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TROPANE ALKALOIDS FROM *SCHIZANTHUS GRAHAMII*

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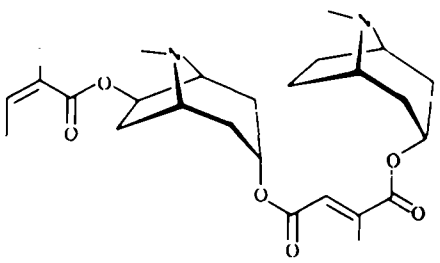
Key Word Index—*Schizanthus grahamii*; Solanaceae; tropane alkaloids; mesaconic, itaconic diesters; 3α-senecioyloxytropane.

Abstract—Three new tropanol dimers of mesaconic and itaconic acids and 3α-senecioyloxytropane, together with other known alkaloids, were isolated from *Schizanthus grahamii*. The structures of the new compounds were established by spectroscopic and chemical transformations including 2-D long-range ^{13}C-^{1}H correlations.

INTRODUCTION

Previous chemical work on *Schizanthus* (Salpiglossideae) has shown that this genus accumulates a number of tropane-derived alkaloids and may thus be considered a typical member of the Solanaceae [1–3]. Additionally, it has been found that the peculiar botanical characteristics of *Schizanthus* are also paralleled by unique structural features of its alkaloids, namely, the utilization of either angelic, tiglic, senecioic or mesaconic acids as well as the synthesis of elaborated hygrine derivatives [C. Labbé and M. Castillo, unpublished results]. In this communication we wish to report the isolation and characterization of new tropane alkaloids from *S. grahamii*, including the dimeric tropanol diesters of mesaconic acid, **1** and **2** and of itaconic acid **3** for which we propose the names schizanthine C, D and E, respectively. The new alkaloid 3α-senecioyloxytropane, together with tropine, (−)-hygroline and (+)-pseudohygroline, already reported from *S. hookeri* [1], were also isolated from this species.



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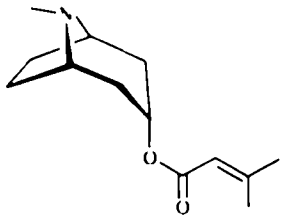

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RESULTS AND DISCUSSION

The ^{1}H NMR data of compounds **1**–**4** are presented in Table 1. The signals of diagnostic value are (a) the chemical shift and multiplicity of the olefinic proton of the corresponding acid or diacids and (b) the position and multiplicities of the skeletal protons at the point of attachment of the ester moiety to the tropane nucleus. Thus, the signals at δ6.74 and 6.72 in **1** and **2**, respectively, indicated the presence of mesaconic acid, whereas the multiplet centred at δ6.04 showed the presence of one angelic acid residue in both compounds. On the other hand, the triplet at δ5.09, integrating for two protons, showed that both tropane units present in **1** ($C_{26}H_{38}N_2O_6$) were linked to an ester group through C-3, whereas the attachment of the third ester residue at C-6 was evident by its characteristic multiplet (*dd*) at δ5.48. The mass spectrum of **1** showed the well established fragmentation pattern of 3,6-diacyloxytropane derivatives and the relative intensities of the ions at *m/z* 238, 222, 138 and 122 (Scheme 1) showed that the angeloyl residue was at C-6 [4, 5].

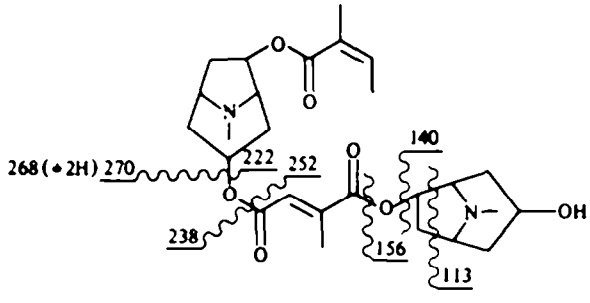
Schizanthine D (**2**) differed from schizanthine C (**1**) by an extra oxygen atom, as shown by EI mass spectrometry ($C_{26}H_{38}N_2O_7$), which also showed that the angelic acid residue was at C-6 of one of the tropane units. A triplet at δ4.06 in the ^{1}H NMR spectrum of **2** indicated a free hydroxyl group at C-3′ (also corroborated by the intense ion at *m/z* 113, not present in **1**), which together with the absorptions and multiplicities shown at δ5.69 (H-6′) 5.46 (H-6) and 5.08 (H-3) defined the substitution pattern exhibited by this alkaloid. The remaining signals in the ^{1}H NMR specta of these compounds were in agreement with the proposed structures (N-Mes, Me-mesaconic, Mes angelic, H-1 and H-5, etc.).

