Pak. j. sci. ind. res., vol. 36, nos. 2-3,Feb.-Mar. 1993

EVALUATION AND COMMERCIAL EXPLOITATION OF ESSENTIAL OIL OF *JUNIPER* BERRIES OF PAKISTAN

MUHAMMAD RAFIQUE, M. HANIF AND F. M. CHAUDHARY

PCSIR Laboratories Complex, Lahore - 54600, Pakistan

(Received May 29, 1992; revised March 11, 1993)

The essential oil of *Juniperus excelsa* M.B. [1] (Syn. *J. polycarpos, C. koch; J. macropoda* Boiss, HK) berries has been studied for its physico-chemical properties and chemical compositon. The oil contains 25 components, the major ones being pinenes (72.5%), myrcene (12.4%), limonene (2.28%), camphene (2.51%). In order to improve the flavour of the oil, it has been recitified, whereby the pinenes contents of the oils has been reduced to 8%, thus making the oil more valuable and acceptable to the market.

Key words: *Juniper* Berries (*Juniperus excelsa* M.B.), Essential oil, Composition, GLC.

Introduction

Common *juniper* is a small to medium tall tree attaining a height of 3.25 m or more. It grows wild in many parts of Europe, Asia and Northern America. The most common north American variety is *Juniperus communis* [2] (Var. depressa pursh).

The common vareity found in Pakistan is *Juniperus excelsa* M.B. mostly found in Baluchistan, Azad Kashmir, Gilgit and Chitral. It grows at an altitude of 2400-3200 m. The area under juniper forests in Quetta region along is estimated to be about 250,000 acres. The *juniper* trees are valued for their fruit called berries. The berries are well known for medicinal purposes, their water extract is used as a cure for chest diseases and as a diuretic [3]. The berries contain from 0.5 to 1.55% a pleasent smelling essential oil. The oil is used as essence for flavouring of beverages and liquors particularly the gin and sloe gin types [4].

The berries ripen around November. The ripe berries are succulent, dark pink in colour, very fragrant and sweet in taste. According to Baluchistan Forest Department, there is an assured supply of more than 50,000 tonnes of dry berries every year from the Quetta region alone. *Juniperus communis* is the commercially available variety in America and European countries and a lot of work appear to have been done on this variety [5-8]. On the other hand a little work has been done on *J. excelsa* and *Juniper berries* oil of Pakistan has not been studied so far. Present studies were undertaken to evaluate the oil for commerical exploitation.

Material and Method

Fully ripe berries (2000 kg) from Quetta region (Ziarat) were collected in November 1981 for experimental investigations. Berries were spread undershade in a thin layer to dry and prevent fermentation.

Extraction of essential oil. The essential oil from berries was obtained by hydro-distillation in 500 kg capacity distillation still built in the workshop of the PCSIR Laboratories Complex, Lahore for this purpose. Extracted oil was collected after every 3-4 hrs of distillaltion period. The oil was dried over anhydrous sodium sulphate.

Gas chromatographic analysis. The gas chromatographic analysis was carried out using Pye-Unicam 104 Gas Chromatograph under the following conditons.

Detector	FID
Column	SCOT Carbowax 20 M, i.d.
Column lenght	25m
Carrier gas	Nitrogen
Flow velocity	26.7 cm/sec.
Split ratio	1:60
Sample size	0.02 µl.
Initial temp.	70° for 5 min.
Increase in temp.	4°/min.
Final temp.	150°
Detector temp.	250°
Injection temp.	300°

The identification of different components was carried out by comparing their retention times and coinjection of standard samples. The percentage of differnt components was determined on the bases of peak area using Pye Unicam DP-88 computing integrator.

Results and Discussion

Ripe berries have a greater contents of oxygenated compounds than green ones and leaves [9]. As the fermentation of ripe berries is very fast, to avoid fermentation, ripe berries after collection were dried at the place of collection. The ripe berries on distillation yield 1.5% pleasant smelling oil, the physico-chemical properties of the oil are given in Table 1. The oil yield is in accordance with the yield reported by Talwar *et al.* [10]

while Mishra *et al.* [11] have reported higher yield of the oil, this difference of yield may be due to the climatic variation. GC analysis of the *J. berries* oil showed, 25 peaks Table 2 corresponding to different compounds but only 18 peaks were identified, α-pinne, β-pinene and myrcence were the major components.

In *J. excelsa* berries oil, four compounds namely Fenchene, Bornyl acetate, α-Humulene and Germacrene have been indentified for the first time. The other fourteen compounds indentified have already been reported by Klein, *et al.* [12], and Talwar [10], Table 2. We could not confirm the presence of α-Thujene and Sabinene already reported by Klein, *et al.* and that of cedrene, aromadendrene, sabinol and linalool reported to be present by Talwar, *et al.* It would be worthwhile to point out that none of the earlier studies reported the presence of so many components in their oil content. This may be ascribed to different geographical conditions prevailing at various places.

As *Juniper berries* oil contains more than 70% pinenes (α- pinene and β-pinene) it has turpentine like off notes and is unacceptable to the consumer as such. In order to remove this undesirable off note the essential oil was recitified using fractionating column as detailed in Table 4.

