Chemical composition of Lavender (*Lavandula int. Grosso*) extracts obtained by Accelerated Solvent Extraction

Petra Strižincová, Michal Jablonský, Aleš Ház, Jozef Šima
petra.strizincova@stuba.sk
Faculty of Chemical and Food Technology, Slovak University of Technology in Bratislava,
Radlinskeho 9, 812 37 Bratislava, Slovak Republic

Abstract — This paper is aimed at evaluating the chemical aspects and marketing prospects of the extraction of Lavender (*Lavandula int. Grosso*) by Accelerated Solvent Extraction (ASE). The extraction was performed at three temperatures (100°C, 120°C and 140°C) and ethanol (96 %v/v) was selected as the polar solvent. The extraction yield was influenced only slightly by the extraction temperature (reaching 17.0 %, 16.5 % and 16.3 % at 140°C, 120°C and 100°C, respectively); its impact on a number and ratio of extractants was more pronounced. As for the chemical composition of the extracts, the major components at 100°C were methyl ursolate (30.50 %) and methyl oleanolate (7.27 %), while at 120°C and 140°C linalyl anthranilate (>31 %) and linalyl acetate (>17 %) dominated. Based on catalogue prices, and taking the costs connected to the extraction and separation of individual extractants into account, some of the components are worth considering also from a marketing viewpoint.

Keywords: extractives, lavender, accelerated solvent extraction, linalyl, market value

1 Introduction

Extraction is one of the ways of gaining effective substances from different types of biomass. Nowadays, the use of extractives obtained from different types of lavender is widespread as described in various scientific publications [1-4]. The reason lies mainly in the broad spectrum of application in the industry due to their antimicrobial [5], antibacterial, anti-inflammatory, antifungal, insecticidal or antioxidant effects. Considering lavender aromatic, medical or pharmaceutical activity, the extracted essential oils are used as an effective remedy in the cosmetics or drugs [3]. Final quantification of extracted compounds is determined by the morphology, plant species, climate, specific part of plants or the location [6]. Garcia-Vallejo et al. [7] showed and proved a difference between chemical composition of the extract from *L. angustifolia* (the most dominant compounds were linalool 25-38 % and linalyl acetate 25-45 %) and chemical composition of the extract from the subspecies *L. angustifolia ssp. Pyrenaica* growing in warmer region in Spanish province (linalool 20-66 %, borneol 6-32 % and camphor 2-14 %). The total yield of extracted substances and their composition depends mainly on the applied extraction techniques. Along with traditional methods of isolating volatile oils such as hydrodistillation [8] and steam distillation [9], innovative techniques such as supercritical fluid extraction (SFE) (with solvents like CO_2, propane, butane etc.) [3] or the microwave-assisted extraction (MAE) are introduced into lab scale and industrial scale practice. The standard laboratory method used for comparison of extracts composition is still soxhlet extraction [10], having long duration and high consumption of solvent disadvantages. Through choosing the extraction technique and setting the operational parameters efficient extraction of different groups of compounds can be achieved. Jianu et al. [11] found out that steam distillation is a suitable method to obtain sesquiterpenes or terpene esters as major compound groups. Contrary, in our research applying Accelerated Solvent Extraction (ASE) technique, presented in this paper, we confirmed the fatty acids esters and terpene esters as the most representative compounds. Lavender is a plant easy to grow, famous because of its blue-violet flowers. It is a member of the Lamiaceae family [5], predominantly full of essential oils. More than 30 lavender species are known [12] differing in their morphological features (leaf and flower shapes and coloration) and, which is a key feature, in their chemical composition [13]. All of these species are divided into 4 basic groups: L. latifolia, L. angustifolia, L. stoechas and L. x intermedia (Lavandin). For our experiment we choose Lavandula int. Grosso belonging to the group Lavandin, a sterile lavender hybrid. Along with L. angustifolia, these are groups primarily cultivated for essential oils. We are aware of the fact that during sample pretreatment (drying) part of volatile compounds evaporate. We investigated, however, the samples pretreated in a similar way as those studied in previous works in order to compare our and literature results reached at alike conditions. ASE technique has become very effective and popular method for obtaining extractives from different types of biomass. In lab scale operation it needs short extraction time and low solvent usage, temperature and pressure are optional. In this paper the results obtained at extrac-

• *Petra Strizincova, Institute of Natural and Synthetic Polymers, Department of Wood, Pulp and Paper, Faculty of Chemical and Food Technology, Slovak University of Technology in Bratislava, Radlinskeho 9*
e-mail: petra.strizincova@tuba.sk

tion of Lavender (*Lavandula int. Grosso*) by ASE performed at three temperatures (100°C, 120°C and 140°C) are presented and discussed from the three viewpoints: total yield of extracted compounds; contents of individual compounds; significance of the process from marketing aspects. It is worth mentioning that at formulation of high-quality cosmetic products, despite their higher price natural components are preferred to synthetic ones.

