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Polyoxypregnane glycosides from the flowers of *Dregea volubilis*

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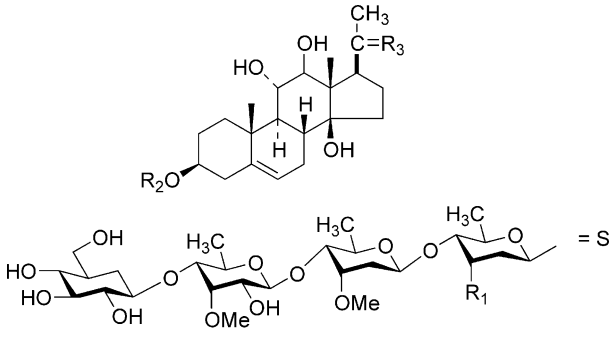
Abstract

Three novel polyoxypregnane glycosides, volubiloside A, B and C (**1–3**), were isolated from the flowers of *Dregea volubilis* Linn., and their structures were elucidated as drevogenin D-3-*O*-β-D-glucopyranosyl (1→4)-6-deoxy-3-*O*-methyl-β-D-allopyranosyl (1→4)-β-D-cymaropyranosyl (1→4)-β-D-cymaropyranoside, drevogenin D-3-*O*-β-D-glucopyranosyl (1→4)-6-deoxy-3-*O*-methyl-β-D-allopyranosyl (1→4)-β-D-cymaropyranosyl (1→4)-β-D-digitoxopyranoside and drevogenin P-3-*O*-β-D-glucopyranosyl (1→4)-6-deoxy-3-*O*-methyl-β-D-allopyranosyl (1→4)-β-D-cymaropyranosyl (1→4)-β-D-cymaropyranoside, respectively, on the basis of extensive NMR experiments, MALDI-TOF MS, and some chemical strategies.

Keywords: *Dregea volubilis*; *Wattakaka volubilis*; *Marsdenia volubilis*; Asclepiadaceae; Polyhydroxypregnane glycoside; Volubiloside A, B and C; Drevogenin D; Drevogenin P; Conduritol A; Quercetin; Quercetin-3-*O*-rutinoside, 2D NMR

1. Introduction

Dregea volubilis (L. f.) Benth. ex Hook. f. (Syn. *Wattakaka volubilis* (L. f.) Stapf., *Marsdenia volubilis* Cooke) (Family: Asclepiadaceae) commonly known as "Jukti" is a tall, woody climber occurring throughout the hotter parts of India. The alcoholic extract of the plant is reported (Chadha, 1976) to show activity on the central nervous system, as well as anticancer activity against sarcoma 180 in the mice. Previous chemical investigations (Yoshimura et al., 1983, 1985) on the ground stems of the plant *D. volubilis* collected from Thailand led to the isolation of a number of C/D-*cis*-polyoxypregnane glycoside derivatives. However, no detailed chemical investigation has appeared on the plant growing in India. The present paper reports the isolation and characterization of three new polyoxypregnane glycosides, named volubiloside A, B and C (**1–3**) along with conduritol A, quercetin, and quercetin-3-*O*-rutinoside from the flowers of the plant.



