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A glycosidic eudesmanolide from *Hyaloseris salicifolia*

Elmira C. de Riscala[a], Silvia Turbay[a], César A.N. Catalán[b,1], Luis R. Hernández[c], Pedro Joseph-Nathan[c,*]

[a]*Cátedra de Química Orgánica, Facultad de Agronomía y Zootecnia, Universidad Nacional de Tucumán. Av. Roca 1900, S.M. de Tucumán, 4000, Argentina*

[b]*Instituto de Química Orgánica, Facultad de Bioquímica, Química y Farmacia, Universidad Nacional de Tucumán. Ayacucho 491, S.M. de Tucumán, 4000, Argentina*

[c]*Departamento de Química, Centro de Investigación y de Estudios Avanzados del Instituto Politécnico Nacional, Apartado 14-740, México, D.F., 07000, Mexico*

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Abstract

The flowers of *Hyaloseris salicifolia* afforded known ivasperin, while the leaves afforded ivasperin and 2-*O*-(6′-*O*-acetyl-β-D-glucopyranosyl)-ivasperin, whose structure was determined by spectroscopic methods. © 2000 Elsevier Science Ltd. All rights reserved.

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1. Introduction

The genus *Dinoseris*, which sometimes is retained separately from *Hyaloseris* (Cabrera, 1977), is currently included in *Hyaloseris* following Espinar (1973) and Bremer (1994). The genus *Hyaloseris* was placed in the subtribe Mutisiinae by Cabrera (1977). However, Hansen (1991) does not consider subtribal classification in his review of the tribe Mutisieae and notes that the distinction between Mutisiinae and Gochnatiinae is artificial. On this basis, Bremer (1994) considers two subtribes in the Mutisieae: Mutisiinae sensu lato and Nassauviinae, indicating that Mutisiinae must be regarded as a very provisional unit. On the other hand, and according to the subtribal classification made by Cabrera (1977), a chemical review of Gochnatiinae (Catalán, Borkosky & Joseph-Nathan, 1996) shows that sesquiterpene lactones are widespread metabolites in Gochnatiinae while they are missing in Mutisiinae, the only known exception being *H. salicifolia*. However, subtribe Nassauviinae is characterized by the presence of trixanes and perezone derivatives.

In order to shed light on the chemotaxonomic placement of *Hyaloseris*, we undertook its chemical study. In a previous study of this plant, where the old synonymy *Dinoseris* was employed (Bohlmann, Zdero, King & Robinson, 1979), polyisoprene, two acetylenes and the eudesmanolide 1β-hydroxyalantolactone were reported. As a result of the present work, we report the isolation of ivasperin (**1**) (Herz & Viswanathan, 1964) and the new glycosidic eudesmanolide **2**, whose structure was determined by spectroscopic methods. This is the second time that a 6-*O*-acetyl-β-D-glucopyranosyl-eudesmanolide is found in nature, since previously only absinthifolide was reported from *Bahia absinthifolia* var. *absinthifolia*, tribe Heliantheae (Pérez C., Nava M. & Romo de Vivar, 1987).

2. Results and discussion

Although leaves and flowers of *H. salicifolia* were

* Corresponding author. Fax: +52-5747-7002.

E-mail address: pjoseph@nathan.chem.cinvestav.mx (P. Joseph-Nathan).

[1] Research Member of the National Research Council of Argentina (CONICET).

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processed separately, ivasperin (**1**) was found in both, while 2-*O*-(6′-*O*-acetyl-β-D-glucopyranosyl)-ivasperin (**2**) was isolated only from the leaves. Ivasperin (**1**) was readily identified by comparison of its ^{1}H- and ^{13}C-NMR spectra ($CDCl_3$) with literature data (Vichnewski, Shuhama, Rosanske & Herz, 1976).

Compound **2**, isolated as a white powder, possesses an α-methylene-γ-lactone ring as indicated by IR bands at 1754 and 1648 cm^{-1} and UV absorption at λ_{max} 201 nm (log ε 3.5, in MeOH). The strong IR band at 3378 cm^{-1} indicates the presence of hydroxyl groups, and the bands at 1724 and 1258 cm^{-1} are indicative of an acetate group.

