Triterpenoid Glycosides of *Corchorus acutangulus* Lam.

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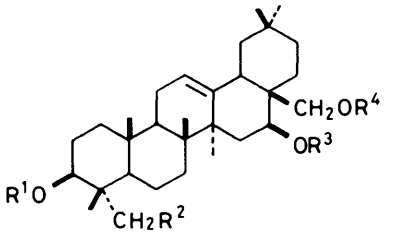
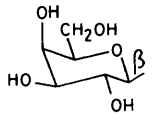
Four new triterpenoid glycosides, corchorusins A, B, C, and D, isolated from the aerial part of *Corchorus acutangulus* Lam. were respectively defined as longispinogenin 3-*O*-β-D-galactopyranoside (**1**), saikogenin F 3-*O*-β-D-galactopyranoside (**12**), 23-hydroxylongispinogenin 3-*O*-β-D-galactopyranoside (**6**), and saikogenin E 3-*O*-β-D-glucopyranosyl(1→2)-β-D-galactopyranoside (**15**) based on their spectral properties and some chemical transformations.

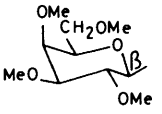
Corchorus acutangulus Lam. (syn. *C. aestuans* Linn.) (Tiliaceae) is a medicinal plant occurring throughout the hotter parts of India. Unlike its sister species *C. capsularis* and *C. olitorious* which yield the jute of commerce in India, it is a wild species rarely used for extracting fibre.[1] The isolation of digitalis glycosides from the plant has been reported.[2] This paper reports the isolation and structure elucidation of four new triterpenoid glycosides of potential biological interest.

The Bu^{n}OH-soluble fraction of an MeOH extract of the leaves of *C. acutangulus* on repeated silica gel column chromatography and reversed-phase h.p.l.c. separation led to the isolation of five triterpenoid glycosides which we have called corchorusins A, B, C, D, and E according to the increasing order of their polarity. Corchorusin E was not obtained in sufficient quantity for its structural study.



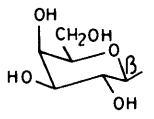
(**1**) R^1 =  , $R^2 = R^3 = R^4 = H$

(**2**) $R^1 = R^2 = R^3 = R^4 = H$

(**3**) R^1 =  , $R^2 = H$, $R^3 = R^4 = Me$

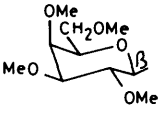
(**4**) $R^1 = R^2 = H$, $R^3 = R^4 = Me$

(**5**) $R^1 = Ac$, $R^2 = H$, $R^3 = R^4 = Me$

(**6**) R^1 =  , $R^2 = OH$, $R^3 = R^4 = H$

(**7**) $R^1 = R^3 = R^4 = H$, $R^2 = OH$

(**8**) $R^1 = R^3 = R^4 = Ac$, $R^2 = OAc$

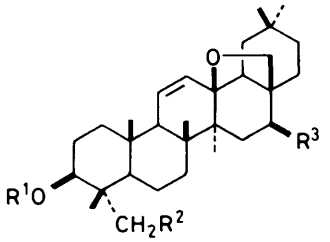
(**9**) R^1 =  , $R^2 = OMe$, $R^3 = R^4 = Me$

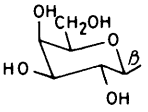
(**10**) $R^1 = H$, $R^2 = OMe$, $R^3 = R^4 = Me$

(**11**) $R^1 = Ac$, $R^2 = OMe$, $R^3 = R^4 = Me$

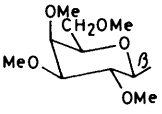
Corchorusin A (**1**) gave positive Liebermann–Burchard and Molish tests, indicating it to be a triterpene glycoside. On acid hydrolysis compound (**1**) yielded a genin (**2**) and a carbohydrate constituent, identified by paper chromatography and g.l.c. as D-galactose by comparison with an authentic sample. The physical data (m.p. and $[\alpha]_D$) of genin (**2**) indicated that it was identical with longispinogenin.[3] The identity was finally confirmed by comparison of its ^{13}C n.m.r. data with those reported[4] for authentic longispinogenin (**2**). The glycoside (**1**) on treatment with sodium hydride–methyl iodide in hexamethylphosphoramide (HMPA) afforded the permethylate (**3**), which showed in its ^1H n.m.r. spectrum a signal at δ 4.20 (1 H, d, *J* 7 Hz) assignable to the 1-H of galactose, thus suggesting the β-configuration ($^4C_1$ conformation) at the anomeric centre of the galactose. The anomeric configuration was further confirmed by the application of Klyne's rule of molecular rotation.[5] The attachment of the sugar unit to 3-OH of compound (**2**) was revealed by hydrolysis of the permethylate (**3**) which resulted in the formation of 2,3,4,6-tetra-*O*-methyl-D-galactose and 16,28-di-*O*-methyl-longispinogenin (**4**) which afforded an acetate (**5**). The ^1H n.m.r. spectrum of compound (**5**) showed a signal at δ 4.85 (1 H, dd, *J* 10.5 and 6 Hz) assignable to the proton at C-3 bearing the AcO group.[6] The linkage was also supported by the ^{13}C n.m.r. data of compound (**1**) which were found to be compatible with the structure shown. The signal assignments were straightforward on comparison with those of compound (**2**) and methyl galactoside.[7] Thus corchorusin A is longispinogenin 3-*O*-β-D-galactopyranoside (**1**). Corchorusin B (**12**) on hydrolysis yielded D-galactose as the sugar constituent and a triterpene agylcone which displayed triple u.v. maxima at 241, 251, and 260 nm, characteristic of a heteroannular diene system, and it was eventually characterised as saikogenin A[8] (**18**) by comparison of its m.p., $[\alpha]_D$, and ^1H n.m.r. data with those of an authentic sample. Compound (**18**) was shown to be an artifact produced during acid hydrolysis of the saponins isolated from *Bupleurum falcatum*.[9] Corchorusin B (**12**) did not exhibit any u.v. absorption above 205 nm, indicating absence of a heteroannular diene system, which ruled out the possibility of compound (**18**) being the genuine aglycone of compound (**12**). Treatment of corchorusin B (**12**) with sodium metaperiodate followed by hydrolysis with ethanolic potassium hydroxide under nitrogen[10] afforded a genin, which was characterised as saikogenin F (**13**) by comparison of its physical and spectral data with those of an authentic sample.[9]

