

# CONSTITUENTS OF THE ESSENTIAL OIL OF ETHIOPIAN *CYMBOPOGON CITRATUS* STAFF

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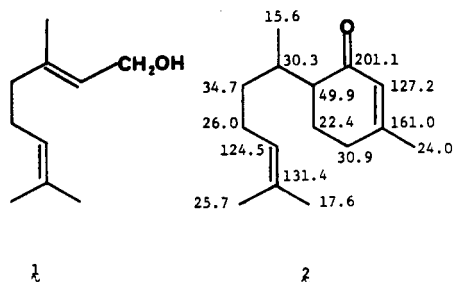
Of the 85 species that constitute the genus *Cymbopogon* (1), about 60 have been examined chemically. *C. citratus* and *C. flexuosus*, cultivated in a number of countries for commercial purposes, have been studied extensively (1). *C. citratus* is also an important medicinal plant (3), and attempts to isolate pharmacologically active ingredients have resulted in the isolation of new triterpenoids (4,5). The oil of *C. citratus* from Brazil (6), Ceylon (7), Phillipines (8), Somalia (9), India (10), and the Congo (11) is reported to contain citral as the main ingredient. Our initial analysis indicated that the Ethiopian highland variety contained geraniol and not citral as the main component. We have subsequently analyzed the oil, and we present herein results that account for over 80% of the essential oil.

## DISCUSSION

Most of the components were identified by comparing gc Rt values with those of standard compounds and by computerized matching of acquired mass spectra with stored NBS spectral library in the data system of the gc/ms. Geraniol (1) and  $\alpha$ -oxobisabolene (2) were isolated, purified, and character-

ized by additional spectroscopic data. The results obtained by analysis of the steam-distilled oil are given in the table. The main component is geraniol (40%). Although oils from different geographical varieties (6-11) of *C. citratus* are reported to contain citral as the main component, the Ethiopian variety that we have examined contains only 13% citral (citral a + citral b). Samples of plant materials collected during different seasons did not show significant variation in the geraniol-citral ratio.

The second major component (12%) is identified as  $\alpha$ -oxobisabolene (2). This compound was isolated from the oil by column chromatography and purified by preparative tlc as described in the experimental section. The identification of this bisabolene derivative is based on its ir,  $^1\text{H}$ -nmr, and ms and on comparison of these data with the reported spectroscopic data for a sesquiterpene recently isolated by Bohlmann *et al.* from *Stevia purpurea* (Compositae) (12-14). The two compounds were found to be identical. The carbon magnetic resonance (cmr) spectrum gave additional confirmation for the assigned structure of this sesquiterpene. The presence of an isopentenyl side chain and an enone grouping was clearly displayed in the cmr spectrum (15). The carbon chemical shift values were initially assigned by comparison with the reported spectra of 6-isopropyl-3-methyl-2-cyclohexenone and citronellal (16). Experiments using cmr and involving selected proton decoupling (17) and polarization transfer sequences (INEPT) (18,19) enabled unambiguous assignment of carbon chemi-



cal shift values as shown in structure 2. The optical rotation measurements, however, were found to have values opposite in sign to those recorded by Bohlmann *et al.* (12) (see Experimental section) indicating an enantiomeric relationship to  $\alpha$ -oxobisabolene isolated from *S. purpurea*.

Menthone and menthol, rarely found in other *Cymbopogon* oils, were found in low quantities. The remaining components are routinely found in the genus (20). The compounds identified constitute over 80% of the oil. The remaining components are mostly monoterpene hydrocarbons with a few sesquiterpenoids, which are presently unidentified.

In summary, it is noted that the composition of the oil of the Ethiopian *C. citratus* is quite different from all other reported geographical varieties and is more comparable in constitution to the oil of *C. nardus* (1).

## EXPERIMENTAL

**GENERAL EXPERIMENTAL METHODS.**—A Perkin-Elmer model 727 infrared spectrophotometer was used to record ir spectra; uv spectra were taken on a Perkin-Elmer 550 recording spectrophotometer;  $^1\text{H}$ -nmr were recorded on a Varian T60-A spectrometer; cmr spectra were taken on a JEOL FX-90Q spectrometer.

**PLANT MATERIALS.**—Plants were collected from Entoto, altitude of ca. 2500 m, just north of Addis Ababa, and were positively identified by Dr. Tewolde Berhan C. Egziabher of the Addis Ababa University Herbarium, where a voucher specimen was deposited. Plants were collected in July 1979.

**ISOLATION AND ANALYSIS OF ESSENTIAL OIL.**—Fresh whole plant material (100 g) was steam distilled to give 4.32 g of light yellow oil (4.32%), density 0.9001,  $[\alpha]_D^{21} + 9.1^\circ$  (neat,  $n_D$  1.4814). The oil (1.0 g) was put on 60 g of silica gel (Merck 60-200 mesh) and successively eluted with 200 ml each of hexane and ethyl acetate. These fractions were concentrated and then analyzed by gc and gc/ms. A Hewlett-Packard model 5710A gas chromatograph with a flame ionization detector was used. Two columns, 10% Carbowax 20M on Chromosorb WHP 100/120 ( $\frac{1}{4}$ " x 5' stainless steel column) and 10% UCON on Chromosorb WHP 8/100 mesh ( $\frac{1}{4}$ " x 5' copper column), were used. Analysis was done either isothermally at 180° or by programming the oven temperature from 70-200° at 2°/min. Nitrogen

TABLE 1. Constituents of Ethiopian *Cymbopogon citratus* oil.

