Novel Steroidal Glycosides from two Indian *Caralluma* species, *C. stalagmifera* and *C. indica*

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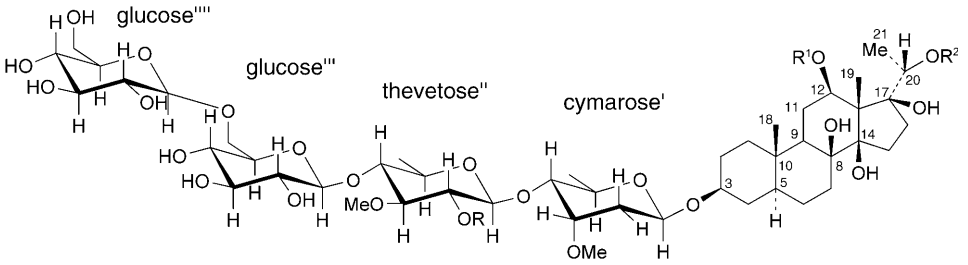
New steroidal glycosides, stalagmosides I–V (**1**–**5**) and indicosides I and II (**7** and **8**), together with the known compounds carumbelloside III, lasianthoside A, and lasianthoside B, were isolated from whole plants of *Caralluma stalagmifera* and *Caralluma indica*, respectively. Their structures were elucidated by extensive NMR spectroscopic studies.

Introduction. – The plants belonging to the genus *Caralluma* (Asclepiadaceae) are thick succulent herbs of which some have been reported to have medicinal properties. According to an Ayurvedic recipe, the juice of *Caralluma stalagmifera* mixed with black pepper is recommended to be taken orally for treating migraine [1]. A decoction of the fresh stems of *C. stalagmifera* is used orally for treating diabetes [2]. *Caralluma tuberculata* (syn. *Boucerosia aucheriana*) is considered a stomachic, carminative, and tonic [3] as well as cure for diabetes and rheumatism [4]. *C. dalzielii* is claimed to be medicinally important in African folk medicine [5]. The genus *Caralluma* is a rich source of steroidal glycosides of the pregnane type [6–9]. Extracts of *C. stalagmifera* were reported to possess anti-inflammatory activity [2].

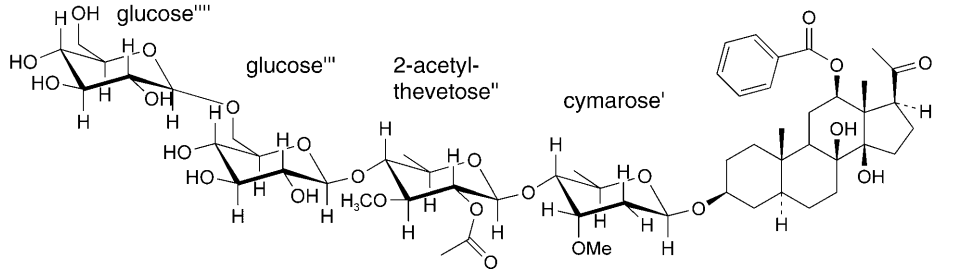
In continuation of our studies on Indian *Carallumas*, we report here on the isolation and structure elucidation of the five new steroidal glycosides **1**–**5** from *C. stalagmifera.* The two novel bisdesmosidic steroidal glycosides **6** and **7** as well as the three known steroidal glycosides **8**–**10** were isolated from *C. indica*. This is the first report on the phytochemical investigation of these two plants.

Results and Discussion. – The BuOH fraction of the whole plants of *Caralluma stalagmifera* yielded the new compounds **1**–**5**, called stalagmoside I–V, after repeated chromatography over normal-phase and reversed-phase (*C-18*) silica gel and prep. HPLC.

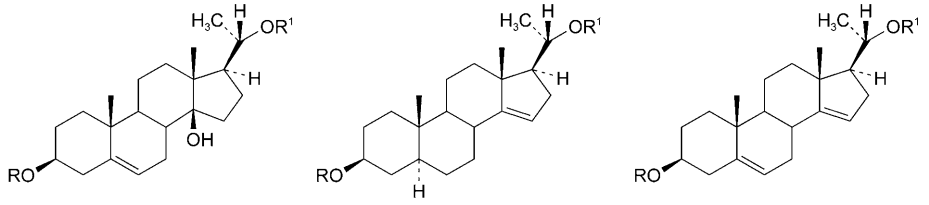
Compound **1** was obtained as a pale yellow amorphous solid. Its molecular formula was determined as $C_{54}H_{84}O_{24}$ by ESI-MS showing an ion at *m/z* 1139.4 ($[M+Na]^+$) in



1 Stalagmoside I	R = H	R^1= H	R^2 = Bz
2 Stalagmoside II	R = H	R^1= Ac	R^2 = Bz
3 Stalagmoside III	R = Ac	R^1 = Ac	R^2 = Bz
4 Stalagmoside IV	R = Ac	R^1 = Bz	R^2 = Bz
11 Hydrolysate	R = H	R^1 = H	R^2= H