2 Material and Methods

2.1 Materials

The lavender (Lavandula int. Grosso) flowers were cultivated in the southern Slovakia (district of Štúrovo). Organic solvents (ethanol, pyridine) were of p.a. quality, supplied by Aldrich and used as received.

2.2 Accelerated solvent extraction (ASE)

Extractions were performed with an accelerated solvent extractor, Dionex ASE 350, where the extraction pressure (1 500 psi) was imposed by an ASE 350 apparatus. The lavender flowers samples were placed into the stainless steel extraction chambers and extracted by ethanol (96 % v/v). Together 3 sequences were accomplished; the duration of each was 10 min. The parameter of static time (time for reaching final temperature) was from 6 to 10 min for all three performed temperatures (100°C, 120°C and 140°C). Finally, the samples were flushed with 50 % v/v ethanol. All extracts were collected in vials.

2.3 Derivatization

The preferred technique of derivatization was methylation with the agent DMF- DMA (N,N-dimethyl formamid + dimethyl acetal) in pyridine. 50 mg of dry sample obtained after evaporation of ethanol was derivatized by 0.5 ml of pyridine and 0.5 ml of DMF-DMA. Process was carried out at 75 °C for 30 min.

2.4 GC/MS analysis

The GC/MS analysis was performed on a gas chromatograph (Agilent 7890 GC) coupled with a mass detector (Agilent 5975 C) which ran electron ionization equipped with a capillary column (HP-5MS, 30 m × 250 µm i.d., 0.25 µm film thickness; Agilent). Helium was used as carrier gas at a rate of 2 mL/min. Chromatograph oven initial temperature was 40°C (held for 2 min), then heating of 10 °C/min to 280 °C. The final temperature was held for 6 min. Recording and evaluation of data was performed using ChemStation Software E 02/01/1177 and identification of compounds using electronic libraries NIST and Wiley.

2.5 Yield of extractives

The yield of extractives (Y, %) was determined after each experiment by drying the samples at 105 °C to a constant weight. The results are expressed on the basis of the dry matter before and after extraction as shown in Eq.1.

$$Y(\%) = \frac{(m_i - m_{ext})}{m_i} 100 \quad (1)$$

where m_i (g) is the mass of the dried lavender flowers before extraction and m_{ext} (g) is the mass of lavender flowers after extraction and drying.

3 Results and Discussion

Total yield of extracted compounds obtained at 100°C, 120°C and 140°C is listed in Table 1.

Tab. 1 Comparison of ASE yields at 3 different temperatures (100°C, 120°C, and 140°C) using ethanol as a solvent

Method	Temperature (°C)	Y (%)
ASE	100	16.3±04
	120	16.5±04
	140	17.0±03

As shown in Tab. 1, the effect of temperature on the yield is not substantial. Based on derivatization there are methyl compounds present in our extracts. Products of derivatization are compounds with similar or closely related structures and properties, though they are not the same as in the original unmodified compounds. Totally, we identified 34 different compounds including saturated hydrocarbons, monoterpenes, oxygenated monoterpenes, fatty acids esters etc. (see data in Table 2). Linalyl anthranilate was the major compound in extracts at 120°C and 140°C, its presence also was confirmed by Jablonsky et al. [2] in their research, but they used different solvent (dichloromethane). Adaszyńska et al. [14] extracted 5 different types of lavender (Munstead, Munstead Strain, Lavender Lady, Elegance Purple and Blue River) where linalyl anthranilate represented from 1.60 to 12.30 % (Blue River) of extracts. In this paper, significant presence of linalyl anthranilate in L. int. Grosso extracts (31.48 and 35.16 %) (Table 2) is demonstrated. Contrary to the total yield of extraction (Table 1), data in Table 2 clearly demonstrate a substantial influence of the temperature of extraction on the final chemical composition of extracts. In the extract obtained at 100°C, methyl ursolate was the major constituent (30.50 %) and it was also identified in the other two extracts, however, its content strongly decreases with extraction temperature. On the other hand, the content of linalyl anthranilate (31.48-35.99 %), linalyl acetate (17.33-19.67 %), camphor (3.88-6.03 %), caryophyllene (1.81-2.13 %), lavandulyl acetate (2.55-3.34 %), myrcene (0.60-0.81 %), methyl linolenate (0.93-1.15 %), β-bisabolene (0.32-0.83 %), γ-muurolene (1.07-1.11 %) and α-muurolene (0.25-0.28 %) increases with the rise of extraction temperature. In addition to the mentioned compounds, fresh flowers contain some biologically active substances such as phytosterols and phenols, including cinnamic acid derivates and flavonoids. This issue is evaluated closer in Nitzsche et al. [15] research.

