

# Supercritical CO<sub>2</sub> Extraction and Fractionation of Lavender Essential Oil and Waxes

E. Reverchon\* and G. Della Porta

Dipartimento di Ingegneria Chimica e Alimentare, Università di Salerno, Via Ponte Don Melillo, 84084 Fisciano (SA), Italy

F. Senatore

Dipartimento di Chimica delle Sostanze Naturali, Università di Napoli, Via D. Montesano 49, 80131 Napoli (NA), Italy

Supercritical CO<sub>2</sub> extraction of essential oil from lavender was performed on a laboratory apparatus as well as in a pilot plant. A two-stage separation procedure was used to induce the fractional separation of the extracts. Detailed GC-MS analysis of the products was performed to assess the best extraction and the best separation conditions. The lavender oil produced by supercritical extraction was compared to the oil obtained by hydrodistillation. The major difference between the two products was reflected in the linalyl acetate content. This compound was found to be 34.7% of the oil produced by supercritical fluid extraction and 12.1% of the hydrodistilled product. This difference can be ascribed to the hydrolysis of part of this compound during hydrodistillation. The oil yield of the extraction process was measured at various extraction lengths. It was modeled using a simple mathematical model.

**Keywords:** *Supercritical extraction; fractionation; lavender; essential oil; linalyl acetate*

## INTRODUCTION

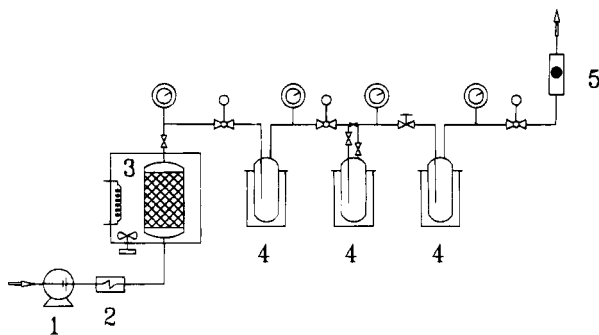
A volatile mixture of terpenes and their derivatives is generally responsible for the characteristic fragrance of vegetable matters. The extraction of essential oil from flowers and leaves represents an attempt to isolate this mixture while preserving the original composition that produces the natural fragrance. Unfortunately, the techniques usually adopted, such as steam distillation and solvent extraction, suffer several limitations in extracting and preserving the composition of natural fragrances. They can produce the degradation of thermolabile compounds, hydrolysis of water sensitive compounds, and solvent contamination. Moreover, steam distillation can produce an incomplete collection of compounds responsible for the fragrance. Indeed, this technique is based on the evaporation of volatile compounds induced by steam (Denny, 1988). Therefore, compounds with low vapor pressure can be not completely extracted by this technique.

Liquid or supercritical CO<sub>2</sub> extractions have been proposed as alternative techniques (Chen and Ho, 1988; Moyler, 1993; Stahl et al., 1987; Reverchon, 1992) to perform the extraction of natural fragrances. Indeed, the process can be performed at low temperatures, CO<sub>2</sub> does not contaminate the products, and it is unlikely that decomposition reactions of any nature could happen in this solvent. Unfortunately, the single-step extraction and separation by liquid or supercritical CO<sub>2</sub> produces the coextraction of nonvolatile compounds: a quasi-solid extract is obtained (Reverchon and Senatore, 1994). However, the possibility of fine tuning the solvent power and the selectivity of the extraction process can be of help in overcoming these problems. Therefore, operating at CO<sub>2</sub> densities lower than approximately 0.6 g/cm<sup>3</sup>, it is possible to exclude all but one of the nonvolatile compound families from the extract. The only exception is represented by paraffins constituting the cuticular waxes. These compounds are

located on the surface of vegetable matter, and although they have a relatively low solubility in supercritical CO<sub>2</sub>, they are coextracted under any process conditions by the simple washing mechanism (Reverchon, 1992). On the contrary, terpenes and their derivatives show very high supercritical CO<sub>2</sub> solubilities (Stahl and Gerard, 1985) but are located inside the vegetable structure and have to overcome a complex mass transfer mechanism before extraction (Reverchon, 1992). Nevertheless, it is possible to eliminate the waxes by inducing the selective precipitation of the extract in two or more separators operated in series (Reverchon, 1992; Reverchon and Senatore, 1992). For this purpose, it is necessary to select adequate pressure and temperature conditions that can produce the selective supersaturation and precipitation of solutes. By this technique, the complete separation of cuticular waxes from the essential oil has been achieved for various vegetable matters such as rosemary, marjoram, chamomile, peppermint, and basil (Reverchon, 1992; Reverchon and Senatore, 1992, 1994; Reverchon et al., 1994a,b).

