

Characteristics of Essential Oil from Basil (*Ocimum basilicum* L.) Grown in Australia

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A commercial sample of essential oil from basil (*Ocimum basilicum* L.) grown in Australia was similar in chemical composition to oil from traditional producing countries, but the sensory properties were considered to be inferior. A small-scale horticultural trial showed that oil yields per unit area of land were similar for three varieties of basil but the composition of the oils differed significantly. Data are provided that show differences in oil composition when basil is dried and also when leaves are extracted by hydrodistillation or supercritical or liquid CO₂. The CO₂ extracts obtained under a limited range of conditions were considered to be of inferior sensory quality when compared with hydrodistilled oil.

Keywords: *Basil; essential oil; critical extraction; carbon dioxide; sensory quality*

INTRODUCTION

Basil essential oil has been used for many years to flavor foods and certain dental and oral products as well as in fragrances (Guenther, 1952). There are several types of basil oil traded commercially. Extracted from different varieties of sweet basil (*Ocimum basilicum* L.), these are known as European (French or sweet basil); Egyptian; Reunion (Comoro); Bulgarian; and Java basil oil (Simon et al., 1990). The oils are conventionally extracted by steam distillation from leaves and flowering tops. Alternatively, the plant material can be extracted using carbon dioxide, under either liquid or supercritical conditions. This technique has been known for some 50 years but has only recently been commercially developed on a limited scale (Moyler and Heath, 1988).

In some regions of Australia there is growing interest in producing essential oils, particularly as a replacement crop in areas where tobacco has been traditionally grown. Australia produces a wide range of essential oils from both native and introduced species including eucalyptus oil, orange oil, lemon oil, tea tree oil, lavender oil, and smaller quantities of sweet fennel, parsley herb, and caraway oils (Lawrence, 1985). In recent years, oil production from peppermint and spearmint has been established in Tasmania and northeast Victoria (Deer, 1993). Other crops successfully grown for commercial oil production include black currant, boronia, and dill (Menary, 1993). As part of a broader investigation into essential oil production this paper reports some preliminary studies on basil.

MATERIALS AND METHODS

Essential Oil. Four commercial samples of basil essential oil were purchased (Australian Botanical Products Pty. Ltd.).

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These were produced from leaves and flowering tops by steam distillation in India, France, Australia, and the Seychelles. One further oil sample, not yet commercially available, was obtained by steam distillation of freshly harvested whole plants grown in a trial plantation at the Ovens Research Station (ORS) in Australia.

Dried Basil. A commercial sample of dried and rubbed basil imported from Egypt (Waters Trading Pty. Ltd.) was used as received (10.0% v/w moisture). This material was used in the studies with carbon dioxide, and its particle size distribution was as follows: <4 and >2 mm, 10.9%; <2 and >1 mm, 68.8%; <1 and >0.5 mm, 18.3%; <0.5 and >0.29 mm, 1.6%; <0.29 mm, 0.4%.

Fresh Plants. Seeds of three varieties of sweet basil (*O. basilicum* L.) were obtained from a local nursery and an overseas private grower. These were Reunion, Large Lettuce, and Anise. Seeds were sown in containers on November 1, 1993, and kept in a glasshouse (temperature controlled within the range 10–25 °C). Seedlings were planted out on January 5, 1994, into a prepared and fertilized field [lime, 4.36 tonne/ha; fertilizer (NPK 20:65:15), 0.135 tonne/ha] at a spacing of 0.4 × 0.6 m. The total area planted was approximately 400 m². After planting, nitrogen was applied (60 kg/ha of NH₄-NO₃). Plants were harvested manually on April 11, 1994, at midflower stage. The bulk of the plants were dried, whole, in a tobacco kiln using forced air circulation at a temperature of 28 °C for 8 days. At the end of this time the moisture content of Reunion was 11.8%, of Large Lettuce was 12.5%, and of Anise was 12.0%. Also, samples of freshly harvested plants were stored in a freezer at -18 °C. We assume that the extracts obtained from this frozen material will be identical with extracts from fresh plants.