5 Stalagmoside V



Caralumagenin Indicagenin Caralasigenin

6 Indicoside I	= Caralumagenin	R = β-Glc*p*''-(1→4)-β-digitalopyranosyl'	R^1 = 3''',4'''-O-dibenzoyl-β-Glc*p*'''
7 Indicoside II	= Indicagenin	R = β-Glc*p*''-(1→4)-β-digitalopyranosyl'	R^1 = 3''',4'''-O-dibenzoyl-β-Glc*p*'''
8 Carumbelloside III	= Caralumagenin	R = β-Glc*p*-(1→4)-β-digitalopyranosyl	R^1 = β-Glc*p*
9 Lasianthoside A	= Caralasigenin	R = β-Glc*p*-(1→4)-β-digitalopyranosyl	R^1 = α-Rha*p*-(1→6)-β-Glc*p*
10 Lasianthoside B	= Caralumagenin	R = β-Glc*p*-(1→4)-β-digitalopyranosyl	R^1 = α-Rha*p*-(1→6)-β-Glc*p*

the positive mode and at *m/z* 1115.4 ([*M*−H]$^-$) in the negative mode. On the basis of NMR spectroscopic evidence (*Tables 1* and *2*), compound **1** was identified as (3β,5α,12β,14β,17α,20*S*)-20-(benzoyloxy)-8,12,14,17-tetrahydroxypregnan-3-yl *O*-β-

Table 1. *^1H-* (600 MHz) *and ^{13}C-NMR* (150 MHz) *Chemical Shifts* δ [ppm] *of the Aglycone Part of Compounds* **1**–**4** *in (D$_5$)Pyridine at 40°*. $SiMe_4$ as internal standard; *J* in Hz[a]).

	1		**2**		**3**		**4**	
	δ(C)	δ(H)	δ(C)	δ(H)	δ(C)	δ(H)	δ(C)	δ(H)
CH_2(1)	38.3	0.95 (*t, J*=12, H_α), 1.75 (H_β)	38.0	0.85 (*t, J*=13.3, H_α), 1.56 (H_β)	37.9	0.85 (*t, J*=13.3, H_α), 1.57 (H_β)	37.9	0.82 (*t, J*=12, H_α), 1.50 (H_β)
CH_2(2)	29.7	1.70 (H_β), 2.02 (H_α)	29.6	1.65 (H_β), 1.96 (H_α)	29.6	1.66 (H_β), 1.97 (H_α)	29.6	1.62 (H_β), 1.93 (H_α)
H–C(3)	76.8	3.87 (H_α)	76.8	3.82 (H_α)	76.8	3.83 (H_α)	76.8	3.82 (H_α)
CH_2(4)	34.7	1.40 (H_β), 1.73 (H_α)	34.6	1.39 (H_β), 1.72 (H_α)	34.6	1.39 (H_β), 1.73 (H_α)	34.6	1.38 (H_β), 1.72 (H_α)
H–C(5)	45.5	1.02 (*t, J*=12, H_α)	45.4	1.00 (*t, J*=12, H_α)	45.3	1.00 (*t, J*=12, H_α)	45.3	1.01 (*t, J*=12, H_α)
CH_2(6)	25.3	1.80 (H_β), 1.12 (H_α)	25.2	1.75 (H_β), 1.10 (H_α)	25.2	1.78 (H_β), 1.13 (H_α)	25.2	1.77 (H_β), 1.14 (H_α)
CH_2(7)	34.7	1.45 (H_α), 2.12 (H_β)	34.4	1.42 (H_α), 2.11 (H_β)	34.4	1.44 (H_α), 2.13 (H_β)	34.5	1.48 (H_α), 2.14 (H_β)
C(8)	76.3	–	76.1	–	76.1	–	76.1	–
H–C(9)	47.8	1.26 (H_α)	46.9	1.31 (H_α)	46.8	1.32 (H_α)	46.7	1.37 (H_α)
C(10)	36.5	–	36.4	–	36.4	–	36.5	–
CH_2(11)	28.9	2.29 (H_β), 1.85 (H_α)	24.5	2.11 (H_β), 1.87 (H_α)	24.5	2.14 (H_β), 1.88 (H_α)	24.8	2.25 (H_β), 1.89 (H_α)
H–C(12)	71.1	3.71	75.1	5.07 (*dd, J*=11.4, 4)	75.1	5.06 (*dd, J*=11.6, 4)	75.5	5.36
C(13)	59.3	–	57.4	–	57.3	–	57.7	–
C(14)	88.5	–	88.6	–	88.6	6.11 (OH)	88.8	–
CH_2(15)	33.9	2.11, 2.08	33.5	2.18, 2.07	33.5	2.19, 2.10	33.6	2.11, 2.23
CH_2(16)	34.0	2.08, 1.99	33.8	2.06, 1.99	33.8	2.08, 2.00	34.1	2.08, 2.08
C(17)	88.5	–	87.8	–	87.8	6.64 (OH)	87.9	–
Me(18)	10.7	1.96 (*s*)	11.7	1.95 (*s*)	11.7	1.97 (*s*)	12.0	2.12 (*s*)
Me(19)	13.1	1.17 (*s*)	12.9	1.10 (*s*)	12.9	1.11 (*s*)	12.8	1.08 (*s*)
H–C(20)	76.3	5.96 (*q, J*=6.6)	75.6	5.20 (*q, J*=6.6)	75.6	5.20 (*q, J*=6.6)	75.4	5.28
Me(21)	15.5	1.65 (*d, J*=6.6)	15.5	1.54 (*d, J*=6.6)	15.5	1.53 (*d, J*=6.6)	15.5	1.48
AcO/BzO at C(12):								
CO			171.2	–	171.2	–	166.9	–
Me or C(1)			22.1	2.08 (*s*)	22.1	2.09 (*s*)	132.0	–
H–C(2,6)							130.1	7.99 (*d, J*=7.2)
H–C(3,5)							128.5	7.12 (*t, J*=7.2)
H–C(4)							132.8	7.38 (*t, J*=7.2)
BzO at C(20):								
CO	165.7	–	166.1	–	166.0	–	165.4	–
C(1)	132.3	–	131.4	–	131.4	–	131.3	–
H–C(2,6)	130.1	8.25 (*d, J*=7.6)	130.1	8.25 (*d, J*=7.2)	130.1	8.27 (*d, J*=7.8)	130.2	7.76 (*d, J*=7.2)
H–C(3,5)	128.5	7.24 (*t, J*=7.6)	128.9	7.35 (*t, J*=7.2)	128.9	7.37 (*t, J*=7.8)	128.3	7.20 (*t, J*=7.2)
H–C(4)	132.6	7.37 (*t, J*=7.6)	133.3	7.49 (*t, J*=7.2)	133.3	7.50 (*t, J*=7.8)	132.9	7.42 (*t, J*=7.2)

