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CHEMICAL COMPOSITION AND ANTIMICROBIAL ACTIVITIES OF THE ESSENTIAL OIL FROM *KUNDMANNIA SICULA L. LEAVES*

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ABSTRACT

This study describes the chemical composition and antimicrobial activity of essential oil of the of fruit of *Kundmannia sicula L* growing wild in jdiouya relizane region. This plant is very used in Algeria and widely used by local people for in traditional medicine for urinary tract infections and to treatement. We proposed to determine the physicochemical, organoleptic and chemical identification of these components. The chemical composition of the essential oil from *Kundmannia sicula* L. leaves was analyzed by GC/GC-MS and resulted in the identification of 5compounds. Other parameters such as refractive index, optical rotation; density, polarimetric deviation; freezing point and Solubility in ethanol are also measured .100% of the total peak areas were identified. The main constituents of the essential oil were the hydrocarbon and oxygen compounds are : 2-BUTENAL, 2-ETHYL97.44%, another important constituent were 2-Propanone, 1,1,3,3-tetrachloro- 2.45 % Other components were present with smaller percent..Isosafrole0.04% Octaverine0.03%, spathulenol 0.02%. The antimicrobial effect for germs causing nosocomial infection.

KEYWORDS: *Kundmannia sicula*, leaves, Essential oil, GC/SM, antimicrobial activity.

INTRODUCTION

Essential oil are very complex mixtures of volatile molecules produced by the secondary metabolism of aromatic and medicinal plants, and can be obtained by different methods, including low- or high-pressure distillation of different plant parts, or the use of liquid carbon

dioxide or microwaves.[1] Several factors can influence the quality and quantity of the extracted product, in particular the soil composition, plant organ, vegetative cycle phase, and climate. EO composition can be divided into two component groups: the main group is usually of terpenoid origin, and the second comprises aromatic and aliphatic components. In general, monoter- penes and sesquiterpenes, as well as their oxygenated derivatives, are the predominant constituents, but phenylpropanoids and both fatty acids and their esters may also be present. Aromatic plants and their EOs have been used since antiquity for their biologic properties (e.g., antibacterial, antifungal, antiparasitic, antiviral, and insecticidal), as well as for cosmetic and medicinal applications.[2]

The aim of the present study was to determine the chemical composition and antimicrobial of essential oil extracted from fruit of *Kundmannia sicula* Collected in wild in jdiouya relizane (West of Algeria). This study will contribute to the valorization of medicinal and aromatic plants of the Algerian flora.

Experimental section

Plant material collection

Collected from jdiouya relizane situated in the West of Algeria in April 2013. 2This plant was identified by botanists of Faculty science. A voucher specimen is deposited in the Herbarium of the Department of Botany and Ecology at the Agronomic Institute under code number 2013-51465.,

Essential oil distillation

The fruits of *Kundmannia sicula* were shade, dried, and stored in a tightly closed container for further use. The essential oils were obtained by hydro-distillation from the plant material using a Clevenger –type apparatus for 3h. The essential oil was dried over anhydrous Na2SO4 and stored in a scaled vial in the dark; at 4°C.[3] The essential oil yield was calculated on a dry weight by gravimetric method.

Analysis of the essential oils

In the essential oil these some parameters are measured the refractive index, density, polarimeter deviation; point of freezing, solubility in ethanol at 90 C; and the acidity.The analyses of the volatile constituents were run on a Hewlett-Packard GC-MS Gas Chromatograph (FID) detectorsystem GC: 5890 series II, MSD 5972) the fused-silica HP-5MS capillary Colum (30 m X 0.25 mm id, film thicknessof 0.25 (µm) was directly coupled

to the MS. The carrier gas was helium, with a flow rate of 1.2 ml/min. Injector port 250°C and oven temperature was programmed as flows:isotherm at 50°C for 1 min,then increased to 280°C at a rate of 5°C/min and subsequently held isothermal for 20 min.Detector280°C, volume injected: 0.1 Ol of 1 % solution(diluted in hexane) and split ratio:1:50. Ionization voltage:70 ev ion source temperature 280°C, mass range: 40-300;mass units scan time 1,5 sec.Software adopted to handle mass spectra and chromatograms was a skin station. The extract composition percentage was calculated from the GC peak area. For retention indices (RI) determination, a hydrocarbon series was chromatographed together with the essential oil on a polar columns, and their retention times were used to convert GC retention values to RI by linear interpolation with those of authentic compounds and literature data [4], and also by computer matching them with the NIST/EPA/NIH MASS SPECTRAL LIBRARY data with those of the published data by Adams.[5,6]

