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# Chemical Composition and Antimicrobial Activity of Essential Oil of *Genista numidica* Spach. and *G. saharae* Coss et Dur

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**Abstract:** The hydrodistilled oils from the aerial parts of *Genista numidica* and *G. saharae*, which are endemic to Algeria, were analyzed by Gas Chromatography-Mass Spectrometry (GC-MS). Sixty nine compounds in the oil of *G. numidica* representing 87% of the total oil and 58 compounds of *G. saharae*, representing 91% of the total oil were identified. The analysis showed that the main constituents of the essential oils are rich in fatty acid. The major constituent are lauric acid (9.1-8.4%), myristic acid (13.5-14.5%), palmitic acid (15.3-32.3%) and linoleic acid (0-2.4%). The effects of these oils on the growth of *Escherichia coli* (ATCC 25922), *Pseudomonas aeruginosa* (ATCC 27853) and *Staphylococcus aureus* (ATCC 25923) were investigated by the diffusion method. The oils showed no significant antibacterial activities.

**Key words:** *Genista numidica*, *G. saharae*, Algeria, essential oil, antimicrobial activities

## INTRODUCTION

The *Genisteae* (Adans) Benth. is a tribe of Fabaceae, basically circum-Mediterranean. Different palaeoclimatic and geological processes have been involved in the evolution of Mediterranean plant communities, including those characterized by *Genisteae* (Pardo *et al*., 2008; Loget and Driessche, 2006).

The *Genisteae* have a great ecological significance in Mediterranean countries. They colonize degraded forests and deforested areas that characterize the landscape (Lopez Gonzalez, 2001).

The genus *Genista*, consisting of 87 species (Martins *et al*., 2005), among these species 23 grow in Algeria (Maire *et al*., 1987; Quezel and Santa, 1962). Taxonomical criteria were based on leaves, branching pattern, size and shape of petals and legume characters (Gibbs, 1966).

All phytochemical analysis on *Genista* has revealed the presence of flavonoids, isoflavones and alkaloids (Martins *et al*., 2005; Southon, 1994; Pistelli *et al*., 1998, 2000; Giachi *et al*., 2002; Rauter *et al*., 2005). Little information about essential oil of *Genista* is available.

The present study aims to determine and to compare the composition of the oils and antimicrobial activity of two endemic species *Genista numidica* and *G. saharae*.

## MATERIALS AND METHODS

**Plant material:** *Genista numidica* and *G. Saharae* are not spiny perennial shrub. These species are endemic, the first species is trifoliate, grows on moist forest floors commonly called Teqtaq, the second is leafless, named Tellegit grows on sandy soils arid. The origin and ecology of the samples are presented in Table 1.

Aerial parts, during flowering stage, were collected at Taza and Bousaada (Algeria) in May 2008. The chemical tests were conducted on February 2009. Voucher specimens were stored in the herbarium of the Department of Biology, Ferhat Abbas University, Algeria.

Table 1: Sites of *Genista* species sampled

| Samples | Locality | Latitude (N) | Longitude (E) | Altitude (m) | Soil |
|---|---|---|---|---|---|
| *G. numidica* | Taza | 36° 43’ | 5° 33’ | 50 | Forest soil degraded |
| *G. saharae* | Bousaada | 35° 12’ | 4° 10’ | 900 | Sand |

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**Essential oil analysis:** The air dried materials were subjected to hydrodistillation for 3 h using a Clevenger-type apparatus. The oils were analyzed by GC on a Hewlett-Packard 5890 GC series II, equipped with FID, fitted with a SE-54 capillary column (25 m×0.25 mm; 0.25 µm film thickness). The column temperature was programmed from 55°C (5 mn) to 210°C (4 mn) at a rate of 6°C $min^{-1}$. The injector and detector temperatures were programmed at 220°C. Helium was used as carrier gas at a flow rate of 0.6 mL $min^{-1}$, split ratio 1:50. The analysis by GC-MS was performed on a Hewlett-Packard GC-MS system (5890, series II; MSD 5971A, Hewlett-Packard), equipped with SE-54 columns, under the following conditions: splitless.