[a]) No multiplicities given in case of signal overlap.

Table 2. *^{1}H-* (600 MHz) *and ^{13}C-NMR* (150 MHz) *Chemical Shifts* δ [ppm] *of the Carbohydrate Moieties of Compounds* **1**–**7** *and* **11** *in (D$_5$)Pyridine at 40°*. $SiMe_4$ as internal standard; *J* in Hz[a]).

	1, **2**, **11**		**3**–**5**			**6**, **7**	
	δ(C)	δ(H)	δ(C)	δ(H)		δ(C)	δ(H)
Cym′ [b]):					Dig′ [c]):		
H–C(1′)	96.0	5.27 (*d*, *J* = 8.3)	96.1	5.24 (*d*, *J* = 9.2)	H–C(1′)	102.7	4.74 (*d*, *J* = 7.7)
CH_2(2′)	37.4	1.89, 2.29	37.2	1.86, 2.28	H–C(2′)	71.5	4.38 (*t*, *J* = 8.4)
H–C(3′)	78.3	4.03 (br. *s*)	77.5	4.00 (br. *s*)	H–C(3′)	85.4	3.56 (*d*, *J* = 9.6)
H–C(4′)	83.7	3.57	84.4	3.47 (*dd*, 9.3, 2.4)	H–C(4′)	76.8	4.30
H–C(5′)	69.4	4.28	68.8	4.21	H–C(5′)	70.4	3.77 (*q*, *J* = 8.0)
Me(6′)	18.8	1.63 (*d*, *J* = 6.8)	18.5	1.46 (*d*, *J* = 6.0)	Me(6′)	17.6	1.58 (*d*, *J* = 6.6)
*Me*O–C(3′)	58.9	3.56	58.7	3.56	*Me*O–C(3′)	59.1	3.67 (*s*)
The″ [d]) or TheAc″:					Glc″ [e]):		
H–C(1″)	106.0	4.68 (*d*, *J* = 7.8)	102.9	4.72 (*d*, *J* = 7.6)	H–C(1″)	105.6	5.10 (*d*, *J* = 7.8)
H–C(2″)	74.7	3.91 (*t*, *J* = 8.8)	73.8	5.35 (*t*, *J* = 8.3)	H–C(2″)	76.1	3.94 (*t*, *J* = 7.5)
H–C(3″)	85.9	3.67	82.8	3.74 (*t*, *J* = 9.0)	H–C(3″)	78.3	4.18
H–C(4″)	82.9	3.85 (*t*, *J* = 8.8)	82.2	3.88 (*t*, *J* = 9.0)	H–C(4″)	71.9	4.14
H–C(5″)	72.0	3.71	72.2	3.75	H–C(5″)	78.3	3.92
Me(6″)	18.6	1.68 (*d*, *J* = 6.0)	18.3	1.67 (*d*, *J* = 6.0)	CH_2(6″)	63.1	4.51 (*d*, *J* = 11.5), 4.32
Me*C*OO–C(2″)			169.7	–			
*Me*COO–C(2″)			21.0	2.10 (*s*)			
MeO–C(3″)	60.6	3.92 (*s*)	59.5	3.77 (*s*)			
Glc‴ [e]):					Glc‴ [e]):		
H–C(1‴)	104.5	4.99 (*d*, *J* = 7.6)	104.6	4.97 (*d*, *J* = 7.8)	H–C(1‴)	100.8	5.08 (*d*, *J* = 7.8)
H–C(2‴)	75.6	3.91	75.6	3.91	H–C(2‴)	73.0	4.21
H–C(3‴)	78.6	4.11	78.6	4.12 (*t*, *J* = 9.0)	H–C(3‴)	77.5	6.25 (*d*, *J* = 8.8)
H–C(4‴)	72.0	4.05	71.8	4.06	H–C(4‴)	71.4	5.99 (*t*, *J* = 8.8)
H–C(5‴)	77.2	4.05	77.4	4.07	H–C(5‴)	75.7	4.22
CH_2(6‴)	70.6	4.27, 4.77	70.5	4.29, 4.76	CH_2(6‴)	62.1	4.24, 4.17
Glc⁗ [e]):					BzO–C(3‴):		
H–C(1⁗)	105.4	5.07 (*d*, *J* = 7.5)	105.4	5.09 (*d*, *J* = 7.8)	CO	166.6	–
H–C(2⁗)	75.4	3.96 (*t*, *J* = 8.0)	75.4	3.97	C(1)	130.4	–
H–C(3⁗)	78.5	4.16	78.4	4.18 (*t*, *J* = 9.0)	H–C(2,6)	130.2	8.12
H–C(4⁗)	71.9	4.12 (*t*, *J* = 9.5)	71.9	4.12 (*t*, *J* = 9.0)	H–C(3,5)	128.7	7.24 (*t*, *J* = 7.8)
H–C(5⁗)	78.3	3.91	78.4	3.91	H–C(4)	133.4	7.35 (*t*, *J* = 7.8)
CH_2(6⁗)	62.9	4.29, 4.45 (*dd*, *J* = 11.4)	63.0	4.30, 4.46 (br. *d*, *J* = 11.6)			
					BzO–C(4‴):		
					CO	166.2	–
					C(1)	130.4	–
					H–C(2,6)	130.2	8.09
					H–C(3,5)	128.7	7.27 (*t*, *J* = 7.8)
					H–C(4)	133.5	7.38 (*t*, *J* = 7.8)

