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LYCHNOPHOLIC ACID AND ITS ACETATE FROM *LYCHNOPHORA MARTIANA**

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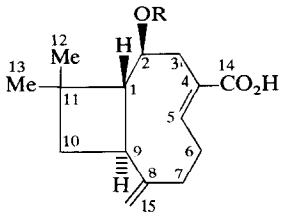
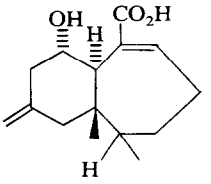
Key Word Index—*Lychnophora martiana;* Compositae; Vernonieae; lychnopholic acid; caryophyllene derivatives; sesquiterpenes.

In the course of our work on Brazilian Compositae we have isolated from the hexane–EtOAc (10:1) extract of *Lychnophora martiana* Gardn. (Vernoniae) [1] two sesquiterpene acid fractions. One of them was identical with lychnopholic acid (**1a**) reported [2] from *Lychnophora affinis* while our work was in progress; the second was a mixture of the acetate **1b** and its isomers (or conformers).



1a R = H
1b R = Ac



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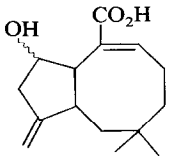

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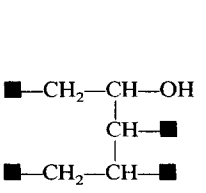
One fraction after recrystallization from hexane–ether and methanol had mp 168°, molecular formula $C_{15}H_{22}O_3$ based on the MS (M^+ *m/e* 250), number of signals in the ^{13}C NMR and integration of the ^1H NMR spectrum. The IR spectrum (KBr) which had bands at 3350, 1680 and 1630 cm^{-1} suggested the presence of an α,β-unsaturated carboxylic acid. The presence of a second double bond, the degree of substitution of the double bonds and the presence of a secondary hydroxyl group and its environment were established by spin-decoupling of the 270 MHz ^1H NMR and selective decoupling of the ^{13}C NMR spectrum (Tables 1 and 2) which will not be detailed here, but led to the four partial structures **A**–**D**. Combination of these structures, which together encompass all atoms of the empirical formula, then led to **1a** (a caryophyllene, proposed stereochemistry based on coupling constants), **2** (a himachalene derivative, proposed stereochemistry based on observed coupling constants) and **3** (representative of a new carbon skeleton) as biogenetically plausible structures.

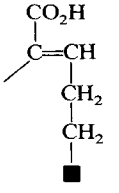
Table 1. 270 MHz ^1H NMR spectra of **1a** and **1b***

	1a	**1b**
H-1	1.84*t* (10)	2.2*m*
H-2	4.07*dt* (6, 10)	5.70*dt* (11, 6)
H-3a	1.98*dd* (12, 10)	2.2*m*
H-3b	2.96*dd* (12, 6)	3.5*m*
H-5	6.34*dd* (10, 7)	6.29*dd* (10, 6)
H-6a	3.17*m* (10, ?)	3.5*m*
H-6b	2.23*m*	2.2*m*
H-7a,b	2.23*m*	
H-9	2.46*dt* (11, 10)	2.89 (11)
H-10a	1.64*tbr* (10)	1.6*m*
H-10b	1.52*tbr* (10)	
H-12†	1.19	1.19
H-13†	1.05	1.05
H-15a	5.03*br*	5.16*br*
H-15b	4.91*br*	4.99*br*

* Run in $CDCl_3$ with TMS as internal standard. All coupling constants established by spin decoupling.
† Intensity three protons.



A



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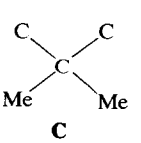
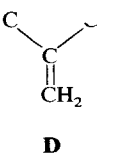


C



D

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Table 2. ^{13}C NMR spectrum of **1a***

Carbon atom	**1a**
1	58.26*d*
2	73.73*d*
3	45.84*t*†
4	128.34
5	146.67*d*
6	30.99*t*
7	34.83*t*
8	153.34
9	45.27*d*
10	42.02*t*
11‡	34.83
12	23.07*q*
13	32.11*q*
14	171.05
15	114.23*t*

* Run at 67.09 MHz in CD_3OD. Unmarked signals are singlets.

† Multiplicity dubious. Possibly a singlet in which case signal must be assigned to C-11.

‡ Assumed to be under 34.83*t* by analogy with ref. [2]. If C-11 is at 45.84 ppm the triplet at 34.83 ppm represents two methylenes.

