

## INTERNATIONAL JOURNAL OF APPLIED BIOLOGY AND PHARMACEUTICAL TECHNOLOGY

Volume: 3: Issue-1: Jan - Mar-2012

**UABPT** ISSN 0976-4550

Accepted: Dec-2011

**Research** Article

## COMPARING ESSENTIAL OIL COMPOSITION AND ESSENTIAL OIL YIELD OF ROSEMARINUS OFFICINALIS AND LAVANDULA ANGUSTIFOLIA BEFORE AND FULL FLOWERING STAGES

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**ABSTRACT:** The chemical composition of essential oils and essential oil yield obtained from *Rosemarinus officinalis* (family Lamiaceae) and *Lavandula angustifolia* (family Lamiaceae) were determined in two harvesting times. Their essential oil was determined by hydro-distillation, and analysed by GC/MS. The results showed that harvesting time had significant effects on the oil content and compositions in both plants. The maximum essential oil percentage was obtained in full flowering stage in rosemary. Also and in lavender maximum linalool percentage (19.2%) was obtained in full flowering, and minimum linalool percentage (0.2%) was shown in the other time. Also the concentration of  $\beta$  – pinene (2.1%),  $\delta$ -3-carene (1.5%),  $\beta$  – phellandrene (6.6%), Camphor(10.6%), Cryptone (0.8%),  $\alpha$ -terpineol (2.3%) and Linalool acetate (1.2%) were higher than befor flowering stage. Therefore the harvesting time have a great importance in the production of essential oil and influenced on the quantity and quality of essential oil. As consequence, the best harvesting time in both medicinal plants was obtained in full flowering stage.

Key worlds: Harvesting time, Essential oil, Lavandula angustifolia, Rosemarinus officinalis.

Abbreviations: RI- Retention Indices, GC-MS - Gas chromatography- mass spectrometry. INTRODUCTION

Lavandula officinalis Chaix. (Synonym: L. angustifolia Mill; L. vera DC.) { family: Labiatae } is an evergreen bushy shrub with straight, woody branches, the lower of which are leafless, putting out numerous herbaceous stems to a height of about 1 meter (Chiej 1984; Wichtl 1994). The plant is native to southern Europe and the Mediterranean area and is commercially cultivated in France, Spain, Portugal, Hungary, the UK, Bulgaria, Australia, China and the USA (Shawl and Kumar 2000). This plant is cultivated primarily for its aromatic inflorescence from which the essential oil is isolated, although its fresh and dried flowers are also marketed (Renaud and Charles 2001). Lavender oil is known for its excellent aroma and is extensively used in the perfumery, flavor and cosmetic industries. The oil is known to possess sedative, carminative, anti-depressive and anti-inflammatory properties (Cavanagh 2005). It was also found to be active against many species of bacteria, including those resistant to antibiotics, such as methicillin-resistant Staphlococcusaureus and vancomycin-resistant Enterococcus (Cavanagh, 2005). Lavender oil was also reported to be an effective antifungal agent against *Aspergillus nidulans* and *Trichophyton mentagrophytes* (Moon et al. 2004).

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The essential oil compositions of lavender grown in different countries were investigated (Tucker and Howell 1984; Adams and Yanke 2007). Essential oils obtained from aromatic plants, are complex of several chemical compounds including terpenes, alcohols, aldehydes and phenols. Lavender oil, obtained from the flowers of Lavandula angustifolia composed mainly of linalyl acetate, linalool, lavandulol, 1, 8cineole, lavandulyl acetate and camphor (Lis-Balchin and Hart 1999). Because of its delightful dour, lavender is one of the most useful medicinal plants and has found wide application in perfumes, colognes, skin lotions and other cosmetics (Paul et al. 2004). In food manufacturing, lavender essential oil is employed in flavoring beverages, ice-cream, candy, baked goods, and chewing gum (Kim and Lee 2002). Recently, with aromatherapy becoming increasingly popular, lavender is used as a relaxant (Lis-Balchin and Hart 1999). The use of aromatherapy as a therapeutic treatment for affective disorders has also been widely reported in historical anecdotal literature (Valnet 1986). The plant is used in different parts of the world for the treatment of several gastrointestinal, nervous and rheumatic disorders (Duke 1989; Leung and Foster 1996). In vitro an cytotoxic activity of lavender oil and its main components linalyl acetate and linalool on human skin cells has been reported (Prashar et al. 2004). Lavender oil also has antioxidant properties (Hohmann et al. 1999). And unlike to many other essential oils used in aromatherapy, the oil is often applied undiluted to the skin.