2.3 Microbial strains

Microbial strains: Antimicrobial activity was carried out according to the disc diffusion assay, tested in vitro against *Escherichia coli, Proteus mirabilis, Klebsiella sp, Staphylococcus saprophyticus* suspensions were adjusted to1×10^{7} CFUmL− (equivalent to 0.5 McFarland).

2.4. Antimicrobial activity

Antimicrobial tests were carried out using the disc diffusion method. The muller-hinton mutriet agar and dimethyl sulfoxide (DMSO) solutions (in ratio 1:25 v.v-1) were vortexes for 2 min and immediately 20 ml were poured into sterile Petri dishes (90 mm diameter) and left to set for 30 min. Paper discs (6 mm diameter) were impregnated aseptically with 3 µl of essential oil at final concentrations of 1-20µ g/ml□ and placed on the inoculated agar surfaces. After aerobic incubation for 24 hours at 37°C, the antimicrobial activity was estimated by measuring the diameters of inhibition zone.[7] The control test by aqueous DMSO alone showed no toxicity in the concentrations used for these bacteria. The antibacterial minimum inhibitory concentrations (MICs) were performed according to the Mueller-Hintonbroth microdilution method in 96 multiwell microtiterplate. The essential oils were dissolved in the aqueous DMSO and the initial concentration was 25µ g/ml□□The initial test concentration was serially diluted two fold .Each well was inoculated with 5 µg/ml of suspension containing 10^{7} CFU/ml of bacteria and incubated for 24hours at 37°C. The

MIC of the tested material was determined as the lowest concentration at which no visible growth of the microorganism had occurred. Each test was carried out in triplicate.

RESULTS AND DISCUSSION

Physicochemical analysis showed an essence green; with pleasant odor .The essential oil yield obtained by the hydro-distillation of dry plant was 2.46 %.

Determination density was obtained by double weighing d = 0.871, the optical activity= - 7.2 by polarimetry and the refractive index n = 1.4664 by an interferometric method. The chromatograms of the essential oil had numerous peaks and many of them were overlapping. Gas Chromatographic analysis was performed under some specific conditions the compounds are listed along with their percent constituents Table 1. Essential oils 29 components were separated on HP- Column and of them were identified as major component representing of 100% of the total Table 2 The identification of chromatogram component demonstrated that this species was characterized by its high rate:

2-BUTENAL, 2-ETHYL97.44%, another important constituent were 2-Propanone, 1,1,3,3-tetrachloro- 2.45 % Other components were present with smaller percent..Isosafrole0.04% Octaverine0.03% , spathulenol 0.02%.

By other findings, essential oils from different parts of an aromatic plant Kundmannia sicula (L.) DC. (Apiaceae) growing in Tunisia. The hydro-distilled essential oils of the leaves and inflorescences with mature seeds (IMS) of K. sicula were analysed for the first time by gas chromatography equipped with flame ionisation detector and gas chromatography coupled with mass spectrometry. Fifty leaves and 47 IMS constituents were identified, accounting for 97.9% and 98.2% of the total oil, respectively. The major compounds identified from the leaves and IMS oils were isocurcumenol (9.9-10.1%), hexadecanoic acid (9.5-10.9%), spathulenol (6.9-3.4%), 10-epi-γ-eudesmol (6.3-5.5%), α- CFU/ml of bacteria and incubated for 24hours at 37°C. The MIC of the tested material was determined as the lowest concentration at which no visible growth of the microorganism had occurred. Each test was carried out in triplicate.