1. Volubiloside A : R_1 = OMe, R_2 = S, R_3= 

2. Volubiloside **B** : R_1 = OH, R_2 = S, R_3= 

3. Volubiloside **C** : R_1 = OMe, R_2 = S, R_3= O

4. Drevogenin D : R_2 = H, R_3 =

5. Drevogenin P: R_2 = H, R_3 = O

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2. Results and discussion

Volubiloside A (**1**) displayed a quasimolecular ion peak at m/z 999 $[M+Na]^+$ in the MALDI-TOF mass spectrum indicating the molecular weight to be 976. This information, together with the data from the elemental analysis and ^{13}C NMR DEPT spectrum, suggested the molecular formula to be $C_{48}H_{80}O_{20}$. The isolate gave positive Liebermann–Burchard and Keller–Kiliani tests indicating the presence of Δ^5-steroid and 2-deoxy sugar(s) moieties in the molecule. The carbon and proton signals of **1** in the NMR spectra were assigned by extensive studies of ^1H–^1H COSY, HETCOR, TOCSY and HMBC spectra (Tables 1 and 2). The ^1H and ^{13}C NMR spectra displayed signals for four anomeric protons at δ 4.98 (*d*, $J=7.8$ Hz), 5.08 (*d*, $J=7.8$ Hz), 5.09 (*dd*, $J=1.7$, 9.4 Hz), and 5.28 (*dd*, $J=1.6$, 9.6 Hz) (labeled A–D respectively) and corresponding carbons at δ 106.5, 104.0, 100.4 and 96.3, respectively (Tables 1 and 2). Acid hydrolysis of **1** afforded an aglycone, identified as drevogenin D (**4**) from its physical and spectral data (Singhal et al., 1980; Yoshimura et al., 1985). Paper chromatography of the acid hydrolyzate revealed the existence of three different kinds of monosaccharides, two of which were identified as glucose and cymarose by direct comparison with authentic samples. Interpretation of the COSY and TOCSY spectra revealed that three of the sugar units belonged to 6-deoxy sugars (systems B, C, and D). The other sugar was identified to be β-D-glucopyranose based on the acid hydrolysis experiment and the all *trans*-diaxial disposition of the ring protons (Table 2). The spin systems of the C and D sugars each contained four methines, one methylene and a terminal methyl group, and both of them displayed a correlation pattern in the COSY spectrum characteristic of 2,6-deoxy sugars (Jia et al., 2002). The location of a methoxy group at C-3 was determined from the long-range HMBC couplings (δ 3.60/78.1 and 4.03/58.9 for C; 3.61/78.0 and 4.03/59.0 for D). In combination with the information from the ^{13}C NMR spectrum and NOESY, the C and D sugar units were identified as 2,6-dideoxy-3-*O*-methyl-β-allopyranose (cymarose).

Table 1
^{13}C NMR chemical shifts of **1–5** in pyridine-d_5[a]

Carbon	1	2	3	4	5			1	2	3
Aglycone part						*Sugar moiety*				
1.	39.8	39.8	40.0	40.2	40.1	Digitoxose:	C-1		96.3	
2.	30.6	30.6	30.7	33.0	32.8		C-2		37.3	
3.	77.8	77.8	77.9	70.6	70.8		C-3		67.8	
4.	39.4	40.0	40.1	44.2	44.1		C-4		83.4	
5.	140.7	140.7	140.7	141.7	141.2		C-5		69.0	
6.	122.3	122.3	122.4	121.8	121.9		C-6		18.6	
7.	28.3	28.3	28.4	28.4	28.2	Cymarose:	C-1	96.3	100.4	96.4
8.	38.2	38.2	37.5	38.5	38.5		C-2	37.3	39.0	37.3
9.	49.9	50.0	49.9	50.2	49.9		C-3	78.0	78.1	78.2
10.	39.5	39.5	39.5	39.6	39.5		C-4	83.2	83.5	83.5
11.	71.6	71.6	71.9	71.8	71.4		C-5	69.0	69.3	69.2
12.	80.5	80.5	78.5	80.7	78.2		C-6	18.6	18.6	18.7
13.	54.1	54.1	55.7	54.2	56.0		–OMe	59.0	58.8	59.2
14.	84.3	84.3	84.9	84.5	85.0	Cymarose:	C-1′	100.4		100.5
15	34.1	34.1	35.3	34.3	34.9		C-2′	37.1		37.5
16.	27.1	27.1	24.5	27.3	24.7		C-3′	78.1		78.3
17.	54.7	54.7	58.8	55.0	58.6		C-4′	83.4		83.5
18.	11.5	11.6	11.0	11.7	10.9		C-5′	69.2		69.4
19.	19.0	19.0	19.1	19.3	19.2		C-6′	18.5		18.4
20.	70.5	70.5	216.7	71.8	216.2		–OMe′	58.9		59.0
21.	23.7	23.7	32.6	23.8	32.5	Allose:	C-1	104.0	103.6	104.2
							C-2	72.5	72.3	72.7
							C-3	83.0	83.0	83.4
							C-4	83.0	83.0	83.4
							C-5	69.3	68.8	69.4
							C-6	18.3	18.3	18.7
							–OMe′	61.8	61.8	61.9
						Glucose:	C-1	106.5	106.5	106.7
							C-2	75.5	75.5	75.6
							C-3	78.4	78.4	78.5
							C-4	71.9	71.9	72.1
C-5	78.4	78.0	78.4							
							C-6	63.0	63.0	63.2

[a]Assignments based upon COSY, TOCSY, HETCOR, NOESY, DEPT and HMBC.