Tab. 2 Chemical composition of Lavandula int. Grosso derivatized extracts obtained by Accelerated Solvent Extraction (ASE) at 100°C, 120°

C, and 140°C by ethanol.

Retention Time (min)	CAS number	Selected compounds	Composition of lavender extracts (%)		
			ASE		
			100°C	120°C	140°C
3.301	470-82-6	Eucalyptol	4.72	3.50	5.54
4.996	115-95-7	Linalyl acetate	-	17.33	19.67
3.523	41720-55-2	Linalool oxide	3.59	0.45	0.67
4.149	76-22-2	Camphor	3.88	4.89	6.03
10.442	531-59-9	Herniarin	-	1.01	-
4.925	20053-88-7	Hotrienol	3.22	-	-
7.013	87-44-5	Caryophyllene	-	1.81	2.13
8.899	1139-30-6	Caryophyllene oxide	2.70	0.65	-
5.329	25905-14-0	Lavandulyl acetate	-	2.55	3.34
14.134	112-61-8	Methyl stearate	-	-	0.25
20.134	112-95-8	Eicosane	0.43	0.30	-
23.695	646-31-1	Tetracosane	1.16	-	-
21.667	593-45-3	Octadecane	2.23	-	-
4.330	507-70-0	Borneol	-	1.81	-
4.481	10482-56-1	α-terpineol	-	0.72	0.72
4.381	562-74-3	Terpinen-4-ol	-	2.45	2.39
9.877	515-69-5	α-bisabolol	-	3.04	2.66
7.235	28973-97-9	Farnesene	-	1.90	1.40
7.820	123-35-3	Myrcene	-	0.60	0.81
12.247	112-39-0	Methyl palmitate	1.54	0.98	1.11
13.871	2462-85-3	Methyl linoleate	-	-	0.31
13.942	301-00-8	Methyl linolenate	-	0.93	1.15
6.357	105-87-3	Geranyl acetate	-	-	1.33
9.464	54274-73-6	(+)-epi-bicyclosesquiphellandrene	-	1.26	1.08
7.901	495-61-4	β-bisabolene	-	0.32	0.83
7.699	30021-74-0	γ-muurolene	-	1.07	1.11
8.052	483-75-0	α-muurolene	-	0.25	0.28
28.258	1721-58-0	Methyl oleanolate	7.27	3.75	-
28.233	35933-00-7	Methyl morolate	5.01	1.80	0.89
29.443	32208-45-0	Methyl ursolate	30.50	12.88	0.65
29.383	1000242-87-7	Methyl p-(phenylethynyl) cinnamate	-	-	5.03
4.996	7149-26-0	Linalyl anthranilate	-	31.48	35.99
24.179	25529-07-1	Methyl ursa-2,12-dien-28-oate	5.39	-	2.29
6.146	2345-26-8	Geranyl isobutyrate	-	-	0.52

