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Chemical composition and antimicrobial activities of the essential oil from leaves of *Solenostemma argel*

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Abstract

This study describes the chemical composition and antimicrobial activity of essential oil of the fruit of*Solenostemma argel* growing in Ain fares Mascara region. This plant is very used in Algeria and widely used by local people for its medicinal properties. We proposed to determine the physicochemical, organoleptic and chemical identification of these components. The chemical composition of the essential oil from *Solenostemma argel* L. leaves was analyzed by GC/GC-MS and resulted in the identification of 29 compounds. Other parameters such as refractive index, optical rotation; density, polar metric 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: Thujone 43, 73%. trans-Sabinene hydrate 10.47% Eugenol 8.41%, 1.8-cineole 7.90% Limonene4.16-% à-PINENE, (-)-3,57%. The antimicrobial effect of *Solenostemma argel* essential oil "in vitro" condition was determined using the agar diffusion method and it was found that it was active which may find its application in future research for the food and pharmaceutical industry.

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Introduction

Essential oils are natural products characterized by a strong odor and formed by aromatic plants as secondary metabolites. The features differentiating these aromatic plants from all others, in spite of the fact that they belong to many different families, are the production of chemically related secondary compounds, the low molecular weight, and the presence of volatile isoprenoids (Tahraoui *et al*, 2007). Numerous species of medicinal plants from Algeria are important aromatic and ornamental plants, as well as being medicinal. In the midst of this new way of life, diseases, stress, and metabolic and psychological disturbances have arisen. Fortunately, research continues, including the search for natural alternatives that may somehow enhance quality of life and reduce damage to health.

Murwan K. *et al* (1998). *Solenostemma argel* is the most important one from the many Egyptian plants which are known to be of potential medicinal value in herbal medicine. *S. argel* is used for the treatment of diabetes and jaundice, purgative properties which may be due to the latex present in the stem parts (El-Kamali H *et al*, 1996). Also, an extract from the leaves of this plant showed fungitoxic activity. (Abd El-Hady *et al*, 1994).

It is used for the treatment of some diseases of liver and kidney and for allergies and as incense in the treatment of measles and anti-inflammatory activity (A. Bouzouita *et al*, 2004), is reputed to alleviate arthritis and rheumatism; it is used to treat earaches and high blood pressure. The plant used in this study, was selected based on its traditional use in Algerian medicine for the treatment of diseases such as hypertension and several types of inflammation (Hassan HA *et al*, 2000).

The aim of the present study was to determine the chemical composition and antimicrobial of essential oil extracted from fruit of *Solenostemma argel* Collected in the region of Ain fares (NorthWest of Algeria) in May 2014. This study will contribute to the valorization of medicinal and aromatic plants of the Algerian floral.

Materials and methods

Plantmaterial collection

The leaves of *Solenostemma argel* collected from Ain fkane situated in the North West of Algeria in may2014.Thisplant 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 2014-51475.

Essential oil distillation

The leaves of *Solenostemma argel* 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. The essential oil yield was calculated on a dry weight by gravimetric method. (Hammiche V., Maiza K., 2006).

Analysis of the essential oils

In the essential oil these some parameters are measured the refractive index, density, polar meter 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) detector system GC: 5890 series II, MSD 5972) the fused-silica HP-5MS capillary Colum (30 m X 0.25 mm id, film thickness of 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 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 chromatographic 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 (Abd El-Hady EK., 1994), and also by computer matching them with the NIST/EPA/NIH MASS SPECTRAL LIBRARY data with those of the published data by Adams (R.P Adams, 2000and Nist, 2004).

Microbial strains

Microbial strains: Antimicrobial activity was carried out according to the disc diffusion assay, tested in vitro against *Escherichia coli, Bacillus cereus*, *Salmonella typhimurium,* and *S. pneumonia* suspensions were adjusted to1×10^{7} CFUmL– (equivalent to 0.5 Mc Farland).

Antimicrobialactivity

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 zone12.

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-Hinton broth microdilution method in 96 multiwell microtiter plate. 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 107 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 0.64 %.Determination density was obtained by double weighing d = 0.887, the optical activity= +6.5 by polarimetry and the refractive index n = 1.4645 by an interferometric method.

Table 1. Physicochemical composition of *Solenostemma argel.*

Specification	*Solenostemma argel*
Density D20	0.887
Refractive index	1.4645
Optical activity N20	+6.5
Solubility in éthanol 90(%)	1 : 3
Freezing Point (°C)	-18
Acidité	0.5

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 2.

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: Thujone 43, 73%, another important constituent were trans-Sabinene hydrate10.47% Chemical analysis of essential oil showed that its major products are Eugenol 8.41%, 1.8-cineole 7.90% Limonene 4.16-% à-PINENE, (-)-3,57%.

Other components were present with smaller percent. This is the first report of the chemical compos ion of *Solenostemma argel* .Thus, further investigations are necessary to study the potential of the essential oil leaves.

Including the preservation of raw and processed food, pharmaceuticals, alternative medicines, and natural therapies (M Zuzarte *et al*, 2011).

This essential oil appears to include a bacterial membrane damage that occurs when the essential oil passes through the cell wall and cytoplasm membrane, and disrupts the structure of their different layers of polysaccharides, fatty acids and phospholipids (Arı´aJose´Abad *et al*, 2015).