Stahl et al. (1987) reported a previous study on lavender extraction in which liquid CO<sub>2</sub> was used. These authors stated that the extract showed high contents of linalool and linalyl acetate. The high percentage of the latter compound is particularly important because the conventional steam distillation process can lead to partial hydrolysis of linalyl acetate. By contrast, liquid CO<sub>2</sub> extraction is performed at high solvent densities that lead to very low extraction selectivities. Therefore, high molecular weight lipophilic compounds contained in the vegetable matter are also extracted (Moyler, 1993). Recently, Adasoglu and Dincer (1994) tried to optimize the supercritical fluid extraction of lavender through a five-level selection of the process variables (response surface methodology).

The aim of this work was to apply supercritical CO<sub>2</sub> extraction and the fractional separation process to



**Figure 1.** Schematic representation of the laboratory apparatus: 1, high-pressure diaphragm pump; 2, heater; 3, extraction vessel; 4, separators; 5, flow measurement.

isolate lavender volatile oil. The oil was then compared to the product obtained by hydrodistillation.

The extraction yield was also measured at various extraction lengths. A mathematical model based on differential mass balances performed along the extraction bed was used to fit the extraction yield data.

## MATERIALS AND METHODS

**Materials.** Dried lavender inflorescence was supplied by Betulla srl (Italy). The flowers were small, single, and unopened. The moisture content was 10.2% on a dry basis. We did not try to comminute them to avoid the loss of volatiles as was observed by Adasoglu and Dincer (1994). Hydrodistillation was performed on the same lot of material.

**Experimental Apparatuses.** Experiments on lavender were performed on a laboratory unit (see Figure 1) based on a 400 cm<sup>3</sup> extraction vessel. It consisted of a high-pressure pump (Milton Roy, Point San Pierre, France, Model Milroyal B) capable of a maximum pressure of 500 bar and a maximum CO<sub>2</sub> flow rate of about 4 kg/h. Three separators operating in series can be used to fractionate the extracts. They were capable of operating at temperatures from -10 to 60 °C and at pressures up to 300 bar. Configuration with one or two separators was also possible. In this study two separators were used: cuticular waxes were precipitated in the first one and the volatile fraction in the second one.

The best overall performance of the process resulted from the extraction performed at 90 bar of pressure and at a temperature of 48 °C. Separation was conducted at 80 bar, -10 °C, and 25 bar, 0 °C, in the first and second separators, respectively. One hundred grams of dried material was used in each run. The extraction was performed for 150 min with a flow rate of 0.8 kg/h. No longer extractions were performed since no more lavender oil was extracted after this extraction time; i.e. the oil contained in the vegetable matrix was exhausted. More details on this apparatus were given in previous works (Reverchon, 1992; Reverchon and Senatore, 1992).

Some experiments were also performed in a pilot plant that mainly consisted of two high-pressure pumps capable of a maximum flow rate of 50 kg/h each and with a maximum operating pressure of 350 bar, one extraction vessel with an internal volume of 20 dm<sup>3</sup>, and three separation stages in series. The pilot plant was operated at the same extraction conditions as the laboratory unit, but separation conditions in the last stage were adapted to allow CO<sub>2</sub> recirculation. Further details on this apparatus are given elsewhere (Reverchon et al., 1994b).

Hydrodistillation was performed for 3 h according to the standard procedure described in the *European Pharmacopoeia* (1975).

**Analytical Procedures.** The fractions extracted were analyzed using a GC-MS apparatus formed by a Varian Model 3400 gas chromatograph (Varian Analytical Instruments, San Fernando, CA) coupled to a Finnigan MAT ion trap detector (ITS 40 Magnum, Finnigan MAT, San Jose, CA). We used a fused silica DB-5 column (J&W, Folsom, CA), 30 m × 0.25 mm

i.d. with 0.25 μm film thickness. The GC conditions were as follows: oven temperature, 50 °C for 5 min, programmed to increase from 50 to 250 °C at rate of 2 °C/min, and subsequently isothermal at 250 °C for 60 min. The percentage composition of volatile oil and of cuticular waxes was computed from GC peak areas without using any correction factor.

The identification of compounds was based on the comparison of retention times and mass spectra with those of the corresponding pure compounds whenever possible. Mass spectra were also compared with National Institute of Standards and Technology (NIST) and WILEY5 mass spectra libraries.

