Phytosterols from the stem bark of *Combretum fragrans* F. Hoffm

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Two sterols, β-sitosterol and stigmasterol, were isolated from the stem bark of *Combretum fragrans.* The identity of these compounds was established by spectral analysis.

Key words: *Combretum fragrans, combretaceae,* β-sitosterol, stigmasterol

INTRODUCTION

Combretum fragrans F. Hoffm belongs to the Combretaceae (Combretum) family. The plant grows in wooded or bushy grassland [1]. The powdered bark is used for the treatment of wounds, diarrhoea, syphilis and gonorrhoea [2] and also in fungal, bacterial and inflammatory conditions [3-4]. Only limited pharmacological studies have been carried out on the plant. Methanolic extracts of *C. fragrans* significantly reduced the activity of the enzyme neuraminidase from *Clostridium chauvoei* in a dose dependent fashion [5]. There are no reports of any compounds isolated from the plant.

METHODOLOGY

Combretum fragrans stem bark was collected from Rarieda in Bondo District, Nyanza Province, Kenya, in October, 2004. Plant identification was done at the Department of Botany Herbarium, University of Nairobi. Voucher specimens were deposited in the same department and the School of Pharmacy, University of Nairobi. The plant material was oven dried at 45 °C, powdered and kept dry at room temperature until use. General phytochemical screening performed on extracts of *Combretum fragrans* showed the presence of saponins, glycosides, flavonoids and tannins in conformity with literature [6-8]. The stem bark yielded 0.72 % of chloroform extract. About 7 g of the extract was introduced into a column containing 80 g of silica gel and eluted using chloroform. One fraction yielded two compounds β-sitosterol and stigmasterol which were further purified by re-crystallization from diethyl ether.

Structure determination

The isolates β-sitosterol and stigmasterol were analysed by use of spectroscopic methods. The ^{1}H and ^{13}C nuclear magnetic resonance (NMR) spectra were obtained using Varian Gemini 200 MHz in deuterated chloroform ($CDCl_3$). Mass Spectrometry (MS) analysis was carried out on Direct Ionization Platform (DIP) on a Fission Platform GC/LC Mass Spectrometer. The spectral data obtained was found to be concordant with that reported in the literature [9-12].

β-Sitosterol

IR (KBr): ν_{max} cm^{-1}, 3446 (H-bonded OH), 2933 (methyl C-H), 2852 (cycloalkane C-H), 1637 (C=C), 1465 (C-H_{def}), 1380 (C–O).

MS: *m/z* (rel. int. %): Base peak 57 (100), 414 (M^+, 25), 412 (2), 399 (7), 396 (10), 381 (7), 354 (2), 329 (13), 303 (12), 301 (4), 273 (8), 255 (12), 231 (9), 213 (12), 163 (12), 159 (16), 149 (12), 147 (13), 145 (19), 133 (16), 121 (12), 119 (17), 107 (20), 105 (21), 97 (35), 95 (34), 85 (41), 83 (44), 71 (62), 69 (57), 55 (79), 43 (88), 41 (44).

^{1}H-NMR (200 MHz, $CDCl_3$): δ 0.68 (3H, s,

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CH_3-18), 0.81 (3H, m, CH_3-29), 0.83, 0.85 (6H, d, CH_3-26 and CH_3-27), 0.91 (3H, d, CH_3-21), 1.00 (3H, s, CH_3-19), 2.25 (2H, m, CH_2-4), 3.53 (1H, m, CH-3), 5.38 (1H, m, CH-6).

^{13}C-NMR (200 MHz, $CDCl_3$): 37.3 (C-1), 31.7 (C-2), 71.9 (C-3), 42.3 (C-4), 140.8 (C-5), 121.7 (C-6), 31.9 (C-7), 31.9 (C-8), 50.2 (C-9), 36.5 (C-10), 21.1 (C-11), 39.8 (C-12), 42.3 (C-13), 56.8 (C-14), 24.3 (C-15), 28.2 (C-16), 56.1 (C-17), 11.9 (C-18), 19.4 (C-19), 36.1 (C-20), 18.8 (C-21), 34.0 (C-22), 26.2 (C-23), 45.9 (C-24), 29.2 (C-25), 19.8 (C-26), 19.0 (C-27), 23.1 (C-28), 12.2 (C-29).