Table 2
^{1}H NMR chemical shifts for the sugar units[a]

Sugar units	1	2		1	2
Digitoxose			*Allose*		
H-1		5.28 *dd* (1.7, 9.6)	H-1	5.08 *d* (7.8)	5.14 *d* (7.8)
H-2		1.88 *m*	H-2	3.80 *dd* like	3.76 *m*
		2.29 *dd* like			
H-3		4.63	H-3	4.45 *t* (2.8)	4.43 *t* (2.75)
H-4		3.48 *dd* (2.5, 9.4)	H-4	3.72 *dd* (2.5, 9.9)	3.68 *dd* (2.5, 9.8)
H-5		4.20 *m*	H-5	4.26 *dd* (6.0, 9.9)	4.27 *m*
H-6		1.33 *d* (6.1)	H-6	1.63 *d* (6.1)	1.58 *d* (6.2)
			OMe	3.84 *s*	3.84 *s*
Cymarose			*Glucose*		
H-1	5.28 *dd* (1.6, 9.6)	5.31 *dd* (1.6, 9.7)	H-1	4.98 *d* (7.8)	4.97 *d* (7.8)
H-2	2.29	2.40 *dd* like	H-2	4.03 *m*	4.03 *m*
	1.89 *m*	1.96 *m*	H-3	4.26 *t* (9.0)	4.26 *m*
H-3	4.03 *m*	4.05 *m*	H-4	4.20 *m*	4.20 *dd* (6.6, 9.4)
H-4	3.47 *dd* (2.5, 9.9)	3.48 *dd* (2.5, 9.4)			
H-5	4.21 *m*	4.28 *m*	H-5	4.0 *m*	3.99 *m*
H-6	1.34 *d* (6.1)	1.49 *d* (6.2)	H-6	4.38 *dd* (5.2, 11.4)	4.36 *m*
OCH_3 (11.5)	3.61 *s*	3.6 *s*		4.54 *dd* (2.1, 11.4)	4.54 *d*
Cymarose					
H-1	5.09 *dd* (1.7, 9.4)				
H-2	2.29				
	1.81				
H-3	4.03 *m*				
H-4	3.45 *dd* (2.5, 9.9)				
H-5	4.15 *m*				
H-6	1.45 *d* (6.0)				
OCH_3	3.60 *s*				

[a]Assignments based upon COSY, TOCSY, HETCOR, DEPT, NOESY and HMBC experiments. Coupling constants (in Hz) are given in parentheses.

The remaining spin-system displayed five ring methines and a terminal methyl group. Starting from the anomeric proton, all of the protons belonging to this spin system were delineated (Table 2) using COSY and TOCSY. The large coupling constants between H-1 and H-2 (7.8 Hz) and H-4 and H-5 (9.9 Hz) indicated a *trans*-diaxial relationship between these protons. A coupling constant of 2.8 Hz between H-2 and H-3 indicated a gauche disposition between these two protons. The same relationship was also true for H-3 and H-4 (2.5 Hz). NOESY showed significant through-space interaction between H-1 and H-5, as well as H-2 and H-4. As in cymarose, a methoxy group was present at C-3 of B as deduced from the long range HMBC correlations. The above information suggested that the system B was 6-deoxy-3-*O*-methyl-β-allopyranose. The identity of the sugar was further confirmed by comparing its ^{13}C NMR chemical shifts with those reported in the literature (Yoshimura et al., 1983). All of the monosaccharides were in a β-configuration as inferred from the *J* values of the respective anomeric protons.