## RESULTS AND DISCUSSION

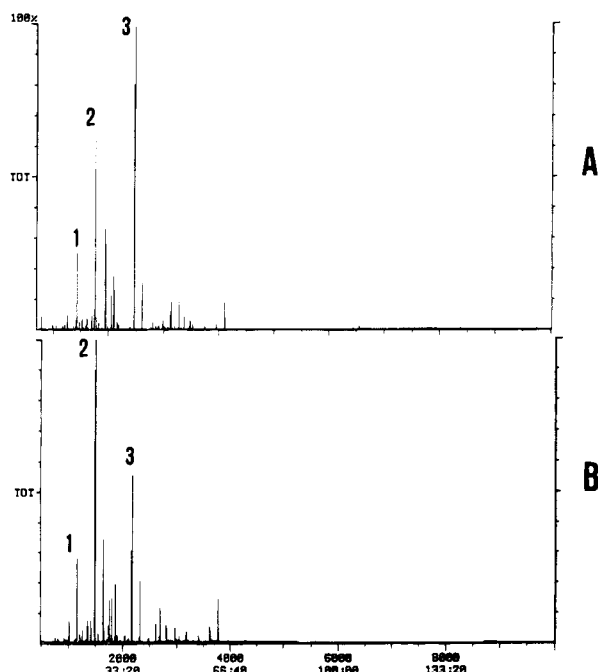
Supercritical CO<sub>2</sub> extraction tests on lavender flowers were performed in the pressure range from 80 to 120 bar and for temperatures between 35 and 60 °C. Extraction conditions characterized by CO<sub>2</sub> densities higher than about 0.6 g/cm<sup>3</sup> were not tested to avoid the coextraction of higher molecular weight compounds. Fractional separation of the extracts was also performed with the scope of selectively precipitating cuticular waxes.

The products obtained at the same extraction conditions on the two supercritical extraction apparatuses showed the same chemical composition. Therefore, in the following any indication as to which apparatus was used for a particular test is omitted. Optimum extraction was obtained at a pressure of 90 bar and a temperature of 48 °C. Optimum conditions were evaluated with respect to the oil composition by performing extraction at different CO<sub>2</sub> densities and analyzing the corresponding extracts by GC-MS. A detailed description of this procedure was published elsewhere (Reverchon, 1992). The optimum condition was the one at which no undesired compounds (higher molecular weight compounds) were coextracted.

Fractional separation of lavender extracts was achieved by reducing pressure and temperature in the first separator with respect to the operating parameters used during supercritical extraction. Lowering of the temperature is particularly effective in producing fractionation between paraffins and terpene compounds. Indeed, these two compound families have opposite solubility behaviors in CO<sub>2</sub> at temperatures lower than 20 °C: paraffin solubility decreases, whereas the solubility of terpene compounds increases (Stahl et al., 1987; Reverchon et al., 1992; Richter et al., 1990; Tufeu et al., 1993). We induced the optimum precipitation of lavender paraffins by cooling the first separator to -10 °C and lowering the pressure to 80 bar. In the second separator, the fragrance compounds were collected by operating at a pressure of 25 bar and a temperature of 0 °C in the laboratory apparatus. In this manner the release of terpenes from the gaseous CO<sub>2</sub> is assured. The chosen value of temperature minimized the loss of volatiles by reducing their vapor pressure.

The maximum lavender oil yield was measured at the end of an exhaustive supercritical extraction run. It resulted in 4.9% by weight of the material charged in the extractor. Approximately the same oil yield was also obtained by hydrodistillation.

In Figure 2A is shown the GC trace of the best composition of lavender oil. The GC trace of the hydrodistilled oil is shown in Figure 2B for comparison purposes. GC peaks have been identified as hydrocarbon terpenes, oxygenated terpenes, sesquiterpenes, and oxygenated sesquiterpenes, i.e., the compounds indicated as responsible for the characteristic smell of



**Figure 2.** (A) GC trace of oil recovered in the second separator. The supercritical CO<sub>2</sub> extraction was performed at 90 bar, 48 °C. (B) GC trace of essential oil obtained by steam distillation: 1, 1,8-cineole; 2, linalool; 3, linalyl acetate.

lavender. Figure 2 gives also a qualitative description of the differences between the two products. A quantitative analysis can be performed from the data in Table 1, in which the identification and the quantitation of all extracted compounds are reported.