The wooden stem-base of the dried or frozen plant was removed, and the leaves and stems were coarsely chopped before being extracted by hydrodistillation.

CO₂ Extraction Procedures. A schematic diagram of the Distillers MG (MG Gas Products Limited, England) supercritical fluid extraction pilot plant is shown in Figure 1. Food grade pure liquid carbon dioxide was provided from a steel cylinder. After cooling (C1), the CO₂ was pressurized by a high-pressure piston pump (P2) to a selected pressure of 68, 102, or 306 atm, which was regulated and checked by a variable pressure indicator controller (PIC2). The pressurized carbon dioxide passed through a heater (H1) to adjust the temperature to 5, 27, or 40 °C and then was allowed to flow up through the vertically mounted 1 L extractor (E1) with a water jacket to maintain the set temperature. The extractor

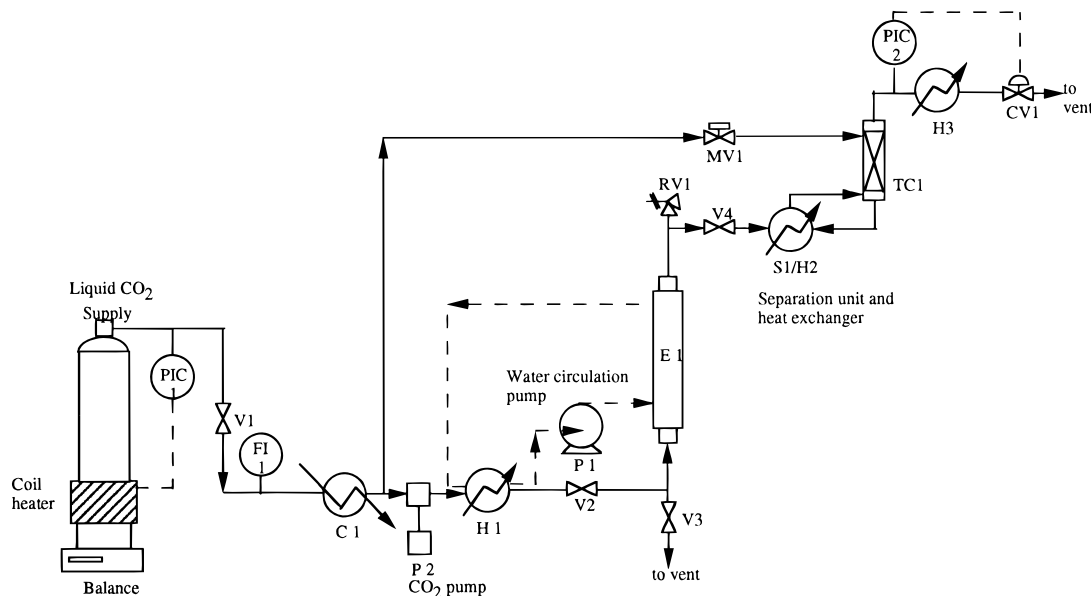


Figure 1. Diagram of supercritical fluid extraction pilot plant.

Table 1. Major Components (Percentage by Area of GC Analysis) in Five Samples of Basil (*O. basilicum*) Essential Oil

major component	sample description and country of origin				
	commercially available				locally produced Reunion basil essential oil (Australia)
	basil essential oil (India)	basil sweet linalool (France)	basil essential oil (Australia)	basil methylchavicol comores (Seychelles)	
linalool	14.2	55.3	34.3	27.7	3.4
methylchavicol	77.5	10.9	34.7	40.2	75.7

was loaded with 150 g of dried, crushed basil leaves in each case. The extract-laden carbon dioxide from the extractor passed through a separation vessel with a glass window (S1/H2), where supercritical or high-pressure liquid CO₂ was depressurized and vented through a packed tailing column (TC1) and vaporizer (H3), leaving the extracted oil in the separation vessel. The CO₂ flow rate was manually adjusted to average 2.5 kg/h. Under the above conditions, extractions were continued for 2 h.

Hydrodistillation Procedures. Hydrodistillation was carried out in a distillation apparatus equipped with a 5 L round-bottom flask. The sample size was 500 or 200 g for frozen or dried material, respectively. The distillation period was approximately 2 h.

