Characteristics of Plants and Plant Extracts from Five Varieties of Basil (*Ocimum basilicum* L.) Grown in Australia

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A horticultural trial on five varieties of basil (*Ocimum basilicum* L.) showed differences in morphological features, growing characteristics, and yields of essential oil produced per unit area of land. Anise basil was the most productive in terms of plant biomass, while Cinnamon basil produced the most essential oil. Gas chromatographic analysis showed considerable differences in the composition of oils among varieties. Seasonal variations had a significant effect on plant growth, essential oil yield, and composition when Reunion and Anise were grown in successive years in the same soil. All varieties were susceptible to cold temperature injury caused by ground frost. Variations in oil composition due to environmental factors appear to be of the same order of magnitude as genetic factors. Data are provided on the yield, composition, and physical appearance of extracts within the same variety obtained by hydrodistillation and with CO_2. The yields were highest using supercritical CO_2 followed by liquid CO_2 and then water. All of the CO_2 extracts were quite similar in chemical composition with respect to volatile components. The hydrodistilled oil contained a larger proportion of lower boiling point hydrocarbons and oxygenated terpenes than the CO_2 extracts. The sensory evaluation of hydrodistilled oil and liquid CO_2 extracts obtained with a one-stage separator under defined conditions indicated that they were quite different products and they would appear to have different end uses.

Keywords: *Australian basil; essential oils; plant extracts; hydrodistillation; sensory evaluation*

INTRODUCTION

Essential oils are volatile substances extracted from odoriferous plants. They are used as flavorings in foods and beverages and as fragrances in pharmaceutical and industrial products. The estimated world production of essential oils in 1984 was about 36 000 tonnes, and of this amount approximately 14 tonnes was from basil (*Ocimum basilicum* L.) (Lawrence, 1985); by 1991 the production had increased to 43 tonnes (Lawrence, 1991).

There are several types of basil oil available on the world market which are obtained from different varieties of sweet basil (Simon *et al*., 1990). They are produced from leaves and flowering tops usually by steam distillation and have been extensively used in food for flavoring confectionary and baked goods; condiments (catsups, tomato pastes, chili sauces, pickles, vinegars); sausages, and meats (Guenther, 1952); and nonalcoholic beverages, ice cream, ices, etc. (Fenaroli, 1975). Basil oil has also found wide application in perfumery, dental, and oral products. In recent years CO_2 extracts of basil have become commercially available, produced in Germany (Flavex Naturextrakte GmbH) and in the United States from imported dried materials (Hunter, 1996).

The essential oil industry in Australia comprises a diverse range of products including eucalyptus, orange, lemon, tea tree and lavender oils and smaller quantities of sweet fennel, parsley herb, and caraway oils (Lawrence, 1985); peppermint and spearmint oils (Deer, 1993); and blackcurrant and boronia absolutes (Hunter, 1996). A range of other species, both native and introduced, are under investigation as potential essential oil crops, particularly in regions of Australia where tobacco has been traditionally grown. We have conducted preliminary studies into the yields, chemical composition, and sensory properties of essential oil from basil (*O. basilicum* L.) grown in northern Victoria (Lachowicz *et al*., 1996), and in this paper we present the results for additional varieties, *viz*. Cinnamon, Dark Opal, and Bush basil, and also report the effect of seasonal variation over two years for the varieties Reunion and Anise.

MATERIALS AND METHODS

Plants Grown at the Ovens Research Station. Seeds of four cultivars of sweet basil (*O. basilicum* L.) were purchased locally from a Canadian supplier (Otto Richter & Sons Ltd, Goodwood, ON). These were Anise, Cinnamon, Dark Opal, and Bush basil. Seeds of Reunion basil were provided by the Ovens Research Station, Agriculture Victoria, from their collection.

Seeds were sown at the beginning of November 1994 into a mixture of pine sawdust, washed sand, horse manure, and mulch (1:1:1:1) and germinated under glass in a controlled environment with the temperature maintained between 15 and 30 °C. Seedlings were planted out on January 13, 1995, into a prepared and fertilized field (5.2% N, 8.7% P, 21.4% K, 0% Mg, and 3.5% S at 0.575 tonne/ha) at a spacing of 0.6 × 0.3 m. The total area planted was approximately 650 m^2.

