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Two New Sterols from *Bolbostemma paniculatum*

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Abstract: Two new sterols were isolated from bulbs of *Bolbostemma paniculatum* (Maxim.) Franquet. Their structures were elucidated as stigmasta-7, 22, 25-triene-3-*O*-nonadecanoic acid ester (**1**) and stigmasta-7, 22, 25-triene-3-*O*-β-D- (6′-palmitoyl) glucopyranoside (**2**) by chemical and spectroscopic methods.

Keywords: *Bolbostemma paniculatum*, sterol, stigmasta-7, 22, 25-triene-3-*O*-nonadecanoic acid ester, stigmasta-7, 22, 25-triene-3-*O*-β-D- (6′-palmitoyl) glucopyranoside.

The bulb of *Bolbostemma paniculatum* (Maxim.) Franquet is a Chinese folk medicine named as "Tu Bei Mu", which is often used for the treatment of tumors as well as for detoxication. $\Delta^{7,16,25}$-Sterol has been isolated from this plant[1]. We report here the isolation and structural elucidation of two new sterols from the chloroform fraction of the methanol extracts of bulbs of *Bolbostemma paniculatum* (Maxim.) Franquet.

The dried and powdered bulbs were extracted with cool MeOH by infiltration. The solvent was removed under reduced pressure, the residue was suspended in water and then partitioned with petroleum ether, $CHCl_3$ and *n*-BuOH successively. The chloroform fraction was chromatographed on silica gel column to afford compound **1** and **2**.

Compound **1** was isolated as colorless crystal, mp. 100~101℃, ESI-MS *m/z*: 690. Its IR spectrum revealed the presence of ester carbonyl (1739 cm^{-1}) and double bond (1650 cm^{-1}). Alkaline hydrolysis of **1** with 5% sodium hydroxide-methanol yielded compound **a** and **b**. The ^{1}H-NMR, ^{13}C-NMR and EI-MS data of **a** are in accord with that of stigmasta-7, 22, 25-triene-3-ol[2]. Compound **b** was identified as nonadecanoic acid by NMR and EI-MS (*m/z*: 298 [M^+]). Consequently the structure of compound **1** is elucidated as stigmasta-7, 22, 25-triene-3-*O*-nonadecanoic acid ester.

Compound **2** was isolated as white waxy solid, mp. 84~86℃, ESI-MS *m/z*: 810. Its IR spectrum revealed the presence of hydroxy (3443 cm^{-1}), ester carbonyl (1734 cm^{-1}) and double bond (1646 cm^{-1}). Alkaline hydrolysis of **2** with 5% sodium hydroxide -methanol yielded compound **c** and **d**. Compound **d** was identified as hexadecanoic acid (palmitic acid) by NMR and EI-MS (*m/z*: 256 [M^+]). The ^{1}H-NMR and ^{13}C-NMR

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spectrum of compound **c** showed the presence of a monosaccharide unit (See **Table 1, 2**). On mineral acid hydrolysis, compound **c** yielded stigmasta-7, 22, 25-triene-3-ol (**a**) and D-glucose. So compound **c** was identified as stigmasta-7, 22, 25-triene-3-*O*-D-glucopyranoside and the configuration was confirmed as β based on the terminal protons signals (δ 4.84, d, *J*=7.6 Hz). On the basis of ^{1}H-^{1}H COSY and HMQC spectra data all the carbons and protons of the glucopyranosyl unit were assigned. The HMBC spectral analysis of compound **2** displayed correlation peak between H-6′ (glucopyranosyl) and the carbonyl of the palmitoyl. Consequently the structure of compound **2** is elucidated as stigmasta-7, 22, 25-triene-3-*O*-β-D- (6′-palmitoyl) glucopyranoside.

