effects of extracts from leaf, stem and root bark of *Schinus molle* against bacterial and fungal plant pathogens were summarized below (Table 3 and 4).

Table 3: Antibacterial activities of leaf, stem and root barks extracts of *Schinus molle*.

Bacteria strains	Plant Part	Mean zone of inhibition (mm) on culture of test bacteria				
		Solvents extract			Control	
		n-Hexane	CF	Ethanol	Positive	Negative
Xanthomonas	Leaf	8.60±0.06	9.33±0.06	8.00±0.20	31.00±0.17	0
	Stem bark	7.00±0.17	9.70±0.06	11.30±0.06		
	Root bark	9.70±0.12	9.30±0.15	11.30±0.06		
Ranstonia	Leaf	3.30±0.06	9.70±0.06	5.70±0.06	9.70±0.06	0
	Stem bark	6.00±0.10	1.70±0.15	6.30±0.12		
	Root bark	9.70±0.12	2.70±0.06	6.70±0.06		

The result revealed that stem and root bark ethanol extracts exhibited relatively higher (11.3mm) inhibition zone against *Xanthomonas campestris*, pv. Campestris. The n- hexane extraction of leaf and stem showed relatively less inhibition against *Xanthomonas* as compared to others. *Ralstonia* showed relatively less inhibition zone (1.7 and 2.7mm) against CF extracts of stem and root. The CF leaf extracts and the n- hexane root extracts showed relatively higher inhibition against *Ralstonia*. As compared to *Xanthomonas*, less inhibition of the plant extracts against *Ralstonia* were observed (Table 3).

Table 4: Antifungal activities of leaf, stem and root barks extracts of *Schinus molle*.

Fungi strain	Plant Part	Mean zone of inhibition(mm) on cultures of test fungi				
		Solvent extracts			Control	
		n-Hexane	CF	Ethanol	Positive	Negative
Aspergillusniger	Leaf	6.30±0.06	5.30±0.056	5.70±0.10	33.70±0.06	0±0.00
	Stem bark	6.30±0.06	5.30±0.21	6.30±0.23		
	Root bark	6.70±0.06	8.00±0.20	6.70±0.12		
Fusariumvert	Leaf	5.70±0.06	1.30±0.06	2.30±0.15	24.30±0.06	0±0.00
	Stem bark	5.30±0.31	1.30±0.06	7.00±0.10		
	Root bark	2±0.100	1.00±0.00	10.00±0.00		

Relatively higher zone of inhibition (10mm) was observed by the ethanolic extracts of the root of *Schinus molle* against *Fusarium verticillioides* followed by the chloroform extracts of the root (8mm) against *Aspergillus niger*. The least zone of inhibition was detected in the chloroform extracts of the root, stem and leaf extracts against *Fusarium verticillioides*. The n-hexane and chloroform extracts of the plant showed relatively higher zone of inhibition against *Aspergillus niger* than *Fusarium verticillioides* (Table 4).

When the overall zone of inhibition was examined, the crude extracts showed relatively higher zone of

inhibition against the tested bacterial species than fungal species. The ethanol extracts of stem and root showed relatively higher zone of inhibition against the tested bacteria than the other solvent extracts. Overall, the maximum inhibition zone of crude extract in the experiment was (12mm).

A report from Rhouma, *et al*, (2009) in the antimicrobial activity of leaf extracts of *Schinus molle* against *Pseudomonas savastanoi* pv. *Savastanoi* showed similar results and the zone of inhibition were between 9-12mm. Another report from (Mehani and Segni, 2013) on the antimicrobial activity of plant extract of *Schinus molle* on four bacterial pathogens (*E. coli, Pseudomonas aeruginosa, Staphyloccus* and *Klebsiella pneumonia*) with a zone of inhibition 13, 12.33, 11.67 and 10.5mm, respectively which is in agreement with the current finding.

This result could be due to the polarity of the compounds which were extracted by each solvent and the ability of extracts to diffuse and dissolve in different culture media used in the study. Absence of antimicrobial activity does not mean that the bioactive compounds are not present in the plant or the plant has no activity against microorganisms. Presence of inadequate quantities of active constituent or constituents in the extract to exhibit the antimicrobial activity can be the reason for the negative results.

Chemical composition of essential oil from leave of *Schinus molle*

The major volatile components of essential oils recovered by hydro distillation and their retention time analyzed by GC/MS of *Schinus molle* were summarized in the Table 5 and Fig 1 below. Analysis of essential oil was carried out by Clarus 600/600 P Gas Chromatograph/Mass Spectrometer (GC/MS).