Plants were harvested manually on April 12, 1995, at midflower stage. They were dried whole in a tobacco kiln using

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Table 1. Weather Conditions in the 1994 and 1995 Growing Seasons at Ovens Research Station, Myrtleford

	monthly rainfall (mm)		mean daily temp (°C)		monthly av lowest temp (°C)		av highest temp (°C)		monthly min temp (°C)		monthly max temp (°C)	
	1994	1995	1994	1995	1994	1995	1994	1995	1994	1995	1994	1995
Jan	45	166	N/A	20.8	11.2	13.6	28.6	28.6	4.0	7.1	35.0	35.0
Feb	183	26	N/A	19.8	14.8	11.8	29.4	28.4	8.0	5.8	35.5	35.8
March	30	0	N/A	15.5	9.4	6.6	25.3	25.5	3.0	0.3	28.5	33.9
April	13	74	N/A	10.3	6.0	4.2	22.3	18.0	2.0	−1.4	29.0	24.3

Table 2. Vegetative Characteristics of Basil Varieties in Two Growing Seasons, 1995 and 1994

	av plant ht at harvest (m)		av plant wt at harvest (kg)		mass of tissue grown per unit area of land[a] (kg m^{-2})		leaf color	
variety	1995	1994	1995	1994	1995	1994	1995	1994
Reunion	0.41	0.45	0.100	0.178	0.560	0.742	green	green
Cinnamon	0.55	–[b]	0.250	–	1.400	–	green	–
Anise	0.61	0.80	0.290	0.499	1.624	2.081	green	green
Dark Opal	0.39	–	0.080	–	0.448	–	dark purplish bronze	–
Bush	0.49	–	0.283	–	1.582	–	light green	–

[a] Calculated from plant spacing and average weight. [b] Not grown in 1994.

forced-air circulation at 28 °C for 8 days. The leaves and stems were separated manually and the leaves used for further analysis. The moisture content of the dried leaves was as follows: 12.1% for Dark Opal; 11.9% for Anise; 12.6% for Cinnamon; 12.5% for Bush; and 11.8% for Reunion basil.

CO_2 Extraction Procedures. Prior to extraction with carbon dioxide (CIG, Melbourne), the dried leaves were ground in a coffee grinder. The particle size distribution of the ground leaves was as follows: <8 and >4 mm, 1.7%; <4 and >2 mm, 10.0%; <2 and >1 mm, 31.7%; <1 and >0.5 mm, 34.0%; <0.5 and >0.29 mm, 9.4%; <0.29 and >0.15 mm, 5.3%; <0.15 mm, 7.9%.

The sample size was 200 g in each case, and extraction was continuous for 1 h. The supercritical fluid extraction pilot plant and other operating parameters are described elsewhere (Lachowicz *et al.*, 1996). We chose conditions on the basis of solubility isotherms of terpenes, oxygenated terpenes, and sesquiterpenes described by Stahl and Gerard (1985), who showed significant differences over the pressure range of 60–120 atm at 40 °C (CO_2 densities from 153 to 723 kg m^{-3}). We used 82 and 150 atm at 40 °C (CO_2 densities of 321 and 784 kg m^{-3}, respectively) and also included nonsupercritical conditions (68 atm at 27 °C; CO_2 density of 693 kg m^{-3}) to cover a similar range of carbon dioxide densities.

CO_2 extracts were quantitatively recovered from the separation unit by washing with hexane (Mallinckrodt, Paris, KY) and dried over anhydrous sodium sulfate (BDH, Kilsyth, Australia). Hexane was evaporated under reduced pressure, and the yield of extract was determined by weighing. CO_2 extracts were stored in airtight containers under nitrogen gas (CIG, Melbourne), in the dark, at 4 °C.

Hydrodistillation Procedures. Hydrodistillation was carried out in an all-glass cohabation distillation apparatus equipped with a 5 L round-bottom flask. Deionized water was used with a sample size of 200 g, and distillation was continued for approximately 1.75 h . Essential oils were dried over anhydrous sodium sulfate and stored in an airtight container under nitrogen, in the dark, at 4 °C.

GC and GC/MS Conditions. Gas chromatography and GC/MS were conducted with a Hewlett-Packard 5890 Series II gas chromatograph and a Hewlett-Packard 5988A mass spectrometer equipped with a fused silica capillary column BP20 (50 m × 0.22 mm) as described previously (Lachowicz *et al*., 1996).

