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PREGNANE GLYCOSIDES FROM *BOUCEROSIA AUCHERIANA**

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Key Word Index—*Boucerosia aucheriana*, Asclepiadaceae, bouceroside-ANC, -ADC, -ANO, ADO, -BNO, -BDO, -BNC, -BDC, -CNO, and -CNC; D-cymarose

Abstract—Ten glycosides named bouceroside-ANC, -ADC, -ANO, -ADO, -BNO, -BDO, -BNC, -BDC, -CNO and -CNC were isolated from *Boucerosia aucheriana* and their structures deduced on the basis of the chemical and spectral evidence. The absolute configuration of cymarose, which was obtained by acidic hydrolyses of seven of the glycosides, was determined by means of HPLC analysis of its carbamoyl derivative on a chiral column as D cymarose

INTRODUCTION

In our continuing studies on the plants of Asclepiadaceae family, a large number of C/D *cis*-pregnane glycosides were isolated [1] Mitsuhashi and his co-workers [2] isolated two C/D *cis*-pregnanes, boucerin and dihydroboucerin, from *Boucerosia aucheriana* and in a previous paper [3], we described the isolation of four glycosides named bouceroside AI, AII, BI and BII from this plant Recently, two pregnane glycosides named caratuberside A and caratuberside B [4] were isolated from *Caralluma tuberculata*, a synonym of *B aucheriana* by Ahmad *et al* [4] In this paper we report the isolation and structural elucidation of 10 glycosides designated as bouceroside-ANC (**1**), ADC (**2**), -ANO (**3**), -ADO (**4**), -BNO (**5**), -BDO (**6**), -BNC (**7**), -BDC (**8**), -CNO (**9**), and -CNC (**10**) from the less polar glycoside fraction of *B aucheriana*. The absolute configuration of cymarose was determined by using a chiral HPLC column [5] comparison with the authentic sample

RESULTS AND DISCUSSION

A crude glycoside mixture was obtained from the chloroform-soluble portion of the methanol extract from the dried aerial part of *B aucheriana* From the less polar fractions of this crude glycoside, 10 glycosides were separated by means of normal and reversed phase gel, especially effective was silver nitrate impregnated silica gel for separating the Δ^5 and 5α-H types of aglycones.

Bouceroside-ANC (**1**) had the molecular formula $C_{49}H_{76}O_{15}$ on the basis of ion peaks at *m/z* 927 $[M + Na]^+$ and 943 $[M + K]^+$ in its field-desorption mass spectrum (FDMS) The 500 MHz ^1H NMR spectrum of **1** showed methyl signals of its aglycone moiety at δ0 81 (3H, *s*, 19-H), 1 23 (3H, *d*, *J* = 6 4 Hz, 21-H), and 1 38 (3H, *s*, 18-H) and esterified methine proton at 4 72 (1H, *dd*, *J* = 12 2, 4 4 Hz). The presence of a benzoyl ester was suggested from the UV maximum (229 nm), aromatic proton signals at δ7 44 (2H, *t*, *J* = 7 3 Hz), 7 56 (1H, *t*, *J* = 7 3 Hz), and 8.05 (2H, *d*, *J* = 7 3 Hz) and the aromatic carbon signals (Table 1) Three anomeric protons at δ4 59 (1H, *d*, *J* = 8 3 Hz), 4 77 and 4 83 (each 1H, *dd*, *J* = 9 8, 2.0 Hz) and anomeric carbon signals at δ96 1, 100 4, and 104 1 were observed The β-linkage of each sugar was revealed by the coupling constants of the respective anomeric protons in the ^1H NMR spectrum From its ^{13}C NMR spectrum (Table 2), compound **1** contained two glycosilated cymaropyranosyl and a terminal 6-deoxy-3-*O*-methyl-allopyranosyl moieties, judging from comparison of the chemical shifts with those of known asclepiadaceous glycosides. In its ^{13}C NMR spectrum. partial relaxation Fourier-transform (PRFT) measurement [6, 7] of **1** showed that 6-deoxy-3-*O*-methyl-allose was the terminal sugar On mild acidic hydrolysis of **1**, an aglycone (**11**), cymarose (**12**), and a biose (**13**) were obtained. The aglycone (**11**) had the molecular formula $C_{28}H_{40}O_5$ on the basis of ion peak at *m/z* 456 (M^+) in its FDMS From its ^1H and ^{13}C NMR spectra, **11** was suggested to be 12-*O*-benzoyl dihydroboucerin, a previously reported compound [3] The NOE experiments supported the stereochemistry at C-17 and C-20 positions deduced from ^{13}C NMR chemical shifts [8] Compound **12** was identified by spectral data and TLC comparison with the authentic sample The biose (**13**) was identified as asclepobiose (6-deoxy-3-*O*-methyl-β-D-allopyranosyl-(1→4)-D-cymarose) [9–11] by specific rotation, ^{13}C NMR spectrum (Table 3), and behavior on TLC As glycosidation shifts [12, 13] were observed at the C-2 (−2.3), C-3 (+6.2), and C-4 (−4.2) positions of **1**, the sugar moiety was linked to the C-3 hydroxyl group of the aglycone Thus, **1** was deduced to be 12-*O*-benzoyl dihydroboucerin 3-*O*-6-deoxy-3-*O*-methyl-β-D-allopyranosyl-(1→4)-β-D-cymaropyranosyl-(1→4)-β-D-cymaropyranoside

Bouceroside-ADC (**2**) had the molecular formula $C_{49}H_{74}O_{15}$ on the basis of an ion peak at *m/z* 925 $[M + Na]^+$ in its FDMS. Compound **2** had the same sugar chain as **1** from its ^1H and ^{13}C NMR spectra, and 6-deoxy-3-*O*-methyl-allose was confirmed as the terminal sugar by PRFT measurement On mild acidic hydrolysis, **2** gave **12**–**14**. The former two compounds were identified

*Part 71 in the series 'Studies on the Constituents of Asclepiadaceae Plant' For part 70 see ref [3].

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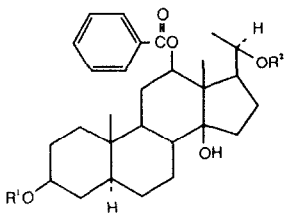
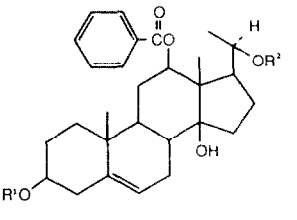
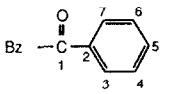


	R^1	R^2
1	Ra	H
3	Rb	H
5	Rb	Ac
7	Ra	Ac
9	Rb	Bz
10	Ra	Bz



	R^1	R^2
2	Ra	H
4	Rb	H
6	Rb	Ac
8	Ra	Ac



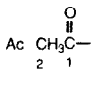


Table 1 ^{13}C NMR chemical shifts of the aglycone moieties of **1**–**4** and their aglycones **11** and **14**

