Journal of
Pharmacognosy & Natural Products

Abebe et al., J Pharmacogn Nat Prod 2018, 4:2
DOI: 10.4172/2472-0992.1000154

Research Article Open Access

Chemical Composition and Antimicrobial Activity of Essential Oil from *Justicia schimperiana*

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Received date: May 02, 2018; **Accepted date:** May 14, 2018; **Published date:** May 17, 2018

Abstract

Background:

Justicia schimperiana is popularly used as folk medicines in Ethiopia.

Objective:

The chemical composition of essential oils from the leaves of *Justicia schimperiana* was analyzed and its antimicrobial activities were evaluated.

Methods:

The essential oil was obtained by steam distillation and its chemical constituents were separated and identified by GC-MS, using normalization method to calculated relative amount. Its antimicrobial activities were evaluated by micro dilution method against *Escherichia coli*, *Bacillus subtilis* and *Candida albicans*.

Results:

From the essential oil of *Justicia schimperiana*, 28 compounds were identified; making up 75.18%, the essential oil of *Justicia schimperiana* has not showed obvious activity against *Escherichia coli*, *Bacillus subtilis* and *Candida albicans*.

Conclusion:

This is the first report on the chemical compositions and antimicrobial activities of essential oils obtained from Ethiopian *Justicia schimperiana*.

Keywords: *Justicia schimperiana*; Essential oil; Antimicrobial activity

Introduction

Traditional medicine has been practiced in virtually all cultures and it has expanded globally and is gaining popularity [1]. In Ethiopia, the knowledge of traditional medicine has been transferred from one generation to another and about 80% of the Ethiopian population is still dependent on traditional medicine, especially the usage of plants [2,3].

Justicia schimperiana (Hochst. ex Nees), belongs to the family Acanthaceae and it is a shrub with branched stems [4]. In Ethiopia, *J. schimperiana* was used for the treatment of stomach complaints, malaria, hepatitis, asthma, jaundice, epilepsy, etc [5]. Essential oils are complex mixtures of volatile substances generally present at low concentrations and they are important components used for their flavor and fragrances in food, pharmaceutical and perfumery industries [6]. Although there are some reports on the pharmacology of *J. schimperiana* mentioned above, but no research on essential oil has been performed [2,5,7]. Here we will focus on the chemical composition and antimicrobial properties of essential oils from this medicinal plant.

Methods and Methods

Plant material

The leaves of *J. schimperiana*, was collected in October 2015, from Addis Zemen, which is a town in Northern-Central Ethiopia, Located in the Debub Gondar Zone of the Amhara Region, on the road connecting Gondar and Bahir Dar. The plant was identified by Amare Seifu Assefa, a botanist from Ethiopian Biodiversity Institute.

Extraction of essential oil

The leaves of *J. schimperiana* (200 g) were coarsely powdered and transferred into a flask with 1.5 L water. Then it was submitted to

steam distillation process, in a Clevenger's apparatus for 8 h. The distillates were saturated with NaCl and extracted with diethyl ether. The organic phase was dried by anhydrous Na_2SO_4 and then recycled the organic solvent at 30°C to give the essential oil. The essential oil was stored at 4°C in refrigerator for further use.

Analysis of essential oil

The GC-MS analyses of the essential oil was carried out using an Agilent 6890 gas chromatograph interfaced with an Agilent 5973N mass spectrometer fitted with a HP-5MS capillary column (30 m × 0.25 mm, 0.25 μm film thickness). Gas chromatography condition: the temperature programmed at 70°C for the first two minutes, increased at a rate of 5°C/minute to 300°C and held isothermal at 300°C for the next 2 minutes; The injector temperature was 250°C; The injected volume was 1 μL; Helium was used as carrier gas; Flow rate was 1 mL/min with a split ratio of 1:20; Mass spectrometry condition: EI ionization mode, 70 eV, scan range 30-500 Amu, ion source temperature was 230°C. Individual components were identified by matching their mass spectra with those of the spectrometer data base (NIST 11). For quantification purposes, relative area percentages were used without the use of correction factors.

Antimicrobial activities

Escherichia coli (ATCC 25922), *Bacillus subtilis* (ATCC 6633) and *Candida albicans* (ATCC 60193) were used for antimicrobial evaluation. Antimicrobial activity assays were performed in 96-well sterilized microplates using a microdilution method described previously [8,9]. The 18-h-old bacterial cultures from *E. coli*, *B. subtilis* were added to LB broth medium (1 L water, 10 g tryptone, 5 g yeast extract and 10 g NaCl) to reach 1×10^5 CFU/ml and the 4-day-old spores from *C. albicans* were added to PDB medium (potato 20%, glucose 2%) to research 1×10^3 spores/mL. The test samples were dissolved in DMSO and their final concentrations were ranged from 0.5 to 512 μg/mL, which were determined by 2-fold serial dilution method. The wells containing test strains and diluted samples were incubated at 37°C (24 h) for bacteria and 28°C (4 days) for fungi. The wells containing a culture suspension and DMSO were run as negative controls. Kanamycin (for bacteria) and nystatin (for fungi) were introduced as positive controls. All experiments were repeated twice. The Minimal Inhibitory Concentration (MIC) was defined as the lowest antibiotic concentration that produced complete growth inhibition of the tested microorganisms.