Extract Treatment. Hydrodistilled essential oils were dried over anhydrous sodium sulfate and stored in an airtight container under an atmosphere of nitrogen gas, in the dark at 4 °C. CO₂ extracts were stored in the same way.

GC and GC-MS Conditions. Gas chromatography was conducted on a Varian 3700 gas chromatograph equipped with a fused silica capillary column BP20 (50 m × 0.22 mm).

Prior to injection, the essential oil and CO₂ extracts were diluted by the addition of 2 volumes of hexane. This was necessary to dissolve the semisolid material obtained when CO₂ was used as solvent. Under the chromatographic conditions we used, only the volatile terpenes and phenols were measured. The long-chain hydrocarbons associated with the waxy fraction were not analyzed because we are interested, at this stage, in the sensory properties of the oil, to which the semisolid materials obtained by CO₂ extraction would not be expected to make a major contribution. Conditions were as follows: sample size, 0.1 μL; initial column temperature, 60 °C, held for 2 min followed by heating at 4 °C min⁻¹ to 200 °C, held at this temperature for a further 20 min; injector temperature, 240 °C, split ratio, about 1:100; FID detector temperature, 240 °C; carrier gas, helium.

GC-MS data were obtained using a gas chromatograph (Hewlett-Packard 5890 Series II) coupled to a mass spectrometer (Hewlett-Packard Model 5988A) fitted with a combined

EI/CI source. Samples of essential oil and extract (0.2 μL) were injected on the above column using a 1:20 split ratio. Data acquisition and reprocessing were performed by a Hewlett-Packard RTE-A data system. In electron impact mode, the electron energy was set at 70 eV for a trap current of 300 mA. In positive chemical ionization mode, methane was used as reagent gas at a housing pressure of 2 × 10⁻⁴ Torr, which relates to a source pressure of 1.1 Torr. The electron energy was set at 200 eV for a trap current of 300 mA. In both ionization modes the source temperature was 200 °C and the interface line temperature was 250 °C.

Identification of components was based on a comparison of the retention times and mass spectra compared with those of pure compounds. The proportions of individual components in the essential oils were obtained by integration of the uncorrected area percent of the detector signal. In this paper we report only the principal components of the oil (>0.1%), although a more comprehensive listing of basil oil components has been published by Vernin and Metzger (1984).

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.

RESULTS AND DISCUSSION

Comparison of Basil Essential Oil Produced in Australia, Europe, India, and the Indian Ocean.

As expected, the major components of the four commercial and the locally grown basil essential oils were linalool and methylchavicol (Lawrence, 1989). The samples showed considerable variation in the content of linalool and methylchavicol (Table 1) and were of two main types: those that contained more than 75% methylchavicol together with small amounts of linalool and those that contained less than 50% methylchavicol

Table 2. Vegetative Characteristics of Basil Varieties

variety	av plant ht at harvest (m)	av plant wt at harvest (kg)	mass of tissue grown per unit area ^a (kg m ⁻²)	leaf color
Reunion	0.45	0.178	0.742	green
Large Lettuce	0.67	0.638	2.660	light green
Anise	0.80	0.499	2.081	green

^a Calculated from plant spacing and average plant weight.

Table 3. Oil Yields of Basil Varieties before and after Drying

variety	yield of essential oil in frozen plants (mL kg ⁻¹)	yield of essential oil from frozen plants per unit area (mL m ⁻²)	yield of essential oil in dried plants (mL kg ⁻¹)
Reunion	3.5	2.6	9.0
Large Lettuce	1.1	2.9	5.2
Anise	1.4	3.0	5.0

together with large amounts of linalool. One sample had less than 11% methylchavicol and 55% linalool. The two Australian samples showed significant variation in chemical composition: one resembled the sample from India, and the other resembled the sample from the Seychelles.