The compounds contained in higher percentages in supercritical and in hydrodistilled oils are 1,8-cineole, linalool, camphor, 4-terpineol,  $\alpha$ -terpineol, and linalyl acetate. They correspond to the most significant compounds indicated by an extensive work performed on several lavender and lavandin cultivars (Tucker et al., 1984). One major difference appeared between the supercritical extract and the hydrodistilled product: the former contained 25.3% linalool and 34.7% linalyl acetate, while the latter contained 35.3% linalool and only 12.1% linalyl acetate. Therefore, there was evidence that linalyl acetate was partly decomposed during hydrodistillation. This confirmed the opinion that the conventional hydrodistillation technique can greatly modify the extract with respect to the starting composition of the vegetable matter. Therefore, with the supercritical extraction and fractionation technique we obtained not only a liquid and low-colored product resembling the conventional steam-distilled essential oil but also a more accurate reproduction of the fragrance contained in lavender.

Organoleptic analysis performed by using the standard panel testing procedure (IFT, 1981) confirmed these analytical results. The oil obtained by hydrodistillation showed an appreciable odor difference compared to the starting lavender, whereas the odor of the oil produced by supercritical extraction was considered by panelists to be not distinguishable from that of the starting materials.

The GC trace of cuticular waxes precipitated in the first separator during the extraction process is shown in Figure 3. These compounds showed higher retention times and were identified as high molecular weight paraffins. There is no overlap between the peaks shown in Figure 2A and in Figure 3: the fractional precipita-

**Table 1. Identification and Quantitation of Compounds Found in Lavender Oil Extracted by SFE and by Hydrodistillation (HD)<sup>a</sup>**

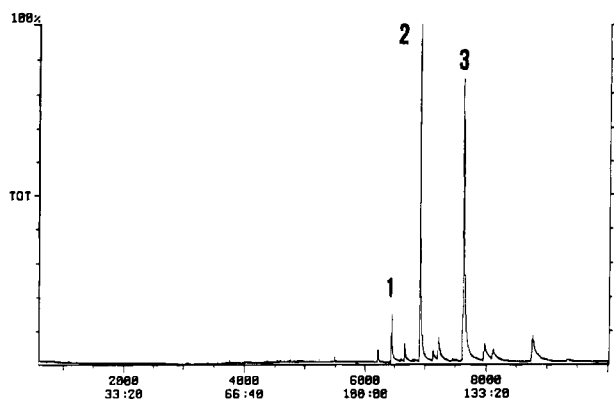
compound	R <sub>t</sub> (min)	SFE%	HD%	waxes%
2-methylcyclopentanol	8.4	0.72		
$\alpha$ -thujene	12.3	tr	0.05	
$\alpha$ -pinene	12.5	0.27	0.26	
camphene	13.4	0.22	0.19	
sabinene	15.3	tr	0.31	
1-octen-3-ol	16.1	tr	0.16	
$\beta$ -pinene	16.6	0.21	1.58	
myrcene	17.0	1.19	tr	
hexyl acetate	18.4	0.15	0.15	
<i>o</i> -cymene	19.0	0.14	0.30	
<b>1,8-cineole</b>	<b>19.3</b>	<b>5.83</b>	<b>6.75</b>	
<i>cis</i> -ocimene	20.2	0.58	0.56	
<i>trans</i> -ocimene	21.0	0.72	0.77	
menth-2-en-1-ol	21.6	0.30	tr	
<i>cis</i> -linalool oxide	22.4	0.83	1.48	
<i>trans</i> -linalool oxide	23.5	0.91	1.57	
<b>linalool</b>	<b>24.6</b>	<b>25.29</b>	<b>35.31</b>	
octen-1-ol acetate	25.6		0.55	
camphor	27.3	7.90	7.81	
borneol	29.2	2.30	2.98	
lavandulol	29.4	0.31	0.55	
4-terpineol	30.1	3.79	3.34	
$\alpha$ -terpineol	31.1	0.46	4.39	
hexyl butyrate	31.4	0.35	0.43	
nerol	34.0	tr	0.47	
geraniol	36.0	tr	0.67	
<b>linalyl acetate</b>	<b>36.1</b>	<b>34.69</b>	<b>12.09</b>	
3,7-dimethyl-2,6-octadien-1-ol acetate	38.4	3.08	4.34	
neryl acetate	43.3	0.33	1.31	
geranyl acetate	44.5	0.71	2.57	
caryophyllene	46.5	1.86	1.30	
bergamotene	48.3	0.19		
<i>cis</i> - $\beta$ -farnesene	49.3	2.23	1.00	
aromadendrene	50.4	1.02	0.46	
$\gamma$ -muurolene	52.5	0.84	0.79	
ledol	56		0.56	
torreyol	60.2	0.54	1.25	
$\alpha$ -bisabolol	62.5	2.09	3.76	
hexenyl ester	77.4			2.22
tetradecene	84.5			0.71
<b>hexadecene</b>	<b>99.5</b>			<b>11.72</b>
octadecene	107.4			10.37
heicosene	110.0			0.33
nonacosane	115.2			5.62
triacontane	119.1			0.21
methylnonacosane	120.4			0.76
<b>hentriacontane</b>	<b>127.2</b>			<b>34.60</b>
methyltriacontane	133.3			7.72
dotriacontane	135.4			3.49
methylhentriacontane	142.1			0.69
<b>tritriacontane</b>	<b>146.4</b>			<b>21.03</b>
tetratriacontane	156.2			0.52

