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A new modified 6,7-secolabdane diterpenoid from *Clutia richardiana*

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Abstract

The aerial parts of *Clutia richardiana* yielded a new modified 6,7-secolabdane diterpenoid derivative, namely 2β-hydroxysaudinolide. The structural assignment and relative stereochemistry of this compound were based on its spectral data, including 2-D NMR experiments, notably the gradient HMBC and NOESY correlations.

Keywords: Clutia richardiana; Euphorbiaceae; Diterpene; Secolabdane; 2β-hydroxysaudinolide

1. Introduction

In previous reports (Muhammad et al., 1994a, 1994b; Mossa et al., 1996) we have described the structure of ten new modified labdane diterpenoids, namely, the 6(7), 9(10)-biseco-6(11), 1(19) bicyclolabdanes, cluytenes A and C and the 6,7-seco-6, 11-cyclolabdanes, saudinolide (**1**), dihydrosaudinolide (**2**), 5β-hydroxyrichardianidines 1 and 2 and cluytenes B, D, E and F, isolated from the aerial parts of *Clutia richardiana* Muell.-Arg.[1] (Euphorbiaceae). Earlier investigations have reported on the isolation of other 6,7-secolabdanes, including saudin (Mossa et al., 1985), richardianidin 1 and richardianidin 2 (Mossa et al., 1988a) from *C. richardiana*, while clutiolide, dihydroclutiolide and isodihydroclutiolide were isolated from *C. abyssinica* (Waigh, Zerihun, & Euerby, 1990). Furthermore, saudin was found to possess a significant hypoglycemic effect in nonalloxanized, rather than alloxanized fasting mice (Mossa, El-Denshary, Hindawi, & Ageel, 1988; Mossa et al., 1996). Examination of the same source has now led to the isolation and characterization of an additional structurally related new diterpenoid, namely, the saudinolide derivative 2β-hydroxysaudinolide (**3**).

2. Results and discussion

The EtOAc precipitate, obtained from the defatted EtOAc extract of *C. richardiana* (see Section 3) (Muhammad et al., 1994a, 1994b; Mossa et al., 1996) was flash chromatographed over silica gel to give, in crystalline form, the minor diterpenoid, 2β-hydroxysaudinolide (**3**), in 0.001% yield. It analyzed for the molecular formula $C_{20}H_{22}O_9$ and was found to have a γ-lactone (ν_{max} 1780 cm^{-1}; δ_c 177.4), a δ-lactone (ν_{max} 1720 cm^{-1}; 172.9) and a monosubstituted furan ring was also present, as previously encountered in all diterpenoids isolated from this plant. The ^1H and ^{13}C NMR spectral data (Table 1) were generally similar to those of the earlier reported 6,7-secolabdane diterpene saudinolide (**1**) (Mossa et al., 1996), except for signals indicating the presence of methylene group at C-3 (δ 36.1, t) and a secondary hydroxy group at C-2 (δ_c 61.9, d; δ 4.42, 1H, m), instead of the double bond at C-2(3) ($\delta_{C\text{-}3}$ 126.2, $\delta_{c\text{-}2}$ 132.3 in **1**).

Since the structure and relative stereochemistry of saudinolide (**1**) were unambiguously established by X-ray crystallography and its ^1H and ^{13}C NMR data were assigned by using 2-D NMR studies (Mossa et al., 1996), the placement of the secondary hydroxyl group at C-2 of **3** was straightforward. A COSY 2-D NMR experiment suggested the presence of the system –CH_2–CH (OH)–CH(O)– (H-3, H-2 and H-1, respectively) in **3** and this was confirmed by a 2-D NMR HETCOR experiment and other ^{13}C NMR data, which showed signals at δ_c 73.6 (d), 61.9 (d), 36.1 (t), 45.0 (s) and 40.4 (s) assigned to C-1–C-4 and C-10, respectively. The placement of the C-2 and

[1] In this paper the genus *Cluytia* is spelled without a 'y', as recommended by Waigh et al. (1990) and Collenette (1998). We have used the name *Cluytia* in our previous reports based on the name adopted by earlier authors (Collenette, 1985; Migahid, 1989). For earlier work on this plant, under *Cluytia richardiana* see Mossa et al. (1985, 1988a, 1988b, 1996) and Muhammad et al. (1994a, 1994b).

