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Research Article

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The antibacterial effect of the sagebrush essential oil (*Artemisia herba-alba* Asso.) of Western Algeria

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ABSTRACT

The essential oil of sagebrush (Artemisia herba-alba Asso.) of western Algeria is subject to a physico-chemical and microbiological study. The extraction of the essential oil was carried out by hydrodistillation using a Clevenger-type apparatus. The yield obtained from the aerial parts of this plant is interesting for industrial exploitation. Analyses of the sagebrush essential oil were carried out to determine the physical and chemical indices. The study of the essential oil activity on the bacterial strains (Escherichia coli, Bacillus subtilis, Staphylococcus aureus) by the technique of Vincent (aromatogram) showed that all of the tested bacterial strains were highly sensitive.

Keywords: Antibacterial activity, Aromatogram, Sagebrush, Essential oil.

INTRODUCTION

Algeria is a country with a diverse aromatic wild flora that is widely used in traditional medicine. Over the past few years, several research groups have focused on the chemical characterization and the biological activities of natural products. Sagebrush (*Artemisia herba-alba* Asso) is a steppe plant from the family *Asteraceae*, known traditionally for its anti-diabetic and antispasmodic effects [1,2]. It is commonly called “Chih” throughout the Middle East and North Africa. The sagebrush is a perennial shrub that forms clumps of 30 to 70 cm, white and woolly, stem numerous and tomentose. The leaves are short, usually pubescent with silvery flower heads with 2-5 sessile or sub - sessile flowers. These latter are hermaphroditic, while the fruit is an achene. The receptacle is naked and the corolla is inserted very obliquely on the ovary [3].

The works carried out, so far, on this species are rather of chemotaxonomic interest [4,5]. The sagebrush is characterized by its richness in essential oils, its high feeding value and its very important ecological role in the fight against desertification [6,7]. With the intent to contribute to a better recovery of this species, this modest study endeavoured to determine the physico-chemical characteristics and the antibacterial efficacy of the sagebrush essential oil vis-a-vis strains originating from patients suffering, for the most part, from urinary tract infections.

EXPERIMENTAL SECTION

Sampling sites of sagebrush steppe are located in the region of El Aricha in Western of Algeria (Figure 1).