The determination of the inter-sugar linkages was accomplished from the following HMBC correlations: H-1 of spin system A (glucose) with C-4 of B (6-deoxy-3-*O*-methyl-β-D-allopyranose); H-1 of B with C-4 of C (cymarose); and H-1 of C with C-4 of D (cymarose). The absolute configurations of the monosaccharides were not determined, but are suggested in keeping with those most commonly encountered among plant steroid glycosides. The attachment of the tetrasaccharide moiety to C-3 of the aglycone was deduced from the long-range correlation shown between H-1 of D with C-3 of the aglycone. In addition, a ^{13}C NMR spectral comparison of **1** with its aglycone **4** revealed a glycosylation shift (Kasai et al., 1977; Seo et al., 1978) in the resonances of C-2 (−2.4 ppm), C-3 (+7.2), and C-4 (−4.8) of the aglycone moiety. These data further supported that the sugar moiety was located only at the C-3 position of the aglycone. Support was also inferred from a phase-sensitive NOESY experiment. Thus H-1 (δ 4.98) of the terminal glucose (A) was correlated with H-4 (δ 3.72) of 6-deoxy-3-*O*-methyl-β-D-allopyranosyl (B), H-1 (δ 5.08) of B with H-4 (δ 3.45) of the inner cymarose (C), H-1 (δ 5.09) of C with H-4 (δ 3.47) of the second cymarose (D), whose anomeric proton (δ 5.28) showed correlation with H-3 (δ 3.89) of the aglycone. From the foregoing evidence, the structure of volubiloside A (**1**) was elucidated as drevogenin D-3-*O*-β-D-glucopyranosyl (1→4)-6-deoxy-3-*O*-methyl-β-D-allopyranosyl-(1→4)-β-D-cymaropyranosyl-(1→4)-β-D-cymaropyranoside.

Volubiloside B (**2**) showed a $[M+Na]^+$ ion peak at *m*/z 985 in the MALDI-TOF MS suggesting a molecular

weight of 962, 14 mass units less than that of **1**. Combined with the results of an elemental analysis and the ^{13}C NMR spectrum, the molecular formula of **2** was deduced to be $C_{47}H_{78}O_{20}$. Comparison of the ^{13}C NMR spectrum of **2** with that of **1** indicated that the aglycone was identical with that of volubiloside A (**1**), but that the compounds differed in the carbohydrate moiety. On acid hydrolysis, **2** furnished drevogenin D (**4**) as the aglycone, and the four monosaccharides were identified as digitoxose, cymarose, glucose and 6-deoxy-3-*O*-methyl allose. The β-linkages of the sugars of **2** were indicated from the coupling constants of the anomeric proton signals at δ 5.28 (*dd*, $J=1.7$, 9.6 Hz, 1-H of digitoxose), 5.31 (*dd*, $J=1.6$, 9.7 Hz, 1-H of cymarose), 5.14 (*d*, $J=7.8$ Hz, 1-H of 6-deoxy-3-*O*-methyl allose), and 4.97 (*d*, $J=7.8$ Hz, 1-H of glucose). Inter-sugar and sugar-aglycone linkages were deduced from the HMBC and NOESY spectra. HMBC correlations were observed between the following carbons and protons in the oligosaccharide moieties of **2**. Thus, H-1 of glucose correlated with C-4 of 6-deoxy-3-*O*-methyl allose, H-1 of 6-deoxy-3-*O*-methyl allose with C-4 of cymarose, and H-1 of cymarose with C-4 of digitoxose. The HMBC relationship between C-3 (δ 77.8) of the aglycone and H-1 of digitoxose and a glycosylation shift ($\delta+7.2$ ppm) indicated that the sugar moiety was linked to C-3 of the aglycone. Thus the structure of volubiloside B (**2**) was established as drevogenin D-3-*O*-β-D-glucopyranosyl (1→4)-6-deoxy-3-*O*-methyl-β-D-allopyranosyl-(1→4)-β-D-cymaropyranosyl-(1→4)-β-D-digitoxopyranoside.