The sensory qualities of basil oil are related to its chemical composition, and Arctander (1960) has identified four main types: 1, sweet basil oil, linalool type; 2, Reunion basil oil, camphor-estragole type; 3, methyl cinnamate type; 4, phenolic type. A full sensory evaluation was carried out on the five basil oils, and they were also classified according to the types described by Arctander (1960). The Indian sample had a predominantly sweet, spicy anisic character with a herbaceous subsidiary note and would be classified as Reunion "exotic" type. The French oil had a predominating sweet spicy, herbaceous note, with a slightly floral and mild anisic subsidiary note with a green and woody back note of the sweet basil type. The Australian oil was predominantly herbaceous and spicy with a slightly camphoraceous subsidiary note and a mild anisic and floral back note. Although the levels of linalool and methylchavicol are balanced in the oil, it did not have the

smoothness and sharpness of the French oil. Therefore, in sensory terms the Australian oil is probably closer to the Reunion type than the sweet type. The Seychelles oil is also of the Reunion type with a predominating sweet anisic, herbaceous note and a slightly camphoraceous back note. The locally produced Reunion type had an initial sweet floral predominating note (reminiscent of lavender) and a herbaceous subsidiary note with an anisic back note. This indicates that although Australian-grown plants can produce oils with a range of chemical compositions resembling those grown in traditionally producing countries, their olfactory qualities are inferior. The reasons for this may be related to growing conditions or methods of extraction.

Yields and Composition of Essential Oil from Basil Grown in Australia. Three varieties of sweet basil (*O. basilicum* L.), Reunion, Large Lettuce, and Anise, were grown in trial plots as described previously and examined for their average fresh plant weight, oil content, and chemical composition. The varieties possessed differences in morphological features and growing characteristics (Table 2).

The amount of essential oil that can be produced from a given area of land is shown in Table 3. Although Large Lettuce produces much more vigorous and larger plants than the other two varieties, the yields of oil from the same area of land were similar.

The oils were extracted from both frozen and dried plants by hydrodistillation and analyzed by GC and GC-MS. The major chemical components in these oils are given in Table 4.

The essential oils hydrodistilled from the frozen plants showed considerable differences in composition. That from *O. basilicum* L. Reunion contained methylchavicol as the main constituent (87%) together with a little linalool (1%). This is similar to the composition of the Comoro type of oil (Lawrence, 1988; Lawrence et al., 1980; Vernin, 1984), which is also known as Reunion type oil (Simon et al., 1990).

Methylchavicol and linalool were the also the principal components in the other two samples but were present in the reverse proportions. The oil from *O. basilicum* L. Large Lettuce contained 46% linalool and 25% methylchavicol, while the oil from *O. basilicum* L.

Table 4. Principal Components (Percent by GC) in Essential Oil from Three Varieties of Basil (*O. basilicum* L.)

component (retention time in min)	frozen			dried		
	Reunion	Large Lettuce	Anise	Reunion	Large Lettuce	Anise
α-pinene (8.18)	0.2	0.4	0.2	0.3	0.4	0.2
camphene (9.13)	0.1	0.1	0.2	0.1	0.1	0.2
β-pinene (10.11)	0.2	0.7	0.2	0.3	0.7	0.2
myrcene (11.42)	0.1	2.0	1.4	0.1	0.6	0.4
α-terpinene (12.03)	0.04	0.2	0.1	0.04	0.05	0.03
limonene (12.59)	0.3	0.9	0.7	0.3	0.3	0.3
1,8-cineole (12.92)	2.0	8.0	1.8	2.6	6.2	1.6
γ-terpinene (14.04)	0.1	0.2	0.1	0.1	0.1	0.03
p-cymene (14.92)	— ^a	0.1	0.05	—	0.1	—
fenchone (19.41)	—	—	0.1	—	—	0.1
camphor (23.63)	0.5	0.4	1.2	0.6	0.5	1.2
linalool (24.01)	1.1	46.2	30.4	1.5	65.4	43.7
β-caryophyllene (26.15)	0.4	0.6	0.4	0.5	0.5	0.2
methylchavicol (28.45)	86.6	25.4	48.6	87.2	13.5	27.3
α-terpineol (29.13)	—	0.8	0.4	—	0.6	0.4
borneol (29.38)	—	0.2	0.5	—	0.1	0.4
citral (30.41)	0.1	0.3	0.6	—	0.2	0.4
citronellol (31.00)	—	0.2	0.1	—	0.07	0.1
geraniol (33.51)	—	0.1	—	—	0.1	—
methyl cinnamate (40.88)	—	—	2.0	—	—	11.2
eugenol (43.77)	—	2.1	—	—	—	—

^a Components not detected.

