Phytochemistry, Vol. 25, No. 8, pp. 1923–1926, 1986.
Printed in Great Britain.

0031–9422/86 $3.00 + 0.00
Pergamon Journals Ltd.

1-HYDROXYPLATYPHYLLIDE, A NORSESQUITERPENE LACTONE FROM *SENECIO GILLIESIANO*

F. H. GUIDUGLI, M. J. PESTCHANKER, M. S. A. DE SALMERON and O. S. GIORDANO

Departamento de Química Orgánica, Facultad de Química, Bioquímica y Farmacia, Universidad Nacional de San Luis, San Luis, Argentina

(*Revised received* 1 *October* 1985)

Key Word Index—*Senecio gilliesiano*; Compositae; norsesquiterpene lactones platyphyllide; 1-hydroxyplatyphyllide; pyrrolizidine alkaloids; retronecine; senecionine.

Abstract—A new norsesquiterpene lactone, 1-hydroxyplatyphyllide, was isolated from roots of *Senecio gilliesiano* together with the previously known platyphyllide and the common pyrrolizidine alkaloids, senecionine and retrorsine. The structures of the products were determinated by spectral and chemical means and by comparison with standards and previously reported data. The genetic pathway of fragmentation by electron impact induced fragmentation of platyphyllide and 1-hydroxyplatyphyllide have been studied with the aid of high resolution measurements and metastable descompositions by the use of linked scanning.

INTRODUCTION

As part of our continuing phytochemical study of the *Senecio* genus we have investigated the constituents of *S. gilliesiano*. In previous papers we have reported the isolation of furanoeremophilanes sesquiterpenoids [1, 2] and pyrrolizidine alkaloids [3–5]. In this paper, we report the isolation of two norsesquiterpene lactones and two pyrrolizidine alkaloids. One of the lactones has been previously isolated from *S. platyphylloides* [6], and the other one is a new compound which was characterized as 1-hydroxyplatyphyllide, while the pyrrolizidine alkaloids were identified as senecionine and retrorsine.

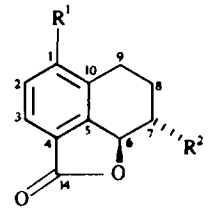
RESULTS AND DISCUSSION

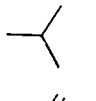
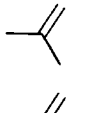
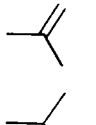
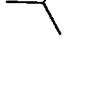
1-Hydroxyplatyphyllide, (**2**), gave a molecular ion at *m/z* 230 (36% relative intensity) which had a formula of $C_{14}H_{14}O_3$ (HRMS: 230.2637, calc.; 230.2629, measured).

The presence was shown of (i) a γ-lactone (IR ν 1760 cm^{-1}; UV $\lambda_{max}^{CHCl_3}$ 312 nm; ^{13}C NMR: δ171.0); (ii) an isopropenyl group (IR: ν 880 cm^{-1} (C=CH_2); ^{13}C NMR: δ109.6, 112.2, 20.4; ^1H NMR showed two broad singlets at δ5.1 and 5.26 attributable at two vinylic protons and 1.85 corresponding to three vinyl methyl protons); (iii) an aromatic system (IR: ν 1610, 1500, 1450, 760 and 740 cm^{-1}; ^{13}C NMR: δ153.9, 116.6, 135.1, 143.9, 148.4 and 124.4; ^1H NMR showed two aromatic protons exhibiting ortho split doublets at δ6.8 and 7.23, J = 8.5 Hz).

The remaining oxygen atom from the molecular formula was assigned to one hydroxyphenolic group on the basis of the following data (IR: ν 3380 cm^{-1}; ^1H NMR: δD_2O exchangeable *br s* at δ6.96). The hydroxyphenolic group was methoxylated (see Experimental) to give 1-methoxyplatyphyllide (**2a**) which was characterized from the following data: IR disappearance of broad band at 3380 cm^{-1}; ^{13}C NMR: δ55.9; ^1H NMR showed a sharp singlet at δ3.90. All these structural features revealed that the substance was based on a norsesquiterpene lactone skeleton like platyphyllide (**4**).

