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ACACIDIOL, A NEW NOR-TRITERPENE FROM THE SAPOGENINS OF *ACACIA CONCINNA**

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Key Word Index—*Acacia concinna*; Leguminosae; acacidiol; nor-triterpene.

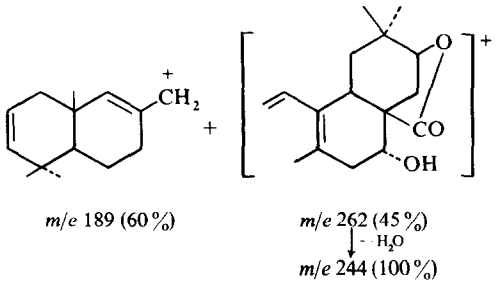
Abstract—From the neutral fraction of the acid hydrolysate of the saponin of *Acacia concinna* pods, acacic acid lactone 3β-acetate and a new nor-triterpene 'acacidiol' have been isolated. The latter is shown to be 28-noroleana-16,18-diene-3β,21β-diol.

INTRODUCTION

The isolation of acacic acid lactone, sapogenin B, machaerinic acid and a new ester aglycone of acacic acid 'acacigenin-B' has been reported [1, 2] from the pods of *Acacia concinna*. Acacigenin-B was found [2] to be a novel 21-hydroxy ester of acacic acid with a hitherto unknown monoterpene acid of tetrahydrofuranoid structure. This paper presents the isolation and identification of two more minor triterpenoid constituents, compounds E and F, from the neutral fraction of the acid hydrolysate of the saponins obtained from the pods.

RESULTS AND DISCUSSION

Compound E ($C_{32}H_{48}O_5$, M^+ 512) crystallized from alcohol as colourless light needles, mp 302–5°, $[\alpha]_D$ 0°. It gave a positive LB test for triterpenes. Its IR spectrum showed the presence of a γ-lactone (1770 cm^{-1}), a hindered hydroxyl (3605 cm^{-1}) and an ester group (1735 cm^{-1}). Its ^1H NMR spectrum showed an acetoxyl at δ 2.05 *s* and a doublet at δ 4.13, 4.18 J = 5 Hz, which is reminiscent of the lactonic proton (21-H) in acacic acid lactone [1]. Compound E gave a monoacetate, $C_{34}H_{50}O_6$, mp 230–32°, $[\alpha]_D$ −25°, which was found to be identical with acacic acid lactone diacetate (**1a**) by mmp and superimposable IR. Hence, compound E was most probably the 3-monoacetate of acacic acid lactone, since the IR signal of the OH at 3605 cm^{-1} readily suggested that the hindered OH at the C-16 position was free. Its MS gave an M^+ ion at 512, and the retro-Diels–Alder fragments at *m/e* 262 (45%) and 244 (100%) for rings D and E and *m/e* 189 (60%) for rings A and B (Scheme 1) clearly showed the presence of a C-16 hydroxyl group confirming compound E to the acacic acid lactone 3β-acetate (**1b**). The identity of compound E (by mmp and IR) with an authentic sample [1] was also confirmed. This is the first report of the isolation of acacic acid lactone monoacetate from a natural source.



Scheme 1. MS fragmentation of compound **1b**.

Compound F, mp 202–04°, $[\alpha]_D$ +82.5°, analysed for $C_{29}H_{46}O_2$. It gave a positive LB test for triterpenes and a yellow colour with TNM showing the presence of unsaturation. Its IR spectrum showed a hydroxyl function at 3400 cm^{-1} (br) and the absence of any carbonyl function. The ^1H NMR spectrum of compound F (Table 1) showed the presence of only seven methyls. There was a multiplet signal integrating for two protons at δ 3.12–3.40 assignable to the α-protons on hydroxyls. The UV spectrum of compound F showed an absorption maximum at 248 nm suggesting the presence of a heteroannular diene. The presence of two olefinic protons in the ^1H NMR spectrum at δ 5.38–5.41 as a multiplet accounted for the two trisubstituted double bonds.