3.1 Comparison of market prices of substances

obtained from L. int. Grosso

The data gathered in Table 2 indicate that purposefully varying the extraction temperature makes it possible to tune the composition of extracts and, in turn, to positively influence the market value of the extracts. Due to the different quality and purity of each extractive the prices on the market are different and strongly variable. Variability depends on the method of isolation or extraction, the quality of final product or the financial requirements. In this paper we refer to prices as of April 2016 published on the portals www.molport.com and www.sigmaaldrich.com. Each substance is represented by a bubble showing the price range (the minimum and maximum market price per 1 kg). The larger the bubble range is, the more variable is the price of a substance on the market. For a better view, we divided extractives into two groups (Fig. 1 and Fig. 2). Also, substances obtained from lavender are divided into classes, which are represented by selected compounds. The most abundant groups of compounds are saturated hydrocarbons (docosane, octadecane, octacosane, heptadecane, pentadecane, tetracosane and eicosane), oxygenated monoterpenes (linalool oxide, terpinen-4-ol, α-terpineol, geranyl acetate, hotrienol, borneol and (+)-camphor), fatty acids esters (methyl dehydroabietate, methyl stearate, methyl linolenate, methyl linoleate and methyl palmitate), terpene esters (linalyl acetate and lavandulyl acetate), oxygenated sesquiterpenes (caryophyllene oxide and α-bisabolol), monoterpenes (myrcene and eucalyptol), sesquiterpenes (caryophyllene and farnesene) and the last one, benzopyrones (coumarin and herniarin), which form the minor group of substances in our research. Fig. 1 refers to the monoterpenes like the group with the lowest prices on average (150 $/kg for myrcene and 610 $/kg for eucalyptol). Generally, linalyl acetate is the one with the lowest price, just 70 $/kg, represented by a light blue bubble in Fig. 1. Saturated hydrocarbons maintain an average price at 1 300 $/kg. The most diverse prices relate to oxygenated monoterpenes ranging from α-terpineol (590 $/kg) up to hotrienol (310 000 $/kg) with the highest quality which is the substance with the highest market price at all. The goal is to extract substances with the highest price and quality in the highest possible yield. Taking this goal into account, 3.22 % yield of hotrienol at 100°C, is a meaningful value. Discovering and applying a mode for obtaining a higher percentage of linalool oxide, borneol, caryophyllene or α-bisabolol could bring certain financial benefits. It is interesting to compare the prices of linalyl acetate (Fig. 1) and lavandulyl acetate (Fig. 2).

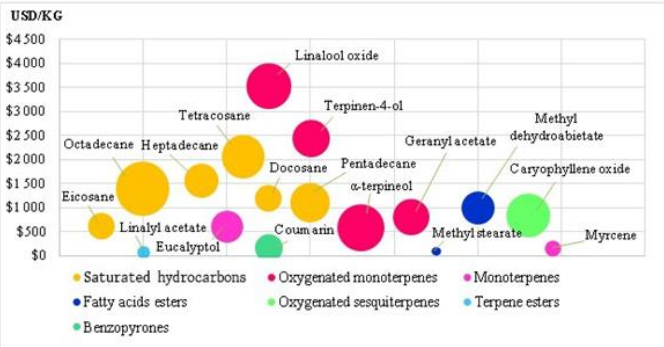

Fig. 1 The market prices range (<4 500 $) of compounds extracted from lavender (Lavandula int. Grosso) by ASE (100 °C, 120 °C and 140 °C) using ethanol as a solvent

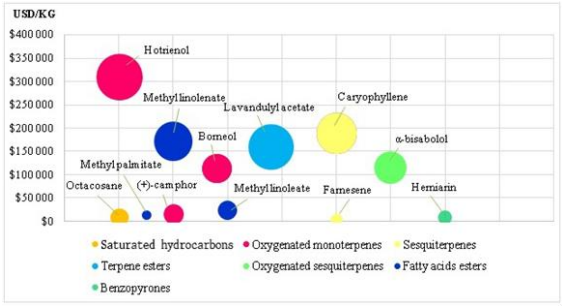

Fig. 2 The market prices range (>4 500 $) of compounds extracted from lavender (Lavandula int. Grosso) by ASE (100 °C, 120 °C and 140 °C) using ethanol as a solvent

While the price of linalyl acetate ranges from 40 to 100 $/kg, lavandulyl acetate price moves between 120 000 and 200 000 $/kg. That is because linalyl acetate, as part of extracts, is normally present in a prevailing ratio resulting in the decrease in market prices, as confirmed by Dušková et al. [4] and Zheljazkov et al. [16]. Based on our research, we can state which compounds are convenient to obtain because of their market prices after choosing the right extraction method and conditions. What we would like to obtain and isolate from the biomass is decisive and determinant factor (it follows an extraction method, temperature, pressure or type of solvent).

4 Conclusions

The lavender is widely use in food, aromatherapy, fragrant and pharmaceutical industries due to its unique chemical composition. Lavender (*Lavandula int. Grosso*), having a sufficiently high content of linalyl compounds, which could be a promising new feedstock for industrial production. By using GC/MS it was found that the examined samples (ASE extracts) contain high quantities of methyl ursolate (30.50 %) at 100 °C and methyl oleanolate (7.27 %) at 120 °C and at 140 °C linalyl anthranilate (>31 %) and linalyl acetate (>17 %). The results show that the chemical compositions in lavender for different ASE conditions are clearly distinguished. The choice of method and extraction conditions is dictated by economics: it is connected with balance between the use and maintenance costs for processing, solid to liquid separation efficiency, and the cost of a given processing method.

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