(C-10), 21.9 (C-11), 39.8 (C-12), 43.6 (C-13), 55.4 (C-14), 23.5 (C-15), 28.2 (**1a**) and 28.8 (**2a**/**3a**) (C-16), 56.4 (C-17), 12.3 (C-18), 13.1 (C-19), 40.7 (C-20). Glucosyl-C: 102.4 (C-1'), 75.4 (C-2'), 78.3 (C-3'), 72.0 (C-4'), 77.4 (C-5'), 63.1 (C-6').

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A FUROSTANOL GLUCURONIDE FROM *SOLANUM LYRATUM**

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Key Word Index—*Solanum lyratum*; Solanaceae; furostanol glucuronide.

Abstract—A new furostanol glucuronide and three known glycosides, SL-0, aspidistrin and methyl proto-aspidistrin, were isolated from the fresh immature berries of *Solanum lyratum*. The structure of the new compound was characterized as 26-*O*-β-D-glucopyranosyl-(22ξ,25*R*)-3β,22,26-trihydroxyfurost-5-ene 3-*O*-α-L-rhamnopyranosyl-(1 → 2)-[β-D-glucopyranosyl-(1 → 3)]-β-D-glucuronopyranoside.

INTRODUCTION

It was previously reported that a furostanol (SL-0), a spirostanol (SL-1) and two steroidal alkaloid glycosides (SL-c, SL-d) were obtained from the stems of *Solanum lyratum* Thunb. and their structures were elucidated [1, 2]. Our continuing study of the fresh immature berries of this plant has led to the isolation of a new steroidal glucuronide (**1**), which was a major component (*ca* 2.8%), along with three known glycosides, SL-0 [1], aspidistrin [3] and methyl proto-aspidistrin [4]. This paper deals with the structural elucidation of compound **1**.

*Part 6 in the series "Studies on the Constituents of *Solanum* Plants". For Part 5 see ref. [2].

RESULTS AND DISCUSSION

Compound **1**, an amorphous powder, $[\alpha]_D - 61.4°$, showed strong absorptions in the IR spectrum due to a carboxyl group (1600 cm^{-1}) and a hydroxyl group (3400 cm^{-1}), but not for a spiroketal function [5, 6] and it was positive to the Ehrlich reagent [7], suggesting a furostanol glycoside structure. Enzymic hydrolysis with almond emulsin gave a spirostanol glycoside (**2**) and D-glucose. Compound **2**, colourless needles, mp > 300°, $[\alpha]_D - 83.4°$, showed absorptions due to a carboxyl group (1600 cm^{-1}) and a characteristic spiroketal ring (920, 900, 865 cm^{-1}) in the IR spectrum, and in the FD mass spectrum the peak at *m/z* 937 originated from $[M + K]^+$. Acid hydrolysis of compound **2** yielded diosgenin together with rhamnose, glucose and glucuronic acid. The EI mass spectrum of the acetate of **2** showed the peaks

ascribable to the peracetylated terminal hexose (*m/z* 331) and methylpentose (*m/z* 273). Compound **2** was subsequently methylated with CH_2N_2 to afford compound **3**, colourless needles, mp 276–277°, $[\alpha]_D$ −83.4°, whose ^{13}C NMR spectrum showed the presence of a methoxycarbonyl group (δ169.8, 52.1). Reduction of compound **3** with sodium borohydride in methanol gave compound **4**, colourless needles, mp 285–286°, $[\alpha]_D$ −82.6°, in which a methoxycarbonyl signal was no longer observed in the ^{13}C NMR spectrum (Table 1). The permethyl ether (**5**) derived from compound **4** by Hakomori's method [8] upon methanolysis furnished methyl 4,6-di-*O*-methyl α-D-glucopyranoside, methyl per-*O*-methyl α-D-glucopyranoside and methyl per-*O*-methyl α-L-rhamnopyranoside, together with diosgenin. Partial hydrolysis of **5** yielded **6**, which was further methylated by the Kuhn method [9] to give compound **7**. Methanolysis of compound **7** afforded methyl per-*O*-methyl α-D-glucopyranoside and methyl 2,4,6-tri-*O*-methyl α-D-glucopyranoside. Thus, together with substantiation of the structures for compounds **2**, **3** and **4**, compound **1** could be represented as 26-*O*-β-D-glucopyranosyl-(22ξ,25*R*)-3β,22,26-trihydroxyfurost-5-ene 3-*O*-α-L-rhamnopyranosyl-(1 → 2)-[β-D-glucopyranosyl-(1 → 3)]-β-D-glucuronopyranoside. The ^{13}C NMR data (Table 1) also unambiguously supported the structures for compounds **2**, **3** and **4**. This seems to be the first example of the isolation of a furostanol glucuronide from a natural source.