[a]) No multiplicities given in case of signal overlap. [b]) Cym = β-Cymaropyranosyl. [c]) Dig = β-Digitalopyranosyl. [d]) The = β-Thevetopyranosyl. [e]) Glc = β-Glucopyranosyl.

glucopyranosyl-(1→6)-*O*-*β*-glucopyranosyl-(1→4)-*O*-*β*-thevetopyranosyl-(1→4)-*β*-cymaropyranoside.

The ^{13}C-NMR and HSQC spectra of **1** showed the presence of 52 C-atoms, namely of 5 Me and 2 MeO, 11 CH_2, 27 CH, and 7 quaternary C-atoms, indicating two degenerated resonances. Complete assignment of ^{1}H- and ^{13}C-NMR resonances of the aglycon portion revealed a pregnane-3,8,12,14,17,20-hexol structure in **1**. Long-range correlations between H–C(20) (*δ* 5.96) and the carbonyl (165.7) function of a benzoyl group indicated the presence of this moiety at C(20). The ^{13}C-NMR data of the aglycon were in very good agreement with data published for (5*α*)-20-*O*-benzoyl-5,6-dihydrosarcostin [10]. However, we found discrepancies for the ^{13}C-NMR assignments of Me(19) and Me(21), with Me(19) assigned a higher *δ* value than Me(21) [10]. Our studies with compound **1** along with **2**–**4** in this series indicated that these assignments have to be reversed. To determine the configuration at C(20), we prepared and studied the debenzoylated compound **11**, for which ^{13}C-NMR data in the C and D rings were in very good agreement with data published for sarcostin (=(3*β*,12*β*,14*β*,17*α*,20*S*)-pregn-5-ene-3,8,12,14,17,20-hexol) [11]. Hence, the aglycone in compound **1** was identified as (5*α*)-5,6-dihydrosarcostin 20-benzoate (=(3*β*,5*α*,12*β*,14*β*,17*α*,20*S*)-pregnane-3,8,12,14,17,20-hexol 20-benzoate).

In the ^{1}H-NMR and HSQC of compound **1**, four anomeric CH signals were identified at *δ* 5.27, 5.07, 4.99, and 4.68, indicating that compound **1** was a tetroside. The large *J* values of the anomeric protons indicated their axial orientation. Complete ^{1}H- and ^{13}C-NMR resonance assignments for the saccharide units were obtained by HSQC-TOCSY, HSQC, DQF-COSY, and HMBC experiments. From the ^{1}H,^{1}H coupling data and the correlations in a 2D-NOESY experiment, the sugars present in **1** were identified as cymarose (=2,6-dideoxy-3-*O*-methyl-*ribo*-hexose), thevetose (=6-deoxy-3-*O*-methylglucose), and two glucose units. The sequence of the saccharide chain and its attachment to the aglycone was determined with an HMBC correlation experiment. Long-range correlations were observed between H–C(1‴″) (*δ* 5.07) and C(6‴) (*δ* 70.6), between H–C(1‴) (*δ* 4.99) and C(4″) (*δ* 82.9), between H–C(1″) (*δ* 4.68) and C(4′) (*δ* 83.7), and between H–C(1′) (*δ* 5.27) and C(3) (*δ* 76.6) of the aglycone. This indicated the sequence of the sugar chain as *β*-glucopyranosyl-(1→6)-*O*-*β*-glucopyranosyl-(1→4)-*O*-*β*-thevetopyranosyl-(1→4)-*β*-cymaropyranose.