Sensory Evaluation Procedures. A full sensory analysis (Jellinek, 1954) was undertaken on the odor profiles of the essential oils and extracts by a trained evaluator. Samples were absorbed onto paper strips and the first odor impressions recorded. Evaluations were repeated after several hours to eliminate olfactory fatigue.

Weather Records. Temperatures and rainfall were recorded daily by meteorological staff from the Ovens Research Station, and details are given in Table 1. Plants were irrigated when the rainfall was inadequate and they showed any signs of water stress (e.g. wilting).

Table 3. Yield of Essential Oil Obtained by Hydrodistillation of Dried Leaves from Five Varieties of Basil

variety	yield of essential oil (mL kg^{-1})	variety	yield of essential oil (mL kg^{-1})
Reunion	11.0	Dark Opal	9.10
Cinnamon	11.98	Bush	3.44
Anise	6.36		

Table 4. Components (Percent by Gas Chromatography) in Hydrodistilled Oil and CO_2 Extracts Obtained from Reunion Basil

			CO_2		
	retention time (min)	hydro-distilled	27 °C, 68 atm	40 °C, 82 atm	40 °C, 150 atm
α-pinene	7.83	0.3	–[a]	–	–
camphene	8.66	–	–	–	–
β-pinene	9.61	0.3	–	–	–
sabinene	9.92	–	–	–	–
myrcene	10.91	–	–	–	–
α-terpinene	11.50	–	–	–	–
limonene	12.09	0.3	–	–	–
1,8-cineole	12.76	2.4	1.1	1.1	1.3
(*Z*)-*β*-ocimene	13.06	0.2	–	–	–
γ-terpinene	13.53	–	–	–	–
(*E*)-*β*-ocimene	13.63	2.2	0.7	1.0	1.4
p-cymene	14.56	–	–	–	–
fenchone	18.91	–	–	–	–
pentadecane	22.14	–	–	–	–
camphor	23.00	1.0	0.8	0.8	0.7
linalool	23.48	1.5	1.1	1.5	1.5
linalyl acetate	23.88	–	–	–	–
endo-fenchol	24.77	0.2	0.2	–	0.2
hexadecane	25.40	–	–	–	–
β-caryophyllene	26.15	0.4	0.5	0.6	0.6
methylchavicol	27.72	82.6	84.2	84.4	84.5
heptadecane	28.37	–	–	–	–
α-terpineol	28.51	–	–	–	–
borneol	28.69	–	–	–	–
octadecane	31.43	–	–	–	–
geraniol	32.74	–	–	–	–
nonadecane	34.34	–	–	–	–
eicosane	36.97	–	–	–	–
methyl cinnamate	39.78	–	–	–	0.2
heneicosane	39.82	–	–	–	–
eugenol	42.56	–	–	–	–
docosane	42.99	–	–	–	–
tricosane	46.84	–	0.3	0.2	0.2

[a] Not detected.

RESULTS AND DISCUSSION

Characteristics of Five Varieties of Basil Grown in Northeastern Victoria. In this study five varieties of basil (*O. basilicum* L.), Reunion, Cinnamon, Anise, Bush, and Dark Opal, were examined for their vegetative characteristics and also yield and composition of their essential oils. Each possessed characteristic mor-

Table 5. Components (Percent by Gas Chromatography) in Hydrodistilled Oil and CO_2 Extracts Obtained from Cinnamon Basil

		CO_2		
	hydrodistilled	27 °C, 68 atm	40 °C, 82 atm	40 °C, 150 atm
α-pinene	0.2	–[a]	–	–
camphene	–	–	–	–
β-pinene	0.2	–	0.1	–
sabinene	0.1	–	–	–
myrcene	–	–	–	–
α-terpinene	–	–	–	–
limonene	0.2	0.1	0.1	–
1,8-cineole	2.4	1.3	1.3	1.2
(*Z*)-β-ocimene	–	–	–	–
γ-terpinene	0.1	–	–	–
(*E*)-β-ocimene	0.4	0.2	0.3	0.1
p-cymene	0.1	–	0.1	–
fenchone	–	–	–	–
pentadecane	–	–	–	–
camphor	0.5	0.4	0.4	0.3
linalool	27.3	21.1	22.7	19.9
linalyl acetate	0.1	0.1	0.1	0.2
endo-fenchol	–	–	–	–
hexadecane	–	–	–	–
β-caryophyllene	–	–	–	–
methylchavicol	6.8	5.7	5.9	4.5
heptadecane	–	–	–	–
α-terpineol	0.5	0.5	0.6	0.4
borneol	0.3	0.4	0.5	0.4
octadecane	–	0.3	0.1	–
geraniol	–	–	–	–
nonadecane	–	0.2	0.1	–
eicosane	–	0.3	0.1	–
methyl cinnamate	28.11	34.1	34.3	35.6
heneicosane	–	–	–	–
eugenol	3.9	–	–	–
docosane	–	–	–	–
tricosane	–	0.2	–	–