C	1	3	11	2	4	14
1	37 3	37 3	37 6	37 5	37 3	37 8
2	30 0 (−2 3)	30 0 (−2 3)	32 3	30 3 (−2 2)	30 3 (−2 2)	32 5
3	76 7 (+6 2)	76 6 (+6 1)	70 5	77 4 (+6 2)	77 4 (+6 2)	71 2
4	35 0 (−4 2)	34 9 (−4 3)	39 2	39 3 (−4 0)	39 3 (−4 0)	43 3
5	44 7	44 6	45 1	139 7	139 7	140 6
6	29 2[a]	29 2[a]	29 2[a]	122 4	122 5	122 0
7	28 2[a]	28 2[a]	28 3[a]	27 8[a]	27 8[a]	27 8[a]
8	40 4	40 3	40 4	36 9	37 0	37 6
9	46 7	46 7	46 7	43 9	44 0	43 6
10	36 1	36 1	36 1	37 5	37 6	37 5
11	27 0[b]	27 2[b]	27 1[b]	26 7[a]	26 7[a]	26 8[a]
12	79 8	79 7	79 9	79 3	79 3	79 2
13	53 4	53 4	53 4	53 3	53 4	53 3
14	85 0	84 9	85 0	85 1	85 2	85 2
15	32 8	32 9	32 8	33 4	33 4	33 4
16	26 5[b]	26 6[b]	26 6[b]	26 4	26 4	26 5
17	53 1	53 1	53 1	53 1	53 1	53 0
18	11 5	11 5	11 6	11 5	11 5	11 6
19	12 2	12 1	12 3	19 5	19 5	19 7
20	70 8	70 8	70 8	70 9	70 9	70 8
21	23 6	23 8	23 7	23 7	23 7	23 7
1′	166 7	166 7	166 7	166 7	166 7	166 7
2′	131 7	131 6	131 7	131 6	131 6	131 6
3′	130 0	130 0	130 0	130 0	130 1	130 1
4′	128 9	128 8	128 9	128 9	128 9	129 0
5′	133 2	133 3	133 3	133 3	133 3	133 3
6′	128 9	128 8	128 9	128 9	128 9	129 0
7′	130 0	130 0	130 0	130 0	130 1	130 1

δ value (ppm) from internal TMS in pyridine-d_5
[a, b] Values in each column may be interchangeable
( ), Glycosidation shifts

by TLC comparison with the authentic samples The molecular formula of **14** was established as $C_{28}H_{38}O_5$ by its high-resolution mass spectrum (HRMS) which was two mass units less than that of **11** In addition to the presence of a benzoyl group (δ7.45, 7 57, and 8 06), olefinic proton and carbon signals were observed at δ5 44 (*br* s), and 140 6 and 122.0, respectively Moreover the 19-methyl signal at δ0 80 in the **11** was shifted to lower field at 1 01 in the spectrum of **14** Thus, compound **14** was assumed to be the Δ^5 derivative of **11**, 12-*O*-benzoyl boucerin [3] Glycosidation shifts of the aglycone carbon signals were observed at the C-2, C-3 and C-4 positions

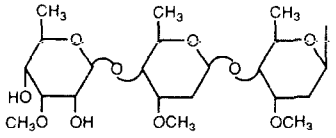
Table 2 ^{13}C NMR chemical shifts of sugar moieties of **1**–**7**, **9** and **10**

	1	**2**	**7**	**10**		**3**	**4**	**5**	**6**	**9**
cym-1	96 1	96 4	96 0	96 0	cym-1	96 0	96 4	96 0	96 3	96 1
2	36 9[a]	36 9[a]	37 0[a]	36 9[a]	2	37 3[a]	37 3[a]	37 3	37 6	37 2[a]
3	78 2	78 1[b]	78 1	78 1[b]	3	77 8	77 9	77 9	77 8	78 0
4	83 3[b]	83 3[c]	83 2[b]	83 4[c]	4	82 7[b]	82 8[b]	82 8[a]	82 8[a]	82 8[b]
5	69 0[c]	69 1[d]	69 0[c]	69 1[d]	5	69 0	69,0	68 9	68 9	69 0
6	18 6	18 6	18 6	18 6	6	18 6[c]	18 6[c]	18 6[b]	18 6[b]	18 6[c]
3-OMe	58 8	58 8	58 8	58 8	3-OMe	58 8	58 8	58 8	58 8	58 8
cym′-1	100 4	100 4	100 4	100,4	ole-1	101 9	101,9	101 9	101,9	101 9
2	37 3[a]	37 3[a]	37,4[a]	37 1[a]	2	37 5[a]	37 6[a]	37 3	37 6	37 6[a]
3	78,2	78 0[b]	78 1	78 0[b]	3	79 2	79,3	79,3	79 2	79 3
4	83,4[b]	83 4[c]	83 4[b]	83 5[c]	4	83 5[b]	83 5[b]	83 5[a]	83,4[a]	83 6[b]
5	69 3[c]	69,3[d]	69 3[c]	69 3[d]	5	72 0	72 1	72 0	72 0	72 1
6	18 6	18 6	18 6	18 6	6	18 9[c]	18 9[c]	18,9[b]	18 9[b]	18 9[c]
3-OMe	58 8	58 8	58 8	58 9	3-OMe	57 0	57 1	57 2	57 1	57,1
allo-1	104 2	104 2	104 2	104 2	allo-1	101 9	101 9	101 9	101 9	101 9
2	73 2	73,2	73 1	73 1	2	73 2	73 3	73 2	73 2	73 3
3	83 9	83,9	83 9	84,0	3	84,0	84 0	83 9	83 9	83 9
4	74 4	74 5	74 4	74,5	4	74 5	74 6	74 5	74 6	74,6
5	70 7	70 7	70 7	70,7	5	70 8	70,9	71 0	70 9	71 1
6	18 6	18 6	18,6	18 6	6	18 6	18 6	18 6	18 6	18 7
3-OMe	62,1	62 1	62 0	62,2	3-OMe	62 0	62 0	62 0	62 0	62 0

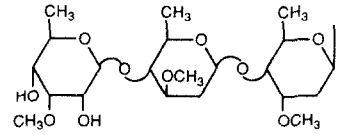
δ value (ppm) from internal TMS in pyridine-d_5 cym, β-cymaropyranose ole, β-oleandropyranose allo, 6-deoxy-3-*O*-methyl-β-allopyranose

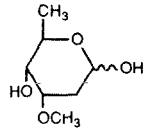
[a–d] Values in each column may be interchangeable.

Ra

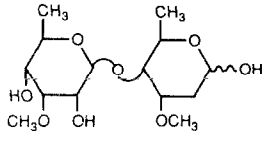


Rb

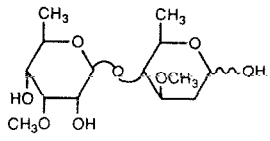




12 D-cymarose



13 asclepobiose



16 pachybiose

Table 3. ^{13}C NMR chemical shifts of **13** and **16**

	13		**16**
cym-1	92 5	ole-1	91 4
2	38 5	2	37 6
3	78 5	3	77 6
4	83 8	4	84 0
5	69 3	5	67 4
6	18 6[a]	6	18 9[a]
3-OMe	58 4	3-OMe	57 2
allo-1	104 2	allo-1	102 2
2	73 0	2	73 3
3	83 9	3	84 0
4	74 4	4	74 6
5	70 7	5	70 9
6	18 9[a]	6	19 2[a]
3-OMe	62 1	3-OMe	62 0

δ value (ppm) from internal TMS in pyridine-d_5
cym, β-cymaropyranose ole, β-oleandropyranose
allo, 6-deoxy-3-*O*-methyl-β-allopyranose
[a]Value in each column may be interchangeable

(Table 1) Therefore, the structure of **2** was deduced to be 12-*O*-benzoyl boucerin 3-*O*-6-deoxy-3-*O*-methyl-β-D-allopyranosyl-(1 → 4)-β-D-cymaropyranosyl-(1 → 4)-β-D-cymaropyranoside