The ^{13}C-NMR spectrum (Table 1) shows 23 signals. Two of them, at δ 170.1 and 20.6, are due to an acetate group, while six signals, at δ 104.6, 75.8, 73.8, 73.4, 70.1 and 63.6, indicated the presence of a glycosidic moiety. These eight signals were assigned to a 6-*O*-acetyl-β-D-glucopyranosyl residue after comparison with data reported for that residue (Yamasaki et al., 1977). The remaining 15 signals of **2** are very similar to those observed for **1**. Due to the insolubility of **2** in $CDCl_3$, the ^{1}H- and ^{13}C-NMR spectra of **1** and **2** were recorded in DMSO-d_6. A comparison of the ^{13}C-NMR spectra of both compounds revealed a close similarity of chemical shifts with the exception of C-2, which in **2** was shifted 12 ppm downfield (Table 1). Therefore, the glucosidic residue of **2** was assigned at C-2. All protonated carbon assignments were confirmed from HETCOR and DEPT experiments.

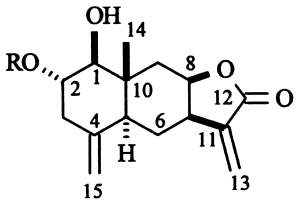
The ^{1}H-NMR spectrum of **1**, in DMSO-d_6, which was readily assigned by comparison with that recorded in $CDCl_3$, is very similar to that of **2**, except for the δ 2.98–4.32 region, where the glucosidic signals appear. The H-1, H-2 and H-7 signals were observed in the same region (Table 1) and were assigned from a COSY experiment and by irradiation of the signals at δ 2.68, 1.73 and 1.18 corresponding to H-3β, H-6α and H-6β, respectively. The signals of the 6-*O*-acetylglucoside residue were assigned with the aid of a COSY experiment and with irradiations starting from the anomeric proton doublet at δ 4.31.

The stereochemical assignments of the individual methylene protons at C-3, C-6 and C-9 for **2**, were made by comparing the observed coupling constant values, with those calculated from the minimum energy conformation structure obtained using the PCMODEL program (Burket & Allinger, 1982). The dihedral angles and coupling constant values calculated are: for CH_2-3: H-2β, H-3α = 173.5° (J = 11.2 Hz); H-2β, H-3β = 56.0° (J = 4.7 Hz); for CH_2-6: H-5α, H-6α = −61.5° (J = 3.0 Hz); H-5α, H-6β = −178.3° (J = 12.5 Hz); H-6α, H-7α = 44.0° (J = 6.0 Hz); H-6β, H-7α = 159.7° (J = 11.0 Hz); for CH_2-9, H-8α H-9α = 45° (J = 4.0 Hz) and H-8α, H-9β = −69.0° (J = 2.5 Hz). These coupling constant values are in good

Table 1
^{1}H- and ^{13}C-NMR spectral data of **1** and **2** (300 and 75.4 MHz, DMSO-d_6, TMS as international standard)[a]

	1		**2**	
	δ H	δ C	δ H	δ C
1	2.94 *d* (9)	82.4	3.16 *d* (9)	80.5
2	3.28 *ddd* (12, 9, 5.5)	69.6	3.34 *ddd* (12, 9, 5.5)	81.6
3α	1.96 *br dd* (13, 12)	42.9	2.05 *br dd* (13, 12)	41.9
3β	2.46 *br dd* (13, 5.5)		2.68 *br dd* (13, 5.5)	
4		146.5		145.9
5	1.90 *br d* (12)	43.3	1.95 *br d* (12.5)	42.9
6α	1.73 *ddd* (13.5, 7, 2.5)	26.3	1.73 *ddd* (13.5, 7, 2.5)	26.1
6β	1.18 *ddd* (13.5, 13, 12)		1.18 *ddd* (13.5, 13, 12.5)	
7	3.08 *ddd* (13, 7, 5)	38.8	3.08[b]	38.7
8	4.57 *ddd* (5, 5, 1.5)	76.3	4.58 *ddd* (5, 5, 1)	76.1
9α	1.47 *dd* (15.5, 5)	37.4	1.53 *dd* (15.5, 5)	37.1
9β	2.38 *dd* (15.5, 1.5)		2.40 *dd* (15.5, 1)	
10		37.6		37.2
11		142.3		142.2
12		169.9		169.8
13a	5.97 *br s*	120.1	5.98 *br s*	120.2
13b	5.74 *br s*		5.76 *br s*	
14	0.63 *s*	12.3	0.67 *s*	12.2
15a	4.82 *br d* (1.5)	108.2	4.82 *br s*	108.9
15b	4.51 *br d* (1.5)		4.55 *br s*	
1′			4.31 *d* (8)	104.6
2′			3.02 *dd* (9, 8)	73.8
3′			3.18 *dd* (9, 9)	75.8
4′			3.05 *dd* (9, 9)	70.1
5′			3.39 *ddd* (9, 7, 2)	73.4
6′a			4.25 *dd* (11.5, 2)	63.6
6′b			4.08 *dd* (11.5, 7)	
AcO			2.02 *s*	170.1
				20.6

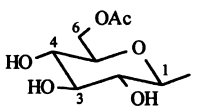
[a] J coupling constants in parentheses.
[b] Overlapped with H-4′ signal.



