

COMPOSITION AND *IN VITRO* ANTIMICROBIAL ACTIVITY OF THE ESSENTIAL OIL OF *Achillea eriophora*

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The *Achillea* species is a chemically polymorphic perennial herb from a genus of complex taxonomy and is widespread throughout Europe, Asia, and North America [1]. Bumadaran is a popular name for several species of *Achillea* in the Persian language. They have been used as anti-inflammatory, antispasmodic, diaphoretic, diuretic, and emmenagogue agents and for treatment of hemorrhage, pneumonia, rheumatic pain, and wounds since ancient times [2]. *Achillea* has a mythological background dating back to Achilles in the Trojan war who used this plant for curing his wounds [1, 3]. These species are also used as medicinal plants for feverish conditions, common cold, and digestive complaints, and are topically used for slow healing wounds, skin inflammations [1], and as veterinary remedy [4].

A survey of the literature revealed a few reports dealing with the chemical composition of *Achillea eriophora* DC. and nothing about its biological activity. This paper describes the chemical composition and antimicrobial activity of this species.

The essential oil of *Achillea eriophora* DC. was analyzed by GC/MS. Thirty-two components were identified representing 98.63% of the total oil (Table 1). 1,8-Cineole (54.93%), linalool (8.92%), α -terpineole (6.66%), and geranyl formate (5.99%) were the main components of the oil. A high number of oxygenated monoterpenes (83.84%), compared with the oil of other *Achillea* species [2–8], are detected, and it seems that *A. eriophora* belongs to a 1,8-cineole rich chemotype (Table 1).

The composition of the essential oil of *A. eriophora* from different locations (at present a few more studies have been done) has been reported before. These reports showed that 1,8-cineole is the major compound, although the amount found is less than that in the present paper. Oxygenated monoterpenes were also characterized as those in our study were observed [7]. The minor component that was identified in this study was a sesquiterpene, chamazulene (0.02%), which has been the object of several studies. Sesquiterpenes are mostly characteristic of taxa of lower (2n–4n) chromosome number.

Numerous studies state that the presence of chamazulene remains characteristics of the members of *Millefolium* group. Only in exceptional cases can references be found describing chamazulene in species outside this group, such as *Achillea wilsoniana* Willd., *A. ageratum* L., or *A. compacta* [3] and now in *A. eriophora*. In the genus *Achillea*, 1,8-cineole exhibits the most frequent appearance among the monoterpenes. It has been described in about one-third of the species and at least in one case as the main component [3].

The *in vitro* antimicrobial tests of the essential oil and ethanol extract of *A. eriophora* resulted in a range of growth inhibition patterns against pathogenic microorganisms. The results were obtained by the disk diffusion method, followed by the measurement of MIC and then MBC for essential oil. According to the disk diffusion method results, all of the microorganisms were inhibited by an amount of 8 μ L/disk of pure essential oil, and none were inhibited by an ethanol extract of 84 μ g/disk, except *Staphylococcus epidermidis* (Table 2).

The most sensitive microorganism against the essential oil was *Staphylococcus aureus*, and the results obtained from *Candida albicans*, *C. kefyr*, *Aspergillus niger*, *A. fumigatus*, and *Pseudomonas aeruginosa* were noticeable. The above-mentioned essential oil possessed antimicrobial activity against *Staphylococcus aureus*, *Aspergillus niger*, and *Bacillus subtilis*, resulting in MIC values of 1 μ L/1 mL of nutrient broth, 0.5 μ L/1 mL for *Escherichia coli*, and 2.5 μ L/1 mL for *Pseudomonas aeruginosa* and *Candida albicans*. MBC values obtained by the above tests show that a concentration of 2.5 μ L/1 mL of this essential oil is bactericidal against *Aspergillus niger* and *Bacillus subtilis*, and for the other four microorganisms *Candida albicans*, *Pseudomonas aeruginosa*, *Escherichia coli*, and *Staphylococcus aureus* the concentration is higher.