The presence of the pair of *ortho*-coupled protons in the aromatic ring allowed only two different arrangements of the hydroxy substituent. Of these the C-3 position could be eliminated since the UV spectrum did not show any shift in the presence of aluminium chloride. Thus the possibility of the presence of a chelated hydroxyl group



	R^1	R^2
1	H	
1a	H	
2	OH	
2a	OMe^{15}	
2b	OH	

was ruled out. Therefore the hydroxyl group must be situated at the C-1 position. This last position is in agreement with the biogenetic pathway proposed by Bohlmann *et al.* [6]. The possibility that the arene oxides function as intermediates during the oxidative metabolism of aromatic substrates to phenols and oxepin derivatives has been described [7, 8]. This oxidative pathway was suggested by Bohlmann *et al.* [6] to explain the biogenesis of senoxepin and liguhodgsonal.

The isolation of 1-hydroxyplatyphyllide agrees with the oxidative pathway proposed by Jerina *et al.* [7, 8] in which the non-enzymatic opening of the 1,2-oxiarene lead principally to the 1-hydroxy derivative.

On the other hand on the basis of the 400 MHz ^{1}H NMR spectrum (see Experimental) of **2**, the relative stereochemistry of H-6 and H-7 could be assigned as *trans* in agreement with the following data: H-6 at δ5.16, *br d*, $J_{6,7}$ = 11 Hz and H-7 at δ2.24, *ddd*, $J_{7,6}$ = 11 Hz, $J_{7,8\alpha}$ = 3.5 Hz and $J_{7,8\beta}$ = 13 Hz. These coupling constants are consistent with a vicinal dihedral angle of approximately 170° between H-6 and H-7. The sign of the optical rotational power of **2** suggests that it has the opposite absolute configurations at C-6 and C-7 to compound **1** [6].

On the other hand the mass spectra of platyphyllide and 1-hydroxyplatyphyllide were particularly interesting. Platyphyllide showed one fragmentation pathway with losses of hydrocarbon fragments (M − Me; M − C_2H_4; M − C_3H_4 and M − C_5H_8), leading to the base peak ($C_9H_6O_2$) at *m/z* 146, from which originated almost exclusively the CO expulsion. This suggests that the fragment at *m/z* 146 was of appreciable stability and it originated in a fragmentation process with arrangement and retention of the oxygen atoms.

The fragmentation pathway of platyphyllide from the base peak at *m/z* 146 resembled closely that of coumarine (**3**) and isocoumarine (**4**) (Scheme 1). The similarity of the mass spectra of coumarine and isocoumarine could be the result of one expulsion to lead to a unique species. Thus expulsion of the lactone CO leads both species to give one unique ion of the benzofurane type at *m/z* 118. In fact, the subspectra of benzofurane is contained in the spectra of both species.

From this data it was necessary to propose one mechanism of rearrangement for platyphyllide that leads to one structure of the isocoumarine or coumarine type. The only plausible rearrangement for the fragment at *m/z* 146 is one which retains the CO bonded to the aromatic ring. This fact, together with the metastable transitions and high resolution data, leads to the proposal of the formation of the *m/z* 146 fragment shown in Scheme 1 which agrees with a pathway of isomerization to isocoumarine.

The validity of the proposed structure was confirmed when the mass spectrum of 1-hydroxyplatyphyllide was considered in which the isocoumarine fragment at *m/z* 162 agreed with the mass spectral studies on 6-hydroxycoumarine. Both compounds yield one unique fragment of the 5-hydroxybenzofurane type at *m/z* 134. The subspectra of 5-hydroxybenzofurane was totally correlated with that of the 1-hydroxyplatyphyllide. All this evidence allows the proposal of a unique mechanism of rearrangement for one lactone.