[a] Not detected.

Table 6. Components (Percent by Gas Chromatography) in Hydrodistilled Oil and CO_2 Extracts Obtained from Anise Basil

		CO_2		
	hydrodistilled	27 °C, 68 atm	40 °C, 82 atm	40 °C, 150 atm
α-pinene	0.2	–[a]	–	–
camphene	0.2	–	–	0.1
β-pinene	–	–	–	–
sabinene	–	–	–	–
myrcene	0.2	–	–	0.1
α-terpinene	–	–	–	–
limonene	0.3	0.1	0.2	0.2
1,8-cineole	1.3	0.6	0.6	0.8
(*Z*)-β-ocimene	–	–	–	–
γ-terpinene	–	–	–	–
(*E*)-β-ocimene	0.4	0.1	0.1	0.2
p-cymene	–	–	–	–
fenchone	0.2	–	0.1	0.1
pentadecane	–	–	–	0.2
camphor	2.5	1.8	1.8	1.8
linalool	27.8	23.7	26.3	24.2
linalyl acetate	–	0.1	0.2	0.2
endo-fenchol	–	–	–	–
hexadecane	–	0.2	0.2	0.5
β-caryophyllene	–	–	–	–
methylchavicol	40.9	31.0	33.0	32.6
heptadecane	–	0.3	–	1.3
α-terpineol	0.4	0.4	0.6	–
borneol	0.7	0.7	0.7	0.7
octadecane	0.2	0.4	0.3	1.1
geraniol	–	–	–	–
nonadecane	–	0.3	0.3	0.9
eicosane	–	0.3	0.2	0.5
methyl cinnamate	3.4	6.0	4.7	3.9
heneicosane	–	–	–	–
eugenol	–	–	–	–
docosane	–	–	–	–
tricosane	–	0.5	0.4	0.3

[a] Not detected.

phological features and exhibited differences in average plant weight and the amount of biomass produced per square meter of land (Table 2). The density of biomass ranged from 0.448 to 1.624 kg m^{-2} with the most productive variety being Anise. However, the yield of this variety was significantly lower than in the previous growing season. This seasonal difference was also observed with Reunion, but the effect was less marked and was attributed mainly to the different prevailing air temperatures in 1995 compared to 1994. In general, the 1995 growing season was cooler, as can be seen from lower mean minimum temperature throughout the growing season and the lower mean maximum in the period toward the end of the season. We observed significant cold temperature injury to plants grown in the 1995 season when they experienced nights with a low of 0.3 °C in March and −1.9 °C in early April (Table 1). Basil is a frost sensitive plant and susceptible to cold temperature injury (Simon *et al.*, 1984). The frosts in March and April of 1995 caused severe damage to the leaves; many plants were burnt brown, while a few had no leaves at all, which explains the decrease of biomass compared to the 1994 season (Table 2). Unpredictable early frost in the Ovens Valley of Victoria where these plants were grown is likely to limit the suitability of basil in this region.

Yields and Composition of Hydrodistilled Oils. Dried leaves of five varieties of basil were subjected to hydrodistillation. The products were liquid oils which ranged from very pale yellow, almost colorless through light yellow, to yellow and the yields varied from 3.44 to 11.98 mL kg^{-1} depending on variety (Table 3). Cinnamon basil contained the highest concentration of essential oil in the dried leaves. The quantity of essential oil obtained per unit area of land ranged from 0.9 to 3.8 mL m^{-2} with the highest yields given by Cinnamon basil.

