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The Effects of Different Drying Methods on Essential Oil Content and Composition and Marketing of *Lippia Citriodora* Kunth

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ABSTRACT

Drying (or dehydration) is the most important phases after producing medicinal plant that have an important role on the quantity of their effective materials. Experiments were carried with random plan form by 3 replications in 2011 and 2012. The object of study was to investigate the effect of different drying methods on medicinal plant essence compounds and percentage such as *Lippie citriodora*. Drying (or dehydration) treatments including drying in the shade for two days (traditional method) and drying by artificial method includes semi-industrial dryer (including metal plates and warm air flow) for 2 days in 40^{c} and industrial dryer (oven) in 50^{c} for 48 hours. Obtained essence of different drying treatments and taking essence by gas chromatograph GC and gas chromatograph linked to Mass spectroscopy GC/MS were investigated in this study. The numbers of compounds were defined in the citriodora plant essence. The highest rate of Spathulenol (%25.18 in first year and 26.2% in second year) was obtained in semi-industrial drying treatment. According to the present study it should be mentioned that main constituents of essence and there was significant variance in each compounds rate. The highest percent of essence observed in drying treatment by semi industrial method (%3.13 in first year and 4.12% in second year). For number of essence extracts, there were no any significant differences between methods.

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INTRODUCTION

Lemon verbena (*Lippia citriodora*) is a medicinal plant, belonging to Verbenaceae family and essential oil has accumulated in its vegetative and reproductive parts. Main essential oil components in this plant are: Geranial, Neral, Limonene, 1,8 Cineole, Spathulenol, Nerol, Geraniol, trans-β-cariofilene and Geranyl acetate (Tutin, 1981; Ocazionez *et al*., 2010; El Hawari *et al*., 2011). Essential oils from *Lippia citriodora* showed a considerable inhibitory effect on dengue virus serotype replication in Vero cells and are virucide and insecticide (Ocazionez, 2010; Amer and Mehlhorn, 2006 a-c). In the other hand dehydration by drying is a traditional method but very widespread for its preservation in order to obtain a new product which, thereafter, will be used in perfumery, confectionery and pharmaceutical manufacturing. Generally, the knowledge of the behavior and the determination of the characteristics of drying product are obtained through experimental tests. Many researches on the mathematical modeling and experimental studies have been conducted on the drying processes of various products such as lippia (Kouhila *et al*., 2001), carrots (Kaya *et al*., 2009), aromatic and medicinal plants (Kavak Akpinar, 2006), apricot (Igual *et al*., 2012), pistachios (Midilli and Kucuk, 2003), apples (Akpinar, 2003), figs (Doymas, 2005), *Lemon aurantium* (Ait Mohamed *et al*., 2005), kiwi (Simal *et al*., 2005), mint, parsley and basil (Kavak Akpinar, 2005), herbs and spices (Janjai and Tung, 2005). All this research proved that the drying air temperature is the main factor in controlling the drying rate. The chemical composition of the essential oil from the leaves of *L.citriodora* has been previously reported (Catalan and De Lampasona 2002; Carnat *et al*., 1999; Sartoratto *et al*., 2004). The post-harvesting process of medicinal plants has great importance in the production chain, because of its direct influence on the quality and quantity of the active principles in the product sold (Rocha *et al*., 2011). A literature search was undertaken on effects of different methods on essential oil content and chemical composition of the essential oil plants, the results showed that drying method had a significant effect on oil content and composition of aromatic plants that reported previously (Al –Tawaha *et al*,2013a; Al –Tawaha *et al*,2013b; Ahmadi *et al*., 2008; Khangholi and

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Rezaeinodehi, 2008; Asekun *et al*., 2006; Okoh *et al*., 2008; Omidbaigi *et al*., 2004; Rao *et al*., 1998). The main objectives of this research are to study the effects of different drying methods on essential oil content and composition of *Lippia Citriodora* Kunth.

Methodology:

Shoot tips and leaf explants (cut in the midrib region) sterilized by soaking in 0.2% mercuric chloride for 5 min, then washed twice with sterilized distilled water gave callus, on Murashige and Skoog (MS) medium impregnated with 4 mg/L 6-benzyl amino purine (BAP). Three methods of drying were used:

1- Semi-industrial dryer (including metal plates and warm air flow) for 2 days in $40^{\dot{c}}$.

2- Industrial dryer (oven) in $50^{\dot{c}}$ for 48 hours.