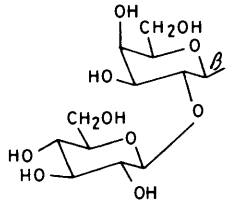
The molecular weight of compound (**12**) was determined by fast-atom bombardment mass spectrometry (FAB m.s.).[11–14] The positive FAB spectrum showed characteristic ions at *m/z* 727, 657, 635, 617, and 455 assigned to $[M\,H + Glyc]^+$, $[M + Na]^+$, $[M + H]^+$, $[(M + H) - H_2O]^+$, and $[(M + H) - gal]^+$ respectively. The negative FAB spectrum showed only three ions, at *m/z* 725, 633, and 471, ascribed to $[(M - H) +$



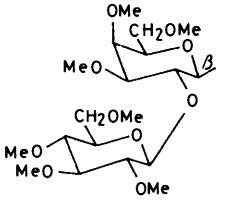
(12) R^1 =  . $R^2 = R^3 = OH$

(13) $R^1 = H$. $R^2 = R^3 = OH$

(14) R^1 =  . $R^2 = R^3 = OMe$

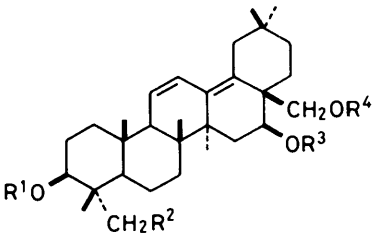
(15) R^1 =  . $R^2 = H$, $R^3 = OH$

(16) $R^1 = R^2 = H$. $R^3 = OH$

(17) R^1 =  . $R^2 = H$, $R^3 = OMe$

glyc]$^-$, $[M - H]^-$, and $[(M - H) -$ galactosyl $+ H]^-$ respectively. The glycoside (**12**) furnished a permethylate (**14**) which exhibited in its ^{1}H n.m.r. spectrum the anomeric proton signal at δ 4.20 (1 H, d, *J* 7 Hz), indicating β-linkage ($^4C_1$ conformation) for the galactose. The permethylate (**14**) on hydrolysis yielded 2,3,4,6-tetra-*O*-methyl-D-galactose and 16,-23,28-tri-*O*-methylsaikogenin A (**19**), characterised by comparison of its physical data with those of an authentic sample.[6] The formation of compound (**19**) established the sugar linkage at C-3 of compound (**12**) as shown. The ^{13}C n.m.r. data of compound (**12**) (Table) also supported the assigned structure. Consequently, corchorusin B is saikogenin F3-*O*-β-D-galactopyranoside (**12**).

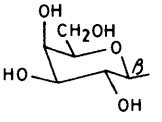
Corchorusin C (**6**) on acid hydrolysis liberated D-galactose and an aglycone (**7**), $C_{30}H_{50}O_4$ (M^+ 474), which on acetylation with acetic anhydride and pyridine furnished a tetra-acetate (**8**) $C_{38}H_{58}O_8$ (M^+ 642). The mass spectra of compounds (**7**) and (**8**) exhibited retro-Diels–Alder fragments characteristic of Δ^{12}-oleanene or ursene skeletons.[15] The locations of two hydroxy groups in the part containing rings A/B and of the other two in rings D/E were indicated by the appearance of the characteristic fragment ions. These data and particularly the ^{13}C n.m.r. δ-values of compound (**7**) (Table) disclosed its structure as being that of 23-hydroxylongispinogenin (**7**). Compound (**7**) is reported to have been prepared from quilaic acid.[16] The physical data of compound (**7**) were comparable with those reported for the synthetic compound. The permethylate (**9**) of compound (**6**) showed in its ^{1}H n.m.r. spectrum only one anomeric proton, at δ 4.20 (1 H, d, *J* 7 Hz), indicating a β-linkage ($^4C_1$ conformation) of the galactose. Acid hydrolysis of the permethylate (**9**) yielded 2,3,4,6-tetra-*O*-methyl-D-galactose and 23-methoxy-16,28-di-*O*-methyl-longispinogenin (**10**) which formed an acetate (**11**) whose ^{1}H n.m.r. spectrum displayed a characteristic signal at δ 4.90 (1 H, dd, *J* 10.5 and 6 Hz) attributable to the proton on C-3 bearing the AcO group. Thus the connecting point of the galactose in corchorusin B (**6**) was demonstrated. The ^{13}C n.m.r. data of compound (**6**) (Table) were also found to be compatible with the structure 23-hydroxylongispinogenin 3-*O*-β-D-galactopyranoside (**6**) deduced by the chemical methods.