Volubiloside C (**3**) showed a peak at *m*/*z* 997 $[M+Na]^+$ in its MALDI-TOF MS two mass units less than **1**. In combination with the information from elemental analysis and DEPT spectrum it was concluded that the molecular formula of **3** was $C_{48}H_{78}O_{20}$. The IR spectrum of the compound displayed an absorption at 1690 cm^{-1} indicating the presence of a carbonyl group. Comparison of the ^{13}C NMR signals of **3** with **1** revealed that the sugar moiety at C-3 was identical, while the aglycone moiety was different. Acid hydrolysis of **3** furnished an aglycone, identified as drevogenin P (**5**) from its physical data (Sauer et al., 1966) and comparison with an authentic sample. Thus the structure of volubiloside C (**3**) was determined to be drevogenin P-3-*O*-β-D-glucopyranosyl(1→4)-6-deoxy-3-*O*-methyl-β-D-allopyranosyl - (1→4) - β - D - cymaropyranosyl(1→4) - β-D-cymaropyranoside.

Repeated chromatographic purification furnished conduritol A, quercetin, and quercetin-3-*O*-rutinoside which were characterized from their physical and spectroscopic data in comparison with those reported (Pollock and Stevens, 1965; Harborne and Mabry, 1985) for authentic samples.

It is noteworthy to mention that the differences between the constituents of the Indian variety and the plant growing in Thailand (Yoshimura et al., 1983, 1985) are mainly in the sapogenin part. In the Indian variety the sapogenin constituents are drevogenins D and P while in that of Thailand the sapogenin constituents are drevogenin A, dervyssogenin K_2, marsectohexol, and derbyssogenin G, and the similarities are more or less the same in the carbohydrate moiety. Moreover, flavonol constituents are found only in the Indian variety.

3. Experimental

3.1. General procedures

All melting points were measured on a Yanagimoto micromelting point apparatus and are uncorrected. ^1H and ^{13}C NMR spectra were recorded at 500 MHz and 125 MHz respectively using a Jeol ECP-500 spectrometer in C_5D_5N with TMS as internal standard. MALDI-TOF MS (positive) were performed on a Perspective Biosystem Voyager DE-STR spectrometer with 2,5-dihydroxybenzoic acid as matrix. Paper chromatography was performed on Whatman paper No. 1 (46×57 cm) with solvent system BuOH–C_5H_5N–H_2O (6:4:3); a saturated solution of aniline oxalate in water was used as staining agent and the spots were visualized after heating at 100 °C.

3.2. Plant material

The plant material was collected from the suburbs of Kolkata, India and identified by Dr. (Ms.) Debjani Basu, Botanist, at Indian Botanic Garden, Howrah, West Bengal, India. A voucher specimen (No. 305) was deposited at the Steroids and Terpenoids Chemistry Department, Indian Institute of Chemical Biology, Kolkata.

3.3. Extraction and isolation

The defatted, air-dried, powdered flowers (2.5 kg) of *D. volubilis* were successively extracted with MeOH (3×5 l) at ambient temperature. The combined MeOH extract was concentrated (500 ml) under reduced pressure and extracted with water-saturated *n*-BuOH (2×1 l). The organic layer was evaporated to dryness under reduced pressure to give a residue (15.2 g). This was chromatographed on silica gel (450 g). Graded elution was carried out with chloroform followed by various mixtures of $CHCl_3$–MeOH (9:1, 17:3, and 4:1). A total of 50 fractions (each 250 ml) were collected and fractions giving similar spots on TLC were combined. Fractions eluted with $CHCl_3$–MeOH (9:1) were combined together and subjected to rechromatography over silica gel (15 g). Earlier fractions (collected in 30 ml lots) eluted with $CHCl_3$–MeOH mixture (93:7) eluted volubiloside A (150 mg) while later fractions eluted a mixture of volubiloside B and volubiloside C. Repeated fractional crystallization furnished volubiloside B (90

mg). The mother liquor was chromatographed repeatedly over silica gel and eluted with various mixtures of $CHCl_3$–MeOH. Later fractions eluted with $CHCl_3$–MeOH (92:8) furnished 45 mg of volubiloside C. Fractions eluted with $CHCl_3$–MeOH (17:3) (2.25 g) was rechromatographed over a column of silica gel (75 gm). Earlier fractions eluted with $CHCl_3$–MeOH (88:12) yielded conduritol A (180 mg) whereas later fractions furnished quercetin (120 mg), and fractions eluted with $CHCl_3$–MeOH (85:15) gave quercetin-3-*O*-rutinoside (60 mg) identified by comparison of their physical and spectral data with those reported in the literature (Pollock and Stevens, 1965; Harborne and Mabry, 1985).