As was the case with biomass, we found that there were yearly variations in the productivity of the land with respect to essential oil for the two varieties Reunion and Anise that we grew in successive seasons. Plants grown in 1995 produced less oil (Reunion, 1.4 mL m^{-2}; Anise, 2.3 mL m^{-2}) compared with the 1994 growing season (Reunion, 2.6 mL m^{-2}; Anise, 3.0 mL m^{-2}) for a given area of land. This was attributed to different weather conditions as previously described. Interestingly, although there was also a decrease in the yield of oil obtained from dried leaves of Anise in the 1995 season, this was not the case for Reunion.

The chromatographic analysis of hydrodistilled oils showed significant differences in their chemical composition (Tables 4−8). The major components were linalool, methylchavicol, and methyl cinnamate. The samples were of four types, *viz*. oils comprised of mostly linalool with little methylchavicol (Bush, Opal); oil with >80% of methylchavicol together with very small amounts of linalool (Reunion); oil that contained approximately equal proportions of linalool and methyl cinnamate together with small amounts of methylchavicol (Cinnamon); and oil with approximately equal proportions of linalool and methylchavicol (Anise).

The major constituents, linalool and methylchavicol, in Reunion oil from plants grown in 1995 and 1994 were present in similar proportions (Table 9), indicating a resistance to weather-induced changes in composition. On the other hand, Anise oil obtained from the 1995 crop contained almost the reverse proportion of methylchavicol and linalool (27.8% linalool and 40.9% me-

Table 7. Components (Percent by Gas Chromatography) in Hydrodistilled Oil and CO_2 Extracts Obtained from Bush Basil

		CO_2		
	hydrodistilled	27 °C, 68 atm	40 °C, 82 atm	40 °C, 150 atm
α-pinene	0.6	0.1	0.1	0.1
camphene	0.2	–[a]	–	–
β-pinene	0.5	0.2	0.1	0.1
sabinene	0.2	–	–	–
myrcene	0.4	0.2	0.1	0.1
α-terpinene	–	–	–	–
limonene	0.5	0.3	0.2	0.3
1,8-cineole	3.0	1.5	1.0	1.4
(*Z*)-β-ocimene	–	–	–	–
γ-terpinene	0.2	–	–	–
(*E*)-β-ocimene	–	–	0.3	–
p-cymene	0.5	0.2	0.1	0.2
fenchone	–	–	–	–
pentadecane	–	0.1	0.2	0.1
camphor	–	0.8	0.7	0.7
linalool	52.0	40.2	32.5	41.1
linalyl acetate	–	0.2	0.2	0.3
endo-fenchol	–	0.4	–	0.5
hexadecane	–	–	0.6	–
β-caryophyllene	–	–	–	–
methylchavicol	–	1.0	15.4	0.6
heptadecane	0.2	0.6	1.6	0.4
α-terpineol	0.7	–	–	–
borneol	–	0.5	0.1	0.5
octadecane	0.3	0.8	1.3	0.5
geraniol	0.9	1.2	0.9	1.1
nonadecane	–	0.6	1.0	0.5
eicosane	–	0.6	0.8	0.4
methyl cinnamate	–	0.5	–	–
heneicosane	–	–	–	0.1
eugenol	–	–	–	–
docosane	–	–	–	–
tricosane	–	0.3	0.5	0.2

[a] Not detected.

thylchavicol) compared with the 1994 crop (43.7% linalool and 27.3% methylchavicol), and the content of methyl cinnamate decreased from 11.2% in 1994 to 3.4% in 1995 (Table 9). These preliminary data, if confirmed in longer studies, indicate that some varieties of basil are more prone to exhibit season alterations in oil composition than others and would be of concern to commercial producers when consistency of product is important.

The genus *Ocimum* exibits a wide variety of morphological characteristics, and 15 distinct varieties or subvarieties of *O. basilicum* L. have been identified (Lawrence *et al.*, 1980). The oil composition seems to fall into four main types which predominate in one or two components. However, even with carefully selected strains of basil there appears to be an approximately 10% variation in the composition of the principal oil components such as methylchavicol, linalool, methyleugenol, and geraniol (Lawrence, 1988). The results presented here, with Anise for example, would seem to indicate that environmental factors can be more important than genetic factors in controlling essential oil composition, although of course they are not independent of each other.