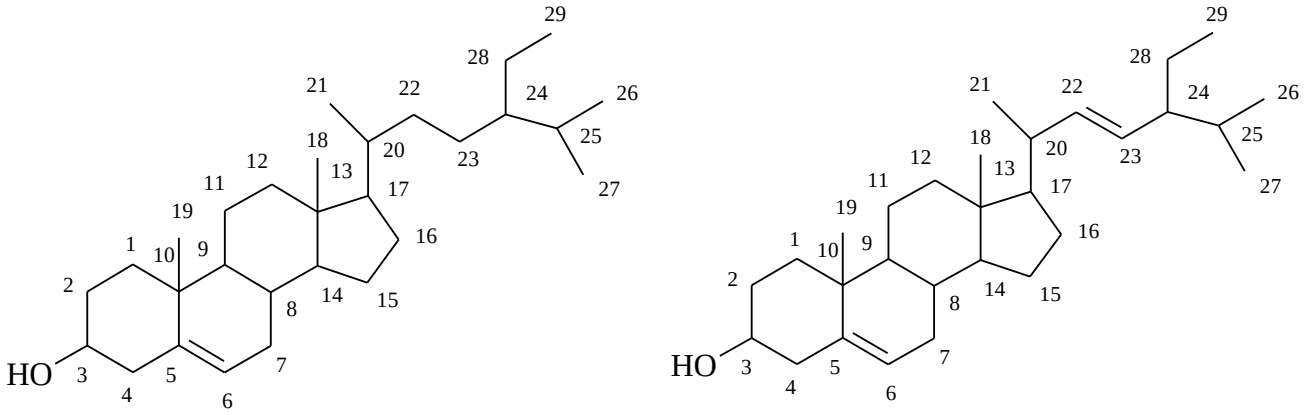
Stigmasterol

IR (KBr): ν_{max} cm^{-1}, 3446 (H-bonded OH), 2933 (methyl C-H), 2852 (cycloalkane C-H), 1637 (C=C), 1465 (C-H $_{def}$), 1380 (C–O).

MS: *m/z* (rel. int. %): Base peak 57 (100), 412 (2.11), 399 (7.2), 396 (10), 381 (7), 354 (2), 329 (13), 303 (12), 273 (8), 255 (12), 213 (12), 163 (12), 159 (16), 147 (13), 145 (19), 133 (16), 121 (12), 119 (17), 107 (20), 105 (21), 97 (35), 95 (34), 85 (41), 83 (44), 71 (62), 69 (57), 55 (79), 43 (88), 41 (44).

^{1}H-NMR (200 MHz, $CDCl_3$): δ 0.68 (3H, s, CH_3-18), 0.81 (3H, m, CH_3-29), 0.83, 0.85 (6H, d, CH_3-26 and CH_3-27), 0.91 (3H, d, CH_3-21), 1.00 (3H, s, CH_3-19), 2.25 (2H, m, CH_2-4), 3.53 (1H, m, CH-3), 5.38 (1H, m, CH-6), 5.00-5.25 (2H, m, CH-22, CH-23).

^{13}C-NMR (200 MHz, $CDCl_3$): 37.3 (C-1), 31.7 (C-2), 71.8 (C-3), 42.4 (C-4), 140.8 (C-5), 121.7 (C-6), 31.9 (C-7), 31.9 (C-8), 50.2 (C-9), 36.6 (C-10), 21.1 (C-11), 39.7 (C-12), 42.4 (C-13), 56.9 (C-14), 24.4 (C-15), 29.0 (C-16), 56.1 (C-17), 12.1 (C-18), 19.4 (C-19), 40.5 (C-20), 21.2 (C-21), 138.3 (C-22), 129.3 (C-23), 51.2 (C-24), 31.9 (C-25), 19.0 (C-26), 21.2 (C-27), 25.4 (C-28), 12.3 (C-29).



β-sitosterol **Stigmasterol**

Figure 1: Chemical structures of β-sitosterol and stigmasterol.

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