Compound **2** was obtained as a white amorphous powder. The molecular formula was determined to be $C_{56}H_{86}O_{25}$. NMR Studies indicated an additional acetyl group in this compound when compared with **1**. This group was found to be present at C(12) as determined by HMBC correlation between H–C(12) (*δ* 5.07) and the carboxyl group of the acetyl unit (*δ* 171.2). Therefore, **2** was identified as (3*β*,5*α*,12*β*,14*β*,17*α*,20*S*)-12-(acetyloxy)-20-(benzoyloxy)-8,14,17-trihydroxypregnan-3-yl *O*-*β*-glucopyranosyl-(1→6)-*O*-*β*-glucopyranosyl-(1→4)-*O*-*β*-thevetopyranosyl-(1→4)-*β*-cymaropyranoside.

Compound **3** was a white amorphous powder. Its molecular formula $C_{58}H_{88}O_{26}$ and NMR data indicated the presence of an additional Ac group when compared with **2**, which was found to be attached to C(2″) of the thevetose unit. Thus, compound **3** was identified as (3*β*,5*α*,12*β*,14*β*,17*α*,20*S*)-12-(acetyloxy)-20-(benzoyloxy)-8,14,17-trihydroxypregnan-3-yl *O*-*β*-glucopyranosyl-(1→6)-*O*-*β*-glucopyranosyl-(1→4)-*O*-2-*O*-acetyl-*β*-thevetopyranosyl-(1→4)-*β*-cymaropyranoside.

Compound **4**, also a white amorphous powder, had the molecular formula $C_{63}H_{90}O_{26}$. The NMR data of **4** indicated a difference in the aglycone portion, whereas the saccharide chain was found to be identical to that of **3**. HMBC Correlations indicated the presence of a benzoyloxy group at H–C(12) instead of an acetyloxy group when compared with **3**. Hence, compound **4** was identified as (3*β*,5*α*,12*β*,14*β*,17*α*,20*S*)-12,20-bis(benzoyloxy)-8,14,17-trihydroxypregnan-3-yl *O*-*β*-

glucopyranosyl-(1→6)-*O*-*β*-glucopyranosyl-(1→4)-*O*-2-*O*-acetyl-*β*-thevetopyranosyl-(1→4)-*β*-cymaropyranoside.

Compound **5**, obtained as white flaky solid, had a molecular formula of $C_{56}H_{84}O_{24}$. In contrast to **1**–**4**, compound **5** was found to be a pregnanone derivative. Its sugar moiety was identical to that of **4**, and the genin showed a benzoyloxy group at C(12). From NMR spectroscopic analysis (*Tables 2* and *3*), compound **5** was identified as (3*β*,5*α*,12*β*,14*β*)-12-(benzoyloxy)-3-{[*O*-*β*-glucopyranosyl-(1→6)-*O*-*β*-glucopyranosyl-(1→4)-*O*-2-*O*-acetyl-*β*-thevetopyranosyl-(1→4)-*β*-cymaropyranoyl]oxy}-8,14-dihydroxypregnan-20-one.

The ether fraction of the whole plant of *Caralluma indica* yielded on repeated chromatography over normal-phase and reversed-phase (*C-18*) silica gel the new compounds **6** and **7**, called indicoside I and II. The three known compounds **8**–**10** were isolated from the BuOH fraction.

Compound **6** was a white amorphous powder. Its molecular formula was determined as $C_{54}H_{74}O_{19}$ by ESI-MS. From ^{1}H- and ^{13}C-NMR spectra, **6** was found to be a bisdesmosidic glycoside of caralumagenin (=(3*β*,14*β*,20*S*)-pregn-5-ene-3,14,20-triol) [12]. An *O*-*β*-glucopyranosyl-(1→4)-*β*-digitalopyranose moiety was found attached at C(3) as well as a glucopyranose moiety at C(20) which was benzoylated at positions C(3‴) and C(4‴). These findings were established by HMBC correlations (H–C(1″) (*δ* 5.10)/C(4′) (*δ* 76.8), H–C(4′) (*δ* 4.30)/C(1″) (*δ* 105.6), H–C(1′) (*δ* 4.74)/C(3) (*δ* 78.5), H–C(20) (*δ* 4.47)/C(1‴) (*δ* 100.8), H–C(3‴) (*δ* 6.25)/*C*OO–C(3‴) (*δ* 166.6), and H–C(4‴) (*δ* 6.01)/*C*OO–C(4‴) (*δ* 166.2)). Hence, compound **6** was assigned as (3*β*,14*β*,20*S*)-20-[(3,4-di-*O*-benzoyl-*β*-glucopyranosyl)oxy]-14-hydroxy-pregn-5-en-3-yl-*O*-*β*-glucopyranosyl-(1→4)-*β*-digitalopyranoside.

Compound **7** was a white amorphous powder. Its molecular formula was determined as $C_{54}H_{74}O_{18}$ by ESI-MS. Complete assignment of ^{1}H- and ^{13}C-NMR resonances revealed a pregnane-3,20-diol structure with a C=C bound between C(14) and C(15). Assignments of all diastereotopic protons of CH_2 groups in the aglycon was achieved by analysis of the homonuclear *J* patterns, NOEs, and intensities in the DQF-COSY plot. The configuration at C(5) was also determined by a NOESY experiment (NOEs H–C(5), H–C(9), H–C(3), and H_{eq} of CH_2(4) and CH_2(6)). As the chemical-shift values of the D-ring C-atoms were identical to lasianthoside A (**9**), the configuration at C(17) and C(20) was determined to be identical in these compounds [12]. The genin in **7** has not been described yet and was named indicagenin (=(5*α*,3*β*,20*S*)-pregn-14-ene-3,20-diol). The saccharide chain in **7** was found to be the same as in compound **6**. Thus, compound **7** was identified as (5*α*,3*β*,20*S*)-20-[(3,4-di-*O*-benzoyl-*β*-glucopyranosyl)oxy]-pregn-14-en-3-yl *O*-*β*-glucopyranosyl-(1→4)-*β*-digitalopyranoside.