3.4. Volubiloside A (*1*)

Colourless needles from MeOH, mp 181–182 °C (dec.), $[\alpha]_D^{25}$ −16.7° (*c* 0.2, MeOH); IR: ν_{max} cm^{-1} (KBr): 3413, 1645, 1373, 1162, 1079 cm^{-1}; MALDI-TOF-MS (positive): *m*/*z* 999 $[M+Na]^+$; ^1H NMR: δ 1.33 (3H, *s*, 19-CH_3), 1.49 (3H, *d*, *J* = 6.6 Hz, 21-CH_3), 1.72 (3H, *s*, 18-CH_3), 2.65 (1H, *m*, 17α-H), 3.55 (1H, *d*, *J* = 9.7 Hz, 12α-H), 3.89 (1H, *m*, 3α-H), 4.15 [1H, *m* (overlapped with other signals), 11β-H] and 5.53 (1H, *d*, *J* = 5.7 Hz, 6-H). (^1H and ^{13}C NMR: Tables 1 and 2). (Found: C, 58.69; H, 8.32; $C_{48}H_{80}O_{20}$ requires: C, 59.00; H, 8.25%).

3.5. Volubiloside B (*2*)

The compound crystallized from MeOH–CH_3CN in needles, mp 178–179 °C (dec.), $[\alpha]_D^{25}$ −17.6° (*c* 0.2, MeOH); IR: ν_{max} cm^{-1} (KBr): 3424, 1646, 1375, 1165, 1080 cm^{-1}; MALDI-TOF-MS (positive): *m*/*z* 985 $[M+Na]^+$; ^1H NMR: δ 1.33 (3H, s, 19-CH_3), 1.49 (3H, *d*, *J* = 6.2 Hz, 21-CH_3), 1.72 (3H, s, 18-CH_3), 2.65 (1H, *m*, 17α-H), 3.55 (1H, d, *J* = 9.4 Hz, 12α-H), 3.89 (1H, *m*, 3α-H), 4.15 [1H, *m* (overlapped with other signals), 11β-H] and 5.53 (1H, d, *J* = 5.7 Hz, 6-H). (^1H and ^{13}C NMR: Tables 1 and 2). (Found: C, 58.52; H, 8.26; $C_{47}H_{78}O_{20}$ requires: C, 58.61; H, 8.16%).

3.6. Volubiloside C (*3*)

The isolate crystallized from MeOH–CH_3CN in fine micro needles, mp 218–220 °C (dec.), $[\alpha]_D^{25}$ +23.8° (*c* 0.14, MeOH); IR: ν_{max} cm^{-1} (KBr): 3414, 1690, 1370, 1164, 1080 cm^{-1}; MALDI-TOF-MS (positive): *m*/*z* 997 $[M+Na]^+$; ^1H NMR: δ 1.26 (3H, *s*, 18-CH_3), 1.33 (3H, *s*, 19-CH_3), 1.49 (3H, *d*, *J* = 6.2 Hz, 21-CH_3), 2.27 (3H, *s*, 21-CH_3), 3.59 (1H, *d*, *J* = 9.4 Hz, 12α-H), 3.87 (1H, *m*, 3α-H), 4.12 [1H, *m* (overlapped with other signals), 11β-H], 5.52 (1H, *d*, *J* = 5.7 Hz, 6-H), 4.96 (1H, *d*, *J* = 7.8 Hz, H-1 of glucose), 5.06 (1H, *d*, *J* = 7.9 Hz, H-1 of 6-deoxy-3-*O*-methyl-β-D-allopyranose), 5.10 (1H, *dd*, *J* = 1.6, 9.2 Hz, H-1′ of cymarose), 5.24 (1H, *dd*, *J* = 1.6, 9.4 Hz, H-1 of cymarose); (^{13}C NMR: Table 1). (Found: C, 58.89; H, 8. 32; $C_{48}H_{78}O_{20}$ requires: C, 59.12; H, 8.06%).