Table 5. Sensory Evaluation of Basil Oils Extracted from Frozen (Fresh) and Dried Plants

source of oil	sensory evaluation
dried Reunion	a somewhat sweet, fresh herbaceous, and mild anisic note with a slightly floral and green subsidiary note; there was a slight camphoraceous and grassy back note; the anisic note became more diffusive after a few minutes on the smelling strip, as did the predominance of the grassy note
frozen Reunion	a sharp, if not somewhat dry, anisic note; the subsidiary notes were herbaceous with a slightly sweet camphoraceous floral back note
dried Large Lettuce	a sharp diffusive clean grassy herbaceous note, with a fruity anisic subsidiary note and a very slightly camphoraceous back note
frozen Large Lettuce	a clean vegetable-type note with a cool herbaceous menthol-like subsidiary note; a slight green and grassy back note
dried Anise	sharp herbaceous and mildly spicy predominating note, with a herbaceous subsidiary note; back notes were slightly fruity
frozen Anise	a grassy herbaceous predominating note with anisic subsidiary notes and slightly balsamic, fruity, and camphoraceous back notes

Table 6. Yields of Oil Obtained by Hydrodistillation and Total Extracts Obtained by Liquid CO₂ and Supercritical CO₂

extraction conditions	yield of extract (% w/w)
hydrodistillation for 2h	0.38
liquid CO ₂ at 5 °C and 68 atm extracted for 2 h	0.22
liquid CO ₂ at 27 °C and 68 atm extracted for 2 h	0.32
supercritical CO ₂ at 40 °C and 102 atm extracted for 2 h	0.51
supercritical CO ₂ at 40 °C and 306 atm extracted for 2 h	0.97

Anise contained 30% linalool and 49% methylchavicol, a result which is similar to that reported by Simon et al. (1990; 30% linalool and 47% methylchavicol). Comparative data for essential oil from Large Lettuce basil were not available to us.

The oils obtained from the dried plants were broadly similar in composition. Drying the plants seemed to have a significant effect on the composition of the oil. In the case of Anise the relative proportions of methylchavicol and linalool were almost reversed (frozen, 49% methylchavicol and 30% linalool; dried, 27% methylchavicol and 44% linalool). Methyl cinnamate concentration increased from 2% to 11% on drying. The reasons for such differences are not clear but cannot be

due to a loss of the more volatile compounds during drying. In Large Lettuce the same trend was observed but to a much smaller degree in that the oil from the dried plants contained less methylchavicol and more linalool. Relative changes in Reunion were small in comparison with the other two varieties.

A full sensory test was conducted on the extracts, and the results are given in Table 5. It was concluded that the organoleptic quality of the oil extracted from the dried leaves was superior to that obtained from frozen leaves.

Composition of Basil Extracts Obtained by Hydrodistillation, by Liquid CO₂, and by Supercritical CO₂. A commercial sample of dried and rubbed basil was extracted by a number of different methods, and the products were analyzed for their principal volatiles.

The physical appearance and odor of extracts obtained according to the different methods were quite varied. They ranged from a yellow oil (hydrodistilled) through yellow to dark green solids (CO₂ extracts). There was more green pigment and the extracts were more solid when supercritical conditions were used. The yields of extracts are given Table 6 and were highest using