Yields and Composition of Extracts Obtained Using Liquid CO_2 and Supercritical CO_2. We did not attempt to control the extractive conditions to optimize the yields and composition of our extracts because we were mainly interested in the effect of plant variety and growing season on volatile terpenoid constituents. We were also interested in the effect of carbon dioxide solvent power and employed three densities in the expectation that our extracts would contain differing proportions of monoterpenes, which have the highest solubility, followed by oxygenated terpenes, sesquiterpenes, waxes, etc. The batch extractions were conducted for a relatively short time (1 h), and under these conditions the extraction could not have been

Table 8. Components (Percent by Gas Chromatography) in Hydrodistilled Oil and CO_2 Extracts Obtained from Opal Basil

		CO_2		
	hydrodistilled	27 °C, 68 atm	40 °C, 82 atm	40 °C, 150 atm
α-pinene	0.8	0.2	0.1	0.2
camphene	0.1	–[a]	–	–
β-pinene	0.9	0.3	0.3	0.3
sabinene	0.4	0.1	0.2	0.2
myrcene	–	0.2	0.2	0.2
α-terpinene	–	–	–	–
limonene	0.4	0.2	0.2	0.2
1,8-cineole	5.5	2.8	2.7	3.1
(*Z*)-β-ocimene	–	–		
γ-terpinene	–	–	–	–
(*E*)-β-ocimene	–	–	–	–
p-cymene	–	–	–	–
fenchone	0.3	–	0.1	0.1
pentadecane	–	–	–	–
camphor	–	1.0	–	–
linalool	57.4	45.9	52.7	46.9
linalyl acetate	0.1	–	0.1	0.1
endo-fenchol	–	–	–	–
hexadecane	–	–	–	–
β-caryophyllene	2.5	2.6	2.5	2.6
methylchavicol	–	2.6	2.4	2.9
heptadecane	–	–	–	–
α-terpineol	0.8	0.6	0.8	0.8
borneol	0.4	0.5	0.4	0.4
octadecane	–	0.3	0.3	0.3
geraniol	0.7	0.9	0.9	0.9
nonadecane	–	–	0.3	–
eicosane	–	0.4	0.2	0.2
methyl cinnamate	0.7	2.3	2.2	2.3
heneicosane	–	–	–	–
eugenol	–	–	–	–
docosane	–	–	–	–
tricosane	–	0.3	–	0.3

[a] Not detected.

Table 9. Chemical Composition (Percent by Gas Chromatography) of Oil Extracted from Two Varieties of Basil Grown in Successive Years

	Reunion		Anise	
	1994	1995	1994	1995
α-pinene	0.3	0.3	0.2	0.2
camphene	0.1	–[a]	0.2	0.2
β-pinene	0.3	0.3	0.2	–
myrcene	0.1	–	0.4	0.2
α-terpinene	0.04	–	0.03	–
limonene	0.3	0.3	0.3	0.3
1,8-cineole	2.6	2.4	1.6	1.3
(*Z*)-β-ocimene	–	0.2	–	–
γ-terpinene	0.1	–	0.03	–
(*E*)-β-ocimene	–	2.2	–	0.4
p-cymene	–	–	–	–
fenchone	–	–	0.1	0.2
camphor	0.6	1.0	1.2	2.5
linalool	1.5	1.5	43.7	27.8
linalyl acetate	–	–	–	–
endo-fenchol	–	0.2	–	–
β-caryophyllene	0.5	0.4	0.2	–
methylchavicol	87.2	82.6	27.3	40.9
α-terpineol	–	–	0.4	0.4
borneol	–	–	0.4	0.7
citral	–	–	–	0.4
citronellol	–	–	–	0.1
octadecane	–	–	–	0.2
geraniol	–	–	–	–
methyl cinnamate	–	–	11.2	3.4
eugenol	–	–	–	–

[a] Not detected.

Table 10. Yields of Oil (Percent w/w) Obtained by Hydrodistillation and Total Extracts Obtained by Liquid CO_2 and Supercritical CO_2

	extraction conditions			
		liquid CO_2	supercritical CO_2	
variety	hydrodistillation	27 °C, 68 atm, 1 h	40 °C, 82 atm, 1 h	40 °C, 150 atm, 1 h
Reunion	1.1	0.75	0.37	1.16
Cinnamon	1.2	1.54	0.79	1.90
Anise	0.6	0.63	0.29	1.23
Dark Opal	0.9	0.86	0.44	1.27
Bush	0.3	0.42	0.27	0.63

exhaustive. The solvent to sample ratio (by mass) we used was only 12.5:1, whereas others (Reverchon, 1992; Reverchon *et al.*, 1993) using similar herbaceous matrices have shown that asymptotic yields are generally only obtained with higher ratios (>25:1) and that higher absolute yields are obtained when the plant material is comminuted to a smaller average particle size than that which we employed.