Bouceroside-BNO (**5**) had the molecular formula $C_{51}H_{78}O_{16}$ on the basis of elemental analysis and FD-MS (*m/z* 969 $[M + Na]^+$ and 946 $[M]^+$) The 500 MHz ^{1}H NMR spectrum of **5** showed methyl signals of its aglycone moiety at δ0 80 (3H, *s*, 19-H) 1 12 (3H, *d*, *J* = 6 3 Hz, 21-H), 1 14 (3H, *s*, 18-H) and 1 93 (3H, *s*, 2''-H), and two esterified methine protons at δ 4 76 (1H, *dd*, *J* = 12 2, 4 4 Hz) and 4 87 (1H, *dq*, *J* = 9 4, 6 3 Hz) Moreover, five aromatic proton signals at δ7 44, 7 56 and 8 09 were observed From the ^{13}C NMR spectrum (Table 4), the two esters were suggested as benzoyl and acetyl Three anomeric proton signals at δ4 48 (1H, *dd*, *J* = 9 8, 2.0 Hz), 4 79 (1H, *d*, *J* = 7 8 Hz) and 4 86 (1H, *dd*, *J* = 9 8, 2 0 Hz), and anomeric carbon signals at δ 96 0 and two 101.9 were observed Thus, compound **5** was also suggested to be a triglycoside composed of terminal 6-deoxy-3-*O*-methyl-allose, **12** and oleandrose from the ^{13}C NMR chemical shifts On mild acidic hydrolysis of **5**, an aglycone (**15**), **12** and a biose (**16**) were obtained Compound **12** was identified by TLC comparison with the authentic sample The biose (**16**) was identified as pachybiose [6-deoxy-3-*O*-methyl-β-D-allopyranosyl-(1 → 4)-β-D-oleandrose] [9, 14] by specific rotation, ^{13}C NMR spectrum (Table 3) and behaviour on TLC The aglycone (**15**) was suggested to be 12-*O*-benzoyl-20-*O*-acetyl dihydroboucerin [3], which was isolated from this plant, by ^{1}H and ^{13}C NMR spectra (Table 4) Though the position of the two esters had been determined by chemical methods, long-range selective proton decoupling (LSPD) measurement [15–17] of **15** gave the same conclusion (Fig 1) In pyridine-d_5, when the proton signal at δ5 14 (12-H) was irradiated, the carbonyl carbon signal of the benzoyl at δ166 7 sharpened and the extent of the signal splitting was decreased simultaneously When the proton signal at δ5 42 (20-H) was irradiated, the carbonyl carbon signal of the acetyl at δ170 2 sharpened and the extent of the signal splitting was decreased As the glycosidation shifts of the aglycone carbon signals were observed at C-2 (− 2 3), C-3 (+ 6 1) and C-4 (− 4 0), the sugar moiety was linked to the C-3 hydroxyl group of the aglycone PRFT measurement of **5** confirmed that 6-deoxy-3-*O*-methyl-allose was the terminal sugar Thus compound **5** was determined as 12-*O*-benzoyl-20-*O*-acetyl dihydroboucerin 3-*O*-6-deoxy-3-*O*-methyl-β-D-allopyranosyl-(1 → 4)-β-D-oleandropyranosyl-(1 → 4)-β-D-cymaropyranoside

Bouceroside-BDO (**6**) had the molecular formula $C_{51}H_{76}O_{16}$ on the basis of the parent ion peak at *m/z* 944 (M^+) in its FDMS On mild acidic hydrolysis of **6**, an aglycone (**17**), **12** and **16** were obtained The latter two compounds were identified by TLC comparison with the authentic samples In the ^{1}H NMR spectrum of **17** in addition to the benzoyl group at δ7 45, 7 57 and 8 11, and the acetyl group at δ1.97, an olefinic proton signal at δ5 44 was observed From its ^{13}C NMR spectrum (Table 4), compound **17** was deduced to be 12-*O*-benzoyl-20-*O*-acetyl boucerin [3] PRFT measurement of **6** showed that 6-deoxy-3-*O*-methyl-allose was the terminal sugar Thus compound **6** was deduced to be 12-*O*-benzoyl-20-*O*-acetyl boucerin 3-*O*-6-deoxy-3-*O*-methyl-β-D-allopyranosyl-(1 → 4)-β-D-oleandropyranosyl-(1 → 4)-β-D-cymaropyranoside

Bouceroside-ANO (**3**) and -ADO (**4**) had the molecular formulae $C_{49}H_{76}O_{15}$ and $C_{49}H_{74}O_{15}$, respectively From their ^{13}C NMR spectra (Table 2), both glycosides have Rb type sugar chain, and 6-deoxy-3-*O*-methyl-allose was confirmed as the terminal sugar by PRFT measurement. On mild acidic hydrolysis, compound **3** gave **11**–**13**, and **4** gave **14**, **12** and **16** respectively Compounds **11**–**13** and **16** were identified by TLC comparison with the authentic samples The glycosidation shifts were observed at the C-2, C-3 and C-4 positions of each aglycone (Table 1) Thus, compound **3** was deduced to be 12-*O*-benzoyl

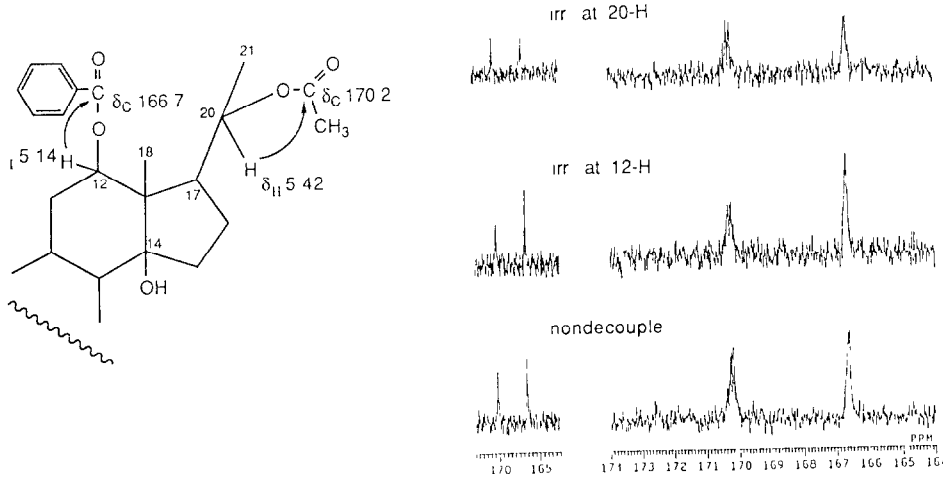
Table 4 ^{13}C NMR chemical shifts of aglycone moieties of **5**, **6**, and **7**, and their aglycones **15** and **17**

C	**5**	**7**	**15**	**6**	**17**
1	37 3	37 2	37 5	37 6	37 5
2	30 0 (−2.3)	30 0 (−2 3)	32 3	30.3 (−2 2)	32 5
3	76 6 (+6 1)	76 6 (+6 1)	70 5	77.2 (+6 1)	71 1
4	34 8 (−4 3)	34 8 (−4 3)	39 1	39.1 (−4.2)	43 3
5	44 6	44 5	45 0	139 7	140 7
6	29 1[a]	29 1[a]	29 2[a]	122 8	122 5
7	28 1[a]	28 1[a]	28 2[a]	27 7	27 8
8	40.8	40 9	40 9	37 6	37.6
9	46 5	46 5	46 6	43 9	43 8
10	36 1	36 0	36 1	37 6	37 5
11	26.8[b]	26 8[b]	26 9[b]	26 7[a]	26 7[a]
12	79 7	79 8	79 7	79 6	79 3
13	52 5	52 5	52 5	52 3	52 4
14	85 5	85 6	85 6	85 7	85 8
15	32 2	32 2	32 3	32 7	32 5
16	25 9[b]	26 0[b]	26 0[b]	25 5[a]	25 6[a]
17	50 7	50 7	50 7	50 6	50 7
18	10 3	10 3	10 3	10 3	10 2
19	12 1	12 1	12 3	19 4	19 4
20	73 9	74.0	74 0	74 0	73 9
21	19 3	19 4	19 4	19.4	19 4
1′	166 7	166 7	166 7	166 6	166 7
2′	131 6	131 7	131 7	131 6	131 6
3′	130 1	130 1	130 1	130 0	130 1
4′	128 9	128 9	128 9	128 9	128 9
5′	133 4	133 3	133 4	133 4	133 4
6′	128 9	128 9	128 9	128 9	128 9
7′	130 1	130 1	130 1	130 0	130 1
1″	170 2	170 3	170 2	170 2	170 2
2″	21 6	21 6	21 6	21 6	21 6

δ value (ppm) from internal TMS in pyridine-d_5

[a] [b]Values in each column may be interchangeable.