**Evaluation of the antibacterial activity:** The antibacterial activity of the oil was carried out by the disc diffusion method, according to the National committee of clinical laboratory standards (Kiehlbauch *et al*., 2000) against three of American Type Culture Collection (ATCC) namely: *Escherichia coli* (ATCC 25922), *Pseudomonas aeruginosa* (ATCC 27853), *Staphylococcus aureus* (ATCC 25923) which were obtained from the Microbiology and Parasitology Laboratory of Ferhat Abbas University Hospital. It was performed using an 20 h culture growth at 37°C and adjusted to approximately 105 CFU $mL^{-1}$. Five hundred microliters of the bacterial suspension was spread on the surface of Muller-Hinton Agar plates. Sterile filter paper disks (Whatman No. 1.6 mm in diameter) containing 10 µL of each dilution of the oil (half, 1/4 and 1/8 v/v in the absolute ethanol) were placed on the surface of the media. The plates were left 30 min at room temperature to allow the diffusion of the oil and then they were incubated at 37°C for 24 h. At the end of this period, the inhibition zones were measured. All the experiments were performed in triplicate. Positive (Gentamycin, 10 µg/disc) and negative controls (10 µL ethanol) were also included in the test.

## RESULTS

The average oils yield of these two species was found to be 0.29% based on the fresh weight, which ranged from 0.27 to 0.31% for the two species respectively.

The composition of the oils differed only quantitatively for *G. numidica* and *G. saharae*. The compounds identified in these oils and their relative proportions are listed in their elution as given in Table 2. A number of 96 compounds were characterized, representing 87% of the total oil of *G. numidica* and 91% of the total oil of *G. saharae*.

Table 2: Chemical composition of essential oils of *Genista numidica* and *G. saharae*

| Compounds | KI | *G. numidica* | *G. saharae* |
|---|---|---|---|
| Hexanal | 801 | | 0.09 |
| 4-hydroxy-4-methyl-pentan-2-one | 839 | 0.19 | 0.12 |
| Benzaldehyde | 963 | | 0.22 |
| Oct-1-en-3-ol | 981 | 2.60 | |
| 2-penthylfurane | 990 | 0.25 | 0.20 |
| Octan-3-ol | 997 | 0.07 | 0.05 |
| 6-méthyl-hepta-3,5-dien-2-one | 1103 | | 0.09 |
| Octanal | 1004 | 0.4 | |
| 3-ethyl-4-methyl-pentan-1-ol | 1022 | 0.38 | |
| Oct-2-en-1-ol E | 1067 | 0.45 | |
| Octanol | 1072 | 0.43 | |
| Linalol | 1099 | 0.20 | 0.33 |
| Nonanal | 1104 | 5.67 | |
| Nonadiènal (2E, 6Z)-thymohydroquinone | 1153 | 0.23 | |
| Nonen-1-al <2E-> | 1160 | 0.32 | 0.16 |
| Nonanol | 1172 | 0.27 | |
| Octanoïc acid (caprilic) | 1175 | | 0.33 |
| Terpinene-4-ol | 1183 | | 0.07 |
| Naphthalene | 1186 | 2.72 | |
| Alpha terpineol | 1197 | | 0.14 |
| Safranal | 1200 | 0.25 | 0.10 |
| Decanal | 1206 | 1.10 | |
| Beta cyclocitral | 1221 | 0.35 | |
| Geraniol | 1250 | | 0.23 |
| Beta-apo-8-carotenal + edulan II | 1258 | 0.23 | |
| 5-penthyl-2(3H)-furanone | 1263 | 0.23 | 0.15 |
| Nonanoïc acid | 1274 | 0.50 | 0.77 |
| Thymol | 1292 | 0.35 | |
| 2-methylnaphtalene | 1296 | 0.21 | |
| Theaspirane A | 1301 | 1.67 | |
| Undecanal | 1308 | 0.26 | |