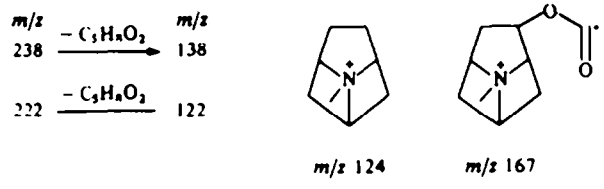
Schizanthine E (**3**), was isomeric with **2**, the main differences in its ^{1}H NMR spectrum being a new set of singlets at δ6.30, 5.72 (1H each) and 3.31 (2H) which indicated the presence of an itaconic acid residue instead

Table 1. ^{1}H NMR data of compounds 1–4 (200 MHz, $CDCl_3$, δ values from TMS*

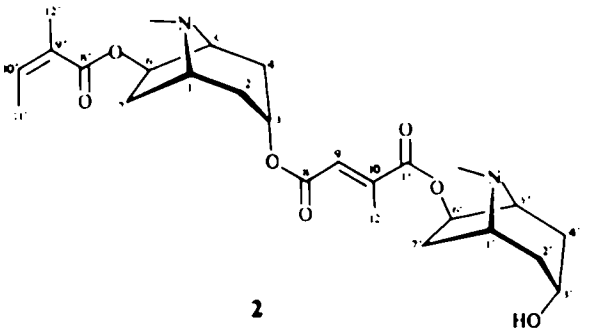
Proton	1	2	3	4
3β	5.09 *t* ($W_{1/2}$ 14)	5.08 *t* (*br*) (4.1)	5.01 *t* (*br*) (4.5)	4.96 *t* (*br*) (5.3)
1	3.18 *s* (*br*) ($W_{1/2}$ 7)	3.18 *s* (*br*) ($W_{1/2}$ 9)	3.18 *s* (*br*) ($W_{1/2}$ 11)	3.09 *s* (*br*) ($W_{1/2}$ 7)
5	3.32 *m* ($W_{1/2}$ 14)	3.30 *m* ($W_{1/2}$ 15)	3.28 *m*	
6α	5.48 *dd* (7.5, 3.2)	5.46 *dd* (7.5, 3.0)	5.42 *dd* (7.5, 3.0)	
7α	2.54 *d*† (7.5)	2.54 *d* (7.7)		
9	6.74 *q* (1.5)	6.72 *q* (1.5)	3.31 *s*	5.60 *m* ($W_{1/2}$ 3.5)
11				1.96 *d* (2.0)
12	2.30 *d* (1.5)	2.27 *d* (1.5)	6.30 *s* 5.72 *s*	1.87 *d* (1.2)
3'β	5.09 *t* ($W_{1/2}$ 14)	4.06 *t* (*br*) (4.0)	4.04 *t* ($W_{1/2}$ 11)	
1'		3.18 *s* (*br*) ($W_{1/2}$ 9)	3.18 *s* (*br*) ($W_{1/2}$ 11)	
5'	3.10 *s* (*br*) ($W_{1/2}$ 10)	3.30 *m* ($W_{1/2}$ 15)	3.28 *m*	
6'		5.69 *dd* (7.4, 3.0)	5.68 *d*‡ (7.5, 3.0)	
7'β		2.70 *dd* (13.6, 7.4)	2.66 *dd* (13.7, 7.5)	
10'	6.04 *qq* (7.1, 1.4)	6.04 *q* (7.1)	6.04 *q* (7.0)	
11'	1.96 *dq* (7.1, 1.4)	1.97 *d* (7.3)	1.94 *d* (7.3)	
12'	1.84 *m* ($W_{1/2}$ 5)	1.85 *m* ($W_{1/2}$ 5)	1.82 *m* ($W_{1/2}$ 5)	
N-Me	2.48 *s*	2.49 *s*	2.43 *s* 2.41 *s*	2.23 *s*

*J values in parentheses.
†*dd*, the other *d* overlapped by N-Me signal.
‡*dd*, the other *d* overlapped by H-12 signal.





Scheme 1. Mass spectral fragmentation of 2.