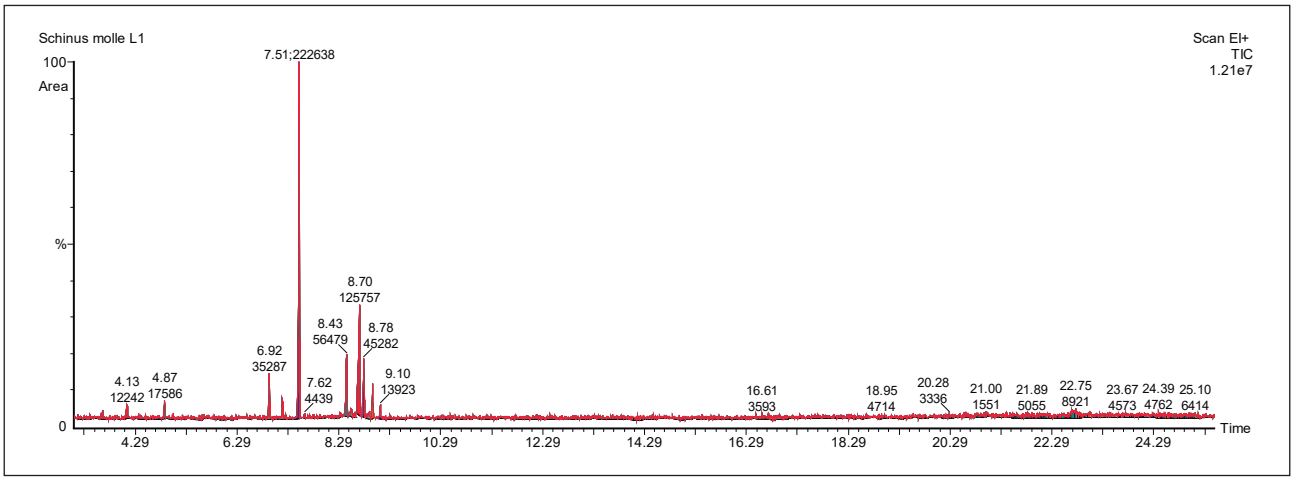

Fig 1: Chromatograms of essential oil from leaf of *Schinus moll*

The result showed three alcohols, three sesquiterpene hydrocarbons, one aromatic hydrocarbon, three oxygenated sesquterpenes (91.78%) and other compounds (8.21%) totally 99.99% of the essential oils were identified from Ethiopian leaf of *Schinus molle* by GC-MS (Table 5). As shown by their proportion given in the Table 5, the essential oils obtained from leaf have been found to be rich in oxygenated sesquterpenes (68.28%), followed by sesquiterpene hydrocarbons (18.33%), and alcohols (5.17%). The published literature on the chemical composition of essential oils of *Schinus molle* growing in Egypt revealed that β- eudesmol (10.34%), elemol (10.27%), β- bisabolenol (5.06%) and epi- α-muurolol (3.29%) as the major oxygenated sesquiterpenes (50%) and ρ-cymene (9.42%) as the most abundant monoterpene hydrocarbons (19.4%) in leaf oil (Dalia *et al*.,2016). However, the present study result on the chemical composition of essential oils of *Schinus molle* growing in Eastern Hararge (Ethiopia) was different. These were hedycaryol (38.62%), β–eudesmol (21.81%) and Ar-turmeroneas (7.85%) the major oxygenated sesquiterpenes (68.28%) followed by γ-Eudesmol (9.80%), γ-elemene (6.12%) and α–cubebene (2.41%) as sesquiterpene hydrocarbons (18.33%).

Table 5: Chemical composition of essential oil from leaves of *Schinus molle*

No	Constituent	RT (min)	%
Alcohols			
1	1-Butyl-2-cyclohexen-1-ol	4.13	2.12
2	(2-Penta-2,4-dienyl-cyclohexyl)-methanol	4.87	3.05
3	(S)-(±)-5-Methyl-1-heptanol	8.96	n.d
Sesquiterpene hydrocarbons			
4	γ-elemene	6.92	6.12
5	γ-eudesmol	8.43	9.80
6	α –cubebene	9.10	2.41
Aromatic Hydrocarbons			
7	1,4- diethylbenzaldeyde	7.18	n.d
Oxygenated sesquterpenes			
8	Hedycaryol	7.51	38. 62
9	β–eudesmol	8.70	21.81
10	Ar-turmerone	8.78	7.85
	Total known identified compounds		91.78
	Alcohols		5.17
	Sesquiterpene hydrocarbons		18.33
	Oxygenated sesquterpenes		68.28
	Others (Unknown or not identified)		8.21