Rosemary, *Rosmarinus officinalis* L. (Lamiaceae) is an aromatic evergreen shrubby herb highly distributed in the Mediterranean region. It is a well-known and greatly valued medicinal herb that is widely used pharmaceutical products and folk medicine as a digestive, tonic, diuretic, diaphoretic and useful for urinary ailments (Chang et al., 1977; Aqel, 1991; Leung and Foster, 1996; Haloui et al., 2000). Multiple studies have been reported on the chemical composition of the essential oils of *Rosmarinus officinalis* belonging to different regions in the world (Khorshidi et al., 2009). Further-more, several extracts, essential oils and chemical constituents isolated from this species demonstrated a number of interesting biological activities such as antioxidant (Inatani et al., 1983; Houlihan et al., 1985; Aruoma et al., 1992, 1996; Haraguchi et al., 2000), and anticarcinogenic (Offord et al., 1995). This biological importance prompted us to re-investigate the chemical constituents of the aerial parts of this species. Earlier work on the chemistry of this species showed that it contains mainly abietane-type diterpenoids including some diterpenoid quinines (Inatani et al., 1983; Nakatani and Inatani, 1983, 1984; Houlihan et al., 1985; Arisawa et al., 1987), however some triterpenoids (Ganeva et al., 1993) were also isolated.

The objective of this study was evaluate the comparing essential oil composition and essential oil yield of *Rosemarinus officinalis* and *Lavandula angustifolia* before and during full flowering stages.

#### MATERIALS AND METHODS

#### **Plant material**

Samples of *Lavandula angustifolia* and *Rosemarinus officinalis* were collected in Eram Garden in 2011. The plant was identified in the Herbarium of Eram Garden in Shiraz, Iran.

#### **Drying Methods**

The drying methods investigated was shade drying. Samples of *Lavandula angustifolia* and *Rosemarinus officinalis* were dried at room temperature and shad.

#### Statistical analysis

Treatments were arranged in a completely randomized design with 2 treatments with three replications. Analysis of variance was performed using the Minitab software and means were separated using Tukey's test ( $p \le 0.05$ ).

#### **Essential oil isolation**

The dried samples of *Lavandula angustifolia* and *Rosemarinus officinalis* were subjected to hydrodistillation in Clevengers apparatus for 3 hour for the extraction of the essential oil and to three replications. The essential oils were separated from the aqueous layer, dried over anhydrous sodium sulfate and calculated average of essential oil yield for three replication. The essential oils were stored at 4°C until analysis by GC-MS.

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#### **Identification of the oil components**

Analysis was carried out using an Agilent-technology chromatograph with HP-5 column ( $30m \times 0.32 \text{ mm}$  i.d.  $\times 0.25 \mu m$ ). Oven temperature was performed as follows:  $60^{\circ}$  C to  $210^{\circ}$  C at  $3^{\circ}/min$ ;  $210^{\circ}$  C to  $240^{\circ}$  C at  $20^{\circ}/min$  and hold for 8.5 min, injector temperature  $280^{\circ}$  C; detector temperature,  $290^{\circ}$  C; carrier gas, N<sub>2</sub> (1 ml/min); split ratio of 1:50. GC-MS analysis was carried out using a with Agilent 7890 operating at 70 eV ionization energy, equipped with a HP-5 MS capillary column (phenyl methyl siloxane,  $30m \times 0.25 \text{ mm} i.d \times 25\mu m$ ) with He as the carrier gas and split ratio 1:50. Retention indices were determined using retention times of n-alkanes that were injected after the essential oil under the same chromatographic conditions. The retention indices for all components were determined according to the method using n-alkanes as standard. The compounds were identified by comparison of retention indices (RRI, HP-5) with those reported in the literature and by comparison of their mass spectra with the Wiley GC/MS Library, Adams Library, MassFinder 2.1 Library data published mass spectra data (Adams 2007; McLafferty 1989; Joulain et al., 2001)

