

Phytochemical and Antioxidant Activity of the Essential Oils of *Satureja briquetii* L. from Morocco

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ABSTRACT

Satureja briquetii L. (Labiatae) species are a well-known aromatic plant which is used to produce essential oils and aromatic water in the mountain regions of Sefrou part of Morocco. In our study, it was aimed to determine phytochemical and antioxidant activities of *Satureja briquetii* L. essential oils *in vitro*. Antioxidant activities of the oils at different concentrations were evaluated using the 1,1-diphenyl-2-picrylhydrazyl (DPPH) radical scavenging. The extraction of essential oils from aerial part of this plant is carried out by hydrodistillation, and yields are 1.75% for dry aerial part and 1.05% for fresh aerial part. The analysis of the essential oils of dry aerial part from *Satureja briquetii* L. by GC/MS identified 57 principles compounds including Spathulenol (9.81%), Verbenone (4.35%), Camphene (3.56%), Himachalene (3.2%) and Cedrene (2.12%). The chromatographic profile of essential oils from fresh aerial part of *Satureja briquetii* L. has 49 constituents representing 96.25% of the essential oil, where the Menth-8-ene (14.99%), Cymene (4.97%) and Carene (4.97%) are major compounds. The Menth-1,4(8)-diene, (1.06%), Cubenol and Longifolene (3.44%) are in minority. Furthermore, the antioxidant activity of the essential oil of *Satureja briquetii* L. was evaluated by the method of DPPH, and showed a significant efficiency in radical DPPH reducing with an IC₅₀ value of $31.027 \pm 0.586 \mu\text{g/ml}$ from essential oils of dry aerial and $35.034 \pm 0.0432 \mu\text{g/ml}$ in essential oils from fresh aerial part.

Keywords: *Satureja briquetii* L., essential oils, GC/MS, hydrodistillation, chemical composition, antioxidant activity.

INTRODUCTION

Medicinal and aromatic plants (MAP) represent a considerable economic interest in perfume, cosmetics and food industries for their antioxidant activities and flavoring, and in pharmacy through their antiseptic, analgesic, anti-inflammatory, and antispasmodic properties¹. Indeed, they constitute an inexhaustible reservoir of folk remedies and a natural source of most currently used drugs². Essential oils (EO) from plants have a plurality of properties mainly due to their complex chemical composition³. These oils have a very broad spectrum of action since they inhibit both the proliferation and synthesis of bacterial toxins and yeast, they act on the biomass and production of pseudomycelium and mold, they inhibit the spore germination, the mycelium elongation and toxin production. EOs also have antiviral, immunostimulant, anti-inflammatory, analgesic actions, and stimulating gastric motility⁴. Currently they are studied to better determine their effectiveness as natural preservatives. Through its geographical position, Morocco is characterized by an ecological diversity, which results in a great diversity of flora and a high rate of endemism⁵. Morocco is a traditional producer of MAP; it is one of the

leading worldwide suppliers of rosemary, verbena, coriander, pennyroyal, thyme and lavande etc.; and an exclusive supplier of several EOs as wormwood, wild chamomile and annual Tansy. Morocco also has an ancestral knowledge of medication by plants, their use for flavoring and preserving food⁶.

In Western medicine 74% of the 121 bioactive plants derived compounds currently in worldwide use were identified via research based on leads from ethnomedicine⁷. In the area of the Balkan Peninsula, different *Satureja* species have been used in traditional medicine to treat bronchitis, skin, respiratory, digestive and urinary inflammation⁸. This has been confirmed by scientific data which pointed out high antimicrobial activity of essential oils isolated from different species of genus *Satureja*^{9,10}. Major active constituents of their essential oils are phenolic compounds, carvacrol and thymol¹¹.

The leaves, flowers and stems of *Satureja* species are used as herbal tea, in production of traditional medicine, to treat various ailments, such as cramps, muscle pains, nausea, indigestion, diarrhea and infectious diseases^{12,13,14}.