Table 2: Continued

| Compounds | KI | *G. numidica* | *G. saharae* |
|---|---|---|---|
| Theaspirane B | 1317 | 1.56 | |
| 4-vinyl-2-methoxy-phenol | 1313 | 0.59 | |
| (Z,E)-1,3,3-trimethyl-7-oxabicyclo[2.2.1] heptane | 1336 | 0.77 | |
| Decanoïc acid | 1370 | 0.43 | 0.78 |
| Beta damacenone E | 1380 | 1.60 | 2.95 |
| Beta bourbonene | 1387 | 0.30 | |
| 2-(1,3-butadienyl)-1,3,5-trimethyl-benzene | 1392 | 0.23 | 0.28 |
| 1,2-dihydro-2,5,8-trimethyl-naphtalene | 1397 | 0.19 | 0.34 |
| 2,2;6,7-tetramethylbicyclo[4.3.0]nona-4,9(1)-dièn-8-ol | 1407 | 0.34 | |
| 2,7-dimethylnaphtalene | 1408 | | 0.22 |
| 4-(2,6,6-trimethylcyclohexa-1,3-dienyl)butan-2-one | 1411 | 0.76 | 0.78 |
| 2,3-dimethylnaphtalene | 1419 | 0.38 | |
| 1-(6,6-dimethyl-2-methylene-3-cyclohexenyl)-buten-3-one | 1428 | 0.12 | |
| 2,6-dimethylnaphtalene | 1429 | | 0.15 |
| Nerylacetone | 1446 | 2.18 | 0.48 |
| Gamma muurolene | 1477 | 0.30 | 0.25 |
| (E)-beta-ionone | 1480 | 1.94 | 0.17 |
| Tridecanal | 1511 | 0.23 | |
| Cyclopentanoate d'ethenyle | 1513 | | 0.56 |
| 1,6,7-trimethyl-naphtalene | 1527 | 0.26 | |
| Elemol | 1551 | | 0.08 |
| Dodecanoïc acid (lauric) | 1579 | 9.11 | 8.429 |
| Megastimatrienone-4 | 1595 | | 0.162 |
| Pseudoionone (E,E) | 1596 | 0.40 | |
| Oxyde de caryophyllene | 1599 | 0.52 | |
| Tetradecanal | 1613 | 0.24 | |
| Megastigmatrienone 2 | 1625 | | 0.094 |
| Dodecanoïc acid trimethylsilyl ester | 1652 | | 0.591 |
| Ethyldibenzothiophene | 1669 | | 0.094 |
| Tetradecanol | 1677 | 0.60 | 0.67 |
| Heptadecane | 1697 | 0.23 | |
| Pentadecanal | 1715 | 1.86 | 0.41 |
| 2,6-diisopropylnaphtalene | 1721 | 0.32 | |
| Diisopropylnaphtalene | 1726 | 0.25 | 0.18 |
| Tétradécaoïc acide (myristic) | 1772 | 13.49 | 14.50 |
| Linolenate de methyle | 1789 | 0.06 | |
| 6,10,14-trimethyl-pentadecan-2-one | 1841 | 1.87 | 4.45 |
| Tetradecanoate de trimethylsilyle | 1846 | | 0.74 |
| Phthalate | 1858 | 0.88 | 0.99 |
| Gamma tetradecalactone | 1885 | | 0.13 |
| Linoleate de methyle | 1886 | 0.37 | |
| Tetradecan-3-en-5-yne | 1891 | 2.33 | |
| Nonadecane | 1899 | 0.21 | 0.26 |
| Fanésylacetone | 1908 | 1.19 | 1.20 |
| Hexadecanoate de methyle | 1923 | 0.33 | |
| Heptadecanol | 1930 | 0.25 | |
| Phytol | 1944 | | 0.21 |
| Iso phthalate | 1953 | 0.30 | |
| n-hexadecanoïc acid (palmitic) | 1989 | 15.34 | 32.32 |
| (E,E)-2,11,15-trimethyl-3-methylenehexadeca-1,6,10,14-tetraene | 2022 | 1.17 | |
| Geranyl linalool | 2023 | | 0.24 |
| Heptadecanoïc acid | 2061 | | 0.20 |
| Eicosane | 2099 | 0.44 | 0.22 |
| Uneincosane | 2099 | | 0.42 |
| Trans phytol | 2110 | 2.60 | 4.56 |
| Octadeca-9,12-dienoïc acid (linoléic) | 2138 | | 2.37 |
| Linolenoate de methyle | 2145 | | 2.01 |
| 9,12,15-octadecatrienoate de methyle | 2146 | 5.67 | |
| Octadecanoïc acid (stearic) | 2171 | | 0.23 |
| Docosane | 2198 | | 0.17 |
| Tricosane | 2299 | 0.25 | 0.55 |
| Tetracosane | 2398 | | 0.23 |
| Pentacosane | 2501 | 0.69 | 1.27 |
| Hexacosane | 2598 | 0.23 | 0.37 |
| Heptacosane | 2700 | | 2.76 |
| Total | | 87.04 | 91.03 |

