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ANALYSIS AND ANTIMICROBIAL ACTIVITY OF THE PLANT *JUNIPERUS COMMUNIS*

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ABSTRACT

Today a medicinal natural product to profit for treatment of numerousness illnesses. Juniperus communis is pharmaceutical plant from erea in Golestan province in Iran. From point of view ecophysiology organs, this species grows on dry, cold-tolerant, variety of soil types including,acidic and calcareous sands, pH of 4.5, which is rated as strongly acid, genuse plant and withdrawal May month, morning time have more effect of chemical materials and antibacterial. That inguiry had tried different organs J. communis point of essensial oil female cone and leaf. I had stady instance some of the bacteria E.coli, Staphylococcus, Sodomunas. That used with equisetum, taraxacum and urtica dioica against UTIs, infection, cancer, and various urinary tracts kidney disorders. In this study ethanolic extracts of fleshy feale cones of plants that collected in May 2005 from natural habitat in Charbagh village (1950 m) from Golestan. Juniperus communis is one most important conifers in Iran and since are most tolerance trees against the cold and drought stesses and have noticeable important in mountainous areas in Iran. Oil from representative samples of population as well as from individual samples was studied. The oils consisted mainly of monoterpene hydrocarbons. Analysis of essentials oil, dried fruits was detected after disitillation by GC-MS.The results indicated that effeciacious materials of this plant were different, significantly. In J. communis 27 essential oils were detected that its highest values were α-pinene and α-cedrol, DETA.3-carene, α-terpinolen, and terpineol-4. In this study.

Keywords: Juniperus communis, α-pinene, α-cedrol, DETA.3-carene, α-terpinolen, and terpineol-4.

INTRODUCTION

Juniper is an evergreen coniferous shrub or small tree occurring throughout the northern hemisphere from Europe to Siberia and grows up to 10m in height [1]. The medicinal portions of the plant are referred to as berries, but they are actually dark blue-black scales from the cones of the tree [2]. Unlike other pine cones, the juniper cones are fleshy and soft [3]. Juniper has a history of medicinal use dating as far back as 1550 B.C. A remedy to treat tapeworm was found [4] in "The Papyrus of Ani" from ancient Egypt, 240 B.C. It is also known that the branches and berries were burned in temples as a part of purification ceremonies. In the Ayurvedic system juniper is believed to not only purify the body but also the aura or subtle body. It also helps to destroy negative astral influences [2]. It was burnt by 15th century herbalists to guard against the plague, in French hospitals to clear the air, and by Native Americans for purification and healing. In more modern times the berries are used to flavour gin by extracting their essential oil and in cooking as well as in herbal medicine [5]. Juniper is an important spice in many European cuisines, especially in Alpine regions, where juniper grows abundantly [6]. It is the only example of a spice in the botanic group of the *coniferae*, and also one of the few examples of spices from cold climatic regions, though the best quality stems from southern European countries[7]. Juniper is much used in the traditional, German speciality *Sauerkraut*. For its preparation, fresh cabbage is preserved by lactic fermentation and seasoned with juniper, caraway and a few bay leaves [8]. The taste then develops during ageing in large wooden barrels. *Sauerkraut* can either be eaten raw (as a kind of salad), or be cooked or fried (often together with small cubes of ham)

to be served as a side dish; there are also dumplings stuffed with it [9]. The ripe, dark blue berries are used for herbal remedies. As it takes 2 to 3 years for the berries to reach full ripeness, both green, unripe berries and dark, blue berries can be found growing together on the same plant [10]. Local Iranian names are Hovars, Avars (in Caraj valley), Ors (in Khorasan), Arduj (in Manjil & Khalkhal), Arbas (in Manjil), Archeh (in Goshankhaneh & Soaldy), Abol (in Bakhtiari), Vors (in Amol & Haraz valley), Archa, Orsa, Qara arsha [11]. The berries are normally harvested in the autumn of their second year when they are blueish-black in colour. They should be dried carefully to preserve the volatile oil. The fresh berries can be made in to syrup. Juniper berries have a bittersweet taste and a hot, drying energy and, because of this characteristic, they should be used sparingly with appropriate balancing herbs [12]. Juniper berries owe their use to an essential oil, content 0.2 to 2% dependent on provenance [13]. Hungarian berries contain 1.2%, German berries only 0.7% and Iran berries 0.9%. The essential oil is mainly composed of monoterpenes: 80% alpha- and beta-pinene, thujene, sabinene, 5% terpinene-4-ol, alpha-terpineol, borneol and geraniol; sesqiterpenes (α- and β-cadinen, caryophyllene) are found in traces. The essential oil from this plant was analyzed for the chemical components [11]. It was also tested for its antibacterial activities against a wide spectrum of Gram-positive and Gram-negative bacteria [14]. In addition, the essential oil was subjected to antifungal and antioxidant testing. The major chemical components obtained were, camphene, *a* -pinene, *B*-pinene, terpinene, limonene, *B*-myrcene, terpinene-4-ol, linalool, *trans*-carryophyllene, *a*-phellandrene, 3-carene, *a*-terpineol and germacrene [15]. The essential oil exhibited notable antibacterial activity against Grampositive and Gram-negative bacteria as well as significant antifungal and antioxidant activity [16]. The major equipments used were clevenger; GC/MS (Varian-3400), other chemicals were of analytical grade. *Juniperus communis* was collected from part of the mountainous in Golestan Chaharbagh of Iran during May-June 2006.