Compound F formed a diacetate, $C_{33}H_{50}O_4$, mp 228–30°. Its ^1H NMR spectrum (Table 1) confirmed the presence of two acetates along with seven methyls. The

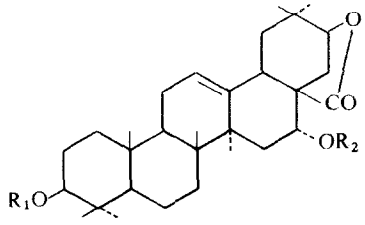
Table 1. ^1H NMR spectra in $CDCl_3$ (TMS as internal standard, chemical shifts in δ)

Compound F (**2a**)	Compound F diacetate (**2b**)	Assignment
0.80, 0.90, 0.96, 1.00, 1.04 all *s*	0.90, 0.92, 1.00, 1.07 all *s*	7C<u>H</u>$_3$
—	2.07 *s*	3,21 OCOC<u>H</u>$_3$
2.60 *m*	—	3,21 O<u>H</u>
3.12–3.40 *m*	—	3-<u>H</u>, 21-<u>H</u>
—	4.48–4.60 *m*	3-<u>H</u>, 21-<u>H</u>
5.38–5.41 *m*	5.44–5.48 *m*	16-<u>H</u>, 19-<u>H</u>

* Part V in the series "Sapogenins of *Acacia concinna*". For Part IV see Anjaneyulu, A. S. R., Rao, M. N., Row, L. R. and Sree, A. (1979) *Tetrahedron* (in press).

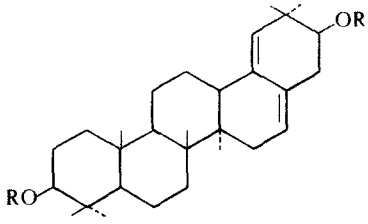
In part from the Ph.D. Thesis of A. Sree submitted to the Andhra University, Waltair, 1977.

presence of two protons at δ 4.48–4.60 *m* assignable to the protons α to the acetoxyls confirmed the nature of the two hydroxyls as secondary and unhindered. The two olefinic protons appeared at δ 5.44–5.48 *m*. A survey of literature showed that compound F was a new nor-triterpene diene diol which is now named as 'acacidiol'.*



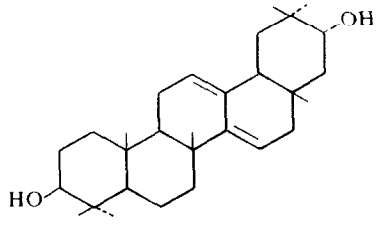
1a $R_1 = R_2 = Ac$
1b $R_1 = Ac; R_2 = H$

The co-occurrence of acacidiol with acacic acid lactone may well be taken to infer that the second hydroxyl could be at the C-16 or C-21 position. The ready formation of a diacetate favours the C-21 position for this hydroxyl in preference to the hindered C-16. Keeping the nor compound and heteroannular diene with two tri-substituted double bonds in view, two alternate structures (**2a**) and (**3**) looked most probable for acacidiol.



2a R = H
2b R = Ac

The MS of acacidiol diacetate, $C_{33}H_{50}O_4$, did not show the M^+ ion at *m/e* 510, but the ion at *m/e* 450 corresponding to $M^+ - 60$ was prominant (base peak) due to loss of an acetic acid moiety. There was further loss of 60 mu due to the loss of a second mole of acetic acid giving rise to the ion at *m/e* 390 (20%). The mass fragments at *m/e* 189 (55%, **4**) and 201 (25%, **5**) comprised the retro-Diels–Alder fragments of rings A/B and D/E. The former ion suggested the presence of the ubiquitous



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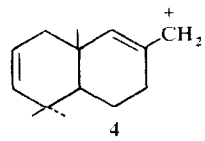

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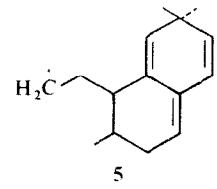

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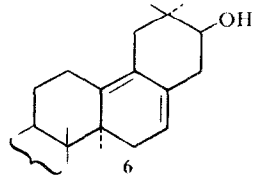
C-3 hydroxyl. The ion at *m/e* 201 was very revealing and accounted for the nor compound with the absence of a methyl group, most probably the C-28 carbon, and also suggested that the second hydroxyl was in rings D/E with the heteroannular diene also in the same part of the molecule, thus favouring the structure **2b** for the diacetate and **2a** for acacidiol.