(18) $R^1 = R^3 = R^4 = H$, $R^2 = OH$

(19) $R^1 = H$, $R^2 = OMe$, $R^3 = R^4 = Me$

(20) $R^1 = R^2 = R^3 = R^4 = H$

(21) R^1 =  , $R^2 = R^3 = R^4 = H$

(22) $R^1 = R^2 = H$, $R^3 = R^4 = Me$

Corchorusin D (**15**) did not show any u.v. absorption above 205 nm. However, on acid hydrolysis it yielded D-glucose and D-galactose as carbohydrate constituents, and a genin (**20**) which displayed three u.v. maxima characteristic of a heteroannular diene. It was apparent, therefore, that the genin (**20**) was an artifact formed during acid hydrolysis of compound (**15**) and it was characterised as saikogenin C (**20**) by comparison of its physical and spectral data with those of an authentic sample.[8] Treatment of compound (**15**) with sodium metaperiodate followed by hydrolysis with ethanolic potassium hydroxide under nitrogen led to the formation of the genuine aglycone, saikogenin E (**16**).

The FAB m.s. of (**15**) was helpful in the determination of its molecular weight. Although the positive FAB spectrum did not show the $[M]^+$ or $[MH]^+$ ion it did exhibit discernible ion peaks at *m/z* 763, 745, 583, 439, 421, and 403 assigned to $[MH - H_2O]^+$, $[MH - 2H_2O]^+$, $[MH -$ (1 hexose + 2 H_2O)]$^+$, $[MH -$ (2 hexose + H_2O)]$^+$, $[MH -$ (2 hexose + 2 H_2O)]$^+$, and $[MH -$ (2 hexose + 3 H_2O)]$^+$ respectively. In the negative FAB spectrum the $[M - H]^-$ ion appeared as the base peak and the other two significant ions at *m/z* 871 and 617 were ascribed to $[M - H +$ glyc]$^-$ and $[M - H + 1$ hexose]$^-$ respectively. It appears that formation of glycerol adduct and frequent losses of 18 units (H_2O) particularly in the positive FAB of compounds of this type are of common occurrence. As both constituents (D-galactose and D-glucose) of the carbohydrate moiety of compound (**15**) are hexoses their sequence could not be determined by the FAB m.s. However, the sequence could be deduced by generation of the prosapogenin (**21**) by partial hydrolysis, which on acid hydrolysis afforded the acid-rearranged aglycone, saikogenin C (**20**), and the only sugar constituent, identified as D-galactose. Consequently it was evident that in compound (**15**) galactose is directly linked to saikogenin E (**16**), and that glucose is present as the terminal sugar. The corchorusin D permethylate (**17**),

Table. ^{13}C N.m.r. chemical shifts (δ_C; ±0.1 p.p.m.) of longispinogenin (**2**), saikogenin F (**13**), 23-hydroxylongispinogenin (**7**), saikogenin E (**16**), corchorusin A (**1**), corchorusin B (**12**), corchorusin C (**6**), and corchorusin D (**15**) measured in C_5D_5N