Indicosides I (**6**) and II (**7**) represent compounds with a rare substitution pattern. Monobenzoylation of sugar units in saponins is known in carumbelloside IV isolated earlier from *Caralluma umbellata*, which has a benzoyloxy group at C(2) of the glucose unit attached to C(20) of the aglycon [13]. However, dibenzoylation of vicinal OH groups as found in compounds **6** and **7** has not been reported for steroidal glycosides, to the best of our knowledge. The steric hinderence caused by this vicinal dibenzoylation is perhaps responsible for an observed hydrolytic degradation of these compounds under aqueous conditions during HPLC. NMR Analysis of the degradation products showed that the dibenzoylated compounds had lost one of their benzoyl groups.

Table 3. *^1H* (600 MHz) *and ^{13}C-NMR* (150 MHz) *Chemical Shifts* δ [ppm] *of the Aglycone Part of Compounds* **5**–**7** *and* **11** *in (D$_5$)Pyridine at 40°*. $SiMe_4$ as internal standard; *J* in Hz[a]).

	5		**11**		**7**		**8**	
	δ(C)	δ(H)	δ(C)	δ(H)	δ(C)	δ(H)	δ(C)	δ(H)
CH_2(1)	38.2	0.90 (*t*, *J*=12, H_α), 1.62 (H_β)	38.4	0.97 (H_α), 1.76 (H_β)	37.8	1.13 (H_α), 1.82 (H_β)	37.4	0.88 (*t*, *J*=12, H_α), 1.70 (H_β)
CH_2(2)	29.6	1.69, 2.01	29.6	1.72 (H_β), 2.03 (H_α)	30.2	1.73 (H_β), 2.13 (H_α)	30.0	1.61 (H_β), 2.04 (H_α)
H–C(3)	76.8	3.86	76.7	3.89	78.5	3.91 (H_α)	77.5	3.90 (H_α)
CH_2(4)	34.6	1.41 (H_β), 1.74 (H_α)	34.6	1.42 (H_β), 1.75 (H_α)	39.4	2.39 (H_β), 2.67 (H_α)	34.9	1.36 (H_β), 1.81 (H_α)
H–C(5) or C(5)	45.4	1.01 (*t*, *J*=12, H_α)	45.5	1.01 (*t*, *J*=12, H_α)	140.0	–	44.4	0.99 (*t*, *J*=12, H_α)
CH_2(6) or H–C(6)	25.3	1.79, 1.12	25.3	1.12 (*d*, *J*=12.6, H_α), 1.80 (H_β)	122.6	5.47 (br. *s*)	30.3	1.24 (H_β), 1.84 (H_α)
CH_2(7)	35.5	1.40, 2.15	34.8	1.44 (H_α), 2.13 (*d*, *J*=12.6, H_β)	28.0	2.04 (H_α), 2.58 (H_β)	28.8	1.24, 1.24
C(8)	76.1	–	76.8	–	37.9	1.99 (H_β)	35.2	2.02 (H_β)
H–C(9)	47.9	1.36 (H_α)	47.5	1.28 (H_α)	46.8	1.24 (H_α)	54.2	0.68 (H_α)
C(10)	36.7	–	36.6	–	37.8	–	35.9	–
CH_2(11)	23.8	2.24, 1.96	28.1	1.93 (H_α), 2.36 (*q*, *J*=12.6, H_β)	21.0	1.43, 1.43	22.0	1.34 (H_β), 1.54 (H_α)
H–C(12) or CH_2(12)	79.1	5.17 (*dd*, *J*=12, 2.4)	71.5	3.86 (H_α)	41.3	1.38, 1.55	42.4	1.33, 2.52 (br. *d*, *J*=9)
C(13)	55.3	–	59.1	–	47.6	–	47.6	–
C(14)	86.5	–	88.6	–	84.8	–	156.4	–
CH_2(15) or H–C(15)	36.0	2.13, 1.98	34.0	2.04[b]), 1.95[b])	33.1	1.75, 1.86	116.3	5.15 (br. *s*)
CH_2(16)	25.1	1.96, 2.16	34.0	2.04[b]), 1.95[b])	25.5	1.95, 1.98	34.0	1.93, 2.16
H–C(17) or C(17)	59.2	3.26 (br. *s*)	89.4	–	56.4	1.97	59.1	2.03
Me(18)	13.0	1.59 (*s*)	11.7	1.88 (*s*)	16.8	1.50 (*s*)	17.1	1.29 (*s*)
Me(19)	13.2	1.16 (*s*)	13.1	1.20 (*s*)	19.5	0.96 (*s*)	12.2	0.71 (*s*)
C(20) or H–C(20)	214.6	–	72.9	4.42 (*q*, *J*=6)	76.9	4.47 (*q*, *J*=6)	74.2	4.33
Me(21)	31.9	2.14 (*s*)	17.6	1.47 (*d*, *J*=6)	18.5	1.41 (*d*, *J*=6)	18.6	1.25 (*d*, *J*=6)
BzO–C(12):								
CO	166.5	–						
C(1)	131.3	–						
H–C(2,6)	129.9	8.30 (*d*, *J*=7.6)						
H–C(3,5)	129.1	7.47 (*t*, *J*=7.6)						
H–C(4)	133.5	7.51						

[a]) No multiplicities given in case of signal overlap. [b]) δ(H) are interchangeable.