n.d. -not detected

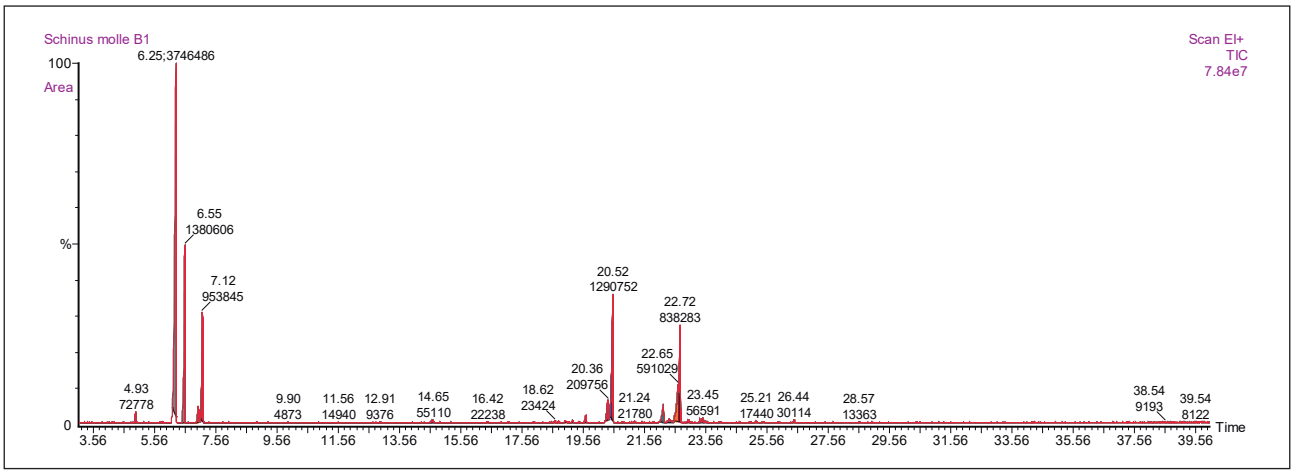

Fig 2: Chromatograms of essential oil from steam of *Schinus molle*

Table 6: Chemical composition of essential oil from stem bark of *Schinusmolle*

No	Constituent	RT (min)	%
Monoterpene hydrocarbons			
1	α-pinene	4.93	0.77
2	β-pinene	6.25	39.57
3	α-Phellandrene	6.55	14.58
4	*p*-Cymene	6.98	1.03
5	d-limonene	7.12	10.07
Sesquiterpene hydrocarbons			
6	α-amorphene	19.64	n.d
7	Germacrene A	20.52	13.63
Oxygenated sesquterpenes			
8	Elemol	20.36	2.22
9	γ-eudesmol	22.17	n.d
10	Agarospirol	22.72	8.85
	Total known identified compounds		90.72
	Monoterpene hydrocarbons		66.02
	Sesquiterpene hydrocarbons		13.63
	Oxygenated sesquterpenes		11.07
	Others (Unknown or not identified)		9.27

n.d. -not detected

According to the GC-MS investigation of the essential oils, it was found that the stem bark of *Schinusmolle* contained, three terpenes (also known as terpenoids or isoprenoids) namely monoterpene (66.02%) as a major component, sesquiterpene hydrocarbons (13.63%) and oxygenated sesquterpenes (11.07%) were identified from the stem bark. Accordingly (Table 5 and Figure 4), β-pinene (39.57%), α-phellandrene, (14.58%), Germacrene A (13.63%), d-limonene (10.07%) and agarospirol (8.85%) were analyzed as major constituents in stem of these species. According to the published literatures, essential oils collected form *Schinus molle* in Tunisia, the species contained elemol (20.7%), 6-epi-shyobunol (20.36%), d-limonene (16.19%) and α-eudesmol (7.01%) (Kasmi *et al.,* 2016). The stem oil 31% of monoterpene hydrocarbons and 43.82% of oxygenated sesquiterpenes with myrcene (15.28%), β-eudesmol (11.79%), elemol (9.79%), limonene (6.43%) and ρ-cymene (5.91%) were obtained as the major volatile components from the plant leaves in Egypt (Dalia *et al*., 2016). The differences of chemical composition in the same plant species might be due to different environmental and geographical factors.

Conclusion

Phytochemical screening results of crude extracts of stem and root bark of *Schinus molle* revealed strong presence of quinones and carbohydrates and moderate presence of flavonoids, alkaloids, steroids and phenols. As compared to chloroform and n-hexane, stem and root bark ethanol extracts exhibited relatively higher (11.3mm) inhibition zone against *Xanthomonas campestris*, pv. Campestris. Relatively higher zone of inhibition (10mm) was observed by the ethanolic extracts of the root of *Schinus molle* against *Fusarium verticillioides* followed by the chloroform extracts of the root (8mm) against *Aspergillus niger*. Furthermore, the leaf extract of the plant were characterized and showed three alcohols, three sesquiterpene hydrocarbons, one aromatic hydrocarbon and three oxygenated sesquterpenes, totally 91.78%) % of the essential oil were identified

by GC-MS. The GC-MS analysis showed, three terpenoids namely monoterpene (66.02%) as a major components, sesquiterpene hydrocarbons (13.63%) and oxygenated sesquterpenes (11.07%) were identified from the stem bark. The finding indicated that essential oils from different parts of *Schinus molle* have a promising potential on inhibiting activity of pathogenic microbes.

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