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TABLE 1. Composition of the Essential Oil of *Achillea eriophora* Aerial Parts

Component	RI ^a	% ^b	Component	RI ^a	% ^b
α -Pinene		1.18	Lonone	1487	Tr.
β -Pinene		1.80	Bicyclogermacrene	1494	0.12
α -Terpinene	1022	0.26	Unknown	1504	0.02
1,8-Cineole	1046	54.93	Unknown	1539	0.04
γ -Terpinene	1066	0.64	Unknown	1551	0.03
<i>p</i> -Mentha-3,8-diene	1075	0.15	Elemol	1557	0.14
α -Terpinolene	1092	0.14	Unknown	1577	0.32
Linalool	1120	8.92	Caryophyllene oxide	1587	2.35
Unknown	1135	0.15	Unknown	1600	0.07
Camphor	1157	3.89	Unknown	1605	0.08
Pinocarvone	1165	0.16	Unknown	1609	0.18
Terpinen-4-ol	1190	2.9	Eremoligenol	1634	0.11
α -Terpineol	1211	6.66	Caryophylla-4(14),8(15)-dien-5, β -ol	1646	0.80
Myrtenol	1216	0.9	1641 β -Eudesmol	1658	0.97
Bornyl acetate	1289	0.08	Unknown	1662	0.07
Geranyl formate	1305	5.99	Unknown	1692	0.23
Unknown	1330	0.03	Chamazulene	1727	0.02
Unknown	1340	0.13	Hexahydrofarnesyl acetone	1847	Tr.
Eugenol	1366	0.09	1,2-Benzendicarboxylic acid	2518	Tr.
Unknown	1376	0.06	Identification, %		98.63
Unknown	1393	0.06	Group components, %		
Z-Jasmone	1409	0.43	Monoterpene hydrocarbons		4.14
Methyl eugenol	1416	0.24	Oxygen-containing monoterpenes		83.47
<i>E</i> -Caryophyllene	1426	5.00	Sesquiterpene hydrocarbons		5.39
α -Humulene	1454	0.18	Oxygen-containing sesquiterpenes		4.77
Alloaromadendrene	1460	0.07	Others		0.93
β -Selinene	1484	0.03			

^aThe retention index of compounds was determined on the HP-5MS.^bPercentages were calculated based on the concentration obtained on the same column.

Tr.: trace.

TABLE 2. Antimicrobial Activity of *A. eriophora* Essential Oil

Microorganism	Inhibition zone ^a , μL ^b			Gentamicin	Ampicillin	Amphotericin-B
	2	4	8			
<i>Staphylococcus aureus</i>	+	+++	++++		++++	
<i>Staphylococcus epidermidis</i>	N.a.	+	+++		++++	
<i>Bacillus subtilis</i>	+	+++	++++		++++	
<i>Enterococcus faecalis</i>	N.a.	+	++		++	
<i>Escherichia coli</i>	++	++	+++	++++		
<i>Pseudomonas aeruginosa</i>	++	+++	+++	++++		
<i>Salmonella typhi</i>	N.a.	++	+++	++++		
<i>Candida albicans</i>	++	+++	++++			++
<i>Candida kefyr</i>	++	+++	++++			++
<i>Aspergillus niger</i>	N.a.	++	+++			++++
<i>Aspergillus fumigatus</i>	N.a.	++	+++			++++

^a+: 1-4 mm; ++: 5-9 mm; +++: 10-14 mm, ++++: >14 mm. N.a.: not active.^bMicroliters of the *A. eriophora* essential oil were applied to the disks.

In general, Gram negative bacteria were more resistant to the essential oils [10]. The reason that the essential oil was active against different microorganisms was that the naturally occurring constituents of this essential oil consist especially of large amounts of eucaliptol (1,8-cineole), borneol, champhor α -pinene, bornyl acetate, terpinen-4-ol, a group of monoterpenes, and eugenol. Borneol and other phenolics were responsible for this activity [1, 3, 7–10].

As a result, it is obvious that this material, according to the results published in different references, has suitable antimicrobial activity against respiratory tract pathogens and opportunistic pathogens such as *Pseudomonas aeruginosa*, and it can be administered in colds and coughs related to airway microbial contamination. On the other hand, it seems that *A. eriophora* essential oil can be used as a natural antimicrobial and food preservative with a low risk from other relevant agents.

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