#### **RESULTS AND DISCUSSION**

# Chemical composition of the essential oil in two time harvesting (before and full flowering stages).

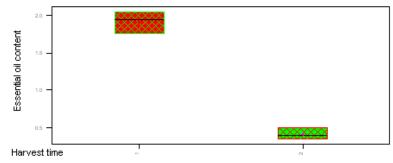
The results obtained by GC-MS analysis of the essential oil of *Lavandula angustifolia* and *Rosemarinus officinalis* are presented in Table 1 and 2. Thirty seven compounds were identified in the essential oils in full flowering and thirty two compounds were identified in the essential oils in before flowering in *Lavandula angustifolia*, but thirty three compounds obtained in *rosemarinus officinalis* in two harvesting time. The main compounds of lavender oil in full flowering stage identified,  $\alpha$ -pinene (1.81%),  $\beta$ -pinene (2.11%), Myrcene (1.60%),  $\delta$ -3-Carene (1.51%),  $\beta$ -Phellandrene (6.56%), 1,8-Cineole (29.03%), Linalool (19.19%), Camphor (10.57%), Borneol(9.32%), Terpinen-4-ol (1.73%),  $\alpha$ -Terpineol (2.33%), Linalool acetate (1.2%),  $\alpha$ -Bisabolol (1.26%), and important compounds of rosemary oil in full flowering stage identified,  $\alpha$ -Pinene (13.2%),  $\beta$ -Pinene (2.0%), 1,8 Cineole (7.1%), Camphor (9.5%), Borneol (8.70%), Linalool (3.7%), and Verbenone (12.1%) were major constituents (Table 1 and 2).

#### The influence of harvesting time on chemical composition in two plants.

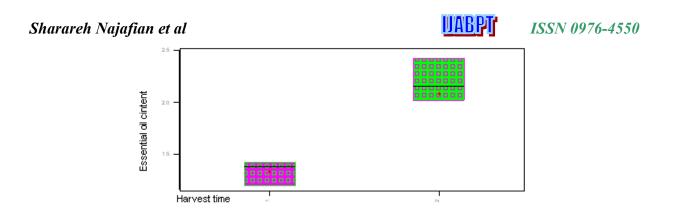
Our results showed that the number of compounds wereas the same in both harvesting times of *Rosmarinus officinalis*, but the effect of harvesting time is different in Lavandula *angustifolia*. In the first time (before flowering stages) there were 32 compounds and in the second time (flowering stages) there were 37 compounds. The data are shown in Table 2.

#### The influence of harvesting time on essential oil yield.

Results showed that harvesting time had a significant effect on essential oil content in tow both plants. The minimum essential oil percentage was obtained in full flowering stage in *Lavandula angustifolia*, But in *Rosmarinus officinalis* from the samples which were that dried in shade in full flowering stage were higher essential oil was obtained as compared to samples that harvested in before flowering stage, and it which was significant (Fig1 and 2).







## Figure 2. The effect of harvest time on essential oil content of Rosemarinus officinalis.

	Before flowering stages		flowering stages
Compounds	RI <sup>a</sup>	Area,%	Area,%
alpha-pinene	932	13.9	13.2
camphene	946	4.8	4.7
Verbenene	961	0.6	0.7
β-Pinene	974	2.0	2.0
3-octanone	979	5.6	4.3
Myrcene	988	2.8	2.7
3-octanol	989	0.6	0.5
alpha-Phellandrene	1002	0.2	0.3
alpha- terpinene	1014	0.6	0.6
p-cymene	1020	1.0	1.2
limonene	1024	3.3	4.4
1,8-cineole	1026	9.2	7.1
gamma-terpinene	1054	0.8	0.8
terpinolene	1086	1.2	1.3
linalool	1095	4.3	3.7
chrysanthenone	1124	1.8	1.1
camphor	1140	10.2	9.5
trans-pinocamphone	1158	0.5	0.5
pinocarvone	1160	0.3	0.3
borneol	1165	8.2	8.7
cis-pinocamphone	1172	1.6	1.5
terpinen-4-ol	1174	1.6	1.6
alpha.terpineol	1186	2.2	2.6
myrtenol	1194	0.4	0.4
2-Methylisoboneol	1203	1.5	3.3
verbenone	1204	11.3	12.1
Cis-p-mentha-1(7),8-dien-	1227	1.0	1.3
2-ol			
Carvone	1239	1.5	2.0
Bornyl acetate	1287	4.5	5.0
Dihydrocarveol acetate	1344	0.3	0.4
Trans-caryophyllene	1417	0.8	1.0
alpha-humulene	1452	0.2	0.3
caryophyllene oxide	1582	0.4	0.6