Results and Discussion

Steam distillation of the leaves of *J. schimperiana*, gave dark brown oil (yield 0.016% w/w). The chemical constituents of the essential oil, along with its retention time and relative percentages, were given in Figure 1 and Table 1.

A total of twenty eight components were identified from *J. Schimperiana* essential oil, representing 75.18% of the total oil. Eucalyptol (24.13%), espatulenol (7.25%), crypton (5.08%) and 1-octen-3-ol (5.06%) were the main constituents. The oil sample was found to be rich in monoterpenoids, including eight compounds, which representing 53.91% of the total identified components.

The essential oil obtained from leaves of *J. schimperiana*, was tested for its antimicrobial activity against *E. coli* (Gram-negative), *B. subtilis* (Gram-positive) and *C. albicans* (fungi). The results (Table 2) indicated that no obvious antimicrobial activity has been observed.

Components	Retention time (min)	Area
1-Octen-3-ol	4.67	5.06%
o-Cymene	5.65	0.74%
Eucalyptol	5.79	24.13%
beta-Terpineol	6.62	0.40%
Epoxylinalool	6.75	2.59%
Linalool	7.40	1.45%
trans-1-Methyl-4-(1-methylethyl)-2-cyclohexen-1-ol	7.96	0.81%
(-)-Terpinen-4-ol	9.39	4.77%
Crypton	9.63	5.08%
2-Methyl-3-phenylpropanal	11.12	2.04%
4-isopropylbenzyl alcohol	12.59	1.30%
Durenol	12.92	1.07%
alpha-Terpinyl acetate	13.85	4.48%
Eugenol	14.32	0.33%
Caryophyllene	15.63	0.28%
alpha-Ionone	15.85	1.12%
beta-Humulene	16.64	0.45%
beta-Ionone	17.29	1.94%
Dihydroactindiolide	18.37	2.79%
Espatulenol	19.45	7.25%
Caryophyllene oxide	19.57	1.53%
beta-Eudesmol	21.12	0.67%
6,10,14-Trimethylpentadecan-2-one	25.21	1.83%
Phytol	30.35	1.38%
Tritetracontane	36.73	0.47%
11-Decyl-tetracosane	38.25	0.29%
Heptacosane	39.71	0.41%
Octadecane	42.48	0.52%

Table 1: Chemical components (%) of the essential oils from *Justicia schimperiana*.

Essential oil	*Escherichia coli*	*Bacillus subtilis*	*Candida albicans*
Justicia schimperiana	>128	>128	>128
Kanamycin	4	4	-
Nystatin	-	-	4

Table 2: Antimicrobial activities of the essential oils from *Justicia schimperiana* (MIC: μg/ml).

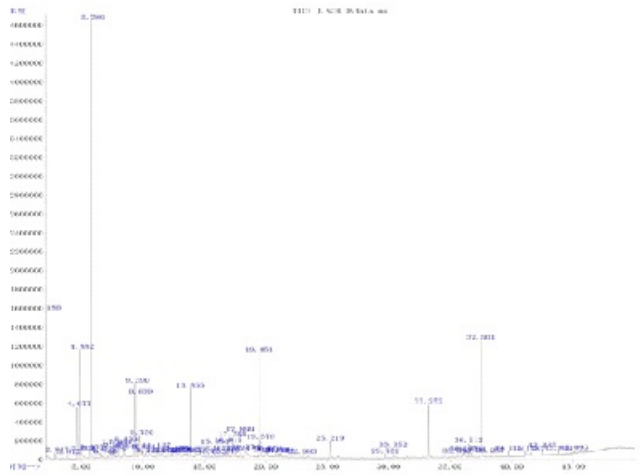

Figure 1: GC-MS profile of an essential oil *Justicia schimperiana*.

Conclusion

This is the first report on the essential oil of *J. schimperiana*, which is an important medicinal plant in Ethiopia. Terpenoids were found to be the major components in the GC-MS analysis of the essential oils. No distinct antimicrobial activities were found for this medicinal plant essential oil and further studies are required on the chemical composition and their bioactivities of this plant.

Acknowledgement

This work was financial supported by the Fundamental Research Funds for the Central University (ZYGX2016J120).

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