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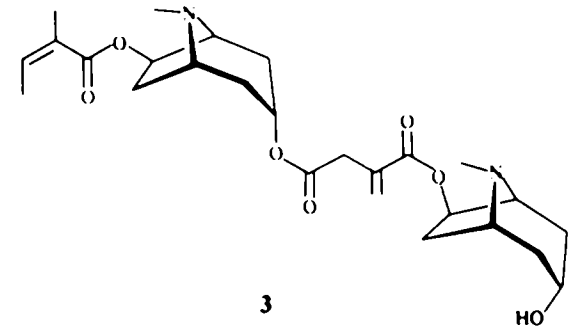

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Table 2. ^{13}C NMR data of compounds 1–6* (50 MHz, $CDCl_3$, δ values from TMS)

C	1	2	3	4	6
1	60.1	59.9	60.0	60.1	59.9
2	35.0†	34.9†	34.9†	36.7	34.4†
3	67.9	67.4	67.7	66.4	67.4
4	35.6†	35.6†	35.7†	36.7	35.8†
5	66.4	66.0	66.1	60.1	66.4
6	79.2	78.9	79.3	25.8	79.2
7	33.5	33.4	33.4	25.8	32.8
8	165.1	164.9	169.8	166.2	176.8
9	126.9	126.6	38.6	116.7	38.1
10	144.8	144.6	134.1	156.5	41.1
11	166.4	166.9	166.1	20.3	171.2
12	14.4	14.5	128.6	27.5	11.7
8′	167.9	167.7	167.9	-	175.2
9′	128.0	127.9	128.0	-	35.9
10′	137.9	137.8	137.9		26.7
11′	15.8	15.8	15.8	-	11.7
12′	20.7	20.7	20.7		16.5
1′	59.2‡	59.8	60.4		59.8
2′	36.7	36.0‡	36.6‡		35.9‡
3′	69.2	63.8	64.3		64.3
4′	36.7	36.9‡	37.4‡	—	37.2
5′	59.2‡	66.1	66.5		65.9
6′	25.9	81.3	80.8	—	80.3
7′	25.9	35.9	35.8		35.8‡
N-Me	40.5	39.9	40.4	40.4	40.9
		40.4	40.5	-	40.1

*DEPT multiplicity.
†,‡ Interchangeable.

of mesaconic acid [6]. The gross structure of 3 was then deduced on the basis of the same reasoning as above.

The ^{13}C NMR spectra of alkaloids 1–4 (Table 2) were in agreement with the proposed structures, the assignments were made taking into account the literature data for simpler tropane alkaloids [7, 8], angelic, senecioic and mesaconic acids [9, 10] and additivity rules.

On the basis of these data alone, it was not possible to assign definitively the diester linkages to the tropane residues present in compounds 1–3. The use of partial hydrolysis to solve this problem was not attempted since the resulting half-esters would also be expected to show similar uncertainties. The specification of the mesaconic ester linkages in schizantine D (2) was resolved by application of a two dimensional long-range ^{13}C–^{1}H chemical shift correlation experiment. This method, which has been successfully applied in the structural determination of ingol esters [11, 12], involves 2-D long-range ^{13}C–^{1}H correlation using polarization transfer from skeletal and acyl protons to the ester carbonyl groups. In principle, all the carbonyl groups together with their point of attachment can be assigned simultaneously. The noise-decoupled ^{13}C NMR spectrum of schizanthine D showed three signals in the ester carbonyl region at δ164.9, 166.9 and 167.7. The angelate carbonyl at 167.7 was readily assigned from the strong correlation observed in the contour plot of the 2-D long-range ^{13}C–^{1}H correlation spectrum (as well as in the corresponding cross-section) with its α-methyl group at δ1.85 [9]. It also showed a weak correlation with the signal at δ5.46 corresponding to H-6 of the tropane nucleus, thereby confirming the previous assignment made on the basis of the mass spectral data. The C-11 carbonyl group of mesaconic ester at δ166.9 was assigned on the basis of the strong 3-bond correlations observed with the protons at δ2.27 and 6.72, assigned to H-12 and H-9, respectively [10]. The remaining carbonyl group at δ164.9 showed a weak 4-bond correlation with the H-12 protons, together with 3-bond correlations with H-9 and with the skeletal H-3 proton at δ5.08, thereby establishing the attachment of the diester group in schizanthine D as depicted in 2. These and other correlations are summarized in Table 3.