EXPERIMENTAL

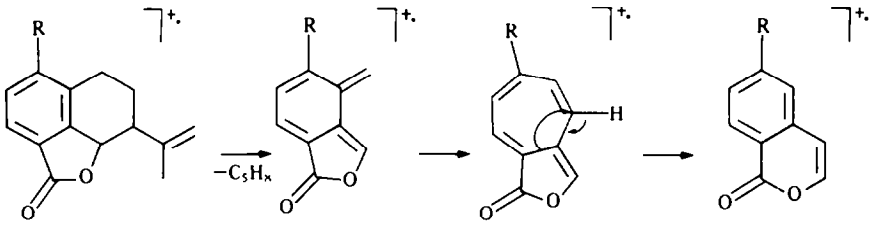
Mps were recorded on a Leitz-hot stage apparatus; the ^{13}C NMR spectra were recorded in a Bruker WP-80 in $CDCl_3$; the ^{1}H NMR spectra were recorded on a Varian EM360A in $CDCl_3$; all the mass spectra were determined utilizing a Varian MAT 112 S spectrometer at 70 eV and 0.7 mA. For the metastable decompositions the scan of the magnetic field strength (B) and the voltage of the electrostatic analyser (U_a) were linked. Thus, for the search of daughter ions U_a/B is maintained constant, while for the search of parents ions U_a/B^2 is constant. For high resolution measurements peak matching was used with an error smaller than 20 ppm and a resolution better than 6000 in the 10% valley definition. The ion source was always kept at 200° and a direct sample inlet between 60 and 150° was used [9].

Plant material. Senecio gilliesiano was collected in June 1984 by L. A. Del Vitto in El Challao, Mendoza, Argentina and identified by L. A. Del Vitto (MERL N°849).

Extraction, purification and isolation. Roots of *S. gilliesiano* (4800 g) were cut into small pieces and extracted with hot MeOH (5 L × 3). The extract was concd *in vacuo*, and the resulting residue (135.8 g) was taken up in 30% citric acid soln and extracted several times with *n*-hexane (35.2 g) and $CHCl_3$ (7.2 g). The acidic extract was made alkaline and worked up in the usual way for the alkaloid extraction [3].

The *n*-hexane extract was chromatographed on silica gel 60 for CC (150 g). Elution was carried out with solvents of increasing polarity, starting with hexane–C_6H_6 (1:1). The collected fractions (100 ml each) were checked by TLC on silica gel 60 HF_{254} (C_6H_6–dioxane–AcOH) (120:20:3) and C_6H_6–EtOAc (18:1 and 9:1), the ones of similar composition were combined and the solvent removed.

Platyphyllide (**1**). The C_6H_6 eluate fractions (17–22) yielded a



R = H —— *m/z* 214 *m/z* 146
R = HO —— *m/z* 230 *m/z* 162

Scheme 1.

Table 1. ^{1}H NMR ($CDCl_3$, 60 MHz) data for compounds **1a**, **2a** and **2b**

Proton	1a	2a	2b
2	7.46 (1H, *dd*, *J* = 8 and 8 Hz)	6.75 (1H, *d*, *J* = 8.5 Hz)	6.78 (1H, *d*, *J* = 8.5 Hz)
3	7.68 (1H, *d*, *J* = 8 Hz)	7.18 (1H, *d*, *J* = 8 Hz)	7.21 (1H, *d*, *J* = 8.5 Hz)
6	5.12 (1H, *d*, *J* = 8 Hz)	5.08 (1H, *d*, *J* = 11 Hz)	5.16 (1H, *d*, *J* = 11 Hz)
12	1.10 (3H, *d*, *J* = 1 Hz)	4.89 (2H, *br s*)	1.12 (3H, *d*, *J* = 1 Hz)
13	1.02 (3H, *d*, *J* = 1 Hz)	1.86 (3H, *s*)	1.02 3H, *d*, *J* = 1 Hz)
15	—	3.93 (3H, *s*)	—
1	7.40 (1H, *d*, *J* = 8 Hz)	—	—
OH	—	—	6.94 (*br s*) D_2O exch.

Table 2. ^{13}C NMR ($CDCl_3$, 20.0 MHz) data for compounds **1**, **2** and **2a**

Carbon	1	2	2a
1	129.6	153.9	146.3
2	122.6	116.6	111.8
3	132.0	135.1	134.3
4	124.5	124.4	124.4
5	143.9	143.9	143.9
6	80.2	81.3	79.3
7	46.1	46.1	45.9
8	25.6	25.0	24.7
9	26.5	27.0	36.6
10	148.5	148.4	150.3
11	133.5	109.4	111.3
12	112.1	112.2	111.9
13	20.4	20.4	20.2
14	170.7	171.0	168.2
15	—	—	55.9

crystalline residue (0.458 g) which was rechromatographed on silica gel 60 H (15 g). The elution was carried out with C_6H_6 and the fractions 7–12 yielded 0.395 g of a crystalline residue which on repeated crystallizations from cyclohexane yielded colourless crystals of compound **1**. The spectral and physical data of compound **1** were identical with those reported for platyphyllide [6]. ^{13}C NMR data of **1** and shown in Table 2. MS *m/z* (rel. int.): 214 $[M]^+$ (44), 146 (100), 118 (82), 90 (14), 89 (11).