Acacidiol gave an isomeric diene in the presence of hydrogen chloride in chloroform. The isomeric diene ($C_{29}H_{46}O_2$) crystallized from alcohol, mp 191–93° and exhibited UV max at 295 nm indicating it was a homo-annular diene. Two structures can be proposed, **6** or **7**, with a calculated value of 293 nm. The former is favoured in



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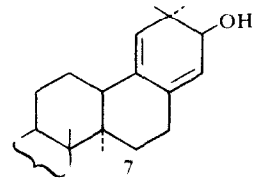

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view of the presence of one of the double bonds in the most stable tetra-substituted C-13, C-18 position.

A few nor compounds with a C_{29} skeleton are also known to coexist with their parent C_{30} compounds [3–6]. In all these cases, a reverse Prins reaction is found to operate to give rise to C_{29} compounds which are believed to be artefacts formed during the acid hydrolysis of the saponins [7, 8]. However, it is known that acacic acid is labile and forms isoacacic acid lactone [1] during the isomerization in acid media with no subsequent decarboxylation. The stability of the lactone ring in acid media may, apparently, be taken to suggest that acacidiol might not be an artefact formed during the acid hydrolysis of the saponins, although this possibility cannot be completely ruled out.

EXPERIMENTAL

Mps were uncorr. IR spectra were recorded in nujol and the UV spectra were taken in EtOH. The NMR spectra were taken in $CDCl_3$ and the values reported in δ downfield to TMS. Optical rotations were taken in $CHCl_3$. For chromatography Acme's Si gel was used.

Neutral genins. The neutral fraction [1] from the acid hydrolysate of the saponin obtained from the pods of *A. concinna* DC. was found to contain 4 spots on TLC (R_f 0.71, 0.69, 0.68 and 0.66; C_6H_6–Me_2CO, 8:2). The genin mixture (3.5 g) was adsorbed on Si gel (10 g) and chromatographed on a Si gel column (40 g) eluted with varying proportions of C_6H_6–Me_2CO

* This compound by oversight was given the name 'acaciol' in the Ph.D. thesis of A.S. This name was already given to an unidentified triterpene (see Varshney, I. P. (1969) *Indian J. Chem.* **7**, 446; Varshney, I. P., Farooq, M. O. and Naim, Z. (1961) *Arch. Pharm. (Weinheim, Ger.)* **294**, 197).

Table 2. Separation of the genin mixture from *Acacia concinna*

Fraction No.	Eluent (C_6H_6–Me_2CO)	Vol. of total fraction (l.)	Weight of compound (mg)	Compound and identification
1	99:1	4.5	—	—
2	98:2	3.0	300	A, mp 242–43°, $[\alpha]_D$ −16°, Sapogenin-B[1]
3	98:2	2.5	100	A + E*
4	97:3	3.2	130	A + E + F
5	97:3	3.0	80	E + F
6	96:4	2.5	100	F
7	95:5	6.0	2000	B, mp. 255–57°, $[\alpha]_D$ +5°, Acacic acid lactone[1]

* Acacigenin-B and machaerinic acid were compounds C and D respectively, from the acidic fraction of the acid hydrolysate of the saponins [1].

mixture collecting 100 ml fractions each time which were monitored by TLC. The fractions were grouped into 7 major fractions (Table 2).

The fractions 3, 4 and 5 contained mixtures of compounds A, E and F which were finally separated into pure compounds by PLC (20 × 20 cm plates, 0.5 mm thick, C_6H_6–Me_2CO, 7.5:2.5). After drying, the plates were sprayed with H_2O and the opaque bands clearly visible were separated, dried and extracted with $CHCl_3$ to give the individual compounds.