( ). Glycosidation shifts



Nondecouple and LSPD spectra

Fig 1

dihydrobouceriη 3-*O*-6-deoxy-3-*O*-methyl-β-D-allopyranosyl-(1→4)-β-D-oleandropyranosyl-(1→4)-β-D-cymaropyranoside, and compound **4** to be 12-*O*-benzoyl boucerin 3-*O*-6-deoxy-3-*O*-methyl-β-D-allopyranosyl-(1→4)-β-D-oleandropyranosyl-(1→4)-β-D-cymaropyranoside

Bouceroside-BNC (**7**) had the same molecular formula $C_{51}H_{78}O_{16}$ as **5** on the basis of ion peak at *m/z* 969 [M+Na]$^+$ in its FDMS From its ^{13}C NMR spectrum (Tables 2 and 4), compound **7** contained **16** as the aglycone moiety and an Ra type sugar chain PRFT measurement of **7** indicated that 6-deoxy-3-*O*-methylallose was the terminal sugar. On mild acidic hydrolysis, **7** gave **16**, **12** and **13**, which were identified by TLC comparison with the authentic samples The glycosidation shifts of the aglycone carbons were observed at C-2 (−2 3), C-3 (+6 1) and C-4 (−4.3) Thus compound **7** was deduced to be 12-*O*-benzoyl-20-*O*-acetyl dihydroboucerin 3-*O*-6-deoxy-3-*O*-methyl-β-D-allopyranosyl-(1→4)-β-D-cymaropyranosyl-(1→4)-β-D-cymaropyranoside

Bouceroside-BDC (**8**) had the molecular formula $C_{51}H_{76}O_{16}$ on the basis of elemental analysis and its fast atom bombardment mass spectrum (FABMS) (*m/z* 967 [M+Na]$^+$ The 500 MHz ^{1}H NMR spectrum of **8** exhibited methyl signals of the aglycone moiety at δ1 00 (3H, *s*, 19-H), 1 14 (3H, *d*, *J* = 6 3 Hz, 21-H), 1 17 (3H, *s*, 18-H) and 1 97 (3H, *s*, 2″-H) and two esterified methine protons at δ4 81 (1H, *dd*, *J* = 12 2, 4 4 Hz, 12-H) and 4.91 (1H, *dq*, *J* = 9 7, 6 3 Hz, 20-H). The presence of a benzoyl group was suggested from the UV maximum (230 nm) and five aromatic protons at δ7 45 (2H, *t*, *J* = 7 3 Hz), 7 58 (1H, *t*, *J* = 7 3 Hz) and 8.11 (2H, *d*, *J* = 7 3 Hz) In addition, an olefinic proton signal was seen at δ5 44 (*br s*) Three anomeric proton signals at δ4 59 (1H, *d*, *J* = 7.8 Hz), 4 76 and 4 85 (each 1H, *dd*, *J* = 9.8, 2 0 Hz) and methoxy methyl signals at δ3.42, 3 45 and 3 66 (each 3H, *s*) were shown in its ^{1}H NMR spectrum On mild acidic hydrolysis, compound **8** gave **17**, **12** and **13**, which were identified by TLC comparison with the authentic samples Thus, compound **8** was presumed to be 12-*O*-benzoyl-20-*O*-acetyl boucerin 3-*O*-6-deoxy-3-*O*-methyl-β-D-allopyranosyl-(1→4)-β-D-cymaropyranosyl-(1→4)-β-D-cymaropyranoside

Bouceroside-CNO (**9**) and -CNC (**10**) had the same molecular formula $C_{56}H_{80}O_{16}$ From their ^{13}C NMR spectra (Table 2 and 5), they contain the same aglycone moiety as boucerogenin II (**18**) [3], but differ from one another in the sugar moiety On mild acidic hydrolysis, compound **9** gave **18**, **12** and **16**, while **10** gave **18**, **12** and **13** Thus, compound **9** was deduced to be boucerogenin II 3-*O*-6-deoxy-3-*O*-methyl-β-D-allopyranosyl-(1→4)-β-D-oleandropyranosyl-(1→4)-β-D-cymaropyranoside, and **10** to be boucerogenin II 3-*O*-6-deoxy-3-*O*-methyl-β-D-allopyranosyl-(1→4)-β-D-cymaropyranosyl-(1→4)-β-D-cymaropyranoside

The chirality of cymarose, which was obtained by acidic hydrolyses of glycosides **1**, **3**–**7** and **9**, was deduced to be D-series by means of HPLC, using a chiral column (SUMIPAX OA-4100) [5].

Table 5 ^{13}C NMR chemical shifts of aglycone moieties of **9** and **10**, and their aglycone **18**

C	9	10	18
1	37 6	37 3	37 8
2	30 0 (−2 2)	30 1 (−2 1)	32 2
3	76 7 (+6 3)	76 5 (+6 1)	70 4
4	34 9 (−4 1)	34 8 (−4 2)	39 0
5	44 7	44 6	44 7
6	29 1[a]	29 2[a]	29 1[a]
7	28 1[a]	28 2[a]	28 1[a]
8	40 4	40 5	40 5
9	46 6	46 5	46 5
10	36 1	36 1	36 0
11	26 9[b]	26 9[b]	26 9[b]
12	79 0	79 0	78 9
13	53 0	52 9	52 8
14	85 7	85 7	85 7
15	32 2	32 1	32 2
16	25 5[b]	25 6[b]	25 5[b]
17	50 4	50 3	50 2
18	10 3	10 3	10 3
19	12 1	12 0	12 1
20	74 8	74 9	74 8
21	19 6	19 7	19 6
1	166 8	166 8	166 7
1″	166 1	166 1	166 0
2′	131 8	131 7	131 7
2″	131 7	131 6	131 7
3′	130 1	130 2	130 1
3″	129 9	130 0	129 8
4′	129 1	129 1	129 1
4″	128 8	128 9	128 8
5′	133 4	133 5	133 4
5″	133 0	133 1	133 4
6′	129 1	129 1	129 1
6″	128 8	128 8	128 8
7′	130 1	130 2	130 1
7″	129 9	130 0	129 8

δvalue (ppm) from internal TMS in pyridine-d_5
[a b]Values in each column may be interchangeable
( ), Glycosidation shifts

EXPERIMENTAL

Mps uncorr ^{1}H NMR spectra were run at 270 and 500 MHz with TMS as the int standard in $CDCl_3$ or pyridine-d_5 soln, ^{13}C NMR spectra were performed at 67 5 and 22 5 MHz in pyridine-d_5 soln UV spectra were obtained in EtOH IR spectra were recorded in $CHCl_3$ CC were carried out on Wakogel C-100 and C-200 for normal phase, and Lobar column LiChroprep RP-8 (40–63 μm) for reversed phase HPLC analyses were performed with a Waters A-4000 pump and 440 absorbance detector at 254 nm using SUMIPAX OA-4100 (5μ, φ4 mm i d × 25 cm) (Sumitomo Chemical Co Ltd) TLC was carried out on a precoated plate Kieselgel 60 F_{254}, Merck with the following solvent systems R_{f1}, MeOH–$CHCl_3$ (1 9), R_{f2}, hexane Me_2CO (1 1), R_{f3}, H_2O–MeOH–$CHCl_3$ (1 3 15, lower layer), R_{f4}, Me_2CO–Et_2O (1 3) Abbreviations used for sugars in this section are as follows cym cymarose, ole, oleandrose, allo, 6-deoxy-3-*O*-methylallose

Plant material Boucerosia aucheriana used in this research was collected and identified by Prof N A Quazilbash of Peshawar University, Pakistan