Dried leaves of all varieties were also extracted with CO_2 under different temperature and pressure conditions, resulting in products which were of two types—liquids (supercritical CO_2 extracts obtained at 82 atm) and semisolids (liquid CO_2 extracts and supercritical CO_2 extracts obtained at 150 atm). The liquid CO_2 extracts were light or dark yellow (orange), while the supercritical CO_2 products obtained at 82 atm were yellow and those obtained at 150 atm were brownish yellow or yellow-brown. The yields were highest using supercritical CO_2 at 150 atm (Table 10), and this may be due to additional coextracted lipids and waxes (Reverchon, 1992; Reverchon *et al.*, 1993).

Tables 4−8 show the composition of extracts from five varieties. The GC peak areas of the identified components in the various extracts range from just <50% to >90% of the total areas because some of the extracts contained a very large number of small peaks of <0.1% relative area and these have not been included in the table.

The method of extraction had a significant effect on the chemical composition of the product. The hydrodistilled oil contained a larger proportion of the lower boiling point hydrocarbons such as α-pinene, camphene, *β*-pinene, sabinene, myrcene, α-terpinene, limonene, α-ocimene, *γ*-terpinene, *β*-ocimene, *p*-cymene, and oxygenated terpenes including 1,8-cineole, fenchone, and camphor. These results confirmed our earlier observations (Lachowicz *et al.*, 1996) and are consistent with results published by Reverchon and Senatore (1992), who reported that rosemary oil obtained by hydrodistillation and supercritical CO_2 extraction yielded substantially the same main compounds, although they were present in different proportions. In comparison with the supercritical CO_2 extracts, the hydrodistilled oil contained a higher proportion of monoterpene hydrocarbons and lower proportion of oxygenated monoterpenes. We did not measure the higher molecular weight compounds present in our extracts because we were mainly interested in the volatiles. Reverchon (1993) has shown that by using two separators, extracts can be separated into material consisting mainly of high molecular weight *n*-paraffins and a low-viscosity liquid that can be considered an essential oil. In our extracts both groups of compounds would be present in mixtures of varying proportions; however, it is possible to achieve a degree of selectivity by changing the density of carbon dioxide.

Comparison of liquid CO_2 extracts with supercritical CO_2 extracts obtained under different temperature and pressure conditions showed that generally within a variety the volatiles were quite similar in composition. On the basis of a chemical analyis of these compounds there appears to be no major advantage of using supercritical carbon dioxide over liquid carbon dioxide. Taking into account production costs, which are lower in the case of extraction with liquid CO_2, further efforts were made to characterize profiles and compare sensory quality of liquid CO_2 products with hydrodistilled oil.

Sensory Quality of Hydrodistilled and CO_2 Basil Extracts. Results from the sensory evaluation of the hydrodistilled oils and liquid CO_2 extracts obtained from the five varieties of basil grown in 1995 are summarized in Table 11. Each variety was quite different in the predominating note, subsidiary note, and backnote. Oil from Cinnamon, Dark Opal, and Bush basil had a hay- or vegetable-like note, which was considered to render them inferior to other basil oils available commercially. The sample of Reunion oil did not have the sweet and spicy anisic character of those usually displayed in “exotic” types. It was rather flat in profile and lacked richness. Also, the oil from Anise basil was considered not sweet and lacking any sharpness in character compared to its commercial counterpart. It was con-