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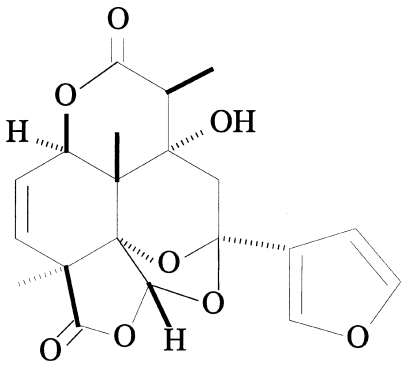
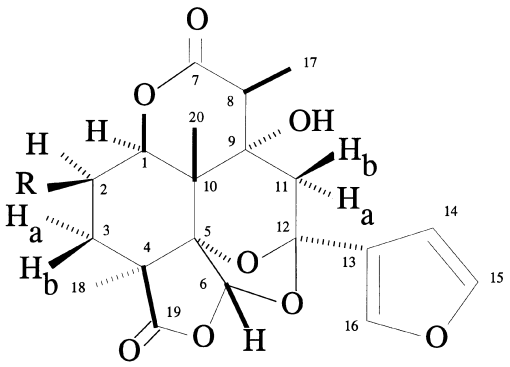


1



2 R =H

3 R = OH

C-9 hydroxy groups was confirmed by a ^{1}H–^{13}C long-range HMBC experiment, which revealed the key three-bond correlations between the signals at δ 1.26 (H-17), δ_c 71.7 (C-9) and 172.9 (C-7) and δ 4.42 (H-2), δ_c 40.4 (C-10) and 45.0 (C-4). Also, the HMBC experiment exhibited the key two bond correlations between H-8 (δ 2.68) and C-9, and H-2 and C-3 (δ_c 36.1). Other HMBC correlations that confirm all the structural features of **3** are summarized in Table 1.

The stereochemical assignments at the centers C-1, C-4, C-6 and C-10 of **3** were inferred from a 2-D NMR ^{1}H–^{1}H NOESY experiment. It showed cross peak between H-1 and H-2 (δ 5.18 and 4.42, respectively), indicating that they were on the same side of the molecule. Furthermore, both H-1 and H-2 showed cross peaks with the proton signals at δ 1.28 and 2.23, which could now be attributed to H-18 and H-3a, respectively. The additional correlations observed between the signals at δ 6.44, 1.86 and 1.22 (H-6, H-3b and H-20, respectively) suggested that all three protons were on the other side of the molecule. The NOESY experiment further showed cross peaks between the signals at δ 1.26 and 2.46 H-17 and H-11b, respectively), indicating that these protons were *cis* with respect to each other; on the other hand H-11a and H-16 (δ 2.52 and 7.98, respectively) were found to be correlated, suggesting they were on the other side of the molecule, as depicted in structure **3**.

3. Experimental

Mp: uncorr; UV: MeOH; IR: KBr; ^{1}H and ^{13}C NMR: 500.13 MHz and 125.77 MHz, respectively, in DMSO, using TMS as an int. standard. Spectral editing (APT and DEPTGL) and 2-D NMR spectra (COSY, HETCOR, NOESY and HMBC) were obtained using standard Bruker software; CIMS: recorded on a Finnigan MAT 300 MS, using CH_4 as ionizing gas; $[\alpha]_D$: at ambient temp. in $CHCl_3$, using a Perkin-Elmer 241 MC polarimeter; TLC: silica gel 60 F_{254}, using $CHCl_3$–Me_2CO (9:1) as solvent, with visualization using 1% vanillin/H_2SO_4 spray reagent. The aerial parts of *C. richardiana* (Collenette, 1998) [1] were collected in Abha, Saudi Arabia, in June 1991. A voucher specimen is deposited at the herbarium of MAPPRC, College of Pharmacy, King Saud University, Riyadh, Saudi Arabia.