Extraction and isolation Dried and powdered aerial parts of *B aucheriana* (420 g) were extracted with MeOH and concd to give an extract (38 g) which was re-extracted with $CHCl_3$ The $CHCl_3$

soluble fraction was dissolved in $CHCl_3$ (80 ml) and the soln poured into hexane (1700 ml) The insoluble portión corresponded to a crude glycoside mixture (10 9 g) which was chromatographed on Wakogel C-100 (100 g) with solvents of increasing polarity from 0 to 30% MeOH–$CHCl_3$ into 6 fractions Fraction 2 (3 74 g) eluated with 2 5% MeOH in $CHCl_3$ was rechromatographed on Wakogel C-200 with 2% MeOH–$CHCl_3$ to separate fraction 2–2 (1 68 g, a mixture of **5–10**) and fraction 2–3 (1.47 g, a mixture of **1–4**) Rechromatography of the fraction 2–3 on silica gel with 2 5% MeOH–$CHCl_3$ and increasing polarity from hexane–Me_2CO (2 1) to hexane–Me_2CO (1 2), and then on reversed-phase gel with H_2O–MeOH (3 17) to separate fraction A (155 6 mg **1** and **2**) and fraction B (100 8 mg **3** and **4**) Fraction A and B were each chromatographed on $AgNO_3$–silica gel with hexane–Me_2CO (2 1 and 3 2) to afford glycosides, **1** (34 2 mg), **2** (45 0 mg), **3** (19 4 mg) and **4** (22 5 mg), respectively Rechromatography of fraction 2–2 on silica gel with 4% MeOH in $CHCl_3$ and hexane–Me_2CO (1 1 and 3·2), and then on reversed-phase gel with H_2O–MeOH (3·17 and 1 4) to separate fraction C (176 6 mg **5** and **6**), fraction D (65 2 mg **7** and **8**) and fraction E (76 7 mg **9** and **10**) Each of fraction C, D and E was chromatographed on $AgNO_3$–silica gel with hexane–Me_2CO (5:2) to afford the following glycosides **5** (65 3 mg) and **6** (30 0 mg) from fraction C, **7** (15 7 mg) and **8** (5.2 mg) from fraction D, **9** (25 5 mg) and **10** (7 0 mg) from fraction E

R_f value· **1** (R_{f_1}; 0 56, R_{f_2}, 0 42), **2** (R_{f_1}, 0 56, R_{f_2}, 0 42), **3** (R_{f_1}, 0.56, R_{f_2}; 0 42), **4** (R_{f_1}, 0 56, R_{f_2}, 0 42), **5** (R_{f_1}, 0 70, R_{f_2}, 0 48), **6** (R_{f_1}; 0.70, R_{f_2}, 0 48), **7** (R_{f_1}, 0 70, R_{f_2}, 0 48), **8** (R_{f_1}, 0 70, R_{f_2}, 0 48), **9** (R_{f_1}, 0 74, R_{f_2}, 0.50), **10** (R_{f_1}, 0 74, R_{f_2}; 0.50).

Boucerosıde-ANC (**1**) Amorphous powder; mp 138 5–142 5° $[\alpha]_D$ −3.2° ($CHCl_3$, *c* 1 02) UV λ_{max}^{EtOH} nm (log ε). 281 (2 95), 278 (3 07), 229 (4 12) IR $\nu_{max}^{CHCl_3}$ cm^{-1} 3400, 1710, 1600, 1580, 1275. FDMS *m/z* 943 $[M+K]^+$, 927 $[M+Na]^+$ ^1H NMR (500 MHz, $CDCl_3$) δ0 81 (3H, *s*, 19-H), 1 22 (3H, *d*, *J* = 6 3 Hz, 21-H), 1.38 (3H, *s*, 18-H), 3 42, 3 44, 3.66 (each 3H, *s*, 3-OMe of sugar moiety), 4 59 (1H, *d*, *J* = 8 3 Hz allo-1-H), 4 72 (1H, *dd*, *J* = 12 2, 4 4 Hz, 12-H), 4 77, 4.86 (each 1H, *dd*, *J* = 9 8, 2 0 Hz, cym-1-H), 7.44 (2H, *t*, *J* = 7 3 Hz, 4′,6′-H), 7.56 (1H, *t*, *J* = 7 3 Hz, 5′-H), 8 05 (2H, *d*, *J* = 7 3 Hz, 3′,7′-H) ^{13}C NMR spectrum see Tables 1 and 2

Acidic hydrolysis of **1** A soln of **1** (28 0 mg) in MeOH (6 ml) was treated with 0 1 M H_2SO_4 (2 ml), and the mixture kept at *ca* 60° for 30 min, then H_2O (6 ml) was added and whole concd to 6 ml. The soln was warmed at 60° for further 30 min, then the soln was extracted with Et_2O (15 ml). The Et_2O layer was washed with 5% $NaHCO_3$ (10 ml) and satd NaCl (10 ml), then evapd to give a syrup, which was chromatographed on silica gel with hexane–Me_2CO (2 1) to afford **11** (4 4 mg). Compound **11** (R_{f_1}, 0.51, R_{f_2}, 0 51), an amorphous powder, mp 128 5–133.5° $[\alpha]_D$ −32° ($CHCl_3$, *c* 0 30) UV λ_{max}^{EtOH} nm (log ε) 281 (3 05), 273 (3.16), 229 (4 24) IR $\nu_{max}^{CHCl_3}$ cm^{-1} 3400, 1710, 1600, 1580, 1275 FD-MS *m/z* 456 $[M]^+$. ^1H NMR (500 MHz, $CDCl_3$) δ0.83 (3H, *s*, 19-H), 1.23 (3H, *d*, *J* = 6 8 Hz, 21-H), 1.39 (3H, *s*, 18-H), 3 61 (1H, *m*, 3-H), 3 83 (1H, *dq*, *J* = 8 4, 6.3 Hz, 20-H) 4 72 (1H, *dd*, *J* = 12.2, 4.4 Hz, 12-H), 7 45 (2H, *t*, *J* = 7 8 Hz, 4′,6′-H), 7 57 (1H, *t*, *J* = 7 8 Hz, 5′-H), 8 06 (2H, *d*, *J* = 7 8 Hz, 3′,7′-H) ^{13}C NMR: see Table 1 The aq. layer was neutralized with satd $Ba(OH)_2$ The ppt was filtered off and the soln evapd to give a mixture of **12** and **13** The sugar mixture (**12** and **13**) was chromatographed on silica gel with H_2O–MeOH–$CHCl_3$ (1 3 18 (v/v), lower layer) to separate **12** (1 5 mg) and **13** (2 0 mg) Compound **12** (R_{f_1}; 0.46, R_{f_2}; 0 41); mp 67.0–71.0° FDMS *m/z* 162 $[M]^+$ Compound **13** (R_{f_3}, 0 31, R_{f_4}; 0 45); $[\alpha]_D$ + 29 8° (H_2O, *c* 0 20) FDMS *m/z* 322 (M^+). ^{13}C NMR see Table 3.

Absolute configuration of cymarose (**12**). A soln of **12** (1 0 mg) in MeOH (2 ml) was allowed to react with Amberlite IR-120 (H^+) at room temp. for 1 hr and then warmed at 60° for 5 min. After removal of the resin by filtration, the reaction mixture was evapd to give a mixture of methyl cymarosides The product was dissolved in dry toluene (0.5 ml) and allowed to react with 3,5-dinitrophenylisocyanate (*ca* 2 mg) in the presence of dry pyridine (0.05 ml) at 60° for 1 hr, then the solvent was evapd off to afford a mixture of carbamates. Purification of the mixture was carried out by preparative TLC [solvent: hexane–EtOAc (2 1)], to collect bands with R_f 0.25 and 0 38 to yield carbamate mixtures The combined mixtures were analysed by a chiral HPLC column SUMIPAX OA-4100 (5μ, φ4 mm i d × 25 cm), mobile phase hexane–EtOH (40 1) flow rate 1 ml/min, monitored by absorption at 254 nm

The other cymaroses obtained from **3–7** and **9**, a mixture of carbamates was prepared and analysed as described above

