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A *NEO*-CLERODANE DITERPENOID FROM *SCUTELLARIA SELERIANA**

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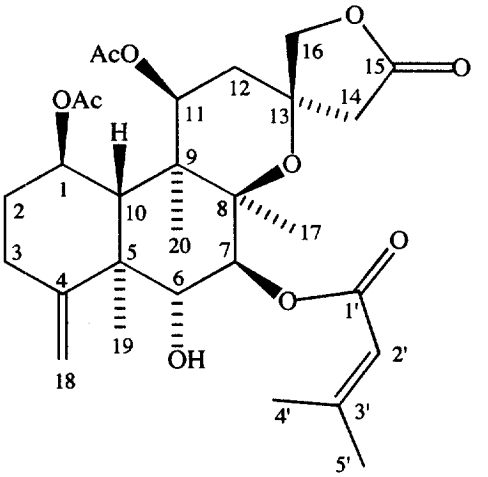
Key Word Index—*Scutellaria seleriana*; Labiatae; *neo*-clerodane diterpenoids; scuteselerin.

Abstract—A new *neo*-clerodane diterpenoid, (13*R*)-1β-11β-diacetoxy-6α-hydroxy-7β-senecioyloxy-8β,13-epoxy-4(18)-neocleroden-15,16-olide (Scuteselerin), has been isolated from the aerial parts of *Scutellaria seleriana*, besides the known flavone oroxylin A. The structure of the new diterpenoid was established by spectroscopic methods. © 1997 Published by Elsevier Science Ltd

INTRODUCTION

Scutellaria L. is a large subcosmopolitan genus of the Labiatae with *ca* 360 currently recognized species [1]. Recently, plants belonging to this genus have attracted much attention due to the interesting biological activities found for some *neo*-clerodane diterpenoids isolated from them, in particular as insect antifeedants [2–4] and as antifungal agents against plant pathogenic fungi [5]. In Mexico, *Scutellaria* is represented by *ca* 32 species, most of them growing in the mountains near the centre of the country [6]. Recently, we reported on the isolation of three *neo*-clerodane diterpenoids, related to ajugarin V, from *S. drummondii* [7]. As part of our ongoing search for diterpenoids from plants of the Labiatae, with potential antifeedant activity [8], we now describe the structure of a new *neo*-clerodane diterpenoid (**1**) isolated from *S. seleriana* Loesen (Subgenus Scutellaria, Section Scutellaria) [9]. The structure of **1** was established from its spectroscopic data and by comparison with related structures.



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RESULTS AND DISCUSSION

Extraction of the aerial parts of *S. seleriana* afforded after extensive chromatographic purification, the flavone oroxylin A [10] and a new *neo*-clerodane diterpenoid (**1**). The mass spectrum of compound **1** indicated a molecular formula of $C_{29}H_{40}O_{10}$. Its IR spectrum showed absorptions for hydroxyl (3525 cm^{-1}) and conjugated double bonds (1648 cm^{-1}). Strong absorption at 1788 cm^{-1} and a broad band at 1730 cm^{-1} were attributed to a γ-spirolactone and ester carbonyls, respectively.

The ^1H and ^{13}C NMR spectra of **1** (Tables 1 and 2) showed signals for two acetate groups (δ_H 1.97, 2.03, 3H each *s*; δ_C 170.9 *s*, 170.2 *s* and 20.5 *q* and 21.6 *q*) and a senecioyloxy moiety (δ_H 5.81, 1H, sept. $J = 2$ Hz, 2.20 and 1.94 3H each, *br d*, $J = 2$ Hz; δ_C 167.3 *s*, 115.3 *s*, 159.5 *s*, 20.5 *q* and 27.6 *q*). In addition, these spectra were consistent with the presence of an exocyclic methylene at C-4 (δ_H 5.15 and 4.85 both *br s*; δ_C 152.1 *s* C-4 and 107.5 *t* C-18) and a methyl group at C-5 (Me-19) (δ_H 1.30 *s*; δ_C 18.2 *q*), instead of the 4α-18 oxirane and the 19-hydroxy or acetoxymethylene groups commonly found in *neo*-clerodane diter-

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Table 1. ^{1}H NMR data for scuteselerin (**1**) 500 MHz, $CDCl_3$, TMS as int. standard)