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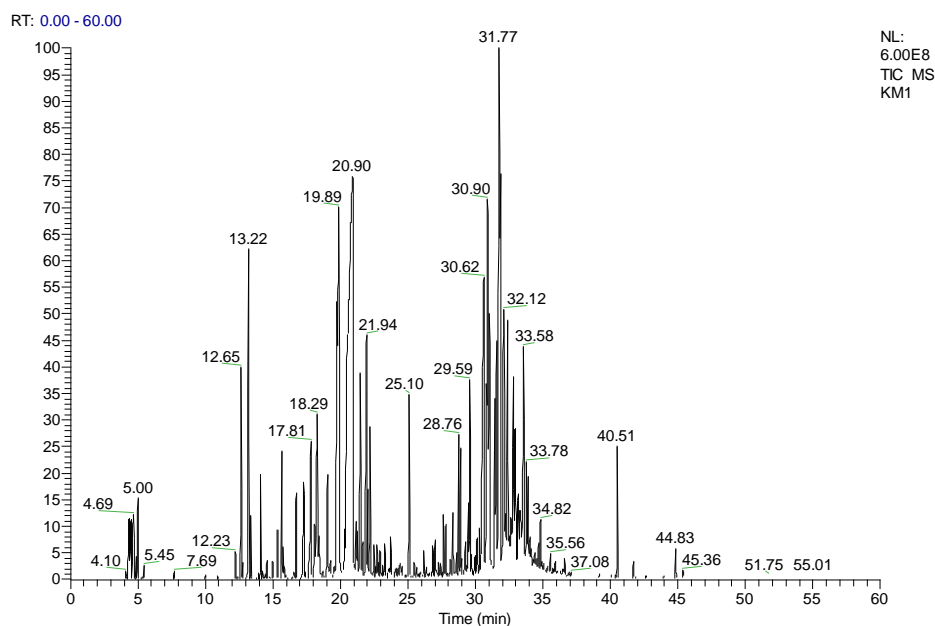


Figure 1: Chromatogram of dry aerial part of *Satureja briquetii* L. by GC/MS.

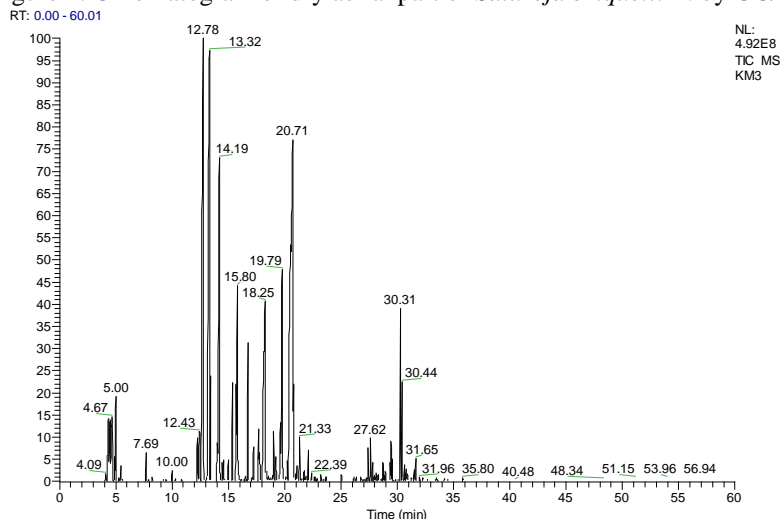


Figure 2: Chromatogram of fresh aerial part of *Satureja briquetii* L. by GC/MS.

MATERIAL AND METHODS

Plant Material

Samples of *Satureja briquetii* L. were collected in May 2016 from karkoura, located between Sefrou and Al Menzel (Morocco). The dry aerial part of *Satureja briquetii* L. was shade dried (25 days) at room temperature.

Essential oil extraction

The extraction of the plant's essential oils is carried out using a Clevenger device, a type of reactor 2000 ml flask. The total duration of the hydrodistillation is 3 hours, with a temperature of 65 °C, and distilled water volume of 500 ml. After calculating the performance, essential oils was stored at 4 °C in the dark¹⁵.

Chromatographic analysis of samples essential oils

For separation and determination of composition of the essential oils of *Satureja briquetii* L., the analysis was performed at the Innovation Center (Fez-Morocco) through a coupling of gas chromatography with mass spectrum (GC/MS) type (Polaris Q) ion in electron impact hatch (AEs) with ionization energy of 70 ev. The column

used is nonpolar capillary column type silica (WCOT Fused Silica) with a stationary phase (CP-SIL5CB), 50 m in length; the column temperature is programmed from 40 to 280 °C at 3 °C/min. The injector temperature is set at 240 °C and the detector (ionization source) is 200 °C. The flow rate of carrier gas (Helium) is set at 1ml/min. The volume of sample injected is 1µl of essential oils diluted in hexane. The components of the essential oils were identified by comparing their mass spectra with those listed in a type library (NIST-MS).