Table 7. Major Identified Components (Percent by GC) in Basil Extract Obtained with Water and Liquid and Supercritical Carbon Dioxide

component (retention time in min)	extraction method				
	hydrodistillation, 1 atm, 2.5 h	liquid CO ₂ , 5 °C, 68 atm, 2 h	liquid CO ₂ , 27 °C, 68 atm, 2 h	supercritical CO ₂ , 40 °C, 102 atm, 2 h	supercritical CO ₂ , 40 °C, 306 atm, 2 h
α-pinene (8.18)	0.6	— ^a	—	—	—
β-pinene (10.11)	0.9	—	—	—	—
myrcene (11.42)	0.5	—	—	—	—
1,8-cineole (12.92)	9.4	0.8	0.9	0.8	1.3
linalool (24.01)	17.2	19.4	17.4	18.5	17.9
β-caryophyllene (26.15)	0.8	1.5	1.4	1.4	1.3
methylchavicol (28.45)	38.4	20.9	17.0	19.8	22.2
α-terpineol (29.13)	0.9	1.7	1.9	1.7	1.4
methyl cinnamate (40.88)	3.9	7.7	10.4	8.5	6.6
eugenol (43.73)	1.0	3.4	4.4	4.0	3.1

^a Components not detected or present at <0.5%.

Table 8. Sensory Evaluation of Basil Extracts Obtained by Hydrodistillation and by Liquid and Supercritical CO₂

method of extraction	sensory evaluation
hydrodistillation	a smooth fresh and diffusive herbaceous note with harmonized cool anisic and slightly balsamic subsidiary notes and warm woody back notes
liquid CO ₂ at 5 °C and 68 atm for 2 h	a smooth warm herbaceous odor, with anisic and slightly balsamic subsidiary notes; woody and slightly camphoraceous back notes
liquid CO ₂ at 27 °C and 68 atm for 2 h	a smooth, warm, haylike note with an anisic and slightly balsamic subsidiary notes; woody and slightly camphoraceous back notes
supercritical CO ₂ at 40 °C and 102 atm for 2 h	a warm herbaceous note with an anisic subsidiary note; woody and slightly camphoraceous back notes
supercritical CO ₂ at 40 °C and 306 atm for 2 h	a fresh herbaceous note with a cool anisic and somewhat balsamic subsidiary note; woody and slightly camphoraceous back notes

supercritical CO₂ followed by liquid CO₂ and then water. This may be due to waxes, epidermal cuticular components, diterpenes, triterpenes, alcohols, acids, and pigments in the CO₂ extracts. Extraction of herbs using solvents above their critical point is known to extract material other than terpenoid components (Reverchon, 1992; Reverchon et al., 1993).

The method by which basil was extracted had a significant effect on the chemical composition of the major volatiles in the extract as shown in Table 7. The hydrodistilled material contained a significantly larger proportion of the lower boiling point hydrocarbons such as α -pinene, camphene, β -pinene, myrcene, α -terpinene, limonene, γ -terpinene, *p*-cymene, and oxygenated terpenes including 1,8-cineole, fenchone, and camphor. This is consistent with a report by Reverchon and Senatore (1992) on rosemary oil in which the hydrocarbons made up a larger proportion of the hydrodistilled oil than they did in the oil fraction in the second separator of their supercritical CO₂ extraction plant. In addition, the CO₂ extracts contained a large number of unidentified high boiling point components with retention times between those of geraniol (33.5 min) and eugenol (43.73 min), which were either present in small amounts or undetectable in the hydrodistilled oil.

The CO₂ extracts were generally quite similar in composition of major components; and differences, where they existed, were found in eugenol and methyl cinnamate content. This has implications for the possible large-scale use of CO₂ since supercritical conditions may not have any major advantage over conditions that are liquid with respect to the carbon dioxide and may enable considerable savings to be made in production costs. However, the sensory evaluation (Table 8) indicated that the quality of the CO₂ extracts obtained under our conditions was inferior to that of the hydrodistilled oil. This was somewhat unexpected since the use of steam distillation and other extraction processes using heat with botanical samples is reported to cause some decomposition associated with unpleasant "still notes" which are not present when carbon dioxide extraction is used (Moyler, 1988). There is no doubt that for many plant materials CO₂ extraction procedures yield products of higher quality than can be obtained by hydrodistillation and which are closer in headspace odor to the original botanical species (Palmer and Ting, 1995); however, our results with basil, although conducted with a limited set of extraction conditions, provide evidence that this cannot be assumed to be the case in all applications.

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