Boucerosıde-ADC (**2**) Amorphous powder, mp 132 0–135.5°, $[\alpha]_D$ −12.5° ($CHCl_3$, *c* 1.00) UV λ_{max}^{EtOH} nm (log ε) 282 (2 98), 278 (3 08), 233 (4.14). IR $\nu_{max}^{CHCl_3}$ cm^{-1}. 3450, 1700, 1600, 1580, 1280, FDMS: *m/z* 941 $[M+K]^+$, 925 $[M+Na]^+$ ^1H NMR (500 MHz, $CDCl_3$). δ1.01 (3H, *s*, 19-H), 1 22, 1 26, 1 28 (each 3H, *d*, *J* = 6.3 Hz, 6-H of sugar moiety), 1 25 (3H, *d*, *J* = 6 8 Hz. 21-H), 1.41 (3H, s, 18-H), 3 42, 3.45, 3 66 (each 3H, *s*, 3-OMe of sugar moiety), 4 59 (1H, *d*, *J* = 7 8 Hz, allo-1-H), 4 76, 4 86 (each 1H, *dd*, *J* = 9 8, 2 0 Hz, cym-1-H), 4.77 (1H, *dd*, *J* = 12 2, 4 4 Hz, 12-H), 5 44 (1H, *br s*, 6-H), 7.45 (2H, *t*, *J* = 7 3 Hz, 4′,6′-H), 7 57 (1H, *t*, *J* = 7.3 Hz, 5′-H), 8 06 (2H, *d*, *J* = 7 3 Hz, 3′,7′-H) ^{13}C NMR· see Tables 1 and 2

Acidic hydrolysis of **2** A soln of **2** (32.0 mg) in MeOH (6 ml) was allowed to react in the same way as **1**, and the products separated to provide **14** (10 8 mg), **12** (1 0 mg) and **13** (3.5 mg) **14** (R_{f_1}, 0.51, Rf_2; 0 51), an amorphous powder, mp 140 0–144 0°, $[\alpha]_D$ −46.4° ($CHCl_3$, *c* 1.08). UV λ_{max}^{EtOH} nm (log ε)· 282 (2 90), 275 (2.96), 230 (3 92) IR $\nu_{max}^{CHCl_3}$ cm^{-1} 3400, 1715, 1600, 1585, 1270 EIMS· *m/z* 454 $[M]^+$, 436 $[M-H_2O]^+$, 418 $[M-H_2O]^+$, 332 $[M-C_6H_5COOH]^+$, 105 $[C_6H_5CO$, base peak$]^+$, 77 $[C_6H_5]^+$ HREIMS: Calcd for $C_{28}H_{38}O_5$. 454.6094, Found 454.2718 ^1H NMR (500 MHz, $CDCl_3$) δ1 03 (3H, *s*, 19-H), 1 25 (3H, *d*, *J* = 6 8 Hz, 21-H), 1 41 (3H, *s*, 18-H), 3 54 (1H, *m*, 3-H), 3.85 (1H, *dq*, *J* = 9 6, 6 4 Hz, 20-H), 4 78 (1H, *dd*, *J* = 12 2, 4 4 Hz, 12-H), 5.44 (1H, *br s*, 6-H), 7.45 (2H, *t*, *J* = 7 3 Hz, 4′,6′-H), 7 58 (1H, *t*, *J* = 7 3 Hz, 5′-H), 8.08 (2H, *d*, *J* = 7.3 Hz, 3′,7′-H) ^{13}C NMR· see Table 1

Boucerosıde-ANO (**3**) Amorphous powder, mp 113.5–116 0°, $[\alpha]_D$ −12.4° ($CHCl_3$, *c* 1 02) UV λ_{max}^{EtOH} nm (log ε) 282 (2.95), 274 (3 05), 230 (4 12) IR $\nu_{max}^{CHCl_3}$ cm^{-1} 3400, 1710, 1600, 1580, 1275 FDMS. *m/z* 927 $[M+Na]^+$. ^1H NMR (500 MHz, $CDCl_3$) δ0 82 (3H, *s*, 19-H), 1.22 (3H, *d*, *J* = 6.8 Hz, 21-H), 1.37 (3H, *s*, 18-H), 3 38, 3 44, 3.66 (each 3H, *s*, 3-OMe of sugar moiety), 4 47 (1H, *dd*, *J* = 9 8, 2.0 Hz, ole-1-H), 4 72 (1H, *dd*, *J* = 12.2, 4 4 Hz, 12-H), 4 79 (1H, *d*, *J* = 7.8 Hz, allo-1-H), 4 86 (1H, *dd*, *J* = 9.8, 2 0 Hz, cym-1-H), 7.44 (2H, *t*, *J* = 7 3 Hz, 4′6′-H), 7.56 (1H, *t*, *J* = 7.3 Hz, 5′-H), 8 05 (1H, *d*, *J* = 7 3 Hz, 3′,7′-H) ^{13}C NMR see Tables 1 and 2

Acidic hydrolysis of **3**. A soln of **3** (15 2 mg) in MeOH (6 ml) was allowed to react in the same way as **1**, and products were separated to provide **11** (2 0 mg), **12** (1 0 mg) and **16** (1 7 mg)

Boucerosıde-ADO (**4**). Amorphous powder; mp 107 5–111 0°, $[\alpha]_D$ −11 8° ($CHCl_3$, *c* 1 03) UV λ_{max}^{EtOH} nm (log ε) 282 (2.87), 275 (2 97), 230 (3 90) IR $\nu_{max}^{CHCl_3}$ cm^{-1} 3400, 1710, 1600, 1580, 1270 FDMS· *m/z* 925 $[M+Na]^+$. ^1H NMR (500 MHz, $CDCl_3$) δ1 01 (3H, *s*, 19-H), 1.23, 1 24, 1 26 × 2 (each 3H, *d*, *J* = 6 3 Hz, 21-H and 6-H of sugar moiety), 1.41 (3H, *s*, 18-H), 3 38, 3.45, 3 66 (each 3H, *s*, 3-OMe of sugar moiety), 4 47 (1H, *dd*, *J* = 9.8, 2 0 Hz, allo-1-H), 4 76 (1H, *dd*, *J* = 12 2, 4.4 Hz, 12-H), 4 86 (1H, *dd*, *J* = 9, 1.5 Hz, cym-1-H), 5 44 (1H, *br s*, 6-H), 7 45 (2H, *t*, *J* = 7 8 Hz, 4′,

6′-H), 7 57 (1H, *t*, *J* = 7 8 Hz, 5′-H), 8 06 (2H, *d*, *J* = 7 8 Hz, 3′, 7′-H) ^{13}C NMR see Tables 1 and 2

Acidic hydrolysis of **4** A soln of **4** (12 1 mg) in MeOH (6 ml) was allowed to react in the same way as **1**, and products were separated to provide **14** (2 5 mg), **12** (1 1 mg) and **16** (1 9 mg)

Bouceroside-BNO (**5**) Amorphous powder, mp 133 5–137 0°, $[\alpha]_D$ +2 4° ($CHCl_3$, *c* 1 02) (Found, C, 62 68, H, 8 25 $C_{51}H_{78}O_{16}$ 3/2H_2O required C, 64 69, H, 8 24) UV λ_{max}^{EtOH} nm (log ε) 281 (2 79), 275 (2 90), 230 (3 97) IR $\nu_{max}^{CHCl_3}$ cm^{-1} 3400, 1710, 1600, 1580, 1275 FDMS *m/z* 946 (M^+), 969 $[M+Na]^+$ ^{1}H NMR (500 MHz, $CDCl_3$), δ 0 80 (3H, *s*, 19-H), 1 12 (3H, *d*, *J* = 6 3 Hz, 21-H), 1 14 (3H, *s*, 18-H), 1 23 (3H, *d*, *J* = 6 3 Hz, 6-H of sugar moiety), 1 26 (6H, *d*, *J* = 6 3 Hz, 6-H of sugar moiety), 1 93 (3H, *s*, 2″-H), 3 39, 3 42, 3 66 (each 3H, *s*, 3-OMe of sugar moiety), 3 55, 3 56, 3 87 (each 1H, *dq*, *J* = 9 4, 6 3 Hz, 5-H of sugar moiety), 4 48 (1H, *dd*, *J* = 9 8, 2 0 Hz, ole-1-H), 4 76 (1H, *dd*, *J* = 12 2, 4 4 Hz, 12-H), 4 79 (1H, *d*, *J* = 7 8 Hz, allo-1-H), 4 86 (1H, *dd*, *J* = 9 8, 2 0 Hz, cym-1-H), 4 87 (1H, *dq*, *J* = 9 4, 6 3 Hz, 20-H), 7.44 (2H, *t*, *J* = 7 3 Hz, 4′,6′-H), 7 56 (1H, *t*, *J* = 7 3 Hz, 5′-H), 8 09 (2H, *d*, *J* = 7 3 Hz, 3′,7′-H) ^{13}C NMR see Tables 2 and 4