<sup>a</sup> SFE conditions: 90 bar, 48 °C. The identification of waxes recovered in the first separator is reported too. R<sub>t</sub> = retention time; % = percentages calculated by GC peak area integration; tr = percentages lower than 0.05%.

tion technique was shown to be very selective, and no paraffins were precipitated together with the oil.

The extraction time can play an important role in determining the extract compositions because the various families of compounds constituting the oil are characterized by different diffusion times. For this reason, the characterization of volatile oil was made using the total quantity of lavender oil extracted during an exhaustive experiment. It was assumed that asymptotic extraction yield corresponded to the complete exhaustion of oil from the vegetable matrix.

The yields of lavender oil and cuticular waxes were determined as a function of extraction time by gravimetric quantitation of the extracts collected in the



**Figure 3.** GC trace of lavender waxes recovered in the first separator: 1, hexadecene; 2, hentriacontane; 3, tritriacontane.

second and first separators, respectively, at the optimum conditions. The maximum oil yield was 4.9% by weight of the charged material. It was assumed to be equal to 100% in the following.

The yield of lavender oil was modeled by using the mathematical model proposed for basil oil extraction by supercritical fluid CO<sub>2</sub> (Reverchon and Sesti Osséo, 1994). This model was based on the integration of differential mass balances performed along the extraction bed. We hypothesized that the external mass transfer resistance was negligible. Moreover, we supposed that the extraction was uniform along the bed and that variations of solute concentration in the fluid phase were negligible. According to these hypotheses, a simplified solution of mass balances is represented by (Reverchon and Sesti Osséo, 1994)

$$Y = 100[1 - \exp(-t/t_i)] \quad (1)$$

where  $Y$  is the normalized oil yield and  $t$  is the extraction time.

Having obtained the value of the internal diffusion time ( $t_i$ ) from eq 1, it is possible to calculate the internal diffusivity ( $D_i$ ) as

$$t_i = \mu(l^2/D_i) \quad (2)$$

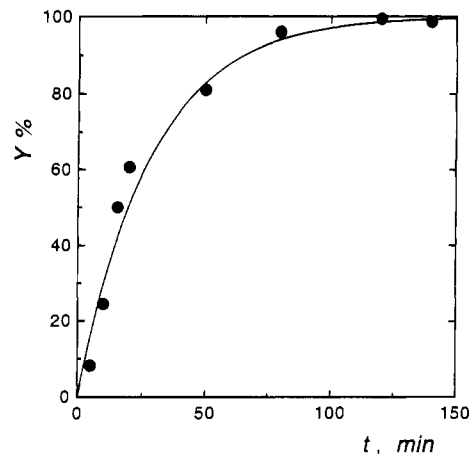
where  $l = V_p/A_p$ ; i.e., it is the ratio between particle volume and particle surface area.

In the case of lavender flowers the hypothesis of spherical particles used for basil oil (Reverchon and Sesti Osséo, 1994) is clearly not applicable since the vegetable matter was not previously comminuted. Because of the characteristic shape of the unopened flowers and on the basis of the mean of experimental measurements, we chose to represent the lavender flowers as hollow cylinders with a mean height of 5 mm, a mean overall diameter of 1.5 mm, and an internal diameter of 1 mm. Moreover, we fixed  $\mu = 1/2$  as suggested by Villermaux (1987) for cylindrical geometry.

The resulting model curve is shown in Figure 4. We used  $D_i$  as the adjustable parameter to fit eq 1 to data. A fairly good superposition of the model to experimental data was obtained for a value of  $D_i$  of  $4.5 \times 10^{-12}$  (m<sup>2</sup>/s).