Hydrogenation of **1**. Compound **1** (0.060 g) was dissolved in dry EtOAc and 0.100 g of 5% Pd on charcoal catalyst grade was added. The mixture was stirred under H_2 atmosphere for 2 hr. Then the mixture was chromatographed on alumina and the resulting product (**1a**) was crystallized in cyclohexane to give 0.042 g of **1a**. HRMS: Calc. for $C_{14}H_{16}O_2$ M_r 216.2802. Found M_r (MS) 216.2813. The ^{1}H NMR data are shown in Table 1. Other significants peaks in the MS *m/z* (rel. int.): 217 (7), 216 $[M]^+$ (39), 188 (20), 187 (6), 173 (9), 159 (6), 149 (10), 148 (95), 147 (9), 146 $[C_9H_6O_2]$ (80), 145 (8), 132 (8), 131 (9), 129 (7), 127 (6), 119 (13), 118 $[C_8H_6O]$ (100), 117 (13), 115 (19), 91 (11), 90 $[C_7H_6]$ (20), 89 $[C_7H_5]$ (15), 77 (8), 69 (49), 63 (8), 51 (9).

1-Hydroxyplatyphyllide (**2**). The C_6H_6–EtOAc (49:1) eluate fractions (29–34) yielded a crystalline residue of 0.637 g which was rechromatographed on silica gel 60 H (15 g), the elution carried out with C_6H_6–EtOAc (49:1). Fractions 8–14 yielded 0.575 g of a crystalline residue which on repeated crystallizations from cyclohexane–C_6H_6 yielded colourless crystals of compound **2**, mp 118–120° (cyclohexane–C_6H_6).

$$[\alpha]^{20} = \frac{589 \quad 578 \quad 546 \quad 436}{+42.7 \quad +45.1 \quad +59.2 \quad +197.5}$$ (MeOH; *c* 6.38 mg/ml).

^{1}H NMR (400 MHz, $CDCl_3$): δ6.80 (1H, *d*, $J_{2,3}$ = 8.5 Hz, H-2), 7.23 (1H, *d*, $J_{3,2}$ = 8.5 Hz, H-3), 5.16 (1H, *br d*, $J_{6,7}$ = 11 Hz, H-6), 2.24 (1H, *ddd*, $J_{7,6}$ = 11 Hz, $J_{7,8\beta}$ = 13 Hz, $J_{7,8\alpha}$ = 3.5 Hz, H-7), 2.16 (1H, *dddd*, $J_{8\alpha,8\beta}$ = 14 Hz, $J_{8\alpha,9\beta}$ = 7.5 Hz, $J_{8\alpha,7}$ = 3.5 Hz, $J_{8\alpha,9\alpha}$ = 8 Hz, H-8α), 1.94 (1H, *m*, H-8β), 2.74 (1H, *ddd*, $J_{9\alpha,9\beta}$ = 17.5 Hz, $J_{9\alpha,8\alpha}$ = 8 Hz, $J_{9\alpha,8\beta}$ = 10 Hz, H-9α), 3.02 (1H, *br dd*, $J_{9\beta,9\alpha}$ = 17.5 Hz, $J_{9\beta,8\alpha}$ = 7.5 Hz, H-9β), 4.89 (1H, *br s*, H-12), 4.88 (1H, *br s*, H-12'), 1.86 (3H, *br s*, H-13). ^{13}C NMR data of **2** are shown in Table 2. Other significant peaks in the MS *m/z* (rel. int.): 231 (7), 230 $[M]^+$ (36), 215 (5), 212 (16), 172 (5), 163 (12), 162 $[C_9H_6O_3]$ (100), 160 (13), 135 (5), 134 $[C_8H_6O_2]$ (55), 115 (5), 106 $[C_7H_5O]$ (5), 78 (6), 77 (5), 51 (5).