Acidic hydrolysis of **5**. A soln of **5** (47 1 mg) in MeOH (6 ml) was allowed to react in the same way as **1**, and the products sepd to provide **15** (22 6 mg), **12** (3 1 mg) and **16** (5 0 mg) Compound **15** (R_{f_1}, 0 61, R_{f_2}, 0 52), amorphous powder, mp 118 5–123 0°, $[\alpha]_D$ +8 0° ($CHCl_3$, *c* 0 50) UV λ_{max}^{EtOH} nm (log ε) 281 (2 73), 275 (2 77), 229 (3 83) IR $\nu_{max}^{CHCl_3}$ cm^{-1} 3400, 1715, 1600, 1580, 1275 FDMS *m/z* 498 (M^+) ^{1}H NMR (270 MHz, $CDCl_3$) δ0 82 (3H, *s*, 19-H), 1 12 (3H, *d*, *J* = 6 2 Hz, 21-H), 1 15 (3H, *s*, 18-H), 1 94 (3H, *s*, 2″-H), 3 61 (1H, *m*, 3-H), 4 76 (1H, *dd*, *J* = 12 2, 4 4 Hz, 12-H), 4 88 (1H, *dq*, *J* = 9, 6 2 Hz, 20-H), 7 45 (2H, *t*, *J* = 7 3 Hz, 4′,6′-H), 7 58 (1H, *t*, *J* = 7 3 Hz, 5′-H), 8 10 (2H, *d*, *J* = 7 3 Hz, 3′,7′-Hz), (270 MHz, pyridine) δ 0 82 (3H, *s*, 19-H), 1 16 (3H, *d*, *J* = 6 Hz, 21-H), 1 50 (3H *s*, 18-H), 2 00 (3H, *s*, 2″-H), 3 83 (1H, *m*, 3-H), 5 12 (1H, *dd*, *J* = 12 2, 4 4 Hz, 12-H), 5 42 (1H, *dq*, *J* = 9 4, 6 Hz, 20-H), 7 54 (2H, *t*, *J* = 7 0 Hz, 4′,6′-H), 7 62 (1H, *t*, *J* = 7 0 Hz, 5′-H), 8 44 (2H, *d*, *J* = 7 0 Hz, 3′,7′-H) Compound **16** $[\alpha]_D$ −10 0° (H_2O, *c* 0 47), FDMS *m/z* 322 [M^+], ^{13}C NMR see Table 3

Bouceroside-BDO (**6**) Amorphous powder, mp 135 5–139 0°, $[\alpha]_D$ −21 0° ($CHCl_3$, *c* 1 05) UV λ_{max}^{EtOH} nm (log ε) 278 (3 04), 272 (3 16), 230 (4 17) IR $\nu_{max}^{CHCl_3}$ cm^{-1} 3400, 1710, 1600, 1580, 1275 FDMS *m/z* 944 [M^+] ^{1}H NMR (500 MHz, $CDCl_3$) δ1 00 (3H, *s*, 19-H), 1 13 (3H, *d*, *J* = 6 3 Hz, 21-H), 1 17 (3H, *s*, 18-H), 1 23 (3H, *d*, *J* = 6 3 Hz, 6-H of sugar moiety), 1 26 (6H, *d*, *J* = 6 3 Hz, 6-H of sugar moiety), 1 97 (3H, *s*, 2″-H), 3 39, 3 45, 3 66 (each 3H, *s*, 3-OMe of sugar moiety), 3 87 (1H, *dq*, *J* = 9 8, 6 3 Hz, 5-H of sugar moiety), 4 48 (1H, *dd*, *J* = 9 8, 2 0 Hz, ole-1-H), 4 79 (1H, *dd*, *J* = 7 8 Hz, allo-1-H), 4 80 (1H, *dd*, *J* = 12 2, 4 4 Hz, 12-H), 4 86 (1H, *dd*, *J* = 9, 1 5 Hz, cym-1-H), 4 91 (1H, *dq*, *J* = 9 8, 6 3 Hz, 20-H), 5 44 (1H, *br s*, 6-H), 7 45 (2H, *t*, *J* = 7 3 Hz, 4′,6′-H), 7 57 (1H, *t*, *J* = 7 3 Hz, 5′-H), 8 11 (2H, *d*, *J* = 7 3 Hz 3′ 7′-H) ^{13}C NMR see Tables 2 and 4

Acidic hydrolysis of **6** A soln of **6** (36 5 mg) in MeOH (6 ml) was allowed to react in the same way as **1**, and the products sepd to provide **17** (8 2 mg), **12** (2 0 mg) and **16** (4 2 mg) Compound **17** (R_{f_1}, 0 61, R_{f_2}, 0 52), amorphous powder, mp 120 0–123 5° $[\alpha]_D$ −10 3° ($CHCl_3$, *c* 0 31) UV λ_{max}^{EtOH} nm (log ε) 281 (2 68), 274 (3 01), 228 (4 14) IR $\nu_{max}^{CHCl_3}$ cm^{-1} 3400, 1715, 1600, 1580, 1275 EIMS *m/z* 496 [M^+], 374, 314 296, 105 [C_6H_5CO, base peak]$^+$ ^{1}H NMR (270 MHz, $CDCl_3$) δ1 01 (3H, *s*, 19-H), 1 13 (3H, *d*, *J* = 6 2 Hz, 21-H), 1 18 (3H, *s*, 18-H), 1 98 (3H, *s*, 2″-H), 3 53 (1H, *m*, 3-H), 4 81 (1H, *dd*, *J* = 12, 4 4 Hz, 12-H), 4 91 (1H, *dq*, *J* = 9, 6 2 Hz, 20-H), 5 43 (1H, *br s*, 6-H), 7 45 (2H, *t*, *J* = 7 3 Hz, 4′6′-H), 7 59 (1H, *t*, *J* = 7 3 Hz, 5′-H), 8 11 (2H, *d*, *J* = 7 3 Hz, 3′,7′-H) ^{13}C NMR see Table 4

Bouceroside-BNC (**7**) Amorphous powder, mp 138 5–141 0°, $[\alpha]_D$ +18 5 ($CHCl_3$, *c* 1 03) UV λ_{max}^{EtOH} nm (log ε) 281 (2 69), 274 (3.04), 230 (4 14) IR $\nu_{max}^{CHCl_3}$ cm^{-1} 3400, 1715, 1600, 1580, 1275 FDMS *m/z* 946 [M^+], 969 $[M+Na]^+$ ^{1}H NMR (500 MHz, $CDCl_3$) δ0 80 (3H, *s*, 19-H), 1 12 (3H, *d*, *J* = 6.3 Hz, 21-H), 1 14 (3H, *s*, 18-H), 1 22, 1 27, 1.28 (each 3H, *d*, *J* = 6 3 Hz, 6-H of sugar moiety), 1 92 (3H, *s*, 2″-H), 3 42, 3 44, 3 66 (each 3H, *s*, 3-OMe of sugar moiety), 4 59 (1H, *d* *J* = 7.8 Hz, allo-1-H), 4 76 (1H, *dd*, *J* = 12 2, 4 4 Hz, 12-H), 4 77, 4 85 (each 1H, *dd*, *J* = 9 8, 2 0 Hz, cym-1-H), 4 88 (1H, *dq*, *J* = 9 2, 6 3 Hz, 20-H), 7 44 (2H, *t*, *J* = 7 8 Hz, 4′,6′-H), 7 57 (1H, *t*, *J* = 7 8 Hz, 5′-H), 8 09 (2H, *d*, *J* = 7 8 Hz, 3′,7′-H) ^{13}C NMR see Tables 2 and 4