Antioxidant activity by the scavenging method of free radical DPPH

The evaluation of the antioxidant activity of *Satureja briquetii* L. essential oils was made by the scavenging method of free radical DPPH (1,1-diphenyl-2-picrylhydrazyl). The absorbance measured at 517 nm is used to calculate the percent inhibition of DPPH radical which is proportional to the anti-radical power of essential oil of *Satureja briquetii*. DPPH molecule (1,1-diphenyl-2-picrylhydrazyl) is characterized by a stable free

Table 1: Chemical composition of EO from dry aerial part from *Satureja briquetii* L.

Pics	RT (min)	Area %	Compounds
1	12,64	1,7	Terpinene
2	13,22	3,56	Camphene
4	15,34	1,1	Menth-1,4(8)-diene
5	21,93	4,35	Verbenone
6	25,11	1,29	Carene
7	28,75	0,85	Longipinene
8	31,76	9,81	Spathulenol
9	32,40	2,12	Cedrene
10	33,57	3,2	Himachalene

Sample 2: fresh aerial part of *Satureja briquetii* L.

Table 2: Chemical composition of essential oils from fresh aerial part from *Satureja briquetii* L.

Pics	RT (min)	Area %	Compounds
1	13,32	14,99	Menth-8-ene
2	15,36	1,06	Menth-1,4(8)-diene
2	15,69	4,97	Cymene
4	15,79	4,97	Carene
5	16,74	1,64	Terpinene
6	30,30	2,13	Longifolene
7	30,44	2,13	Cubenol

Table 3: Comparison of the chemical composition between the essential oils from dry aerial part and from fresh aerial part from *Satureja briquetii* L.

Samples	Compounds	Major constituents	Yields
Essential oils from dry aerial part	Terpinene	Spathulenol Verbenone Camphene Himachalene	1.75
	Camphene		
	Menth-1,4(8)-diene		
	Verbenone		
	Carene		
	Longipinene		
	Spathulenol		
	Cedrene		
	Himachalene		
	Menth-8-ene		
Essential oils from fresh aerial part	Menth-1,4(8)-diene	Menth-8-ene	1.05
	Cymene	Cymene	
	Carene	Carene	
	Terpinene		
	Longipinene		
	Cubenol		

radical with an absorption band in methanol solution centered at about 517 nm, in presence of electron donor, DPPH is reduced to 1,1-diphenyl-2-hydrazine DPPH2 thus the violet color is changed to yellow due to the presence of picryl.

In order to evaluate the antioxidant potential through free radical scavenging by tested sample, the change in optical density of DPPH radicals is monitored. The solution of DPPH is prepared (0.0042g in 200 ml of methanol). The sample extract of *Satureja briquetii* L. is prepared at a concentration of 1mg/ml in methanol and is diluted with

methanol and 2 mL of DPPH solution is added. After 30 min, the absorbance is measured at 517 nm.

Percent inhibition of DPPH

The percentage of the DPPH radical scavenging is calculated using the equation as given below¹⁶:

$$\text{DPPH (\%)} = \frac{(\text{DO control}) - (\text{DO spl})}{(\text{DO control})} \times 100$$

DO control: optical density of the negative control tube.

DO spl: optical density of the sample.

IC50 determination

The IC₅₀ value is the concentration which provides 50% of the activity of DPPH and that constitutes the antioxidant activity of essential oils determined graphically from the curve of the percentage inhibition versus concentration of EO¹⁷. This value is compared to that found in the reference antioxidant, ascorbic acid.

RESULTS AND DISCUSSION

Comparison of essential oils yield

The extraction from the dry plant gives a yield of 1.75 ± 0.07% which is higher compared to that obtained from fresh plant (1.05 ± 0.05%). If we compare the yields of our plant's essential oils with a plant produced in another region of Morocco, we note that the essential oils yield obtained for our study of dry plant (1.75 %) and from fresh plant (1.05%), is relatively high to the work done by essential oils by¹⁸ of 1.3% for dry plant of *Satureja briquetii* L. and a yield of 1.35 from *Satureja atlantica*¹⁸.

Chemical composition

Sample 1: dry aerial part of *Satureja briquetii* L.

The chromatographic profile of essential oils from dry plant of *Satureja briquetii* L. shows relative abundance of different compounds based on their output time in minutes. The first peaks appear after 14 minutes, the majority of components are grouped between 14 and 30 minutes with varying abundances.