Carbon	(2)	(1)	(13)	(12)	(7)	(6)	(16)	(15)
1	38.8	39.3	38.3	38.4	38.4	38.6	38.5	38.7
2	26.8	26.1	25.8	25.6	26.5	25.5[a]	26.6	25.7[a]
3	78.8	88.6	75.3	82.1	76.5	82.3	78.5	88.9
4	38.9	38.9	42.1	43.3	41.9	43.0	39.1	39.7
5	55.2	55.8	48.8	47.3	49.2	47.4	54.4	55.5
6	18.3	18.4	17.6	17.4	18.5	17.9	17.9	17.9
7	32.6	33.2	31.4	31.4	32.6	32.4	31.7	31.9
8	39.8[a]	40.0[a]	41.9	42.0	40.0[a]	40.0	41.8	42.2
9	46.9	47.0	52.8	52.9	47.1	46.9	52.7	53.0[b]
10	37.0	36.7	36.2	36.2	37.0	36.5	36.3	36.3
11	23.5	24.0	133.0	132.2	23.6	23.6	132.5	132.0
12	122.3	122.3	129.8	130.7	122.6	122.3	130.1	131.0
13	143.2	143.7	84.2	84.0	143.2	143.6	84.0	83.9
14	43.7	43.8	45.3	45.6	43.8	43.7	45.9	45.7
15	36.0	36.5	35.3[a]	36.2[a]	36.3	36.3	35.7[a]	36.0
16	67.5	66.8	64.5	64.2	67.5	66.7	64.3	64.2
17	40.2[a]	40.7[a]	46.4	46.7	40.5[a]	40.6	46.2	46.9
18	44.7	44.5	52.0	52.0	44.6	44.4	51.8	52.2[b]
19	46.9	47.0	37.5	37.7	47.0	46.9	37.9	37.9
20	30.7	30.8	31.5	31.4	30.9	30.7	31.5	31.5
21	33.8	34.1	34.5[a]	35.7[a]	33.8	33.9	34.2[a]	34.8
22	26.2	26.3	25.3	25.6	26.0	25.9[a]	25.2	26.5[a]
23	28.0	28.1	69.8	64.2	71.3	64.7	27.6	27.9
24	16.0	16.8	11.3	12.7	11.4	13.1	15.7	16.0
25	15.6	15.5	18.4	18.5	16.0[b]	16.0[b]	17.8	18.0
26	16.7	16.8	19.5	19.7	16.7[b]	16.8[b]	19.5	19.8
27	26.8	26.9	20.7	20.7	26.8	26.9	20.6	20.8
28	70.8	69.0	72.6	72.8	70.0	68.9	72.3	72.9
29	33.0	32.9	33.5	33.5	33.1	33.0	33.8	33.6
30	23.9	23.8	23.8	23.8	24.1	23.9	23.7	23.8
gal-1		106.8		105.7		105.7		105.6
gal-2		72.8		73.0		72.8		81.5
gal-3		75.0		74.9		74.9		71.8
gal-4		69.9		69.9		69.7		69.9
gal-5		76.1		76.0		76.0		76.5
gal-6		62.0		62.0		61.9		62.0
glc-1								105.0
glc-2								75.2
glc-3								77.9
glc-4								71.8
glc-5								77.6
glc-6								62.8

[a,b] Signals may be interchanged within each vertical column. gal = galactose, glc = glucose.

prepared as described for other permethylates, on acid hydrolysis liberated 2,3,4,6-tetra-*O*-methyl-D-glucose and 3,4,6-tri-*O*-methyl-D-galactose as sugar moieties, and 16,28-di-*O*-methylsaikogenin C (**22**), identified by comparison of its physical data with those of an authentic sample.[6] The ^1H n.m.r. spectrum of compound (**17**) exhibited signals assignable to two anomeric protons, at δ 4.28 (1 H, d, *J* 7 Hz) and 4.72 (1 H, d, *J* 7 Hz), indicating two β-linkages ($^4C_1$ conformation). The ^{13}C n.m.r. spectrum of the permethylate (**15**) (Table) also supports the structure shown. The signals of saikogenin E (**16**) were assigned by comparison with literature data.[4] Assignments of the signals of compound (**15**) were made by comparison with the ^{13}C data of both compound (**21**) and methyl sugars[7] using known chemical-shift rules[17] and glycosylation shifts.[18,19] Thus corchorusin D was characterised as saikogenin E 3-*O*-β-D-glucopyranosyl(1→2)-β-D-galactopyranoside (**15**).

It is noteworthy that saikosaponins containing various saikogenins as aglycones and different combinations of fucose, glucose, and rhamnose, isolated so far only from Japanese *Bupleurum falcatum* L.[6,20–22] (Umbelliferae), have been reported to have anti-inflammatory[23] and antiviral[24] activity. *Corchorus acutangulus* appears to be the only alternative plant so far, of a different genus and family, which contains saikosaponin-like compounds. Moreover, the activity profile of the corchorusins are of interest in view of their structural similarity with the saikosaponins.

Experimental

M.p.s are uncorrected t.l.c. was carried out on silica gel 60 HF_{254} (E. Merck) with the following solvent systems: (A) benzene–$CHCl_3$–EtOAc (1 : 2 : 2); (B) Water (2 ml)–$CHCl_3$ (60 ml); MeOH was then added till the solution became clear. Paper chromatography (p.c.) for sugars was done on Whatman paper No. 1 with solvent system (C): Bu^nOH–C_5H_5N–water (6:4:3); other solvent systems are detailed in the text. A saturated solution of aniline oxalate in water was used as staining agent. G.l.c. was performed on a Hewlett-Packard model 5730A instrument with the following columns: (i) ECNSS-M, 3% on Gaschrome at 190 °C for alditol acetates; and (ii) OV-225 on Gaschrome Q at 195 °C for partially methylated alditol acetates. Optical rotations were measured on a Perkin-Elmer automatic polarimeter; i.r. spectra were recorded in Nujol

mulls on a Perkin-Elmer model 177 instrument. ^{1}H N.m.r. spectra were recorded on a JEOL FX-100 (99.6 MHz) instrument for $CDCl_3$ or C_5D_5N solutions. ^{13}C N.m.r. spectra were recorded on a JEOL FX-100 Fourier-transform spectrometer operating at 25.05 MHz, for $CDCl_3$ or C_5D_5N solutions with tetramethylsilane as internal standard. Electron-impact mass spectra were recorded on a Hitachi model RMU-6L mass spectrometer. Fast-atom bombardment mass spectra were obtained on a Kratos MS-80 RFA mass spectrometer, operating with a potential of 5—8 kV applied to the xenon gun with glycerol as matrix. H.p.l.c. analysis was performed on a spectra-Physics model 8000B instrument with a Spherisorb S-10-ODS reversed-phase column (25 cm length, i.d. 10 mm) and a Micromeritics 771 refractive-index detector. Light petroleum refers to the fraction boiling in the range 60—80 °C.