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Table 1. Physicochemical composition of *Kundmannia sicula*

Specification	***Kundmannia sicula***
Density D20	0.871
Refractive index	1.4664
Optical activity N20	-7.2
Solubility in ethanol90%	1 : 4
Freezing Point (°C)	-16
Acidité	0.4

Table 2. The major identified components in essential oil from *Kundmannia sicula L.*analyzed by GC-MS technique with retention indices on HP-5MS capillary Column

N	**Volatile componds**	**Ri**	**%**
1	2-BUTENAL, 2-ETHYL-	791	97.44
2	2-Propanone, 1,1,3,3-tetrachloro-	1146	2.45
3	.Isosafrole	1296	0.04
4	spathulenol	2207.6	0.02
5	Octaverine -	3156	0.03

The permeability of the bacterial membrane, constituents key elements that influence the diffusion and the action of the essential oil into the cell and cause their damage.[9] E*scherichia coli*, (inhibition zone: 18.5mm, MIC: 10.50µg/ml ,the results for *Proteus mirabilis*(inhibition zone 10.5mm MIC: 6.50µg/ml) and *Klebsiella sp*, (inhibition zone: 17.5mm, MIC: 4.50µg/ml) and *Staphylococcus saprophyticus*, (inhibition zone:11.30 mm, MIC: : 4.50µg/ml)*Kundmannia sicula* essential oil found in Relizane region may be regarded as 2-BUTENAL, 2-ETHYL chemotype and The potential for thedevelopment of leads

from these Essential oil is continuing to grow, particularly in the area of nosocomial infection.

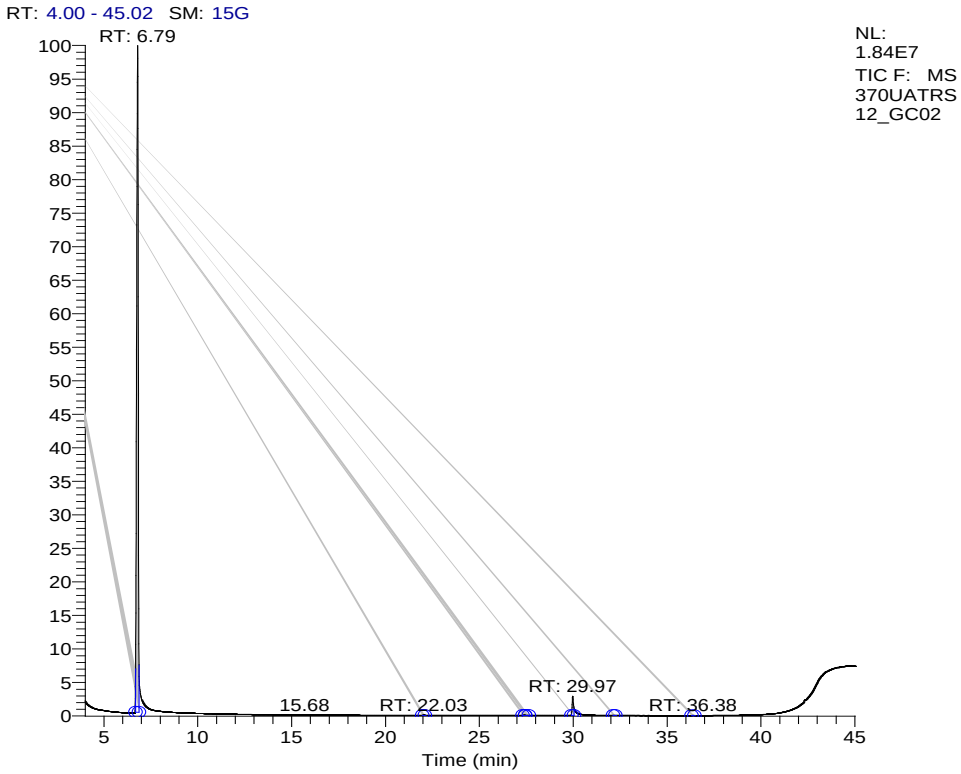

Figure 1.Gas Chromatogram (GC-FID) of essential oil of *Kundmannia sicula L*.

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