3- Traditional (in shade by local temperature) were investigated. The shade-drying occurred at room temperature (Soysal and Oztekin, 2001). 50 gr of shoot dry matter was added to 1000 cc of water and by Clevenger; essence extracted and then by GC/MS the chemical components of the essence was determined. The essential oils were analyzed by gas chromatography-mass spectrometry (GC/MS). Thermo Finnegan Trace 2000 GC/MS, made in the USA, was employed with a HP-5MS capillary column (30 m long and 0.25 mm wide, and a 0.25 µm of film thickness) at a 250°C of injector chamber. The initial column temperature was at 120°C for 5 min then raised to 280°C at the rate of 10°C/min. Helium was used as a carrier gas at a rate of 35 ml/min. MS parameters were as follows: ionization energy, 70eV; ion source temperature, 200°C; voltage, 3000 v; and mass range, 30 to 600. The compositions of the essential oil were identified by comparison of their retention indexes, retention times and mass spectra with those of authentic samples in Wiley library (Shibamoto, 1987; Adams, 2001). Experiments were arranged in a complete randomized design and three replications during 2011 and 2012. 100 plants were used for each trial. All data were subjected to analysis of variance (ANOVA) using the statistical computer package SAS and treatment means were separated using Duncan's multiple range test at $P < 0.05$ level.

Results:

The content of essential oil isolated from the aerial parts of *Lippa citriodora* which were dried under different conditions and results of them presented in Table 1 and 2. Result clearly showed that the oil content linked to the drying method. Indeed, in this survey the maximum mean of essential oil obtained from semi industrial of extraction. The analysis of variance showed that the different drying methods (traditional, semi industrial and industrial) had a significant effect on percentage of essence. The most essence in two years (3.2%-3.5%) was obtained in semi industrial method and had significant difference between 3 drying method (table2). Twenty- nine compounds were identified in the essential oils in all methods and there was no significant difference. The identified constituents with their respective percentages are summarized in table 2. The main constituents of the oils in traditional method that made 77% of whole of essence were Geranial (20.81% and 21.1%), Spathulenol (15.56% and 15.8%), Neral (13.83% and 14.1%) in first and second year respectively, in semi industrial method components made 85% of whole of essence and main of them were: Geranial (12.03% and 12.1%), Spathulenol (25.18% and 26.2%), Neral (9.03% and 9.2%) in first and second year respectively. Components that extracted in industrial method made 80% of essence and contained Geranial (20.97% and 22.1%), Spathulenol (16.58% and 17.2%), Neral (13.98% and 14.2%) in first and second year respectively. In our study, Neral, Geranial and Spathulenol were also identified at high percentages.

Table 1: Analysis of variance of essential oil components of *Lippia Citriodora*

Components	Source of Variation	D.F	First Year		Second Year	
			M.S	F	M.S	F
Nerol	Between Group	2	0.148	113.195**	0.15	114**
	Inter Group	6	0.001		0.009	
Neral	Between Group	2	23.85	3035.75**	24.1	3076.2**
	Inter Group	6	0.008		0.009	
Geranial	Between Group	2	78.55	12971.418**	80.1	13100.4**
	Inter Group	6	0.006		0.007	
1, 8 Cineol	Between Group	2	3.6	1450.46**	3.8	1650.1**
	Inter Group	6	0.002		0.004	
Linalool	Between Group	2	0.11	97.069*	0.15	95.2*
	Inter Group	6	0.001		0.001	
1-octen-3-el	Between Group	2	0.49	1303.265*	0.51	1341.2*
	Inter Group	6	0.001		0.002	
Cedren (α)	Between Group	2	0.009	3.84 ns	0.008	2.8ns
	Inter Group	6	0.002		0.001	

ns,* and **: Non significant, significant at the 5% and 1% levels of probability, respectively.

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Table 2: Essential oil components percentage in drying methods.

Components	First year						Second year					
	Traditional		Industrial		Semi industrial		Traditional		Industrial		Semi industrial	
1- Octen-3-ol	0.3	b	0.26	b	0.97	a	0.28	b	0.27	b	0.98	a
Limonene	0.45	a	0	b	0	b	0.49	a	0.49	a	0	b
1,8- Cineole	0.4	b	0.51	b	2.4	a	0.52	b	0.52	b	2.4	a
Linalool	0.4	b	0.34	b	0.72	a	0.38	b	0.38	b	0.71	a
Nerol	0.4	b	0.9	a	0.8	a	0.45	b	0.87	a	0.78	a
Neral	14.1	a	14.2	a	9.2	b	13.8	a	13.98	a	9.03	b
Geranial	21.1	a	22.1	a	12.1	b	20.8	a	20.97	a	12.03	b
α - Terpineol	1.2	a	1.1	a	1.45	a	1.11	a	1.11	a	1.46	a
Geraniol	0	b	0.28	a	0	b	0	b	0.29	a	0	b
Geranyl acetate	1.4	a	1.32	a	1.72	a	1.31	a	1.35	a	1.73	a
Caryophyllene - (E)	1.4	a	1.48	a	1.64	a	1.43	a	1.49	a	1.64	a
(6-methyl-5-)Hepten-2-one	0.3	b	0.67	a	0.64	a	0.32	b	0.31	b	0.65	a
Eugenol	0	b	0	b	0.34	a	0	b	0	b	0.35	a
Aromadendrene	0.45	b	0.4	b	0.78	a	0.42	b	0.46	b	0.79	a
Terpinen-4-ol	0	b	0	b	0.74	a	0	b	0	b	0.73	a
(ar-)Curcumene	6.2	a	5.9	b	7.8	a	6.08	a	6.3	a	7.85	a
Cubebol	1.6	a	1.2	a	2.1	a	1.59	a	1.66	a	1.93	a
α - Cadinene	0	b	0	b	0.96	a	0	b	0	b	0.97	a
Nerolidol (E)	2.5	a	2.8	a	2.46	a	2.54	a	2.63	a	2.47	a
Spathulenol	15.8	b	17.2	b	26.2	a	15.5	b	16.5	b	25.1	a
Guaia-3,10(14)- diene(9,11-epoxy-)	2.8	a	2.89	a	1.54	b	2.69	a	2.79	a	1.55	b
β – Cedrene epoxide	0	b	0	b	2.9	a	0	b	0	b	2.91	a
(epi- α)Cadinol	2.2	b	2.9	b	4.1	a	3.01	a	3.09	a	4.12	a
Santalol (z-alpha-)	0	b	0	b	1.25	a	0	b	0	b	1.24	a
Khusinol	0.6	b	0.77	b	1.16	a	0.67	b	0.67	b	1.15	a
Phytol	0.6	b	0.5	b	1.5	a	0.7	b	0.51	b	1.52	a
Damascone(Z-alpha)	0.9	a	0.9	a	0	b	0.93	a	0.91	a	0	b
Alpha – Cadinol	1.1	a	1.2	a	0	b	1.07	a	1.14	a	0	b
Delta – Cadinene	0.3	a	0.3	a	0.42	a	0.28	a	0.29	a	0.43	a