This chromatogram identified 57 constituents representing 96.25% of the essential oil, with a 1.75% yield, including Spathulenol (9.81%), Verbenone (4.35%), Camphene (3.56%), Himachalene (3,2%) and Cedrene (2,12%) (Table 1).

The chromatographic profile of essential oils from fresh aerial part of *Satureja briquetii* L. has 49 constituents representing 96.25% of the essential oil (between 12 and 21 minutes) with a yield of 1.05%, where the Menth-8-ene (14.99%), Cymene (4.97%) and Carene (4.97%) are major compounds. The Menth-1,4(8)-diene (1,06%), Cubenol and Longifolene (2,13%) are in minority (Table 2).

Other study¹⁸ indicate that essential oils of dry aerial part of *Satureja briquetii* L. characterized by the presence of the borneol with 27.64% as a major component followed by the β-bisabolene (9.58%), α-pinene (6.97%), linalool (6.77) and Camphene (5.73%).

Antioxidant activity of essential oils *Satureja briquetii* L.

The antioxidant activity of plants is mainly contributed by the active compounds present in them. The DPPH radical scavenging is a sensitive antioxidant assay and is widely used to evaluate antioxidant activities in a relatively short time compared with other methods. Antiradical activities of *Satureja briquetii* essential oils was measured by this

Table 4: DPPH radical scavenging of essential oil from *Satureja briquetii* L.

Sample	DPPH IC50 (µg/mL)		Essential oils from fresh aerial part
	Essential oils from dry aerial part	Ascorbic acid	
<i>Satureja briquetii</i> oils	31.027 ± 0.586	27.165 ± 0.841	35.034 ± 0.0432

method. The linear curve of DPPH allows us to determine the IC50 value of *Satureja briquetii* L. essential oils. As shown in Table 4, essential oil of *Satureja briquetii* possessed an interesting DPPH-scavenging activity, showing an IC50 of 31.027 ± 0.586 µg/ml in essential oils from dry aerial part and IC50 of 35.034 ± 0.0432 µg/ml in essential oils from fresh aerial part. The results of this study confirm the antioxidant activity of the essential oil of *Satureja briquetii* L. of Sefrou region in Morocco.

Indeed, the essence of *Satureja briquetii* L. and vitamin C could reduce the DPPH radical resulting in a change in the color of DPPH solution in methanol with values of 31.027 ± 0.586 µg/ml in essential oils from dry aerial part, IC50 of 35.034 ± 0.0432 µg/ml in essential oils from fresh aerial part and 27.165 ± 0.841 µg/ml, respectively. The absorbance of ascorbic acid (antioxidant standard) was measured under the same conditions as the samples. These results show that the essential oil of *Satureja briquetii* L. has antioxidant activity close to that of than vitamin C.

CONCLUSION

The present study was conducted to investigate the chemical composition and antioxidant activity of essential oils of *Satureja briquetii* L. from Morocco. The extraction of the essential oils by Cleverger gives a yield of 1.75% from dry plant; which was more important than that obtained from the fresh plant (1.05%). The chromatograms of the essential oils of dry plant of *Satureja briquetii* L. show that Spathulenol (9.81%), Verbenone (4.35%), Camphene (3.56%), Himachalene (3,2%) and Cedrene (2,12%) are the main constituents of the 57 identified. The chromatographic analyzes of essential oils from fresh aerial part from *Satureja briquetii* L. highlighted the predominance of the Menth-8-ene (14.99%), Cymene (4.97%) and Carene (4.97%), while the Menth-1,4(8)-diene (1,06%), Cubenol and Longifolene (2,13%) are in minority. The results obtained by the method of DPPH confirm the antioxidant potential of the essential oils of *Satureja briquetii* L. from Sefrou region of Morocco. Indeed, the essence of *Satureja briquetii* L. from dry aerial part, fresh aerial part and vitamin C could reduce the DPPH radical in solution of DPPH methanol with values of 31.027 ± 0.586 µg/ml, 35.034 ± 0.0432 and 27.165 ± 0.841

respectively, these results show that the essential oils of *Satureja briquetii* L. has antioxidant activity, near to that of vitamin C. The results of this study can contribute to the enhancement of essential oils of this plant by the local production of this species. The antioxidant activity also suggests application prospects in the fields of food, cosmetics, pharmaceutical and of herbal medicine. The results of this study could contribute to the valorization of this Moroccan aromatic and medicinal plant. This study can be considered as an important source of information on chemical properties and antioxidant power of the essential oils of Moroccan *Satureja briquetii* L.

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