3.7. Acid hydrolysis of *1–3*

A sample of **1**, **2**, or **3** (30 mg) was heated in 1 M HCl (3 ml, dioxane–water 1:1) at 75 °C for 1.5 h on an oil bath, cooled, and 3 ml water was added. Dioxane was distilled off under reduced pressure and the solution was extracted with EtOAc (5 ml×3). The EtOAc layer was washed with water (3 ml×4), dried and the residue was crystallized from methanol–ether to furnish drevogenin D, mp 223–225 °C, $[\alpha]_D^{25}$ −7.8° (*c* 0.14, MeOH) for **1** and **2** (Found: C, 68.69; H, 9.41; calc. for $C_{21}H_{34}O_5$: C, 68.82; H, 9.35%); and drevogenin P, mp 208–210 °C, $[\alpha]_D^{25}$ −32.8° (*c* 0.18, MeOH) for **3**. (Found: C, 69.34; H, 8.94; calc. for $C_{21}H_{32}O_5$: C, 69.20; H, 8.85%). The acid hydrolyzate was worked up as described by Mahato et al. (1989). The monosaccharides were identified as glucose and cymarose for **1**; glucose, cymarose, and digitoxose for **2**; and, glucose and cymarose for **3** by paper chromatography on comparison with authentic samples. The monosaccharide 6-deoxy-3-*O*-methyl-β-allose for **1–3** was confirmed from extensive NMR studies (vide results and discussion) and comparison of its ^{13}C NMR data with those reported in the literature (Yoshimura et al., 1983).

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References

Chadha, Y.R., 1976. The Wealth of India, Raw Materials, Vol. 6. Council of Scientific & Industrial Research, New Delhi. pp. 564–565.

Harborne, J.B., Mabry, T.J., 1985. The Flavonoids, Advances in Research. Chapman and Hall, London.

Jia, Z., Koike, K., Sahu, N.P., Nikaido, T., 2002. Triterpenoid Saponins from Caryophyllaceae Family. In: Atta-Ur-Rahman (Ed.), Studies in Natural Products Chemistry, Vol. 26. Elsevier, Amsterdam, pp. 3–61.

Mahato, S.B., Sahu, N.P., Roy, S.K., Pramanik, B.N., 1989. Cardiac glycosides from *Corchorus olitorius*. Journal of Chemical Society, Perkin Transaction I, 2065–2068.

Kasai, R., Suzuo, M., Asakawa, J., Tanaka, O., 1977. Carbon-13 chemical shifts of isoprenoid-β-D-glucopyranosides and -β-D-mannopyranosides. Stereochemical influences of aglycone alcohols. Tetrahedron Letters 175–178.

Pollock, J.R.A., Stevens, R., 1965. Dictionary of Organic Compounds, Vol. 2. Eyre & Spottiswoode, London.

Sauer, H.H., Weiss, Ek., Reichstein, T., 1966. Die struktur der drevogenin. 2. mitteilung struktur von drevogenin P. Glykoside und aglykone, 278 Mitteilung. Helvetica Chimica Acta 49, 1632–1655.

Seo, S., Tomita, Y., Tori, K., Yoshimura, Y., 1978. Determination of the absolute configuration of a secondary hydroxy group in a chiral secondary alcohol using glycosidation shifts in carbon-13 nuclear magnetic resonance spectroscopy. Journal of American Chemical Society 100, 3331–3339.

Singhal, S., Khare, M.P., Khare, A., 1988. Tenasogenin, a pregnane ester from *Marsdenia tenacissima*. Phytochemistry 19, 2431–2433.

Yoshimura, S., Narita, H., Hayashi, K., Mitsuhashi, H., 1983. Studies on the constituents of Asclepiadaceae plants LVI, Isolation of new antitumor-active glycosides from *Dregea volubilis* (L). Benth. Chemical and Pharmaceutical Bulletin 31, 3971–3983.

Yoshimura, S., Narita, H., Hayashi, K., Mitsuhashi, H., 1985. Studies on the constituents of Asclepiadaceae plants LIX. The structures of five new glycosides from *Dregea volubilis* (L). Benth. Chemical and Pharmaceutical Bulletin 33, 2287–2293.