Similar experiments were not performed with schizanthine E; structure 3, however, can be considered the most likely on the basis of the following observations. A comparison of the ^{1}H NMR data of tropane alkaloids esterified with α,β-unsaturated acids with that of saturated analogues, showed that in the latter examples, H-3 absorbs normally at 0.05–0.1 ppm higher field than in the former. The triplet at δ5.01 assigned to H-3 in 3 would indicate that C-3 was attached to the unconjugated carbonyl group of the itaconic ester (*cf.* corresponding values in 1 and 2). This assumption was reinforced by examination of 5, the tetrahydro derivative obtained by hydrogenation of 3, which showed almost the same value for H-3, whereas H-6′ was now shifted 0.07 ppm upfield, as expected. Furthermore, the physical and spectral data of 5 were identical in all respects with those of 6, also obtained by hydrogenation of 2 (see Experimental).

Basic hydrolysis of 2 and 3 gave (+)-(3*R*,6*R*)-(tropan-3α,6β-diol) [13] which defined their absolute configuration as depicted in the formulae.

3α-Senecioyloxytropane (4) was also isolated and its structure readily assigned on the basis of its spectroscopic properties and by synthesis.

Table 3. ^{13}C–^{1}H long-range correlations in schizanthine D (2)*

C	δ	Correlations
8'	167.7	H-12', H-6
11	166.9	H-9, H-12
8	164.9	H-12, H-9, H-3
10	144.4	H-9, H-12
10'	139.0	H-12', H-11'
9'	127.6	H-12, H-11'
9	126.6	H-12, H-9 (direct)
6'	81.1	H-6' (direct), H-4', H-1'
6	78.9	H-4, H-1
3	67.3	H-5, H-1, CH_2 at 1.7
3'	63.8	H-5', H-1', CH_2 at 1.6
5	65.9	N-Me, H-1, H-3
5'	66.0	N-Me, H-1', H-3'
1'	59.9	N-Me, H-5', H-3'
1	59.8	N-Me, H-5, H-3
12'	20.5	H-12' (direct)
12	14.3	H-12 (direct), H-9

*Experimental details of 2-D long-range of C H correlations were the same as those described in ref. [11].

EXPERIMENTAL

Schizanthus grahamii Gill was collected in Rengo, VI Region, in December. Voucher specimens are kept at the Faculty of Sciences (Universidad de Chile) Herbarium.

Isolation of alkaloids. Dried ground plant material (8.1 kg) was Soxhlet extracted with petrol and then percolated with EtOH at room temp. The EtOH extract was partitioned between acid and base in the usual manner to yield 50 g of basic material. This residue was subjected to a countercurrent distribution, in 12 funnels, between $CHCl_3$ (moving phase) and McIlvaine buffer, pH 7.1. The first five funnels contained tropine (3.3 g), (−)-hygroline (1.1 g) and (+)-pseudohygroline (1.7 g) [1]. The contents of funnels 6–9 were not analysed. The weak bases (funnels 10–12, 26 g) were partitioned again at pH 6 to yield four fractions (1.3, 1.8, 4.0 and 12 g, respectively). The third fraction was chromatographed on Al_2O_3 (Type 60/E) and eluted with mixtures of increasing polarity of petrol and EtOAc.

Schizanthine C (1). Colourless oil (70 mg). $[\alpha]_D^{25}$ − 17.2 (EtOH, *c* 0.32). IR ν_{max}^{film} cm^{-1}: 3020, 1705, 1640, 1245. ^{1}H NMR (Table 1). ^{13}C NMR (Table 2). EIMS (70 eV, direct inlet) *m/z* (rel. int.): 474.2736 ($C_{26}H_{38}N_2O_6$, calc. 474.2728) [M]$^+$ (14), 254.1378 ($C_{13}H_{20}NO_4$, calc. 254.1365) (7), 238 (2.5), 222.1523 ($C_{13}H_{20}NO_2$, calc. 222.1554) (17), 208.1289 ($C_{12}H_{18}NO_2$, calc. 208.1242) (21), 138.0928 ($C_8H_{12}NO$, calc. 138.0938) (15), 124 (100), 122 (31), 96 (38), 95 (76), 94 (94), 84 (28), 83 (39).