Terpenoid	% of Oil	Method of Identification
Borneol . . . . .	5.0 <sup>a</sup>	gc
Camphor . . . . .	0.2 <sup>a</sup>	gc/ms
$\Delta^3$ -Carene . . . . .	0.1 <sup>b</sup>	gc/ms
Citral-a (geranial) . . . . .	10.2 <sup>a,b</sup>	gc/ms
Citral-b (neral) . . . . .	3.3 <sup>a,b</sup>	gc/ms
Citronellol . . . . .	0.1 <sup>a</sup>	gc
Fenchone . . . . .	0.3 <sup>a</sup>	gc
Geraniol . . . . .	40.2 <sup>a,c</sup>	gc,ms,ir,nmr
Linalool . . . . .	3.4 <sup>a</sup>	gc/ms
Menthol . . . . .	0.5 <sup>a</sup>	gc
Menthone . . . . .	0.2 <sup>a</sup>	gc
Myrcene . . . . .	0.1 <sup>a</sup>	gc
Nerol . . . . .	4.5 <sup>a</sup>	gc
Ocimene . . . . .	0.2 <sup>b</sup>	gc/ms
$\alpha$ -Pinene . . . . .	0.1 <sup>a</sup>	gc/ms
Terpinolene . . . . .	0.1 <sup>a</sup>	gc/ms
$\alpha$ -Oxobisabolene . . . . .	12.1 <sup>b,c</sup>	gc,ms,ir,nmr
Unidentified . . . . .	19.4	

<sup>a</sup>Percentages were determined by absolute calibration method (gc).

<sup>b</sup>Percentages were determined by internal standardization method (gc).

<sup>c</sup>Percentages are based upon isolated compounds.

was used as carrier gas. A Finnigan model 3200F spectrometer interfaced to a model 6100 data system was used to perform gc/ms analysis. A 50-m glass capillary column coated with SP1000 was used. Identification was made by comparison of Rt values with those of standard compounds and by computerized matching of acquired mass spectra with stored NBS mass spectral library in the data system of the gc/ms.

**ISOLATION OF  $\alpha$ -OXOBISABOLENE (2).**—We chromatographed 2 g of the oil with 80 g of silica gel and first eluted it with 600 ml of hexane followed by 600 ml of ethyl acetate. The concentrated ethyl acetate fraction was rechromatographed on 40 g of silica gel and eluted with 10% ethyl acetate in hexane. The second 25-ml eluate was found to contain ca. 80% of 2. An analytically pure sample was obtained by preparative tlc on 1 mm silica gel by using 20% ethyl acetate in hexane as developing solvent. This compound,  $\text{C}_{15}\text{H}_{24}\text{O}$ , showed ir;  $\nu$  max (neat) 1675, 1450, 1380, 1220, 880  $\text{cm}^{-1}$ ;  $\lambda$  max ( $\text{CHCl}_3$ ) 230 (log  $\epsilon$  4.1);  $^1\text{H}$ -nmr, 60 MHz ( $\text{CDCl}_3$ )  $\delta$  5.73 (q, 1H,  $J=1$  Hz ring vinyl H), 5.06 (t, 1H,  $J=7$  Hz side chain vinyl H), 1.92 (d, 3H,  $J=1$  Hz, ring  $\text{CH}_3$ ), 1.66 (s, 3H, side chain vinyl  $\text{CH}_3$ ), 1.60 (s, 3H, side chain vinyl  $\text{CH}_3$ ), 0.79 ppm (d, 3H,  $J=7$  Hz, side chain  $\text{CH}_3$ ) [Lit. (12)  $\delta$  5.86 (q, 1H,  $J=1$  Hz), 5.11 (t, 1H,  $J=7$  Hz), 1.93 (d, 3H,

$J=1$  Hz), 1.68 and 1.60 (s,s, 3H,3H);  $^{13}\text{C}$  cmr, 22.5 MHz ( $\text{CDCl}_3$ ), see structural formula **2**; ms (70 eV  $m/e$ , rel. int.): 220 ( $\text{M}^+$ ), 137 (100), 110 (58), 135 (36), 99 (39), 95 (33), 82 (18), 138 (9). Optical rotation measurements in chloroform ( $c=4.59$ ) gave the following data:  $[\alpha]^{21} -37.0^\circ +34.6$  ( $\lambda=589$ ),  $+37.2$  (578),  $+44.8$  (546), and  $+108.9$  (436 nm); [Lit. (12),  $[\alpha]^{24}$  (589),  $-39.3$  (578),  $-47.3$  (546), and  $-129.0$  (436 nm),  $c=3.7$  chloroform].

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