In addition to the novel compounds **6** and **7**, the three known steroidal glycosides carumbelloside III (**8**), lasianthoside A (**9**), and lasianthoside B (**10**) were isolated from the BuOH fraction of *C. indica*. These compounds were isolated before from *C. umbellata* [13] and *C. lasiantha* [12].

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Experimental Part

1. *General.* CC = Column chromatography. TLC: precoated silica gel *60* F_{254} and silica gel *RP-C-18* plates (both *Merck*), detection by spraying the plates with vanillin/H_2SO_4 reagent followed by heating. HPLC (anal. and prep.): *Shimadzu* model *LC-8A* on *YMC-pack*, *R&D ODS* column (250 × 4.6 mm, 250 × 20 mm) and UV detector *Shimadzu SPD-10AVP*. Melting points: *Tempo* melting-point apparatus; uncorrected. UV Spectra: *Elico-SL-164* double-beam UV/VIS spectrophotometer; λ_{max} in nm. IR Spectra: *Perkin-Elmer* spectrometer, *BX series*; in cm^{-1}. NMR: ^1H-, ^{13}C-, and 2D-NMR experiments with *Varian-Unity-Inova-600* spectrometer, experimental parameters according to [14], δ in ppm, *J* in Hz. Mass spectra: ESI *via* flow injection analysis on a LC/MS system consisting of an *Agilent-1100* HPLC system and an *Agilent-G1946D-1100* single quadrupole mass spectrometer (*SL* type); in *m/z*.

2. *Plant Material*. *C. stalagmifera* C. E. C. FISCHER was collected from Kakatiya University campus, Warangal, India, in the first week of August 2002. *C. indica* N. E. BROWN was collected from Bommavaram of Nellore district, Andhra Pradesh, India, in January 2003. The plants were identified by Prof. *V. S. Raju*, Department of Botany, Kakatiya University. Voucher specimens of the plants are deposited in the University College of Pharmaceutical Sciences, Kakatiya University.

3. *Extraction and Isolation*. The fresh whole plant (3.5 kg) of *C. stalagmifera* was chopped, crushed, and extracted with EtOH (10 l) at r.t. for seven days. The extract was filtered and the filtrate flash evaporated. The concentrated extract (40 g) was dispersed in H_2O (500 ml) and extracted successively with toluene, AcOEt, and BuOH. The extracts were evaporated: 8.70 g from the toluene extract, 3.12 g from the AcOEt extract, and 8.90 g from the BuOH extract.

The extract from BuOH was subjected to CC (silica gel *C-18*, MeCN/H_2O 1:1): *Fractions A – D. Fr. A – C*, on repeated CC (normal-phase silica gel, $CHCl_3$/MeOH/H_2O 80:20:2; silica gel *C-18*, MeCN/H_2O 40:60), yielded 12 mg of stalagmoside I (**1**), 120 mg of stalagmoside II (**2**), and 428 mg of stalagmoside III (**3**). *Fr. D*, on prep. HPLC (MeCN/H_2O 40:60, 20 ml/min) gave 1.9 mg stalagmoside of IV (**4**, t_R 26.7 min) and 33 mg of stalagmoside V (**5**, t_R 33.7 min).

The fresh whole plant (10 kg) of *C. indica* was processed in a similar way as described for *C. stalagmifera*. The EtOH extract (410 g) was dispersed in 1 l of H_2O and fractionated successively with Et_2O, AcOEt, butanone, and BuOH.

The Et_2O fraction (40 g), on repeated CC (normal-phase silica gel, $CHCl_3$/MeOH/H_2O 90:16:1; silica gel *C-18*, MeOH/H_2O 75:25), yielded 380 mg of indicoside I (**6**) and 20 mg of indicoside II (**7**). The BuOH fraction (23 g), on CC (silica gel, AcOEt/MeOH/H_2O 75:15:10), yielded the three known steroidal glycosides carumbelloside III (**8**) and lasianthoside A and B (**9** and **10**, resp.).

Stalagmoside I (= *(3β,5α,2β,14β,17α,20S)-20-(Benzoyloxy)-8,12,14,17-tetrahydroxypregnan-3-yl* O-*β-Glucopyranosyl-(1→6)*-O-*β-glucopyranosyl-(1→4)*-O-*6-deoxy-3*-O-*methyl-β-glucopyranosyl-(1→4)-2,6-dideoxy-3*-O-*methyl-β*-ribo-*hexopyranoside*; **1**): Pale yellow amorphous solid. M.p. 176–180°. UV (MeOH): 210. IR (KBr): 3431, 2928, 1701, 1654, 1280, 1078. ^1H- and ^{13}C-NMR: *Tables 1* and *2*. ESI-MS: 1139.4 ($[M+Na]^+$), 1115.4 ($[M-H]^-$).