Table 11. Sensory Evaluation of Products Obtained by Hydrodistillation (HD) and Liquid CO_2 (LCO_2) Extraction from Five Varieties of Basil

source of oil/ extract	sensory evaluation
1995 Reunion HD oil	cool herbaceous predominating note with a mildly sweet anise subsidiary note and very slight woody backnote
1995 Cinnamon HD oil	cool slightly grassy herbaceous predominating note with a green and fruity subsidiary note and a slightly earthy wood backnote
1995 Anise HD oil	warm herbaceous predominating note with a cool diffusive anise subsidiary note and a somewhat spicy, woody backnote
1995 Dark Opal HD oil	sweetish vegetable-like herbaceous predominating note with a slight green subsidiary note and very slight camphorous woody backnote
1995 Bush HD oil	warm smooth hay-like herbaceous predominating note with a woody subsidiary note and earthy, anise, camphor backnote
1995 Reunion LCO_2	sweet anise predominating note with a herbaceous and floral subsidiary note and a very slightly woody backnote
1995 Cinnamon LCO_2	green herbaceous predominating note with slightly peppery anise subsidiary note and a woody backnote
1995 Anise LCO_2	slightly hay-like herbaceous predominating note with a cool anise subsidiary note with a slight woody backnote
1995 Dark Opal LCO_2	smooth cool anise predominating note with a herbaceous subsidiary note and a green, very slightly woody backnote
1995 Bush LCO_2	warm herbaceous predominating note with a woody subsidiary note and a green, very slightly anise backnote
1994 Reunion LCO_2	sharply sweet anise predominating note with a herbaceous and floral subsidiary note and very slight woody backnote
1994 Anise LCO_2	grassy hay-like herbaceous and cool anise predominating note with a very slight floral subsidiary note and a woody and slightly camphorous backnote

cluded that none of the essential oils obtained from 1995 crop would have significant commercial value.

Among the liquid CO_2 extracts, that obtained from Opal basil was regarded to have the best sensory quality. However, the olfactory profile was inferior to that of an Egyptian CO_2 extract available commercially (CO_2 - Extract Basil - type no. 2007.010 produced by Flavex Naturextrakte GmbH), which is obtained by a two-stage separation process and considered to be a true essential oil since it is free of waxes. We did not fractionate our extracts, and so they would contain waxes that should not, however, affect their olfactory profile. Therefore, the relatively inferior quality of our Opal extract could be due to a different balance of terpenoid consitituents or to the presence of other compounds which are absent from the commercial product. The presence of higher molecular weight components in the Opal extract could also be deduced from the fact that at room temperature it was viscous and was not completely liquid.

The hydrodistilled oils and liquid CO_2 extracts possess quite different sensory properties (Table 11). A comparison was made of the sensory quality of essential oil from Reunion and Anise basil grown in 1995 and in 1994. The 1995 Reunion oil was more balanced than that from 1994. Although the predominating notes of the 1995 oil were herbaceous, there was more balance in the anisic subsidiary notes, unlike the 1994 oil which was floral and herbaceous.

The 1995 Anise essential oil had lost the fruity backnotes that the 1994 Anise displayed. The complex of the 1995 Anise was more woody than the 1994 Anise. An anise subsidiary note was detectable in the 1995 oil, which was absent in the 1994 Anise oil. However, the predominating note of the 1995 Anise was still herbaceous.

Thus, the poor growing season in 1995 correlated with an improvement in the sensory properties of the Reunion oil compared with the previous season, whereas the Anise oil seemed to be adversely affected. However, it was concluded that none of these oils possessed the sweetness, balance, and richness in profile of commercial oils produced in traditionally producing countries.

A comparison of liquid CO_2 extracts from Reunion and Anise basil grown in the two years showed that the 1995 extract had much better quality than the 1994 extracts. The 1995 extracts had smoother profiles than their corresponding 1994 extracts and also had a better appearance. Sensory profiles of extracts obtained in 1995 were closer to those of commercially available products; however, they would still not be considered commercially acceptable. Sensory quality of analyzed liquid CO_2 extracts was superior to that obtained by hydrodistillation.

In general, with some exceptions the extraction of aromatic materials from plant material using liquid carbon dioxide results in essential oils which more closely resemble the aroma and taste of the botanical starting material than the steam-distilled or hydrodistilled equivalent. Hydro- and steam-distilled essential oils also differ from liquid carbon dioxide extracts in terms of market potential due to the high production costs of the latter. Liquid carbon dioxide extracts are not considered direct substitutes to steam-distilled or hydrodistilled essential oils; rather, they are specialty products (Hunter, 1996) employed in the creations of flavors for food and cosmetic products in cases which require an especially fine sensory profile.

This study showed that yield, chemical composition, and sensory quality of basil oil depends on several factors including variety, climatic conditions, and method of extraction and that the variation in these factors is not always predictable.

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