3.1. Extraction and isolation of diterpenoids

The initial isolation procedure for the diterpenoids obtained from cold defatted EtOAc extract of *C. richardiana* was as previously described (Muhammad et al., 1994a, 1994b; Mossa et al., 1996). Elution with petroleum ether (60–80°C)–EtOAc (1:1) from silica gel column yielded 2β-hydroxysaudinolide (**3**, 80 mg) as plates, R_f 0.35, using $CHCl_3$–Me_2CO (9:1) as solvent.

3.2. 2β-Hydroxysaudinolide (3)

Colorless plates from petroleum ether/EtOAc and (off-white) needles from hot EtOAC; mp 232°C; $[\alpha]_D$ +2.8° (*c* 0.14, $CHCl_3$): UV λ_{max}^{MeOH} nm: 220 (4.30), 265 (3.90) nm; IR ν_{max} (KBr) cm^{-1}: 3510 (OH), 1780 (γ-lactone), 1720 (δ-lactone), 1510, 1450, 1380 (br), 1200, 1190, 1050, 970, 860, 830, 735 cm^{-1}. ^{1}H and ^{13}C NMR: Table 1. CI-MS *m/z* (rel.int.): 435 $[M^+ +29]^+$ (10), 470 $[MH]^+$ $[C_{20}H_{22}0_9+H]^+$ (100), 389 $([MH]^+ -H_20)$ (18), 167 (5), 111 (5) and 101 (10).

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Table 1
NMR Spectral data of 2β-hydroxysaudinolide (**3**)[a]

C/H	δ ^{13}C	δ ^{1}H	HMBC correlations[b]	NOESY correlations
1α	73.6 d[b]	5.18 d (7.7) [d]	C-20	H-2, H-3a, H-18
2α	61.9 d	4.42 m	C-3, C-4, C-10	H-1, H-3α, H-18
3α	36.1 t	2.23 dd (8.8, 14.3)	C-1, C-2, C-4, C-18	H-2, H-18
3β	–	1.86 dd (7.2, 14.3)	–	H-20
4	45.0 s	–	–	–
5	91.0 s	–	–	–
6β	100.6 d	6.44 s	C-5, C-12	H-20
7	172.9 s	–	–	–
8α	48.8 d	2.68 brq (7.9)	C-7, C-9, C-10, C-17	C-9–OH
9	71.7 d	–	–	–
10	40.4 s	–	–	–
11α	41.4 t	2.52 d (15.2)	C-9, C-10, C-12	H-14, H-16
11β	–	2.46 d (15.2)	-	H-17
12	108.4 s	–	–	–
13	125.9 s	–	–	–
14	109.5 d	6.49 br d (1.0)	C-15, C-16	H-11α
15	144.8 d	7.70 t (1.0)	C-12, C-13, C-16	–
16	142.6 d	7.98 br s	C-12, C-13, C-15	H-11α
17β	15.6 q	1.26 d (7.9)	C-7, C-8, C-9	H-11β
18α	18.4 q	1.28 s	C-3, C-4, C-5	H-1, H-2, H-3α
19	177.4 s		–	–
20β	17.5 q	1.22 s	C-1, C-5, C-9, C-10	H-3β, H-6β
OH	–	5.38 br d (4.3)	–	–
	–	4.73 s		–

[a] Spectra recorded at 500.13 (^{1}H) and 125.77 (^{13}C) MHz.
[b] From gradient HMBC experiments optimized for $^{n}J_{CH}$ = 8.0 or 5.0 Hz.[c]Multiplicities of carbon signals were determined by APT and DEPT experiments, also aided by 2-D NMR COSY and HETCOR experiments.
[d] Values in parentheses are coupling constants (J) in Hz.

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