H	δ (ppm)	J (Hz)
1	5.44 *dt*	2.5, 5.5
6	4.18 *dd*	4.5, 10
7	5.18 *d*	10
10	2.40 *d*	5.5
11	5.41 *dd*	4, 13
12α	1.79 *dd*	4, 13
12β	2.36 *t*	13
14 pro *S*	2.84 *d*	17
14 pro *R*	2.68 *d*	17
16A	4.28 *d*	9.5
16B	4.16 *d*	9.5
3H-17	1.36 *s*	
H-18-pro *Z**	5.15 *br s*	
H-18-pro *E**	4.85 *br s*	
3H-19	1.30 *s*	
3H-20	1.15 *s*	
2′	5.81 *sept* (1H)	2
3H-4′	2.20 *br d* (Me)	2
3H-5′	1.94 *br d* (Me)	2
OAc	1.97 *s*	
	2.03 *s*	

* Distinguished by NOESY spectrum.

penoids from *Scutellaria species* [11–14]. Scuteselerin (**1**) shares these features with the recently described scutebaicalin and scutedrummonin from *S. baicalensis* [11] and *S. drummondii* [7], respectively. The chemical shift observed for H-18 pro-*Z*, in compound **1** (δ_H 5.15), indicated the presence of a hydroxyl group located at C-6. A double doublet (J = 4.5 and 10 Hz) at δ 4.18 was assigned to the geminal proton of this function i.e. H-6, since it was transformed into a doublet (J = 10 Hz) upon addition of D_2O. The coupling constant of this signal indicated an *equatorial* orientation for the hydroxyl group. Inspection of a Dreiding model of **1** indicated that the hydroxyl group at C-6 exerts a deshielding effect on H-18 pro-*Z*, thus accounting for its chemical shift. A doublet at δ 5.18 (J = 10 Hz) was attributed to a geminal proton of an ester group and assigned to H-7. The coupling constant of this proton indicated an antiperiplanar disposition relative to H-6 and therefore an *equatorial* orientation for the ester moiety. Two one-proton, partially overlapped, signals at δ 5.44 (*dt*, 1H, J = 5.5 and 2.5 Hz) and δ 5.41 (*dd*, 1H, J = 4 and 13 Hz) were also assigned to the geminal protons of two additional ester groups. COSY experiments led us to locate these functionalities at the C-1 and C-11 positions.

Table 2. ^{13}C NMR data for scuteselerin (**1**) (125 MHz, $CDCl_3$, TMS as int. standard)

C	δ	C	δ
1	72.6 *d*	16	79.1 *t*
2	25.5 *t*	17	20.2 *q*
3	26.9 *t*	18	107.5 *t*
4	152.1 *s*	19	18.2 *q*
5	44.0 *s*	20	17.8 *q*
6	72.0 *d*	1′	167.3 *s*
7	74.5 *d*	2′	115.3 *d*
8	83.1 *s*	3′	159.5 *s*
9	43.9 *s*	4′	20.5 *q*
10	44.8 *d*	5′	27.6 *q*
11	71.9 *d*	OAc	170.9 *s*
12	34.5 *t*		20.5 *q*
13	77.4 *s*		170.2 *s*
14	42.6 *t*		21.6 *q*
15	174.1 *s*		

* Assignments confirmed with the aid of HMBC and HMQC spectra.

Other relevant signals in the ^{1}H and ^{13}C NMR spectra of **1** were those due to a γ-13-spiro 15,16-lactone moiety and a 8-13 ether bridge (Tables 1 and 2). These functional groups are frequently found in other *neo*-clerodane derivatives found in several *Scutellaria* spp [11, 13–16]. Two singlets at δ 1.36 and 1.15 (3H each) were assigned to Me-17 and Me-20, respectively.

The location of the senecioyloxy substituent in compound **1** was established from the HMBC spectrum, which showed a cross-peak of correlation through three bonds between the carbonyl carbon of the senecioyloxy group (δ 167.3 *s*) and the proton at δ 5.18 (H-7). The two acetate substituents must, therefore, be located at the C-1 and C-11 positions.