Schizanthine D (2). Colourless oil (243 mg). $[\alpha]_D^{25}$ − 9.7 (EtOH, *c* 0.584). IR ν_{max}^{film} cm^{-1}: 3600, 3005, 1720, 1645, 1265. ^{1}H NMR (Table 1). ^{13}C NMR (Table 2). EIMS *m/z* (rel. int.): 490.2626 ($C_{26}H_{38}N_2O_7$, calc. 490.2576) (7) [M]$^+$, 270.1363 ($C_{13}H_{20}NO_5$, calc. 270.1387) (2.7), 268 (3), 252.1202 ($C_{13}H_{18}NO_4$, calc. 252.1170) (1), 238.1409 ($C_{13}H_{20}NO_3$, calc. 238.1376) (4), 222.1533 ($C_{13}H_{20}NO_2$, calc. 222.1572) (13), 167.1044 ($C_9H_{13}NO_2$, calc. 167.1143) (10), 156.1075 ($C_8H_{14}NO_2$, calc. 156.1125) (10), 138.0920 ($C_8H_{12}NO$, calc. 138.0940) (21), 122 (46), 113 (80), 96 (52), 95 (86), 94 (100).

Schizanthine E (3). Colourless oil (60 mg). $[\alpha]_D^{25}$ − 2.3 (EtOH, *c* 0.086). IR ν_{max}^{film} cm^{-1}: 3660, 3002, 1715, 1705, 1640, 1245. ^{1}H NMR (Table 1). ^{13}C NMR (Table 2). EIMS *m/z* (rel. int.): 490.2675 ($C_{26}H_{38}N_2O_7$, calc. 490.2665) (3) [M]$^+$, 270 (4), 268 (3), 239.1481 ($C_{13}H_{21}NO_3$, calc. 239.1432) (3), 222.1510 ($C_{13}H_{20}NO_2$, calc. 222.1527) (10), 221 (5), 156.1073 ($C_8H_{14}NO_2$, calc. 156.1122) (8), 140 (10), 138 (16), 122 (28), 113 (81), 96 (61), 95 (53), 94 (100).

3α-Senecioyloxytropane (4). Colourless oil (287 mg). IR ν_{max}^{film} cm^{-1}: 3005, 1700, 1650, 1040. ^{1}H NMR (Table 1). ^{13}C NMR (Table 2). EIMS *m/z* (rel. int.): 223.1529 ($C_{13}H_{21}NO_2$, calc. 223.1498) (5) [M]$^+$, 222 (11), 208 (5), 140.1059 ($C_8H_{13}NO$, calc. 140.1043) (13), 124.1117 ($C_8H_{14}N$, calc. 124.1110) (100), 123 (12), 122 (16), 111 (18), 110 (26), 96 (33), 94 (55). Esterification of 80 mg of tropine with 0.8 ml of senecioyl chloride (reflux, 3 hr) gave 45 mg of a product identical in all respects to compound 4.

Tetrahydroschizanthine E (5). Obtained by hydrogenation of 2 (30 mg), disolved in EtOAc (20 ml), over Pd–charcoal (10%), and work up in the usual manner. Colourless oil. IR ν_{max}^{film} cm^{-1}: 3660, 3600, 1720, 1240. ^{1}H NMR (200 MHz, $CDCl_3$): δ0.88 (*t*, *J* = 7.3, H-11'), 1.11 (*d*, *J* = 6.9, H-12), 1.19 (*d*, *J* = 6.9, H-12'), 2.45 (*s*, 6H, N-Me), 3.08 (*m*, 2H, H-1, H-1'), 3.27 (*m*, 2H, H-5, H-5'), 4.03 (*t*, *br*, H-3'), 4.99 (*t*, *br*, H-3), 5.40 (*m*, H-6) 5.60 (*m*, H-6'). EIMS *m/z* (rel. int.): 494 (3.2) [M]$^+$, 272 (9), 270 (4.4), 224 (16), 140 (20), 138 (18), 122 (30), 113 (100), 96 (27), 95 (74), 94 (75).

Tetrahydroschizanthine D (6). IR, ^{1}H NMR and EIMS as 5. ^{13}C NMR (Table 2).

(+)-(3R,6R)-*Tropan-3α,6β-diol.* A soln of 22.1 mg of schizanthine D (2) in 4 ml of EtOH H_2O containing 80 mg of $Ba(OH)_2$ was heated under reflux during 10 hr. The reaction mixture was evaporated to dryness and the solid residue sublimed (170°, 0.1 mm) affording 6.8 mg of crystalline product. $[\alpha]_D^{25}$ + 11.02 (EtOH, *c* 0.136) (lit. + 24.0) [13].

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