Table 3: Antibacterial activity of *Genista numidica* and *G. saharae* oil *in vitro*

| | Inhibition zone (mm) | | | | | | |
|---|---|---|---|---|---|---|---|
| | 1/2 | | 1/4 | | 1/8 | | |
| Strains (C) v/v | Num | Sah | Num | Sah | Num | Sah | Gen |
| *Escherichia coli* ATCC 25922 | - | 8 | - | 8 | - | - | 32 |
| *Pseudomonas aeruginosa* ATCC 27853 | - | 7.9 | - | 6.6 | - | - | 20 |
| *Staphylococcus aureus* ATCC 25923 | - | 8.7 | - | 6.8 | - | 6.2 | 30 |

num = *Genista numidica*; sah = *Genista saharae*; Gen. = Gentamycine (10 µg/disk); Inhibition zone (diameter of the disk, 6 mm, include), values represent average of three determination

The qualitative difference of the oil of these two species is very low; however the quantitative difference is important. The composition of essential oil from *Genista numidica* and *G. saharae* is slightly different.

This oil is very rich in fatty acids; lauric acid (14.32-8.46%), myristic acid (11.45-4.98%), palmitic acid (18.63-26.4%) and linoleic acid (3.08-11.71%). The essential oils of both species are very low in terpenoids.

Many plant derived essential oils are known to exhibit antimicrobial activity against a wide range of bacteria. The in-vitro antimicrobial activities of the essential oil of this species were not reported previously. The antibacterial activity of the *Genista* species essential oils in comparison with Gentamycin is shown in Table 3.

The bacteria tested were resistant to the studied essential oil concentrations. The half dilution of *Genista saharae* oils decreases the density of the growth of *Escherichia coli* ATCC 25922, *Pseudomonas aeruginosa* ATCC 27853 and *Staphylococcus aureus* ATCC 25923, on a halo of 7.9-8.7 mm. *G. numidica* shows no bacterial activity.

## DISCUSSION

The antibacterial activities of fatty acids have been well known for many years (Walters *et al*., 2004; Agoramoorthy *et al*., 2007). Some fatty acids have been demonstrated to be bactericidal to important pathogenic microorganisms including antibiotic resistant *S. aureus* (Sun *et al*., 2003; Shin *et al*., 2007). Free fatty acids can be regarded as potential bactericides (Hinton and Ingram, 2000; Lee *et al*., 2002; Mbandi *et al*., 2004). The natural concentrations are not sufficient to cause the total inactivation of pathogens (Sado-Kamdem *et al*., 2008). The results showed that *S. aureus* been most sensitive compared to *Echerichia coli* and *P. aeruginosa.* The antibacterial activity is absent for both species with 1/8 dilution. The reduced growth is due to the presence of fatty acids as demonstrated by Shin *et al.* (2007) on *S. aureus* and *P. aeruginosa*, this action is proportional to concentration of fatty acids in oil (Sylvain *et al*., 2009).

In brief, the essential oils analysis carried out on two species (*Genista numidica* and *G. saharae*) showed the presence of faty acids variability within species. They emphasize the abundance in fatty acids; lauric acid, myristic acid, palmitic acid and linoleic acid. The essential oils of *Genista* species show no important biological activity.

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