#### ACKNOWLEDGMENT

This work was partly developed in the framework of Progetto Strategico "Tecnologie Chimiche Innovative", CNR, Roma, Italy.



**Figure 4.** Lavender oil yield at various extraction times. The continuous line represents the best fit of data by eq 1. ●, Experimental results.

#### LITERATURE CITED

- Adasoglu, N.; Dincer, S. Optimization of SCFE of Essential Oil from Turkish Lavender Flowers by Response Surface Methodology. *Proceedings of the 3rd International Symposium on Supercritical Fluids*; Perrut and Brunner, Eds.; Strasbourg, 1994; Vol. 2, pp 329–334.
- Chen, C. C.; Ho, C. T. Gas Chromatographic Analysis of Volatile Components of Ginger Oil Extracted with Liquid Carbon Dioxide. *J. Agric. Food Chem.* **1988**, *36*, 322–328.
- Denny, E. F. K. Steam Distillation of the Subcutaneous Essential Oils. *Flavours and Fragrances: A World Perspective; Proceedings of the 10th International Congress of Essential Oils*; Lawrence, Mooknerjee, and Willis, Eds.; Elsevier Science Publishers: Amsterdam, 1988.
- European Pharmacopoeia*; Maisonneuve: Sainte-Ruffine, France, 1975; Vol. 3, pp 68–71.
- IFT. Sensory Evaluation Guide for Testing Food and Beverage Products. *Food Technol.* **1981**, *35*, 50–57.
- Moyler, D. A. Extraction of Essential Oils with Carbon Dioxide. *J. Flavour Fragrance* **1993**, *8*, 235–245.
- Reverchon, E. Fractional Separation of SFE Extracts from Marjoram Leaves: Mass Transfer and Optimization. *J. Supercrit. Fluids* **1992**, *5*, 256–261.
- Reverchon, E.; Senatore, F. Isolation of Rosemary Oil: Comparison between Hydrodistillation and Supercritical CO<sub>2</sub> Extraction. *J. Flavour Fragrance* **1992**, *7*, 227–230.
- Reverchon, E.; Senatore, F. Supercritical Carbon Dioxide Extraction of Camomile Essential Oil and Its Analysis by Gas Chromatography-Mass Spectrometry. *J. Agric. Food Chem.* **1994**, *42*, 154–158.
- Reverchon, E.; Sesti Osséo, L. Modelling the Supercritical Extraction of Basil Oil. *Proceedings of the 3rd International Symposium on Supercritical Fluids*; Perrut and Brunner, Eds.; Strasbourg, 1994; Vol. 2, pp 189–196.
- Reverchon, E.; Ambruosi, A.; Senatore, F. Isolation of Peppermint Oil Using Supercritical CO<sub>2</sub> Extraction. *J. Flavour Fragrance* **1994a**, *9*, 19–23.
- Reverchon, E.; Sesti Osséo, L.; Gorgoglione, D. Extraction of Basil Oil: Characterization of Products and Process Modelling. *J. Supercrit. Fluids* **1994b**, *7*, 185.
- Richter, M.; Sovová, H.; Kucera, J. Solubility of Monoterpenes in Supercritical CO<sub>2</sub>. *2nd International Symposium on High Pressure Engineering, Erlangen*; 1990; p 305.
- Stahl, E.; Gerard, D. Solubility Behaviour and Fractionation of Essential Oils in Dense Carbon Dioxide. *Perfum. Flavo.* **1985**, *10*, 29–33.
- Stahl, E.; Quirin, K. W.; Gerard, D. *Verdichtete Gase zur Extraction und Raffination*; Springer Verlag: Berlin, 1987.
- Tucker, A. O.; Maciariello, M. J.; Howell, J. T. A. A Preliminary Analysis of Some Lavender and Lavandin Cultivars. *Perfum. Flavor.* **1984**, *9*, 49–60.

Tufeu, R.; Subra, P.; Plateaux, C. Contribution to the Experimental Determination of the Phase Diagrams of Some (Carbon Dioxide + a Terpene) mixture. *J. Chem. Thermodyn.* **1993**, *25*, 1219.

Villermaux, J. Chemical Engineering Approach to Dynamic Modelling of Linear Chromatography. *J. Chromatogr.* **1987**, *406*, 11–26.

Received for review November 23, 1994. Accepted March 17, 1995.<sup>⊗</sup>

JF940663Y

---

<sup>⊗</sup> Abstract published in *Advance ACS Abstracts*, May 1, 1995.