Isolation of the Glycosides.—The air-dried powdered aerial part of *C. acutangulus* (1 kg) was successively extracted with light petroleum, chloroform, and methanol. The methanolic extract, on removal of the solvent under reduced pressure, yielded a viscous dark brown mass (40 g). This was extracted with Bu^{n}OH and the extract was washed with water to remove inorganic impurities, then evaporated to dryness under reduced pressure to give a viscous mass (25 g). This extract was chromatographed on silica gel (500 g). Graded elution was effected with light petroleum, followed by light petroleum–chloroform (50:50) and chloroform–methanol (various ratios, see below). A total of 104 fractions (each 250 ml) was collected and fractions giving similar spots on t.l.c. were combined. Fractions 39—45, eluted with $CHCl_3$—MeOH (90:10), yielded crude β-sitosterol glucoside (0.4 g). Fractions 46—52 [$CHCl_3$–MeOH (90:10)] furnished crude corchorusin A (0.7 g). Fractions 53—58 [$CHCl_3$–MeOH (85:15)] yielded a mixture of corchorusins B and C (1.2 g), and fractions 67—95 [$CHCl_3$–MeOH (80:20)] afforded crude corchorusin D (0.6 g). Fractions 96—104 [$CHCl_3$–MeOH (80:20)] yielded a mixture of corchorusins D and E (0.9 g). The mixture of corchorusins B and C was effectively separated into its two constituents by h.p.l.c. on a reversed-phase Spherisorb S-10-ODS column with solvent system MeOH–water (70:30). Thus corchorusin B (0.5 g) and corchorusin C (0.3 g) were obtained. Although the mixture of corchorusin D and E could be separated by h.p.l.c., corchorusin E was not obtained in a sufficient quantity for further analysis.

β-Sitosterol Glucoside.—The crude β-sitosterol glucoside was purified by rechromatography and crystallised from $CHCl_3$–MeOH to afford crystals (0.25 g), m.p. 287—289 °C (decomp.); $[\alpha]_D$ −37° (*c* 0.5 in pyridine).

Corchorusin A (**1**).—The crude corchorusin A, on further purification by rechromatography followed by crystallisation from MeOH–$CHCl_3$, yielded *microneedles* (0.45 g), m.p. 282—284 °C; $[\alpha]_D$ +22.5° (*c* 0.25 in MeOH) (Found: C, 69.6; H, 9.7. $C_{36}H_{60}O_8$ requires C, 69.64; H, 9.74%).

Hydrolysis of Corchorusin A (**1**).—Compound (**1**) (0.2 g) was hydrolysed with 2M HCl in aq. MeOH (50 ml) at water-bath temperature for 4 h. The usual work-up followed by chromatographic purification on a silica gel column gave longispinogenin (**2**), which was crystallised from MeOH as prisms (70 mg), m.p. 218—220 °C; $[\alpha]_D$ +67° (lit.,[3] m.p. 222—223 °C; $[\alpha]_D$ +68°). Its i.r., ^{1}H n.m.r., and m.s. data were comparable to those of an authentic sample.

The filtrate from the hydrolysate was neutralised with Ag_2CO_3, then filtered, and a portion of the filtrate was concentrated under reduced pressure and tested for carbohydrate by paper chromatography with the solvent system Bu^{n}OH–C_5H_5N–water (6:4:3). Only one spot, corresponding to D-galactose, was obtained. The other portion of the concentrated filtrate was reduced with $NaBH_4$ and worked up in the usual manner. The residue was acetylated with Ac_2O–pyridine (1:1) at water-bath temperature for 1 h, dried *in vacuo*, purified by chromatography over silica gel, and subjected to g.l.c. analysis on column (i). Only one peak, corresponding to D-galactitol acetate, was obtained.

Permethylation of Corchorusin A (**1**) *and Hydrolysis.*—A solution of compound (**1**) (100 mg) in HMPA (12 ml) was treated with NaH (500 mg) and MeI (12 ml) at room temperature for 3 h. The reaction mixture was extracted with diethyl ether, and the extract was washed with water, dried over anhydrous Na_2SO_4, and evaporated to yield a gummy residue, which was purified by chromatography over silica gel to give the permethylate (**3**) (85 mg) as a white powder, m.p. 116—118 °C; i.r. (no hydroxy absorption) δ($CDCl_3$) 0.84 (3 H, s), 0.89 (3 H, s), 0.92 (3 H, s), 0.96 (3 H, s), 1.00 (3 H, s), 1.04 (3 H, s), 1.16 (3 H, s), 3.28, 3.36, 3.40, 3.52, 3.56, and 3.64 (6 × OMe), 4.20 (1 H, d, *J* 7 Hz, 1-H of galactose unit), and 5.20 (1 H, t-like, 12-H).

