RESEARCH REPORT

The Analysis of the Essential Oil of *Ocimum basilicum* L. growing in Kenya

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The essential oil of flowering tops and leaves of *Ocimum basilicum* L. (Labiatae) obtained from Migori, Migori District, Kenya has been extracted by steam distillation for varying times and analysed using GC and GC-MS. The optimum time has been found to be 6 hours with the yield of the oil and geraniol content as 0.71% and 90.66%, respectively. Other constituents include camphor, linalool, geranyl isobutyrate, cedrol, eugenol, ledol and ß-selinene.

Ocimum basilicum L. (Labiatae) is commonly known as sweet basil (Kiswahili - *kirimbasi*). It is an erect annual herb with branched stems. The leaves are distinctly petioled, ovate and membranous (Charles and Simon, 1990). It is a native of India. In Kenya it is found growing in sandy plains, open dry locations, in valleys and along roads. Polymorphism in the species *basilicum* is responsible for the very great number of subspecies, varieties and forms. As a result, the essential oil of *Ocimum basilicum* (basil oil) is classified according to the chemical composition and geographical source (Guenther, 1974). The four major types of basil oil are European, Reunion, methyl cinnamate and eugenol types.

The essential oil of *Ocimum basilicum* varies from one geographical region to another; Zola and Garnero (1973) have shown that geographical regions have an impact on the constituents of the oil. Basil oils from France, Italy and Morocco were found to contain linalool as the main constituent. The French and Italian samples contained a large amount of methyl chavicol whereas the Moroccan oil had a high eugenol content. The European basil oil contains mainly cineole, linalool and methyl chavicol while the Reunion type contains mainly ß-piinene, cineole, camphor and methyl chavicol. Charles and Simon (1990) have analysed, using GC-MS, the essential oils of *Ocimum basilicum* obtained by hydrodistillation and steam distillation extraction methods. The yield of the oil was consistently higher from steam distillation than from hydrodistillation. However, a similar number of compounds were recovered from both methods. The compounds identified were terpineol, linalool, cineole, citral, eugenol, geraniol, methyl chavicol and citronellol. Preliminary work by Maitai and Talalaj (1984) on *O. basilicum* growing in East Africa revealed the major component as methyl chavicol.

Ocimum basilicum oil is used in medicine (basil camphor) due to its antibacterial properties, in flavouring foods and in dental and oral products. For *Ocimum basilicum* growing in Kenya, there is no data showing the chemical composition of the essential oil, the aim of the present work is to establish the quantity and quality of the oil obtained from *Ocimum basilicum* growing in Migori. The reference compounds were obtained from Aldrich Company Limited, England. All the solvents and other reagents used were purchased locally and were of analytical

grade.

The specific gravity, refractive index and optical rotation were determined at 20°C±0.1 by using pycnometer, refractometer (Atago Company Limited; model 1T) and polarimeter (Instruments for Research and Industry; model 554 Bs)respectively. A 0.5% solution of the oils in n-hexane was used to determine optical rotation.

GC was performed on a Hewlett Packard 5790A gas chromatograph fitted with an FID detector and Hewlett Packard 3393A electronic integrator. A Hewlett Packard-20M (Carbowax 20M) 50m x 0.2mm i.d. x 0.1 μ m film thickness column with nitrogen as carrier gas at 40 ml/min was used. The temperature was programmed from 60°C (7min) to 220°C (10 min) at 10° C/min.

GC-MS was performed on a VG Analytical 12-250 instrument equipped with a Hewlett Packard 5790A GC and a data system. The column and column temperature programme that were used for the GC analysis were used for GC-MS determinations. EIMS spectra were recorded at 70eV at a source temperature of 200°C. Injector was used in the splitless mode.

The flowering tops and leaves of *Ocimum basilicum* were collected early in the morning from Migori, Migori District in December 1989. They were packed in polythene bags and covered tightly to avoid loss of essential oils and then transported to Nairobi. The taxonomic identification of the plant was established by Mr. Simon Mathenge and a voucher specimen was deposited in the herbarium of the Botany Department, University of Nairobi, Nairobi.

The flowering tops and leaves were cut into small pieces using a pair of scissor.
Five batches each of 240g of flowering tops and leaves and 1000 ml water were steam distilled using a modified Clevenger apparatus. The distillation time was varied from four hours up to a maximum of twenty hours. The yield of oil was expressed on moisture-free basis. The essential oils were dried over anhydrous sodium sulphate and stored in sealed ampoules at 0°C until use.

Component identity was established by retention time comparison with reference compounds and peak enhancement by co-injection of the essential oil with known compounds. Library MS searches, published data and/or available mass spectra collections were also used for peak identification and correlation.