Acidic hydrolysis of **7** A soln of **7** (15 1 mg) in MeOH (6 ml) was allowed to react in the same way as **1**, and the products were separated to provide **15** (3 1 mg), **12** (1 1 mg) and **13** (2 5 mg)

Bouceroside-BDC (**8**) Amorphous powder, mp 103 5–106 0°, $[\alpha]_D$ +2 1° ($CHCl_3$, *c* 0 32) (Found C, 61 18, H, 7 97 $C_{51}H_{76}O_{16}$ 3/2H_2O required C, 64 89, H, 8 11) UV λ_{max}^{EtOH} nm (log ε) 281 (2 57), 274 (2 65), 230 (3 75) IR $\nu_{max}^{CHCl_3}$ cm^{-1} 3400, 1715, 1600, 1580, 1280 FABMS *m/z* 967 $[M+Na]^+$ ^{1}H NMR (500 MHz, $CDCl_3$) δ1 00 (3H, *s*, 19-H), 1 14 (3H, *d*, *J* = 6 3 Hz, 21-H), 1 17 (3H, *s*, 18-H), 1 22, 1 27, 1 28 (each 3H, *d*, *J* = 6 3 Hz, 6-H of sugar moiety), 1 97 (3H, *s*, 2′-H), 3 42, 3 45, 3 66 (each 3H, *s*, 3-OMe of sugar moiety), 3 85, 3 91 (each 1H, *dq*, *J* = 9 3, 6 3 Hz, 5-H of sugar moiety), 4 59 (1H, *d*, *J* = 7 8 Hz, allo-1-H), 4 76 (1H, *dd*, *J* = 9 8, 2 0 Hz, ole-1-H), 4 81 (1H, *dd*, *J* = 12 2, 4 4 Hz, 12-H), 4 85 (1H, *dd*, *J* = 9, 1 5 Hz, cym-1-H), 4 91 (1H, *dq*, *J* = 9 7, 6 3 Hz, 20-H), 5 44 (1H, *br s*, 6-H), 7 45 (2H, *t*, *J* = 7 3 Hz, 4′,6′-H), 7 58 (1H, *t*, *J* = 7 3 Hz, 5′-H), 8 11 (2H, *d*, *J* = 7 3 Hz, 3′,7′-H)

Acidic hydrolysis of **8** A soln of **8** (1 4 mg) in MeOH (3 ml) was allowed to react in the same way as **1**, and the products (**17**, **12** and **13**) were identified by TLC comparison with the authentic samples

Bouceroside-CNO (**9**) Amorphous powder, mp 143 5–147 0°, $[\alpha]_D$ −7 5° ($CHCl_3$, *c* 0 96) UV λ_{max}^{EtOH} nm (log ε) 282 (3 40), 275 (3 46), 228 (4 60) IR $\nu_{max}^{CHCl_3}$ cm^{-1} 3400, 1715, 1600, 1580, 1275 FDMS *m/z* 1031 $[M+Na]^+$, 1008 $[M]^+$ ^{1}H NMR (500 MHz, $CDCl_3$) δ0 78 (3H, *s*, 19-H), 1 15 (3H, *s*, 18-H), 1 23 (3H, *d*, *J* = 6 3 Hz, 21-H), 3 38, 3 44, 3 66 (each 3H, *s*, 3-OMe of sugar moiety), 3 55, 3 87 (each 1H, *dq*, *J* = 9 8, 6 3 Hz, 5-H of sugar moiety), 4 47 (1H, *dd*, *J* = 9 8, 2 0 Hz, ole-1-H), 4 79 (1H, *d*, *J* = 7 8 Hz, allo-1-H), 4 85 (1H, *dd*, *J* = 9 8, 2 0 Hz, cym-1-H), 4 91 (1H, *dd*, *J* = 12 2, 4 4 Hz, 12-H), 5 25 (1H, *dq*, *J* = 9 8, 6 3 Hz, 20-H), 7 21, 7 44 (each 2H, *t*, *J* = 7 8 Hz, 4′,6′ and 4″,6″-H), 7 48, 7 61 (each 1H, *t*, *J* = 7 8 Hz, 5 and 5″-H), 7 83, 8 06 (each 2H, *d*, *J* = 7 8 Hz, 3′,7′ and 3″ 7″-H) ^{13}C NMR see Tables 2 and 5

Acidic hydrolysis of **9** A soln of **9** (20 3 mg) in MeOH (6 ml) was allowed to react in the same way as **1**, and the products sepd to provide **18** (6 5 mg), **12**, (1 5 mg) and **16** (3 6 mg) Compound **18** (R_{f_1}, 0 65, R_{f_2}, 0 58) amorphous powder, mp 133 5–137 5°, $[\alpha]_D$ −5 7° ($CHCl_3$, *c* 0 65) UV λ_{max}^{EtOH} nm (log ε) 281 (3 15), 278 (3 23), 260 (3 26), 256 (3 35), 229 (4 35) IR $\nu_{max}^{CHCl_3}$ cm^{-1} 3400, 1715, 1600, 1580, 1275 EIMS *m/z* 438 [$M-C_6H_5COOH$]$^+$ 316 [$438-C_6H_5COOH$], 298 [$316-H_2O$], 105 [C_6H_5CO, base peak]$^+$ ^{1}H NMR (270 MHz, $CDCl_3$) δ0 79 (3H, *s*, 19-H), 1 16 (3H, *s*, 18-H), 1 23 (3H, *d*, *J* = 6 3 Hz, 21-H), 3 60 (1H, *m*, 3-H), 4 93 (1H, *dd*, *J* = 12 2, 4 4 Hz, 12-H), 5 26 (1H, *dq*, *J* = 9 8, 6 3 Hz, 20-H), 7 21, 7 45 (each 2H, *t*, *J* = 7 7 Hz, 4′,6′ and 4″,6″-H), 7 50, 7 65 (each 1H, *t*, *J* = 7 7 Hz, 5′ and 5″-H), 7 84, 8 06 (each 2H, *d*, *J* = 7 7 Hz, 3′,7′ and 3″,7′-H) ^{13}C NMR see Table 5

Bouceroside-CNC (**10**) Amorphous powder, mp 114 0–117 5°, $[\alpha]_D$ +8° ($CHCl_3$, *c* 0 60) UV λ_{max}^{EtOH} nm (log ε) 281 (3 53), 273 (3.60), 228 (4 65) IR $\nu_{max}^{CHCl_3}$ cm^{-1} 3400, 1715, 1610, 1590, 1280 FABMS *m/z* 1031 $[M+Na]$, ^{1}H NMR (270 MHz, $CDCl_3$) δ0 78 (3H, *s*, 19-H), 1 14 (3H, *s*, 18-H), 1 22 (3H, *d*, *J* = 8 0 Hz, 21-H), 3 42, 3 44, 3 66 (each 3H, *s*, 3-OMe of sugar moiety), 3 84 3 91 (each 1H, *dq*, *J* = 9 8, 6 Hz, 5-H of sugar moiety), 4 59 (1H, *d*, *J*

= 8 Hz, allo-1-H), 4 76, 4.85 (1H, *dd*, *J* = 9.5, 2.0 Hz, cym-1-H), 4.91 (1H, *dd*, *J* = 12, 4 Hz, 12-H), 5.25 (1H, *dq*, *J* = 10, 6.4 Hz, 20-H), 7 24, 7.45 (each 2H, *t*, *J* = 8 Hz, 4′,6′ and 4″,6″-H), 7 48, 7 62 (each 1H, *t*, *J* = 8 Hz, 5′ and 5″-H), 7 83, 8.06 (each 2H, *d*, *J* = 8 Hz, 3′,7′ and 3″,7″-H) ^{13}C NMR: see Tables 2 and 5.

Acidic hydrolysis of **10**. A soln of **10** (1 8 mg) in MeOH (6 ml) was allowed to react in the same way as **1**, and products (**18**, **12** and **13**) were identified by TLC comparison with the authentic samples

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