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

N	Volatile componds	Ri	Area %
1	Naphthalene	200	0.07
2	Brocide	620	0.25
3	b-PINENE, (-)-	705	0.25
4	Retinene	722	0.20
5	à-PINENE, (-)-	724	3.57
6	.DELTA.3-Carene	727	0.22
7	.B 3-Catenine	747	2.88
8	Cyclopentasiloxane, decamethyl	865	0.85
9	Terpinolene	865	1.80
10	Limonene	948	4.16
12	Thujone	1011	43.73
13	Dolcymene	1042	0.10
14	Eucalyptol	1059	0.12
15	1.8-cineole	1059	7.18
16	3-Carene	1172	0.12
17	1-Amino-1-ortho-chlorophenyl-2-(2-quinoxalinyl)ethane	1332	0.77
18	Benzenamine, 2,4,5-trimethyl	1332	0.19
19	Chrysanthenone Chrysanthenone	1379	0.48
20	Eugenol	1392	7.90
21	-Carene	1539	0.07
22	Santolina triene	1867	2.08
23	Santolina	1877	0.62
24	Bornylene	1995	0.20
25	Aceto-sterandryl	2124	10.47
26	trans-Sabinene hydrate	2139	8.41
27	Pnacide	2168	0.85
28	benzaldehyde2,3,5,trymethoxy	2620	0.98
29	Neoserpin	4428	0.38
Total			100

Table 3. Inhibition zone (mm) using direct contact technique in agar medium and MIC (µg/mL) for the essential oil using micro dilution method in 96 multiwall micro liter plate.

Microorganism	Diameter of inhibition zones (mm)	MIC (µg/mL)
S. pneumoniae(G-) ATCC 27853	14.5±0.14	19.5
Escherichia coli ATCC 25922	18.5±0 ,10	10.5
Salmonella typhimurium TCC14028	17.30 ± 0.93	14.5
Bacillus cereus ATCC-6633	14.5±0.4	06.0

E*scherichia coli*, (inhibition zone: 18.5mm, MIC: 10.50µg/ml the results from *Bacillus cereus* (inhibition zone 14.5mm MIC: 6.50µg/ml) and *Salmonella typhimurium,* (inhibition zone:17.30 mm, MIC: 14.5 µg/ml. and *S. pneumoniae*, (inhibition zone: 19.5mm, MIC: 7.50µg/ml) tm *Solenostemma argel* essential oil found in Mascara region may be regarded as Thujone chemo type and The potential for the development of leads from these Essential oil is continuing to grow, particular lying they are noninfectious diseases. The information summa-razed here is intended to serve as are ferrous tool to researchers in all fields of ethno pharmacology and natural product chemistry it's that may lead to the development of new antibacterial drugs.

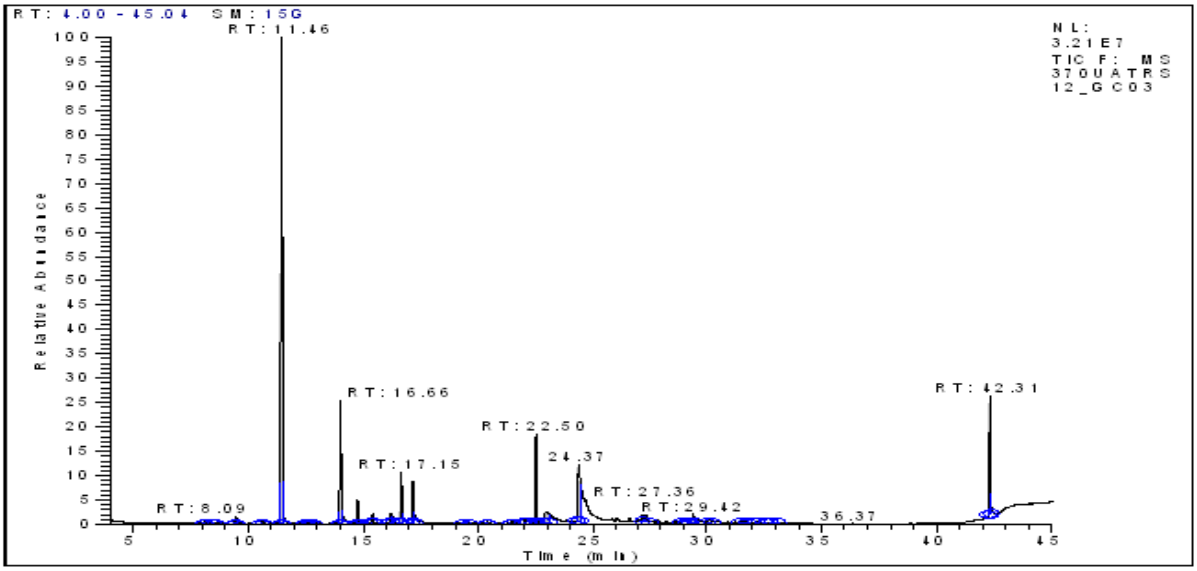

Fig. 1. Gas Chromatogram (GC-FID) of essential oil of *Solenostemma argel* collected in May 2014.

Conclusion

The chemical composition of the essential oil from *Solenostemma argel* L. Leaves was analyzed by GC/GC-MS and resulted in the identification of 29 compounds.

The main constituents of the essential oil were Thujone43, 73%. trans-Sabinene hydrate10.47% trans-Sabinene hydrate 10.47% Eugenol 8.41%, 1.8-cineole 7.90% Limonene 4.16-% à-PINENE3,57%.

The antimicrobial effect of *Solenostemma argel* essential oil "in vitro" condition was determined using the agar diffusion method and it was found that it was active which may find its application in food.

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