Steam distillation of flowering tops and leaves yielded a yellow oil with a warm spicy aroma.

The percentage yields of oil from different batches of *Ocimum basilicum* flowering top and leaves, refractive index, optical rotation, specific gravity and percentage yield of cis- and trans- geraniol in the oils extracted for 4, 6, 8, 12 and 20 h are listed in Table 1.

The major components in the oil of *Ocimum basilicum* analysed are trans-geraniol (39.54-51.04%) and cis-geraniol (38.29-50.35%). This contrasts the observation by Maitai and Talalaj[5] that the major components of East African basil oil are linalool (30-40%) and methyl chavicol (61%). However, geraniol is an isomer of (±)- linalool and is optically inactive and has a higher boiling point and specific gravity than (±)-linalool. According to Guenther (1974), this oil

cannot be classified as the European or Reunion, Methyl cinnamate or eugenol type.

Table 2 lists the constituents identified with relative percentages in the oil obtained for 6 h of steam distillation.

Table 1. Percentage yield of oil, cis- and trans-geraniol and physical properties of the essential oils of *Ocimum basilicum*.

Time extraction (h)	% yield (v/w)	Refractive index at 2° C±0.1	Optical rotation 20° ± 0.1	Specific gravity 20° ± 0.1	% Cis-geraniol	% trans-geraniol
4	0.58	1.473	-6°	0.860	50.35	46.16
6	0.71	1.473	-4°	0.886	47.27	43.39
8	0.75	1.474	0°	0.925	39.54	39.54
12	0.81	1.474	-2°	0.954	39.51	42.11
20	0.83	1.476	-3°	0.960	38.29	51.04

In line with Zola and Garnero's findings that geographical regions have an impact on the constituents of the essential oil of *O. basilicum*, the constituents in the oil of *Ocimum basilicum* growing in Migori District are expected to be different from those growing in other regions (Zola and Garnero, 1973). Moreover, geraniol has been reported in the basil oil from Egypt by Charles and Simon (1990), though the report did not indicate the percentage content of geraniol.

Considering that geraniol is the major compound of the basil oil analysed, it is justified to classify the oil from Migori District as the geraniol type. There is a general decrease in the amount of geraniol extracted with increase in time of extraction for 4, 6 and 8 h. The highest decrease is from 6 h (90.66%) to 8 (79.08%); and there is an increase of 7.71 in the amount obtained from 12 h to 20 h. Six hours can be considered to be the optimum time for maximum yield, since the total geraniol content is 90.66% and the yield 0.71%. Although the geraniol content for 4 h is 96.51%, the yield is lower (0.58%). The yield of the oil for 6 h of extraction is higher than that of *O. basilicum* from France (0.1%), America (0.24%) and Reunion (0.03%), (Guenther, 1974) but it is lower than that reported by Maitai and Talalaj (1-2%).

Since the yield of basil oil is considerably high, it can be used as a source of geraniol. Geraniol is used as an aromatic isolate indispensable in the compounding of rose scents and perfumes. Geraniol can readily be isolated from the oil of basil by reacting it with anhydrous calcium chloride, and then decomposing the complex so formed with water into geraniol and calcium chloride. The separated geraniol is rapidly washed with warm water and steam distilled.

The analysis of essential oil of *Ocimum basilicum* growing in Migori District shows that it has a considerable yield and should be exploited for the production of geraniol as this is of great importance in many industries. However, further investigations on this plant in different

geographical regions, seasonal variations and time of harvest are required to have a conclusive information on the essential oil content.

Table 2. Constituents of essential oil of *Ocimum basilicum*.

Constituent	Rt (min.)	% Total	Identification method
Camphor	9.51	3.24	ms, pe
(±)-Linalool	10.28	0.68	ms, pe
cis-Caryophyllene	11.00	t	ms
4-Terpineol	11.19	t	ms
trans-Citral	12.38	2.00	ms
cis-Farnesol	13.23	t	ms
cis-Geranyl isobutyrate	13.43	0.34	ms
trans-Geranyl isobutyrate	14.14	1.01	ms
cis-Geraniol	15.26	47.27	ms,pe
trans-Geraniol	15.29	43.39	ms,pe
ß-pinene	16.37	t	ms,pe
Cedrol	17.49	t	ms,pe
Eugenol	18.17	0.20	ms,pe
Ledol	18.26	t	ms
Torreyol	18.34	t	ms
ß-Selinene	18.52	0.22	ms
9-Octadecenal	19.40	t	ms

t=trace amount, less than 0.1% of oil. ms=EIMS matching with library spectrum

pe=peak enhancement by co-injection with an authentic sample

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