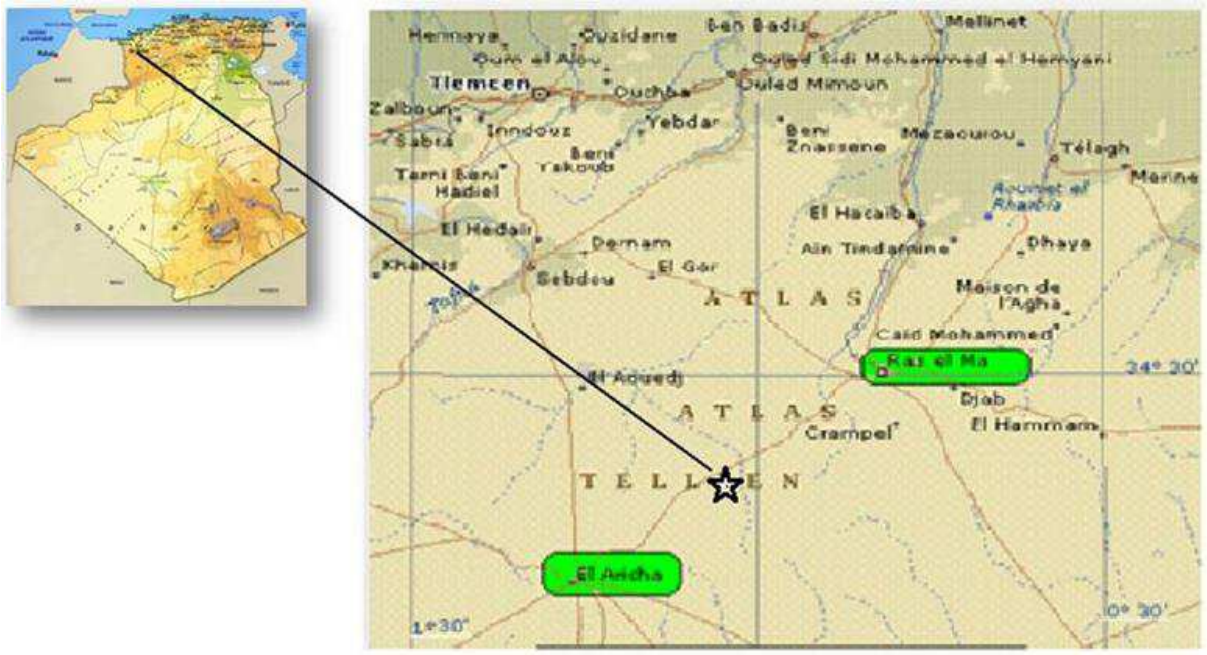

Figure 1: Location map of sampling site (Scale: 1 / 25 000)

The harvest of the sagebrush aerial parts (*Artemisia herba-alba* Asso) was collected in May 2012 from a homogeneous population (Figure 2).

Figure 2: Stand and plant of sagebrush (*Artemisia herba-alba* Asso)

Biomass of the studied species was dried for 10 days under shelter at room temperature laboratory (25-28 °C). The essential oil was obtained by hydrodistillation of the dried leaves using the Clevenger-type apparatus [8]. The water fraction was separated from the organic phase by using dichloromethane. The solvent was removed at room temperature under reduced pressure on a rotary evaporator yielding the oils. The oils obtained were stored under nitrogen in a sealed vial in the dark at 4 °C.

The biological activities of essential oils of *Artemisia herba-alba* Asso were tested against *Bacillus subtilis* (ATCC 6633), *Staphylococcus aureus* (ATCC 25923) and *Escherichia coli* (5044172), that were isolated from microbiological analyses at the hospital laboratory of Sidi Bel-Abbes country.

The technique used in our research is the method of Vincent (aromatogram) which is believed to permit the examination of the sensitivity and resistance of bacteria to essential oils, in a reliable and reproducible manner. [9]. Stressing the sensitivity and resistance of the antimicrobial agents, they were put on a culture based on Mueller-Hinton agar [10]

The emulsification is performed with Tween 80 to 10% to promote contact germ / compound. The dilutions were prepared by aseptically adding a quantity of essential oil in the solution of Tween 80 in order to have a final volume of 60 µl.

The sensitivity of germs tested with essential oils was characterized by the formation of a clear circle (zone of inhibition) around the disks containing these oils. To this effect, the inhibitory effect of essential oils was evaluated by determining, in millimeters, the diameter of the inhibition zone formed [11,12]. The minimum inhibitory concentration (MIC) is the lowest concentration of essential oil to which we do not observe a visible bacterial growth after incubation for 24 h at 37°C [13].

RESULTS AND DISCUSSION

The essential oil of sagebrush is brown and has a strong aromatic odor. The results of the physical and chemical analysis are summarized in Table 1.

Table 1 : chemical and physical values of the essential oil of sagebrush in El Aricha (western Algeria)

Properties	Results
Density relative to 20°C	0,84
Miscibility with ethanol	1v/3v
Ph	6,82 (at 18,9 °C)
Salinity	0
R.S.S (Rate of substances in Solution)	15 mg/l
Conductivity	28,76. 10^{-2}
Resistance	34769,23 Ω
Acid value	11,78

Compared to the results obtained from the sagebrush steppe of El Aricha region (Algeria), the sagebrush steppe of Guercif (Morocco) and Matmata (Tunisia) provide low yields of the order of 0.62% and 0.65%, respectively [14,15]. Such difference can be explained by the environmental factors involved in the development of the species (altitude, climate, the soil).

The organoleptic characteristics of the sagebrush essential oil (*Artemisia herba-alba* Asso) reveal its consistency. Miscibility of the oil with ethanol indicates its solubility in alcohols [16].

The value of the acid indicates the presence of free carboxyl group in the structure of the essential oil of sagebrush (*Artemisia herba-alba*. Asso) [16].

The complexity in essential oils is due to terpene hydrocarbons as well as their oxygenated derivatives, such as alcohols, aldehydes, ketones, acids and esters [17,18],

Evaluation of the antibacterial activity of the sagebrush essential oil
After 24 hours of incubation for the three strains tested, the results are shown in Table 2.