The permethylated product (**3**) (30 mg) was hydrolysed on being refluxed with 2M-HCl in aq. MeOH (10 ml) for 3 h. The reaction mixture was cooled, and evaporated to dryness under reduced pressure, the residue was dissolved in water, and the mixture was filtered. The filtrate was worked up in the usual way and the product obtained was subjected to g.l.c. analysis on column (ii). Only one peak was detected, and was identified as that of 2,3,4,6-tetra-*O*-methyl-D-galactitol diacetate (R_t 1.19 min) by comparison with an authentic sample. The residue on chromatographic purification yielded the *partially methylated aglycone* (**4**), m.p. 154—156 °C; $[\alpha]_D$ +66.0° (*c* 0.5 in $CHCl_3$); δ($CDCl_3$) 0.76 (3 H, s), 0.84 (3 H, s), 0.88 (3 H, s), 0.92 (3 H, s), 0.96 (3 H, s), 1.20 (3 H, s) (together 6 × Me), 3.28 (3 H, s, OMe), 3.36 (3 H, s, OMe), and 5.32 (1 H, t-like, 12-H) (Found: C, 78.5, H, 11.0. $C_{30}H_{50}O_3$ requires C, 78.55; H, 10.99%).

The partially methylated compound (**4**) (10 mg) furnished the acetate (**5**) with Ac_2O (0.5 ml) and pyridine (0.5 ml) at 100 °C for 2 h. Work-up in the usual way yielded the *acetate* (**5**), m.p. 144—146 °C; $[\alpha]_D$ +60.6° (*c* 0.6 in $CHCl_3$) (Found: C, 77.2; H, 10.7. $C_{34}H_{56}O_4$ requires C, 77.22; H, 10.67%).

Corchorusin B (**12**).—This was crystallised from MeOH as *microprisms*, m.p. 278—280 °C; $[\alpha]_D$ +74.3 (*c* 0.6 in MeOH); FAB m.s. (positive) *m/z* 727 $[M$H + glyc$]^+$ (4%), 657 $[M$ + Na$]^+$ (6), 635 $[M + H]^+$ (18), 617 $[M + H - H_2O]^+$ (100), 599 $[M + H - 2\ H_2O]^+$ (29), 455 [617 − gal]$^+$ (10), 437 [455 − H_2O]$^+$ (58), and 419 [437 − H_2O]$^+$ (20); FAB m.s. (negative) *m/z* 725 $[M - H +$ glyc$]^-$ (22%), 633 $[M - H]^-$ (100), and 471 [633 − gal]$^-$ (4) (Found: C, 68.1; H, 9.2. $C_{36}H_{58}O_9$ requires C, 68.11; H, 9.21%).

Acid Hydrolysis of Corchorusin B (**12**).—Compound (**12**) (150 mg), on hydrolysis with 2M-HCl in aq. MeOH (45 ml) under reflux for 4 h, yielded the acid-rearranged aglycone, saikogenin A (**18**), which was crystallised from MeOH in needles, m.p. 288—289 °C (decomp.); $[\alpha]_D$ −52° (*c* 0.5 in pyridine) {lit.,[8] m.p. 287—290 °C; $[\alpha]_D$ − 43.3° (pyridine)}.

The filtrate of the hydrolysate was worked up to afford D-galactose, identified by paper chromatography and g.l.c. in the usual way.

Isolation of the Genuine Sapogenin Saikogenin F (**13**).—A solution of the glycoside (**12**) (200 mg) in 95% EtOH (20 ml) at 20 °C was stirred and treated dropwise with a solution of $NaIO_4$ (200 mg) in water (20 ml). The mixture was kept at room temperature overnight, diluted with water, and extracted with EtOAc, and the extract was evaporated under reduced pressure.

The residue (160 mg) was refluxed with 3% KOH in 85% aq. EtOH (8 ml) under N_2 for 1 h. The mixture was diluted with water, cooled, acidified to pH 3 with dil. HCl, and extracted with EtOAc. The extract was washed successively with 5% aq. $NaHCO_3$ and water, dried over Na_2SO_4, and evaporated to give crude saikogenin F (90 mg) which, on purification by preparative t.l.c. (p.l.c.) on silica gel G, afforded pure saikogenin F (**13**), m.p. 248—250 °C; $[\alpha]_D$ +63° (*c* 0.6 in MeOH). The identity of compound (**13**) was confirmed by comparison (m.p., mixed m.p., co-t.l.c.) with an authentic sample.

Permethylation of Corchorusin B (**12**) *and Hydrolysis.*—The procedure of permethylation was the same as described for the methylation of compound (**1**). Compound (**12**) (60 mg) gave the permethylate (**14**) (47 mg) as a white powder, m.p. 128—130 °C; δ($CDCl_3$) 0.70 (3 H, s), 0.89 (6 H, s), 1.00 (3 H, s), 1.08 (3 H, s), 1.27 (3 H, s) (together 6 × Me), 3.17 (1 H, d, *J* 8 Hz, 28-H), 3.92 (1 H, d, *J* 8 Hz, 28-H′), 3.32 (3 H, s), 3.36 (3 H, s), 3.40 (3 H, s), 3.52 (3 H, s), 3.56 (3 H, s), 3.60 (3 H, s) (together 6 × OMe), 4.20 (1 H, d, *J* 7 Hz, 1-H of galactose), 5.40 (1 H, dd, *J* 10 and 3 Hz, 11-H), and 5.88 (1 H, d, *J* 10 Hz, 12-H).