Methylation of compound **2**. Compound **2** (0.15 g) was dissolved in dry Et_2O and CH_2N_2 was added slowly, followed by the usual work-up to give 1-methoxyplatyphyllide (0.135 g) **2a**. HRMS: Calc. for $C_{15}H_{16}O_3$: M_r 244.2906, Found M_r (MS) 244.2898. The ^{1}H NMR data of **2a** are shown in Table 1, the ^{13}C NMR data in Table 2. Other significants peaks in the MS *m/z* (rel. int.): 245 (5), 244 $[M]^+$ (27), 177 (13), 176 (100), 148 (5), 147 (45), 120 (12), 119 (7), 118 (18), 115 (6), 91 (8), 90 (16), 89 (9), 77 (8), 63 (5), 51 (6).

Hydrogenation of compound **2**. Compound **2** (0.07 g) was dissolved in dry EtOAc and 0.100 g of 5% Pd on charcoal catalyst grade was added. The mixture was stirred under H_2 atmosphere for 2 hr. The mixture was chromatographed on deactivated alumina and the resulting product was crystallized in cyclohexane–C_6H_6 to give 0.035 g of **2b**. HRMS (Calc. for $C_{14}H_{16}O_3$ M_r 232.2796. Found M_r (MS) 232.2791). The ^{1}H NMR data of **2b** are shown in Table 1.

6-Hydroxycoumarine. MS *m/z* (rel. int.): 163 (10), 162 $[M]^+$ (100), 135 (8), 134 (75), 106 (10), 105 (14), 79 (6), 78 (28), 77 (16), 67 (7), 66 (5), 63 (5), 54 (7), 53 (13), 52 (11), 51 (23).

Coumarine (**3**). MS *m/z* (rel. int.): 146 $[M]^+$ (100), 118 (55), 90 (21), 89 (20).

Isocoumarine (**4**). MS *m/z* (rel. int.): 146 $[M]^+$ (100), 118 (81), 90 (33), 89 (30).

The extract containing the crude alkaloids (3.87 g) was chromatographed on a column (0.35 × 20 cm) of silica gel 60 H (65 g). Fractions of 25 ml were collected. The solvent system $CHCl_3$–MeOH–NH_3 (85:14:1), was used. According to the results of TLC, fractions 8–14 (1.28 g) were combined as were fractions 19–25 (1.76 g).

From the first fractions senecionine was identified by comparison with an authentic sample. From the fractions 19–25

retrorsine was identified by comparison with authentic sample and previously reported data.

Acknowledgements—The authors wish to express their thanks to Ing. L. A. Del Vitto (IADIZA, Mendoza) for his collaboration in the collection of plant specimens and for the botanical identification, to Ing. D. L. Anderson (INTA, Villa Mercedes, San Luis) for his botanical advice, to Dr. E. Rúveda and M. G. Sierra (IQUIOS, Rosario) for their ^{13}C NMR spectral determinations, to Dr. F. Bohlmann for the ^{1}H NMR 400 MHz determination and to Tec. E. Strazza for his technical assistance. This work was supported by grants from CONICET and SUBCYT. M. J. Pestchanker wishes to thank CONICET for a fellowship.

REFERENCES

1. Ascheri, M. S. and Giordano, O. S. (1980) *An. Asoc. Quim. Argentina* **68**, 105.
2. Salmeron, M. S. A., Kavka, J. and Giordano, O. S. (1983) *Planta Med.* **47**, 221.
3. Pestchanker, M. J., Ascheri, M. S. and Giordano, O. S. (1985) *Planta Med.* **2**, 165.
4. Pestchanker, M. J., Ascheri, M. S. and Giordano, O. S. (1985) *Phytochemistry* **24**, 7, 1622.
5. Pestchanker, M. J. and Giordano, O. S. (1986) *J. Nat. Prod.* (in press).
6. Bohlmann, F., Knoll, K. H., Zdero, C., Mahanta, P. K., Grenz, M., Suwita, A., Ehlers, A., Le Van, N., Abraham, W. R. and Natu, A. A. (1977) *Phytochemistry* **16**, 965.
7. Jerina, M. D., Daly, J. W. and Witkop, B. (1968) *J. Am. Chem. Soc.* **90**, 6523.
8. Jerina, M. D., Daly, J. W., Witkop, B., Zaltzmann-Niremberg, P. and Udentifriend, S. (1968) *J. Am. Chem. Soc.* **90**, 6525.
9. Guidugli, F. H., Pestchanker, M. J., Kavka, J. and Joseph-Nathan, P. (1984) *Org. Mass Spectrom.* **19**, 10.