At this stage a communication appeared [2] on the structure elucidation by X-ray crystallography of lychnopholic acid from *L. affinis* as **1a**. While no mp was given in this publication the NMR data reported by Raffauf *et al.* [2] were sufficiently similar to those of our compound to warrant a direct comparison which established identity in all respects except mp. The authentic sample kindly supplied by Professor P. W. LeQuesne had been isolated from the mother liquors of **1a** from *L. affinis* and melted at 181–182°, somewhat lower then the mp 183–185° of the material isolated originally (private communication from Prof. LeQuesne who also suggested that small traces of impurities markedly affect the mp of **1a**). TLC behavior, IR and 270 MHz ^1H NMR spectra were identical. However on the basis of our decoupling experiments we would interchange the assignments of H-1 and H-3a (caryophyllene numbering) given in ref. [2]. Our ^{13}C NMR spectrum in CD_3OD duplicates that of ref. [2] taken in $CDCl_3$–$(CD_3)_2SO$, if the expected minor solvent shifts are taken into account.

A somewhat less polar fraction was a mixture of several acetates (NMR spectrum). Repeated purification by TLC gave material which consisted primarily of the acetate **1b** as indicated by the NMR spectrum (Table 1). Other constituents of this fraction were stereoisomers and/or conformers of **1b** (see Experimental). Whether caryophyllene derivatives like **1a** are characteristic constituents of *Lychnophora* species will have to be demonstrated by further study of the genus.

EXPERIMENTAL

Lychnophora martiana Gardn. was collected by Dr. Hermógenes de Freitas Leitão Filho in Conceição do Mato Dentro (Serra do Cipó), Minas Gerais, Brazil, in December 1973. The aerial parts (9 kg) were extracted with hexane–EtOAc (10:1) and the extract was worked up in the usual manner [3]. The crude gum (100 g) was chromatographed over Si gel (700 g), 400 ml fractions being eluted in the following order: 1–14 (C_6H_{14}–EtOAc, 20:1), 15–25 (C_6H_{14}–EtOAc, 10:1), 26–35 (C_6H_{14}–EtOAc, 5:1), 36–52 (C_6H_{14}–EtOAc, 2:1), 53–58 (C_6H_{14}–EtOAc, 1:4:1), 59–65 (EtOAc), 66–75 (EtOAc–EtOAc, 5:1), 76–78 (EtOAc–EtOH, 2:1), 79–84 (EtOAc–EtOH, 1:4:1) and 85–89 (EtOh). Fractions 26–28 whose NMR spectra indicated the presence of isomeric acetates were semisolid (460 mg). Recrystallization of fraction 28 from C_6H_{14}–Et_2O (4:1) gave material of mp 158–164°, which had IR bands at 3400 (br), 1710 and 1660 (strong) cm^{-1} and whose NMR spectrum indicated the presence of a mixture of isomeric acetates. Purification by PLC (EtOAc–C_6H_6, 1:3) gave material whose main component (~80%) exhibited the NMR spectrum listed in Table 1. Clearly visible signals of the minor component appeared at δ 5.99*dd* (12,6, H-5), 5.3*m*, 5.27*br* (=CH_2), 3.65*m*, 3.15*m*, 2.03 (Ac), 1.17 and 1.08 ppm (two methyl singlets). This suggested that the contaminant might be the C-8 epimer or the isocaryophyllene analog of **1b**, or that the NMR spectrum indicated the presence of a conformational equilibrium. A third isomeric constituent of fractions 26–28 had signals at δ 6.28*dd* (12, 6, H-5), 5.11*m* (H-2), 4.96 and 4.83 (=CH_2), 3.3*m*, 3.06*dd*(12, 6), 2.2*m* (4–5 H's), 2.62*q* (10), 2.06*q* (10), 1.99 (Ac), 1.83*dd* (12, 10), 1.5*m* (2p), 1.08 and 0.99 (methyl singlets) and was contaminated by, or in equilibrium with, material displaying visible singlets at δ 6.15*m* (H-5), 5.06 and 4.92 (=CH_2), 3.17*m*, 2.89*m*, 2.00 (Ac), 1.09 and 0.96 (methyl singlets). Fractions 37 (107 mg) and 38 (600 mg) were semicrystalline. Recrystallization and purification by PLC (C_6H_6–EtOAc, 1:19) gave crystalline material, mp 168°, MW (MS) 250, IR bands (KBr) at 3350, 1680 and 1630 cm^{-1} which exhibited the ^1H and ^{13}C NMR signals shown in Tables 1 and 2. IR spectra and 270 MHz ^1H NMR spectra of this material and authentic lychnopholic acid were superimposable. Methylation with MeI in Me_2CO–K_2CO_3 afforded a gummy methyl ester (IR spectrum) which appeared to be an equilibrium mixture of conformers or isomers (NMR spectrum). Acetylation or benzoylation gave inhomogeneous material. An attempt at oxidation with P_2O_5–DMSO [4] resulted in recovery of starting material.

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