The permethylated product (**14**) (35 mg) was hydrolysed with 2M-HCl in aq. MeOH (10 ml) and worked up in the usual way. The carbohydrate constituents obtained from the filtrate was identified as 2,3,4,6-tetra-*O*-methyl-D-galactose (p.c. and g.l.c. with authentic sample). The residue obtained on purification by silica gel chromatography afforded the partially methylated aglycone, 16,23,28-tri-*O*-methylsaikogenin A (**19**) (25 mg), m.p. 176—177 °C; $[\alpha]_D$ −63° (*c* 0.25 in $CHCl_3$) (lit.,[6] m.p. 177—179 °C; $[\alpha]_D$ −62°).

Corchorusin C (**6**).—This was crystallised from MeOH as flakes (180 mg), m.p. 220—222 °C; $[\alpha]_D$ +25.6° (*c* 0.6 in MeOH) (Found: C, 67.9; H, 9.5. $C_{36}H_{60}O_9$ requires C, 67.89; H, 9.50%).

Hydrolysis of Corchorusin C (**6**).—The glycoside (**6**) (70 mg) was hydrolysed with 2M-HCl in aq. MeOH (15 ml) and the mixture was worked up as usual. The sugar constituent was identified as D-galactose (g.l.c.) and the aglycone, 23-hydroxylongispinogenin (**7**), was crystallized from MeOH–$CHCl_3$ as microneedles (28 mg), m.p. 253—255 °C; $[\alpha]_D$ +46° (*c* 0.5 in pyridine) (lit.,[16,25] m.p. 254—258 °C; $[\alpha]_D$ +47.6°). The amorphous tetra-acetate (**8**), prepared in the usual way from compound (**7**) with Ac_2O and pyridine, had $[\alpha]_D$ +59° (*c* 0.26 in $CHCl_3$); *m/z* 642 (M^+, 3%), 582 (M^+ − AcOH, 3), 522 (M^+ − 2 AcOH, 38), 462 (M^+ − 3 AcOH, 4), 390 (M^+ − 3 AcOH − CH_2OAc, 5), 274 (16), 201 (100), and 187 (30).

Permethylation of Corchorusin C (**6**) *and Hydrolysis.*—Compound (**6**) (75 mg) was permethylated, as described previously, to yield the permethylate (**9**) (60 mg) as a white powder, m.p. 118—120 °C; δ($CDCl_3$) 0.72 (3 H, s), 0.88 (3 H, s), 0.89 (3 H, s), 0.93 (3 H, s), 0.96 (3 H, s), 1.20 (3 H, s), (together 6 × Me), 3.28 (3 H, s), 3.32 (3 H, s), 3.33 (3 H, s), 3.37 (3 H, s), 3.52 (3 H, s), 3.53 (3 H, s), 3.58 (3 H, s) (together 7 × OMe), 4.20 (1 H, d, *J* 7 Hz, 1-H of galactose), and 5.28 (1 H, t-like, 12-H).

The permethylated product (**9**) (30 mg) on hydrolysis followed by the usual work-up afforded 2,3,4,6-tetra-*O*-methyl-D-galactose and 23-*methoxy*-16,28-*di*-*O*-*methyl*-*longispinogenin* (**10**) (14 mg), m.p. 136—138 °C; $[\alpha]_D$ +48° (*c* 0.34 in $CHCl_3$) (Found: C, 76.7; H, 10.9. $C_{33}H_{56}O_4$ requires C, 76.69; H, 10.92%). On acetylation with AC_2O and pyridine compound (**10**) yielded the monoacetate (**11**), m.p. 140—142 °C; $[\alpha]_D$ +72° (*c*, 0.62 in $CHCl_3$); δ($CDCl_3$) 4.90 (1 H, dd, *J* 10.5 and 6 Hz, 3-H) (Found: C, 75.2; H, 10.5. $C_{35}H_{58}O_5$ requires C, 75.22; H, 10.46%).

Corchorusin D (**15**).—This compound was crystallised from MeOH as microneedles (0.45 g), m.p. 210—212 °C; $[\alpha]_D$ +40 °C (*c*, 1.2 in MeOH); FAB m.s. (positive) *m/z* 765 (5%), 764 (12), 763 [*M*H − H_2O]$^+$ (21), 762 (4), 761 (4), 747 (2), 746 (5), 745 [*M*H − 2 H_2O]$^+$ (10), 658 (1), 657 (2), 655 (1), 583 [*M*H − (glc + 2 H_2O)]$^+$ (5), 513 (12), 455 (14), 453 (21), 451 (12), 441 (19), 440 (38), 439 [*M*H − (glc + gal + H_2O)]$^+$ (100), 438 (19), 437 (30), 436 (9), 435 (16), 423 (28), 422 (37), 421 [*M*H − (gal + glc + 2 H_2O)]$^+$ (93), 419 (23), and 403 [*M*H − (glc + gal + 3 H_2O)]$^+$ (26); FAB m.s. (negative) *m/z* 872 (42%), 871 [*M* − H + glyc]$^-$ (84), 781 (25), 780 (50), 779 [*M* − H]$^-$ (100), 778 (8), 777 (16), 709 (12), 671(12), 655 (12), 653 (8), 619 (12), 617 [*M* − H − gal]$^-$ (16), 531 (16), 367 (25), 365 (21), and 363 (29) (Found: C, 64.6; H, 8.8. $C_{42}H_{68}O_{13}$ requires C, 64.59; H, 8.78%).

