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**RESEARCH ARTICLE**

# Comparison of *Juniperus oxycedrus* subsp. *oxycedrus* and *J. oxycedrus* subsp. *macrocarpa* berries essential oils from Turkey

Muhammet Buyukbayram* and Sonay Gurer

Parkim R&D Department, Parkim Parfume Plastic & Chemical Ltd., 41400, Kocaeli, TURKEY

*Corresponding author. Email: Muhammet.buyukbayram@parkimgroup.com

**Abstract**

*Juniperus oxycedrus* L. and related taxa are within the natural habitat in Turkey. In this present study, essential oils from *J. oxycedrus* subsp. *oxycedrus* and *J. oxycedrus* subsp. *macrocarpa* berries, collected from different provinces of Turkey were obtained by hydrodistillation and analyzed for their chemical compositions. GC/MS analyses revealed α-pinene (19.9-47.8%), β-myrcene (44.6-10.3%) and germacrene D (15.5-13.5%) as main constituents of the berries, respectively. The olfactory evaluation showed characteristic woody, greenish, herbal, turpentine-like and balsamic sweety nature suggesting their uses especially in men's cosmetics.

**Keywords**: *Juniperus oxycedrus*, *Juniperus macrocarpa*, Berry essential oil, Olfactory

## Introduction

The genus *Juniperus* (Cupressaceae), well known juniper, is a very common botanical species in northern hemisphere. The family is represented with around 70 subspecies. *Juniperus* sp. consists of a wide variety in especially Mediterranean countries and is used for its wood as construction material because of resistance to pests and other pathologies (Arapi et al., 2012). *J. oxycedrus* and *J. macrocarpa* are two of the 14 species that comprise the section Juniperus throughout the world (Medini et al., 2010). Both *J. oxycedrus* and *J. macrocarpa* are most distinctive subspecies of *Juniperus* family as their attractive volatile and non-volatile chemical composition (Adams, 1998; Barrero et al., 1993). Because of their volatile and non-volatile chemical composition diversity, they have attractive olfactive effects. *Juniperus* exists in Turkey by 7 species and 14 taxa. *Juniperus oxycedrus* has two subspecies – subsp. *oxycedrus* and subsp. *macrocarpa* – in Turkey (Alan et al., 2016).

*Juniperus* pseudo fruits, female cones – improperly called ''berries'' are used as a spice, mainly in European cuisine; they are used in Northern European and particularly Scandinavian cuisine to impart a sharp, clear flavour to meat dishes (Taviano et al., 2013).

Berries of *J. oxycedrus* have widely been used as medicinal herbal in folk medicine in order to threat gastrointestinal disorders, common colds, as expectorant in cough, to treat calcinosis in joints and as diuretic to pass kidney stone, against urinary inflammations, hemorrhoids, and as hypoglycemic; leaves and berries are applied externally for parasitic disease (Akkol et al., 2009; Loizzo et al., 2007; Sezik et al., 1997). In addition, *Juniperus* subspecies are used for various purposes, such as sweating antiseptic, and as a tar source. These species also contain oleoresins, a triterpene known as 'resin cadinene' and phenols (guaiacol and cresol derivatives) and they are used in the treatment of some skin ailments among many other uses (Digrak et al., 1999; Baytop, 1984).

The aim of this work was to extract the essential oils from the berries, compare their chemistry and olfactory properties for possible utilization in various cosmetics.

## Materials and Methods

### Plant material

Air dried *J. oxycedrus* subsp. *oxycedrus* and *J. oxycedrus* subsp. *macrocarpa* berries were obtained from south of Turkey, Antalya and Isparta, in May, 2018.

### Isolation of essential oils

The oils were obtained by hydrodistillation for 3 hours using a Clevenger apparatus. The essential oils were stored at 4 °C in the dark until analyzed. The oils were analyzed by GC/MS using Agillent GC-MS system. The essential oils were obtained from two type berries (*J. oxycedrus* and *J. macrocarpa*) in 1.6% and 1.9% yields, respectively.

### Gas chromatography - mass spectroscopy (GC-MS) analysis

The GC/MS analysis was carried out with an Agilent 5975 C GC/MS system. 95% Dimethylpolysiloxane HP 5 MS column (30 m x 0.25 mm x 0.25 µm film thickness) was used with helium as carrier gas (0.8 mL/min). GC oven temperature was kept at 60 °C for 10 minutes and programmed to 130 °C at a rate of 4 °C/minutes, and kept constant at 220 °C for 1 min. and then programmed to 170 °C at a rate of 1.5°C/minutes, then programmed to 270 ºC at a rate of 20 ºC/minutes and kept constant 10 minutes. Split ratio was adjusted 30:1. The injector temperature was at 250 °C. Mass range was from *m/z* 35 to 400.