R

1 **H**

2



agreement with the experimentally observed values (Table 1).

A derivative of ivasperin (**1**) having a methylbutyrate residue at C-1 and an endocyclic double bond at C-3/4 has been reported from *Wunderlichia mirabilis* (Bohlmann, Ludwing, Jakupovic, King & Robinson, 1984), which is placed in the subtribe Gochnatiinae, while *H. salicifolia* is placed in the subtribe Mutisiinae. In view of these facts, it is clear that the splitting of tribe Mutisieae into subtribes and the natural position of the genera among the subtribes needs much further investigation.

3. Experimental

3.1. General

For the separation of mixtures, HPLC with a differential refractometer detector was used. The column employed was a Beckman ODS Ultrasphere (5 μm, 250 × 10 mm i.d.). Retention time (R_t) was measured from the solvent peak.

3.2. Plant material

Aerial parts of *H. salicifolia* (Griseb.) Hieronymus were collected at the flowering stage on 9 September 1996 on Highway 308, 5 km north La Merced, Catamarca province, Argentina. A voucher specimen (LIL 601271) is on deposit in the Herbarium of the Instituto Miguel Lillo, Tucumán, Argentina.

3.3. Extraction and isolation

Flowers (735 g) were extracted (2×) with $CHCl_3$ (3.5 l) at room temperature for 7 days to give 17.0 g (2.3%) of crude extract, which was suspended in EtOH (135 ml) at 55°C, diluted with warm H_2O (100 ml) and extracted (3×) successively with hexane (150 ml) and $CHCl_3$ (150 ml). Evaporation of the $CHCl_3$ extract in vacuo furnished 4.8 g of residue which was chromatographed over silica gel (180 g) using $CHCl_3$ with increasing amounts of EtOAc (0–100%); 125 frs. were collected. Frs. 52–76 (1.10 g) which showed one major spot on TLC were combined, washed several times with Et_2O, and recrystallized from EtOAc to give 651 mg of ivasperin (**1**). A portion (103 mg) was processed by HPLC (MeOH–H_2O 2 : 1, 1.7 ml min^{-1}) to give 88 mg of pure **1** (R_t 5.1 min), mp 162–164°C, reported 157–159°C (Vichnewski et al., 1976).

Leaves (820 g) were extracted (2×) with $CHCl_3$ (4 l) at room temperature for 7 days to give 35.6 g (4.3%) of crude extract, which was suspended in EtOH (320 ml) at 55°C, diluted with warm H_2O (214 ml) and extracted (3×) with hexane (300 ml) and $CHCl_3$ (400 ml). Removal of solvent from the $CHCl_3$ fraction in vacuo furnished 3.9 g of residue, which was chromatographed over silica gel (125 g) using $CHCl_3$ with increasing amounts of EtOAc (0–100%) with 97 frs. being collected. Fractions 29–60 (1.45 g) showed one major spot on TLC and were combined; the residue obtained after solvent evaporation was washed several times with Et_2O and recrystallized from EtOAc to give 931 mg of ivasperin (**1**), identical to the sample obtained above. Fractions 84–87 (117 mg) gave a solid residue which after several washing with ether and finally with EtOAc afforded 59 mg of crystalline 2-*O*-(6′-*O*-acetyl-β-D-glucopyranosyl)-ivasperin (**2**), uncorrected mp 223–227°C; UV λ_{max} (MeOH) nm (log ε): 201 (3.5); IR ν_{max} (KBr) cm^{-1} 3378, 3080, 1754, 1724, 1670, 1648, 1258; $[\alpha]_{589}$ + 42°, $[\alpha]_{578}$ + 42°, $[\alpha]_{546}$ + 50°, $[\alpha]_{436}$ + 91°, $[\alpha]_{365}$ + 149° (*c* 0.97, $CHCl_3$); EIMS (direct inlet) 20 eV *m/z* (rel. int.): 450 $[M\text{-}H_2O]^+$ (1), 432 $[M\text{-}H_2O\text{-}H_2O]^+$ (1), 407 (1), 296 (15), 293 (46), 275 (23), 247 (78), 229 (100), 219 (30), 201 (29), 183 (28), 98 (26), 43 (24).

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