Discussion:

Results obtained are in agreement with previous reports in Roman chamomile (Omidbaigi *et al*., 2003; Rushing *et al*., 2003) and Thyme (Venskutonis, 1997; Sefidkon *et al*., 1999; Argyropoulou *et al*., 2007). Therefore, we investigated Neral, Spathulenol and Geranial in three drying methods in two years. Results showed that maximum Spathulenol percentage (25.18%-26.2%) was obtained in semi industrial and maximum Neral and Geranial percentage was shown in industrial method of drying. The most essence percentage was made in semi industrial drying method from %3.13 and 4.12% in first and second year respectively (figures 1, 2). Industrial method because few time required is the good method but had negative effect on essential oil components (Hoseini nejad *et al*., 2002). In this research varying methods not had significant effect on quantity of component that Sefidkon *et al*., (2006) reported same results on *Satureja hortensis*. All other components remained more or less unchanged. In accordance with our results, there are similar reports from other researchers about other medicinal plants (Ahmadi *et al*., 2008; Khangholi and Rezaeinodehi, 2008; Asekun *et al*., 2006; Okoh *et al*., 2008; Omidbaigi *et al*., 2004; Rao *et al*., 1998).

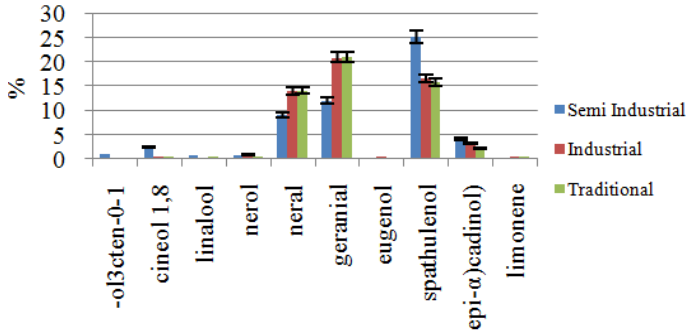

Fig. 1: Effect of drying methods on main of essential oil components in first year.

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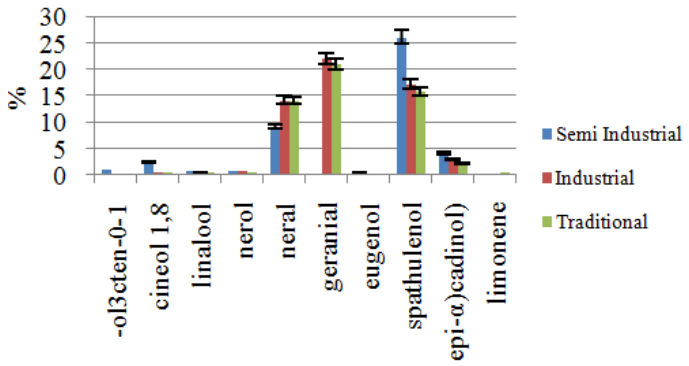

Fig. 2: Effect of drying methods on main of essential oil components in second year.

Conclusion:

Aim of this research was evaluation and determine of quality and quantity of essential oil components of *Lippia Citriodora*. Between 3 methods in this study, it could be concluded that semi industrial method is suitable for highest essential oil quantity. The most of components in all methods were Neral, Spathulenol and Geranial. The highest Spathulenol and most of essence was obtained in semi industrial method. Limonene that is mono terpen, in semi industrial method decreased and no made but in other methods was remained. It seems that most of essential oil components on more than of 30$^{°C}$ declined.

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