Figure 1 Structure of compound **1**

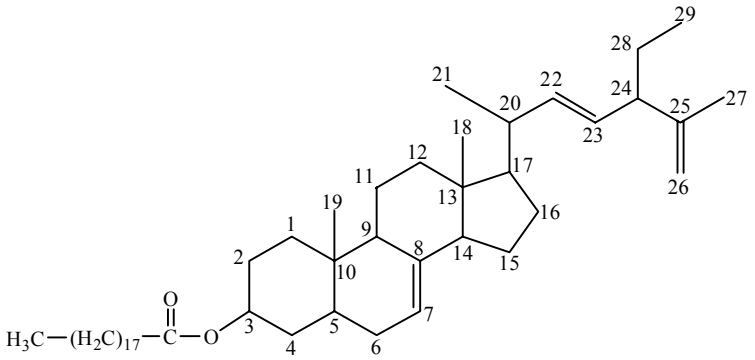

Figure 2 Structure of compound **2**

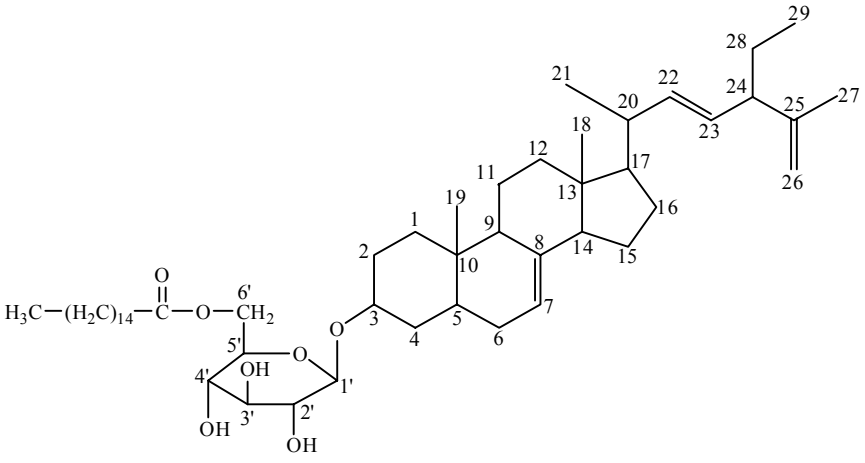

Table 1 ^{13}CNMR data of compounds **1**, **2** and **a** ($CDCl_3$, 100 MHz, δ ppm)

No.	**a**	**1**	**2**
1	37.2	34.8	34.4
2	38.0	36.9	37.3
3	71.0	73.1	73.5
4	39.5	39.4	39.5
5	40.4	40.5	40.5
6	31.5	31.9	31.9
7	117.5	117.4	117.4
8	139.5	139.4	139.5
9	49.5	49.3	49.5
10	34.2	34.2	34.3
11	23.0	23.0	23.0
12	29.6	29.7	29.7
13	43.3	43.3	43.3
14	55.1	55.1	55.1
15	28.2	28.3	28.2
16	21.5	21.5	21.6
17	55.9	55.9	56.0
18	12.0	12.1	12.1
19	13.0	12.9	13.0
20	40.3	40.1	40.4
21	20.9	20.9	21.0
22	137.0	137.0	136.9
23	130.2	130.2	130.3
24	52.0	52.0	52.0
25	148.5	148.5	148.4
26	109.5	109.6	109.6
27	20.2	20.2	20.2
28	25.7	25.7	25.8
29	12.1	12.1	12.1
Glu-1′			101.3
2′			76.3
3′			79.1
4′			70.5
5′			73.8
6′			63.6
Fat acid-1″		173.4	174.1
2″		33.9	34.6
CH_3-		14.1	14.0
Others CH_2-		22.7~29.7	22.5~31.5

Table 2 ^{1}HNMR data of compounds **1**, **2** and **a** ($CDCl_3$, 400 MHz, δ ppm)

No.	**a**	**1**	**2**
3	3.59 (1H, m)	4.72 (1H, m)	3.91 (1H, m)
7	5.16 (1H, br, m)	5.17 (1H, br, m)	5.16 (1H, br, m)
18	0.55 (3H, s)	0.56 (3H, s)	0.56 (3H, s)
19	0.80 (3H, s)	0.83 (3H, s)	0.81 (3H, s)
21	1.02 (3H, d, 6.4)	1.04 (3H, d, 6.8)	1.04 (3H, d, 6.8)
22	5.25 (1H, dd, 8.0, 15.2)	5.26 (1H, dd, 7.6, 15.2)	5.25 (1H, dd, 7.6, 15.2)
23	5.19 (1H, dd, 8.0, 15.2)	5.21 (1H, dd, 7.6, 15.2)	5.22 (1H, dd, 7.6, 15.2)
24	2.42 (1H, q, 7.4)	2.44 (1H, q, 7.2)	2.44 (1H, q, 7.2)
26	4.70 (2H, m)	4.72 (2H, m)	4.72 (2H, m)
27	1.65 (3H, s)	1.67 (3H, s)	1.67 (3H, s)
29	0.84 (3H, t, 7.4)	0.86 (3H, t, 7.4)	0.86 (3H, t, 7.4)
Glu-1′			4.84 (1H, d, 7.6)
2′			3.85 (1H, m)
3′			4.12 (1H, m)
4′			3.95 (1H, m)
5′			3.93 (1H, m)
6′			4.70 (1H, dd, 6.0, 12.0) 4.90 (1H, d, 12.0)
Fat acid-2″		2.28 (2H, t, 7.6)	2.33 (2H, t, 7.6)
CH_3-		0.90 (3H, t, 6.8)	0.90 (3H, t, 6.6)

References

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