EXPERIMENTAL

Oil extraction and analysis

The dray powder of plant materials were steam distilled for 1.5 h in full glass apparatus. The oils were isolated using a Clevenger- type apparatus. The extraction was carried out for 6-8-h in 500 round bottom flask. The GC/MS unit consisted of Varian- 3400 gas chromatograph coupled to a Saturn II ion trap detector. The column was same as of the GC under the same conditions stated above. The constituents were identified by comparison of their mass spectra with those in the computer library and with authentic compounds. The identifications were confirmed by comparison of their retention indices with those of authentic compounds or with literature data. The essential oil was subjected to GC/MS analysis, antibacterial testing.

Chemical composition of the essential oils

The results obtained by GC–MS analysis of the essential oil of *Juniperus communis* is presented in Table1, respectively. Thirty compounds were identified in the essential oil of *Juniperus communis*, respectively. As a result of GC–MS analyses, *Juniperus communis* contained α-pinene (46.63%) and α-cedrol (12.36%), DETA.3-carene (9.85%), α-terpinolen (4.64%), and terpineol-4 (2.86%), were the major compounds of *Juniperus communis* oil.

Antibacterial testing

Test organisms: The test organisms were selected on the basis that they cause a lot of infections in humans. Both Gram-positive and Gramnegative bacterial species were selected as test organisms. The organisms were obtained from the Department of Pharmacy at the University of Guilan, who in turn obtained them from Rasht as pure characterized strains [19].

Inoculation procedure: The isosensitest broth was inoculated aseptically with the appropriate microorganism 24 h before testing. This was to ensure that the bacteria fully adapted to the broth. The procedure was repeated for each bacterial species. The inoculated bacterial strains were incubated at 25 ◦C for 24 h. The procedure was repeated for each bacterial species [20].

Antibacterial activity

Essential oils were diluted with absolute alcohol to produce the following concentrations: 10, 20, 50 and 100% (v/v). Agar was melted in a steam bath set at 30 ◦C to prevent solidification. Four Petri dishes were pre-inoculated with the appropriate bacteria in the following manner. 1 mL of the bacterial suspension was pipetted into the appropriately labeled Petri dish to which 25 mL of molten agar was then added followed by thorough mixing of the bacteria and molten agar. The agar was allowed to set for 1 h. Four 4mm wide holes were then made in the agar using a borer [21]. Oil (25µL)

of a specific concentration was introduced into each of the holes in an appropriately labeled Petri dish using a sterile micropipette. Gentamicin (10 g/mL) was used as a positive control and absolute alcohol as a negative control. The dishes were then incubated at 25 ◦C for 24 h after which zones of inhibition were measured and recorded. The zone of inhibition was taken to be the diameter of the zone visibly showing the absence of growth including the 4mm hole.

RESULTS AND DISCUSSION

Three compounds, representing more than 99.99% of the essential oil were identified in species.

Table-1: Chemical composition of *Juniperus communis* essential oil

NO.	Compound	RI	percentage
1	α–pinene	939	46.63
2	α–fenchene	950	0.25
3	Sabinene	974	0.58
4	β–pinene	980	1.38
5	Myrcene	991	1.52
6	DETA.3-carene	1011	9.85
7	Limonene	1030	1.90
8	Terpinolene	1060	2.45
9	Terpine ol -4	1089	2.86
10	α-terpineol	1110	0.94
11	Carvone	1136	0.74
12	Carvacrol	1154	0.83
13	γ-terpinene	1183	0.50
14	α-terpinolen	1198	4.64
15	α-amorphene	1205	0.99
16	β–caryophyllene	1226	1.14
17	α–humulene	1238	0.95
18	Germacrene-D	1254	1.75
19	α–muurolene	1261	0.89
20	β–cadinene	1288	1.43
21	β–elemene	1294	0.89
22	Junipene	1300	0.73
23	α–cedrol	1309	12.36
24	γ–cadinene	1337	1.25
25	δ–cadinene	1339	0.90
26	α–cadinene	1342	1.10
27	α–cadinol	1347	0.54

Antibacterial activity: As can be noticed from Table 2, the essential oil exhibited notable antibacterial activity against Bacterial species *E.coli, Staphylococcus and Sodomunas* bacterial tested. The essential oil from this plant exhibited antibacterial.These activities may be attributed to the presence of α-pinene, β-pinene, cymene, limonene, *B*-myrcene, terpinene-4-ol, linalool, *trans*-carryophyllene, *a*-phellandrene, 3-carene, *a*-terpineol and germacrene found in *Juniperus communis* essential oil.

Table-2: Antibacterial activity of *J. communis*

Bacterial species Source	Inhibition zone diameter (mm)					Gentamycin (10 µg/mL) (positive control)
	0%	10%	20%	50%	100%	
Staphylococcus aureus NCIB 6751	0	2.1	4.8	5.0	3.5	2.1
Pseudomonas aeruginosa NCIB 950	0	0	0	0	4.5	4.5
Escherichia coli NCIB 8879	0	6.1	7.2	8.3	2.2	2.7

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