Two fractions of the oil depending upon their terpenes contents were separated. From the G.C. analysis Table 3, it was observed that in the 2nd fraction pinenes decreased to 8% percent while other components increased proportionately. The deterpenated oil exhibits a sweeter and better note and

TABLE 1. PHYSICO-CHEMICAL PROPERTIES OF THE *JUNIPER* BERRIES OIL.

Yield	1.5%
Colour	Greenish yellow to yellow
Sp. gravity at 20	0.8349
Refractive index at 20	1.476

TABLE 2.

Components	Percentage in original oil	Reported by		
		Klein *et.al*[12]	Talwar *et.al*[10]	Pradeep *et al.*[11]
α-Thujene	-	0.9	-	-
α-Pinene	64.4	38.4	17.0	71.38
Fenchene	Traces	-	-	-
Camphene	2.51	0.5	-	-
Sabinene	-	0.6	25.2	-
Myrcene	12.4	11.5	-	20.28
β-Pinene	8.14	1.5	-	-
α-Phellandrene	0.07	0.9	-	-
α-Terpinene	0.16	0.15	10.9	-
p-Cymene	0.09	0.65	-	-
Limonene	2.28	9.0	-	5.57
γ-Terpinene	0.27	0.4	-	1.5
Terpinolene	0.08	0.5	1.6	-
Cedrene	-	-	11.6	-
Aromadendrene	-	-	7.9	-
β-Cadinene	0.52	-	6.9	-
γ-Cadinene	0.165	-	-	-
Sabinol	-	-	1.7	-
Terpinene-4-01	0.712	-	1.4	-
Linalool	-	-	0.75	-
Bornyl acetate	0.302	-	-	-
α-Humulene	0.105	-	-	-
Germacrene	0.5	-	-	-

TABLE - 3 GLC ANALYSIS OF THE *JUNIPER* BERRIES ESSENTIAL OIL AND FRACTION 1 AND 2.

Component	Original oil (%)	Fraction-I (%)	Fraction-2 (%)
α-Pinene	64.4	68.923	8.114
Fenchene	Traces	0.136	-
Camphene	2.51	0.311	-
β-Pinene	8.14	1.481	0.665
Unidentified	1.62	1.393	0.043
Myrcene	12.4	23.97	27.889
α-Terpinene	0.164	-	0.184
Limonene	2.28	2.623	14.134
Phelandrene	0.072	-	0.456
γ-Terpinene	0.269	-	1.933
p-Cymene	0.089	-	1.324
Terpinolene	0.082	-	2.851
Bornyl acetate	0.302	-	1.582
Unidentified	0.5	-	-
Unidentified	0.5	-	-
Terpineol-4	0.712	-	4.319
Caryophyllene	0.12	-	-
Unidentified	1.1	-	1.153
α-Humulene	0.105	-	1.447
Unidentified	0.5	-	0.537
Germacrene	0.5	-	0.929
Unidentified	0.5	-	0.33
β-Cadinene	0.52	-	5.18
γ-Cadinene	0.165	-	-
Unidentified	0.5	-	-

TABLE 4.

Quantity of oil fractionated	No of fraction	Reflux ratio	Fraction distilled
1000 ml	1	1:20	400 ml
	2	1:30	450 ml
1000 ml	1	1:20	380 ml
	2	1:30	450 ml

thus has become more acceptable to the market and comparable to the oil of *J. communis* [6,7]. In the light of theses studies it is suggested that if a distillarly is installed in the field 750,000 kg of oil could be obtained which could be a good source of foreign exchange.

References

1. E. Nasir and Y.J.Nasir, *Flora of Pakistan* (PARC, Islamabad, No.178-86), (1987), pp. 20.
2. E. Guenther, *The Essential Oils* (D.Van Norstrand Company. Inc., Toronto, New York, London, 1965), 4th ed, vi. 370.
3. R.N.. Chopra, *Indigenous Drugs of India* (UN Dhur and Sons, Calcutta, India, 1958), 2nd ed., pp. 195-6.
4. E. Guenther, *The Essential oils* (D.Van Norstrand Co., New York, Toronto, London, 1965),4th ed., vi, 381.
5. Giorgio Bonanga and C. Guido, Galletti, Ann. Chim. (Rome), **75** (3-4), 131 (1985).
6. A. Proenca Da Cunha and L.R. Odete Roque, J. Essent. Oil Res., **1** (1), 15 (1989).
7. Gaston Vornin, Boniface, Christian, Metzzer, Jacques, Claude, Ghiglione, Ahmed Hammoud, Suon Kim Nuor, Fraisse Daniel and Parkanyi Cyril, Phytochemistry, **27** (4), 1061 (1988).
8. D. Lamparsky and Klimes, 1., Perfume Kosmet, **66**, 553, 558 (1985).
9. N. Gelsomini, V. Vidrich, P. Fusi and M. Michelozzi, J. High Resolut. Chromatogr. Commun., **11** (2), 218 (1988).
10. Y.P. Talwar, M.C. Nigam and K.L. Handa, Perfumery Essential Oil Record, **53**, 822 (1962).
11. P. Mishra, B.R. Gandhe and R.A.K. Riechst, Aromen, Kosmet, **34** (1-2), 10 (1984).
12. E. Klein and H. Farnow, Dragoco Rept., (**11**), (**10**), 223 (1964).