Hydrolysis of Corchorusin D (**15**).—Compound (**15**) (90 mg), on hydrolysis with 2M-HCl in aq. MeOH (15 ml) under reflux for 5 h followed by the usual work-up, yielded D-galactose and D-glucose (identified by g.l.c. of the alditol acetates with authentic samples) as sugar constituents, and saikogenin C (**20**) (30 mg), m.p. 289—291 °C; $[\alpha]_D$ −46.5° (*c* 0.4 in pyridine) (lit.,[17] m.p. 291—294 °C; $[\alpha]_D$ −45.8°).

Permethylation of Corchorusin D (**15**) *and Hydrolysis.*—Permethylation of compound (**15**) (80 mg) following the procedure as described for corchorusin A yielded the permethylate (**17**) as a white powder (68 mg), m.p. 118—120 °C; δ($CDCl_3$) 0.80 (3 H, s), 0.92 (6 H, s), 1.00 (3 H, s), 1.04 (3 H, s), 1.08 (3 H, s) (together 6 × Me), 3.36 (3 H, s), 3.40 (3 H, s), 3.42 (3 H, s), 3.48 (3 H, s), 3.50 (3 H, s), 3.56 (3 H, s), 3.60 (3 H, s), 3.64 (3 H, s) (together 8 × OMe), 3.18 (1 H, d, *J* 8 Hz, 28-H), 3.92 (1 H, d, *J* 8 Hz, 28-H′), 4.28 (1 H, d, *J* 7 Hz, 1-H of galactose), 4.72 (1 H, d, *J* 7 Hz, 1-H of glucose), 5.40 (1 H, dd, *J* 10 and 2 Hz, 11-H), and 5.88 (1 H, d, *J* 10 Hz, 12-H).

The permethylate (**17**) (35 mg) was hydrolysed in refluxing 2M-HCl –aq. MeOH for 3 h. Work-up in the usual way afforded a mixture of alditol acetates obtained from the carbohydrate fractions, which was subjected to g.l.c. analysis on column (ii). The peaks corresponding to 3,4,6-tri-*O*-methyl-D-galactitol diacetate (R_t 2.14 min) and 2,3,4,6-tetra-*O*-methyl-D-glucitol diacetate (R_t 1.00 min) were identified by comparison with authentic samples. The partially methylated aglycone obtained was purified by silica gel chromatography to afford 16,28-di-*O*-methylsaikogenin C (**22**) (12 mg), m.p. 138—139 °C; $[\alpha]_D$ −44° (*c* 0.8 in $CHCl_3$) lit.,[6] m.p. 138—140 °C; $[\alpha]_D$ −44°).

Genuine Aglycone Saikogenin E (**16**) *from Corchorusin D* (**15**).—A solution of corchorusin D (**15**) (150 mg) in 95% EtOH (8 ml) was treated with a solution of $NaIO_4$ (150 mg) in water (5 ml) at 15 °C. The mixture was kept at room temperature overnight, diluted with water, and extracted with EtOAc. The extract was washed with water, dried over anhydrous Na_2SO_4, and evaporated to give the intermediate product (135 mg), which was refluxed with 3% KOH in 85% aq. EtOH (10 ml) under N_2 for 1 h. The mixture was diluted with water, acidified to pH 3 with dil. HCl, and extracted with EtOAc. Usual work-up of the extract, and crystallisation of the residue from EtOAc, afforded saikogenin E (**16**) (95 mg), m.p. 288—290 °C; $[\alpha]_D$ +112° (*c* 0.45 in MeOH), identified by comparison with an authentic specimen (mixed m.p., i.r. and m.s.).

Prosapogenin (**21**) *and Saikogenin C* (**20**).—Corchorusin D (**15**) (50 mg) was hydrolysed with 0.75M-H_2SO_4 in EtOH (6 ml) on a steam-bath for 20 min. The solution was cooled, and diluted with water (55 ml), and the precipitate was collected by filtration and then subjected to p.l.c. [solvent system $CHCl_3$–MeOH (4:1)]. The prosapogenin (**21**) (4 mg) and saikogenin C (**20**) (4.5 mg) were thus isolated in the pure state. Compound

(**21**) crystallised from $CHCl_3$–MeOH as microneedles, m.p. 252–255 °C; $[\alpha]_D$ −44.2° (*c* 0.34 in MeOH) (Found: C, 69.9; H, 9.50. $C_{36}H_{58}O_8$ requires C, 69.87; H, 9.45%). On acid hydrolysis it provided saikogenin C (**20**) and D-galactose.

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