## Table1.comparing essential oil composition Rosemarinus officinalis

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	Befor	e flowering	Full flowering	
Compounds	RI <sup>a</sup>	Area,%	RI <sup>a</sup>	Area,%
α-thujene	924	0.1	931	0.2
α-pinene	934	2.0	939	1.8
Camphene	949	1.0	946	0.6
Sabinene	974	0.2	969	0.9
B - pinene	979	0.8	974	2.1
Myrcene	983	0.2	988	1.6
α-phellandrene	991	0.6	1002	0.2
δ- 3-carene	1012	0.4	1008	1.5
P-Cymene	1023	0.3	1020	0.5
β-phellandrene	1026	1.4	1025	6.6
Limonene	1030	2.2	-	-
1,8-Cineol	1041	41.0	1031	29.0
Cis-B-Ocimene	-	-	1032	0.6
Trans-B-cimene	_	-	1044	0.2
γ-Terpinene	_	_	1054	0.7
Terpinolene	1074	0.3	1086	0.3
Linalool	1105	0.2	1095	19.2
$\alpha$ - Campholenal	-	-	1122	0.2
Trans-pinocarveol	1130	0.5	1135	0.2
Camphor	1147	0.5	1141	10.6
Borneol	1156	14.5	1165	9.32
Terpinene-4-ol	1177	20.4	1174	1.7
P-Cymen-8-ol	-		1179	0.3
Cryptone	1185	0.1	1183	0.8
$\alpha$ -Terpineol	1189	0.5	1186	2.3
N-Hexylbutanoate	1193	0.8	1191	0.3
Myrtenol	1199	2.4	1194	0.2
Methyl Chavicol	1205	0.5	-	-
Iso Bornylformate	1226	0.9	1227	0.5
Cumin aldehyde	1234	1.5	1238	0.3
Carvone	1246	1.0	1239	0.2
Linalool acetate	1253	0.4	1254	1.2
Lavandulyl acetate	1298	0.3	1288	0.3
Geranyl acetate	1389	0.6	-	-
(E)-Caryophyllene	1422	0.5	1417	0.4
(E)-B-Farnesene	1454	-	1454	0.7
α-Amorphene	1485		1485	0.4
Caryophylene oxide	1591	1.3	1582	0.6
$\alpha$ - Cadinol	1652	0.9	1652	0.8
α -Bisabolol	1685	_	1685	1.3

Table2.comparing essential oil composition lavandula angustifolia.

#### Conclusion

Results showed that harvesting time (before and full flowering stages) had a significant effect on essential oil content of *Lavandula angustifolia* and *Rosmarinus officinalis*. The best harvesting time in this research was full flowering stage in *Rosmarinus officinalis* because of the maximum essential oil percentage (Fig6),. And in lavender oil, one of the important compounds of oil is Linalool.

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Several investigations (Barazandeh, 2002; Figueiredo et al., 1995; Nogueria and Romano., 2002) on the essential oils of various lavandula species showed that linalool was the most important compounds of these plants. Therefore, we investigated linalool in lavender. Results showed that maximum linalool percentage (19.2%) was obtained in full flowering, and minimum linalool percentage (0.2%) was shown in the other time. Also the concentration of  $\beta$ -pinene (2.1%),  $\delta$ -3-carene (1.5%),  $\beta$  – phellandrene (6.6%), Camphor (10.6%), Cryptone (0.8%),  $\alpha$ -terpineol (2.3%) and Linalool acetate (1.2%) were higher than befor flowering stage. Therefore the harvesting time have a great importance in the production of essential oil and influenced on the quantity and quality of essential oil. As consequence, the best of harvesting time in both medicinal plants was obtained in full flowering stage.

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