Table 2 : Measures the diameter of the inhibition circle of the three stocks tested

Bacterial strains	*Escherichia coli*	*Staphylococcus aureus*	*Bacillus subtilis*
The diameter of inhibition (mm)			
Witness (Tween 80)	0	0	0
Concentration of HE (%)			
100%	9	10	6
75%	3	2	2
50%	2	2	1,5
25%	1	1	1

The biological activity is manifested by the appearance of the inhibition circle of the bacterial growth around the disks containing the essential oil at different concentrations. Each circle a bright area shows the destruction of pathogens and provides an accurate indication of the antibacterial activity of the oil studied. The reading is done by measuring the diameter of the inhibition circles observed. The diameters obtained differ from one bacterium to another ranging from 1 mm to 10 mm. The product tested (Tween 80) has no inhibitory effect on the three tested strains (Figure 3).

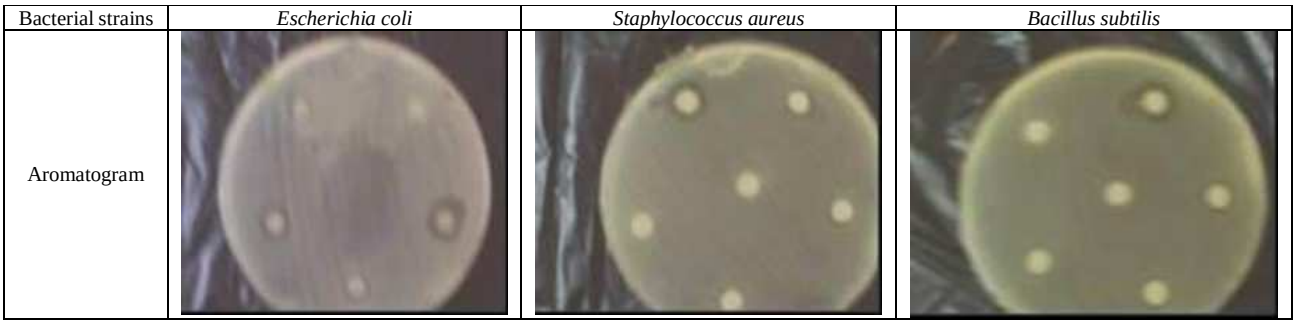

Figure 3: Aromatogram essential oil of sagebrush (*Artemisia herba-alba* Asso) carried out on three bacterial strains

The results indicate a very high sensitivity for the three bacterial strains to the essential oil of sagebrush (Table 2).

Minimum inhibitory concentration (MIC) values of the essential oil of sagebrush are equal for all bacteria studied. The antibacterial activity of the analyzed essential oil can be attributed mainly to its major constituent.

The numerical results and graphs of the essential oil of *Artemisia herba-alba* Asso demonstrate the antibacterial effect vis-à-vis *Escherichia Coli*, *Staphylococcus aureus* and *Bacillus subtilis*.

This antibacterial action can be explained by the fact that the plant produces various secondary metabolites belonging to certain classes known to have this type of activity such as terpenoids and phenolic compounds [19,20,21].

The antimicrobial activities of *Artemisia herba-alba* can be attributed to the presence of camphor, 1.8-cineole and thujone [22,23]. In addition, other minor components such as borneol have been also reported to have antimicrobial potential [24,25].

CONCLUSION

The procurement of essential oil of El Aricha sagebrush steppe by hydrodistillation provided a high yield compared with other works. The results of physico-chemical analysis carried out on the essential oil of sagebrush showed its consistency and purity. Tests of bacterial sensitivity to different concentrations of the essential oil of *Artemisia herba-alba* Asso have revealed the presence of antibacterial activity due to the chemical nature of its constituents and, in particular, its major constituents.

To verify that the effect is specific to the essential oil of sagebrush, emulsions were made with Tween 80 to 10%, to avoid any interaction between the effect of essential oil and that of the emulsifiers. This work confirms conclusively the antibacterial activity of sagebrush through the real effectiveness of its essential oil.

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