Identification of compound E: acacic acid lactone 3β-acetate (**1b**). Compound E crystallized from EtOH as colourless light needles, mp 302–04°, $[\alpha]_D$ 0° (*c*, 1.0). It gave a positive LB test for triterpenes. (Found: C, 74.58; H, 9.11. $C_{32}H_{48}O_5$ requires: C, 74.96; H, 9.44%). IR ν_{max}^{Nujol} cm^{-1}: 3605 (OH), 1770 (γ-lactone), 1735 (OAc), 1250, 1210, 1180, 1100, 1040, 995 and 950. ^1H NMR: δ 0.88, 0.95, 0.98, 1.10 and 1.22 (*s*, 7 CH_3), 2.05 (*s*, $OCOCH_3$), 4.13, 4.18 (*d*, *J* = 5 Hz, 21-H), 4.45 (*m*, 3-H, 16-H) and 5.42 (*m*, 12-H). MS *m/e* (rel. int.): M^+ 512 (10), 497 (3), 452 (20), 263 (15), 262 (45), 245 (20), 244 (100), 203 (22), 201 (15), 189 (60), 187 (10), 177 (80) and 176 (40).

Compound E acetate: acacic acid lactone diacetate (**1a**). Compound E (50 mg) on acetylation with Ac_2O–Py at 100° on a steam-bath for 3 hr gave an acetate which crystallized from MeOH as colourless needles, mp 230–32°, $[\alpha]_D$ −25° (*c*, 1.0). (Found: C, 73.13; H, 9.10. $C_{34}H_{50}O_6$ requires: C, 73.61; H, 9.08). IR ν_{max}^{Nujol} cm^{-1}: 1780 (γ-lactone), 1735 br (OAc), 1260, 1230, 1200, 1180, 1160, 1140, 1100, 1060, 1020, 990 and 920. Mmp with authentic acacic acid lactone diacetate was undepressed.

Compound F: acacidiol (**2a**). Compound F crystallized from EtOH–$CHCl_3$ as colourless small prisms, mp 202–04°, $[\alpha]_D$ +82.5° (*c*, 0.8). It gave a positive LB test for triterpenes. (Found: C, 81.8; H, 10.61. $C_{29}H_{46}O_2$ requires: C, 81.63; H, 10.87%). IR ν_{max}^{Nujol} cm^{-1}: 3400 br (OH), 1620, 1450, 1385, 1360, 1340, 1200, 1130, 1090, 1070, 1050, 1030, 990, 970, 910, 820 and 790. UV λ_{max} nm: 248 (log ε 3.21).

Compound F diacetate: acacidiol diacetate (**2b**). Compound F (50 mg) on acetylation with Ac_2O–Py at 100° for 3 hr gave an acetate which crystallized from EtOH as colourless light needles, mp 228–30°, $[\alpha]_D$ +38°. (Found: C, 77.38; H, 9.95. $C_{43}H_{50}O_4$ requires: C, 77.60; H, 9.87%). IR ν_{max}^{Nujol} cm^{-1}: 1730 (OAc), 1370, 1345, 1240, 1205, 1030, 990, 972 and 810. MS *m/e* (rel. int.): M^+ 510 (not observed), 452 (3), 451 (40), 450 (100), 435 (3), 390 (20), 201 (25), 190 (40), 189 (55), 187 (10), 177 (40) and 175 (30).

Isomerization of compound F. Compound F (**2a**, 25 mg) was dissolved in pure $CHCl_3$ (10 ml) and dry HCl gas was passed through for *ca* 10 min. The solvent was removed *in vacuo* and the product crystallized from EtOH as colourless small needles of isocompound F (**6**, 20 mg), mp 191–93°, UV λ_{max} nm: 295 (log ε 3.84). (Found: C, 81.48; H, 10.61. $C_{29}H_{46}O_2$ requires: C, 81.63; H, 10.87%).

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