The relative stereochemistry depicted in **1** was firmly established from its NOESY spectrum. The H-14 pro-*S* showed a strong NOE cross-peak with Me-17 indicating a 13*R* stereochemistry. On the other hand, the H-14 pro-*R* exhibited NOE correlations with H-11 and H-12α, indicating a β-orientation for the acetate group at C-11. While H-6β axial correlated only with H-10 and H-18 pro-*Z*, the H-7α *axial* showed cross peaks with the Me-19, Me-20 and Me-17 protons. These facts in addition to the cross-peak of the Me-20 protons with H-1, indicated that H-7, Me-20, Me-17, Me-19 and H-1 were on the same side of the decalin and H-10 and H-6β *axial* were on the opposite one. These results established a *cis* junction between the B and C rings. Moreover, the NOESY spectrum showed a correlation between Me-19 with a complex signal which could be assigned by the COSY spectrum to H-2α. These results are in agreement with the relative stereochemistry proposed for scuteselerin (**1**). The chemical shifts observed, in the ^{13}C NMR spectrum of **1** for the Me-19 and Me-20 (Table 2) indicated a *trans* fusion for the A/B rings [17]. The coupling constants found for H-10 and H-1 (Table 1) and the NOE correlation between the Me-19 protons and H-2α, led us to establish that the A ring was in a boat conformation. A careful inspection of a Dreiding model of **1** indicated that in a chair conformation the acetate groups located at C-1 and C-11 must be very close in space, producing a strong electronic repulsion. This repulsion is attenuated in a boat conformation.

The absolute stereochemistry of **1** was not ascertained. However, on biogenetic grounds, we can

assume that **1** belongs to the *neo*-clerodane series like other diterpenoids isolated from *Scutellaria* spp whose absolute configuration was firmly established by X-ray diffraction analysis [4, 7, 16] or the CD exciton chirality method [11, 18].

From a chemotaxonomic point of view, it is of interest to note that scuteselerin (**1**) lacks the oxygenated substituent at C-19 commonly found in several *neo*-clerodane diterpenoids from European *Scutellaria* spp. Compound **1** shares this feature with scutebaicalin from *S. baicalensis* [11] and with the *neo*-clerodane diterpenoids isolated from the Mexican species *S. drummondii* [7] and the Chinese plant *S. rivularis* [19–20].

EXPERIMENTAL

General. Mps: uncorr.; EIMS: 70 eV, direct inlet; UV MeOH; ^{1}H and ^{13}C NMR: 500 and 125 MHz, respectively, $CDCl_3$, TMS as int. standard. *Scutellaria seleriana* was collected in the State of Queretaro (México) in August 1995. A voucher specimen (PT19185) was deposited at the herbarium of the Instituto de Biologia UNAM.

Extraction, fractionation and isolation of scuteselerin from Scutellaria seleriana. Dried and powdered aerial parts of *S. seleriana* (300 g) were extracted × 2 with Me_2CO (7 l) for 5 days at room temp. The extracts were combined and the solvent removed *in vacuo* to yield 6.8 g of a gummy residue which was partitioned between MeOH–H_2O (4:1) and C_6H_6–petrol (1:1). The less polar phase was concd *in vacuo* to yield 3.64 g of residue. The aq. MeOH fr. was concd *in vacuo*, H_2O was added and the mixt. was extracted with EtOAc. The organic extract was dried and the solvent removed to yield 1.48 g of a gum, which was subjected to vacuum chromatography over silica gel. Mixts of petrol–EtOAc of increasing polarity were used as eluents. From the frs eluted with petrol–EtOAc (4:1) oroxylin A (5 mg) was isolated. The physical data obtained (mp, MS, IR and ^{1}H NMR) were identical with those published in the literature [10]. Elution with petrol–EtOAc (3:2) afforded 8 mg of scuteselerin (**1**).

Scuteselerin. Amorphous solid, mp 104–105°; $[\alpha]_D$ −63.6° ($CHCl_3$; *c* 0.305); IR $\nu_{max}^{CHCl_3}$ cm^{-1}: 3525, 1788, 1730, 1648, 1452, 1375, 1252, 1229, 1026; UV λ_{max}^{MeOH} nm (log ε): 205 (4.21), 219 (4.23); ^{1}H NMR: Table 1; ^{13}C NMR: Table 2; MS *m/z* (rel. int.): 550 (2), 549 (5), 548 (10), 510 (10), 488 (30), 470 (5), 448 (10), 432 (20), 433 (70), 389 (20), 388 (50), 373 (35), 328 (30), 313 (30), 283 (25), 219 (50), 213 (48), 201 (90), 173 (60), 159 (38), 105 (93), 83 (100), 55 (8), 43 (15). $C_{29}H_{40}O_{10}$ requires $[M]^+$ at *m/z* 548.

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