### Characterization of components

Identification of the essential oil components was carried out by comparison of their relative retention times with those of authentic samples or by comparison of their relative retention indices (RRI) to series of *n*-alkanes. Computer matching against commercial (Wiley GC/MS Library) and *in-house* "Parkim Fragrance House GC/MS Library" built up by genuine compounds and components of known oils was used for the identification of oil components.

### Olfactory evaluation panel

Olfactive parameters of the essential oils were evaluated by trained perfumers. Both essential oils, *J. oxycedrus* subsp. *oxycedrus* and *macrocarpa,* olfactive notes were determined for their characteristic odors and their usage area were classified with some features such as diffusion rate and deep of the olfactive note.

## Results and Discussion

The oils were analysed by GC-MS, *J. oxycedrus* subsp*. oxycedrus* berries essential oil showed 15 compounds representing 97.3 percent of the oil. α-Pinene (19.9%), β-myrcene (44.6%) and germacrene D (15.5%) were characterized as main constituents.

*J. oxycedrus* subsp*. macrocarpa* berry essential oil showed overall 18 compounds representing 100 percent of the oil. α-Pinene (47.8%), sabinene (7.2%), β-myrcene (10.3%) and germacrene D (13.5%) were characterized as main constituents.

Table 1. Composition of the essential oils of *J. oxycedrus* subsp*. oxycedrus* and *macrocarpa* berries

| ***RI*** | ***t*R** | **Compounds** | **Relative percentage (%)** | |
|---|---|---|---|---|
| | | | ***J. oxycedrus* subsp*. macrocarpa*** | ***J. oxycedrus* subsp*. oxycedrus*** |
| 1087 | 8.5 | α-Pinene | 36.5 | 22.7 |
| 1196 | 11.08 | Sabinene | 6.7 | 3.4 |
| 1222 | 11.15 | β-Pinene | 2.4 | - |
| 1275 | 12.2 | β-Myrcene | 18.3 | 41.8 |
| 1314 | 14.8 | Limonene | 2.2 | 2.9 |
| 1360 | 12.8 | β-Phellandrene | - | 0.6 |
| 1502 | 23.6 | α-Cubenene | 2.0 | 0.3 |
| 1617 | 25.82 | Isobornyl acetate | - | 0.2 |
| 1526 | 29.218 | α-Copaene | 0.8 | - |
| 1542 | 24.95 | Unidentified sesquiterpene | - | 0.5 |
| 1711 | 31.52 | β-Caryophyllene | 2.1 | 3.1 |
| 1731 | 32.69 | β-Farnesene | - | 0.7 |
| 1753 | 32.57 | α-Humulene | 1.8 | 2.8 |
| 1776 | 33.371 | α-Amorphene | 1.5 | - |
| 1702 | 31.175 | Germacrene D | 13.2 | 15.1 |
| 1865 | 36.548 | α-Muurolene | 1.3 | 0.7 |
| 1902 | 42.371 | γ-Cadinene | 1.6 | 1.8 |
| 1963 | 41.11 | δ-Cadinene | 2.4 | - |
| 2264 | 41.75 | epi-α-Cadinol | 0.9 | - |
| 2638 | 41.84 | α-Cadinol | 0.7 | 0.7 |
| 1849 | 35.46 | Manoyl oxide | 3.8 | - |
| 1856 | 36.48 | Abietadiene | 1.8 | - |

RI: Retention indices. Rt: Retention times in minutes. %; calculated from the GC-MS chromatograms; main constituent of the unidentified sesquiterpene is γ-terpinene.

In this present study, we investigated the essential oil chemical composition of berries of *J. oxycedrus* subsp*. oxycedrus* and subsp*. macrocarpa* from south of Turkey. Sezik et. al (2005) performed similar study with leaves of subsp*. oxycedrus* and subsp*. macrocarpa* was collected in Turkey. Another study was performed by Lesjak et al. (2014) as antimicrobial activity of subsp. *macrocarpa*. In this study samples were collected as leaves and berries. Berries samples had GC-MS data similar to ours.

Another parameter of our study was based on olfactive evaluation of subsp. *oxycedrus* and subsp. *macrocarpa* berries essential oils. *J. oxycedrus* berries essential oil consisted of α-pinene, β-myrcene and germacrene D, respectively. These components redounded olfactive results of *J. oxycedrus* subsp. *oxycedrus* essential oil with a turpentine-like juniper odor; fresh balsam, woody terpene. *J. macrocarpa* berries essential oil of our sample was mostly consisting of α-pinene, β-myrcene and germacrene D, however, percentages were different from that of *J. oxycedrus* subsp. *oxycedrus*, demonstrated in Table 1. The top note has a turpentine type odour with the woody dryness, and *Amyris balsamifera* and the sweetness of juniper berries combined with earthy elements of patchouli. Dry woody notes absolutely dominate the heart and as the oil dries out, one is flooded with intense balsamic sweetness.

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