Stalagmoside II (= *(3β,5α,12β,14β,17α,20S)-12-(Acetyloxy)-20-(benzoyloxy)-8,14,17-trihydroxypregnan-3-yl* O-*β-Glucopyranosyl-(1→6)*-O-*β-glucopyranosyl-(1→4)*-O-*6-deoxy-3*-O-*methyl-β-glucopyra-*

nosyl-(1→4)-2,6-dideoxy-3-O-*methyl-β*-ribo-*hexopyranoside*; **2**): White amorphous solid. M.p. 196–198°. UV (MeOH): 210. IR (KBr): 3500, 2950, 1717, 1458, 1081, 715. ^{1}H- and ^{13}C-NMR: *Tables 1* and *2*. ESI-MS: 1181.5 ($[M+Na]^+$), 1157.4 ($[M-H]^-$).

Stalagmoside III (=*3β,5α,12β,14β,17α,20S)-12-(Acetyloxy)-20-(benzoyloxy)-8,14,17-trihydroxypregnan-3-yl* O-*β-Glucopyranosyl-(1→6)*-O-*β-glucopyranosyl-(1→4)*-O-*2*-O-*acetyl-6-deoxy-3*-O-*methyl-β-glucopyranosyl-(1→4)-2,6-dideoxy-3*-O-*methyl-β*-ribo-*hexopyranoside*; **3**): White amorphous solid. M.p. 193–195°. UV (MeOH): 218. IR (KBr): 3448, 2925, 1735, 1654, 1451, 1376, 1240, 1070, 716. ^{1}H- and ^{13}C-NMR: *Tables 1* and *2*. ESI-MS: 1223.5 ($[M+Na]^+$), 1199.5 ($[M-H]^-$).

Stalagmoside IV (=*(3β,5α,12β,14β,17α,20S)-12,20-Bis(benzoyloxy)-8,14,17-trihydroxypregnan-3-yl* O-*β-Glucopyranosyl-(1→6)*-O-*β-glucopyranosyl-(1→4)*-O-*2*-O-*acetyl-6-deoxy-3*-O-*methyl-β-glucopyranosyl(1→4)-2,6-dideoxy-3*-O-*methyl-β*-ribo-*hexopyranoside*; **4**): White amorphous solid. ^{1}H- and ^{13}C-NMR: *Tables 1* and *2*. ESI-MS: 1285.5 ($[M+Na]^+$), 1261.5 ($[M-H]^-$).

Stalagmoside V (=*3β,5α,12β,14β)-12-(Benzoyloxy)-3-{[*O-*β-glucopyranosyl-(1→6)*-O-*β-glucopyranosyl-(1→4)*-O-*2*-O-*acetyl-6-deoxy-3*-O-*methyl-β-glucopyranosyl-(1→4)-2,6-dideoxy-3*-O-*methyl-β*-ribo-*hexopyranosyl]oxy}-8,14-dihydroxypregnan-20-one*; **5**): White flakes. M.p. 178–180°. UV (MeOH): 206. IR (KBr): 3448, 2927, 1718, 1279, 1070, 712. ^{1}H- and ^{13}C-NMR: *Tables 2* and *3*. ESI-MS: 1163.4 ($[M+Na]^+$), 1139.5 ($[M-H]^-$).

Indicoside I (=*3β,14β,20S)-20-[3,4-Di*-O-*benzoyl-β-glucopyranosyl)oxy]-14-hydroxypregn-5-en-3-yl-3* O-*β-Glucopyranosyl-(1→4)-6-deoxy-3*-O-*methyl-β-galactopyranoside*; **6**): White amorphous solid. M.p. 128–129°. IR (KBr): 3418, 2938, 1731, 1653, 1452, 1070. ^{1}H- and ^{13}C-NMR: *Tables 2* and *3*. ESI-MS: 1049.4 ($[M+Na]^+$), 1025.4 ($[M-H]^-$).

Indicoside II (=*3β,5α,20S)-20-[(3,4-Di*-O-*benzoyl-β-glucopyranosyl)oxy]pregn-14-en-3-yl* O-*β-Glucopyranosyl-(1→4)-6-deoxy-3*-O-*methyl-β-galactopyranoside*; **7**): White amorphous solid. M.p. 182–185°. IR (KBr): 3448, 2929, 1718, 1654, 1560, 1071. ^{1}H- and ^{13}C-NMR: *Tables 2* and 3. ESI-MS: 1033.4 ($[M+Na]^+$), 1009.4 ($[M-H]^-$).

4. *Mild Basic Hydrolysis of* **3**. Compound **3** (20 mg) was hydrolyzed with 0.1N NaOH by heating at 40° for 1 h. The deacylated product (*(3β,5α,12β,14β,17α,20S)-8,12,14,17,20-Pentahydroxypregnan-3-yl* O-*β-Glucopyranosyl-(1→6)*-O-*β-glucopyranosyl-(1→4)*-O-*6-deoxy-3*-O-*methyl-β-glucopyranosyl-(1→4)-2,6-dideoxy-3*-O-*methyl-β*-ribo-*hexopyranoside*; **11**) was extracted with BuOH. ^{1}H- and ^{13}C-NMR: *Tables 2* and *3*.

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