EXPERIMENTAL

General. Mps are uncorr. ^{13}C NMR spectra were taken at 68.0 MHz; chemical shifts are given in δ-values with TMS as internal standard. Chromatographic columns were packed with Bondapak C_{18} and silica gel (Merck 60) and TLC plates were precoated with silica gel (Merck 60 F_{254}).

Extraction and isolation. The MeOH extract of the fresh immature berries (1.4 kg) of *Solanum lyratum* Thunb. was concd *in vacuo* to give a residue, which was treated with refluxing MeOH to separate it into the soluble portion (37.7 g) and the insoluble material (77.0 g). A part (5.5 g) of the insoluble portion was subjected to Bondapak C_{18} CC (solvent 60% MeOH) to afford compound **1** (2.8 g), whereas a part (500 mg) of the insoluble portion was chromatographed over silica gel ($CHCl_3$–MeOH–H_2O, 7:3:0.3) to give SL-0 (5 mg), aspidistrin (10 mg) and methyl proto-aspidistrin (25 mg).

Compound **1**. An amorphous powder, R_f 0.02 ($CHCl_3$–MeOH–H_2O, 7:3:0.5), $[\alpha]_D^{24}$ −61.4° (H_2O; *c* 1.00), IR ν_{max}^{KBr} cm^{-1}: 3400 (OH), 1600 (COO^-).

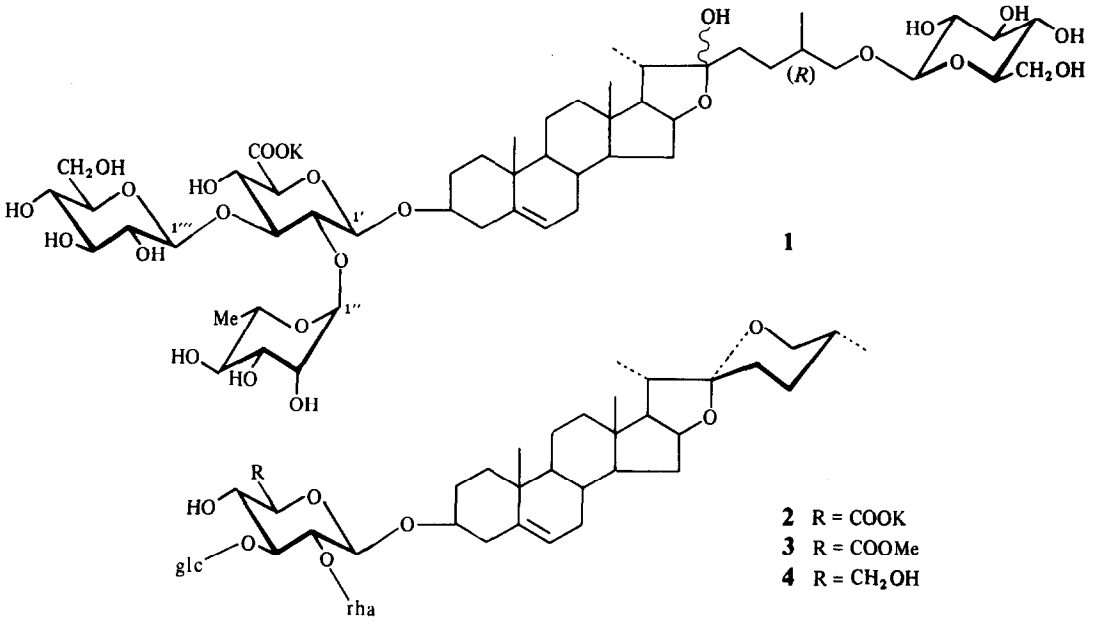
Enzymic hydrolysis of compound **1**. A mixture of **1** (300 mg) and almond emulsin (60 mg) in H_2O (10 ml) was incubated at 40° for 10 hr and evaporated *in vacuo* to dryness to give a residue. The MeOH-soluble portion was subjected to Bondapak C_{18} CC eluting with 60% MeOH to afford D-glucose and a spirostanol glycoside (**2**), R_f 0.11 ($CHCl_3$–MeOH–H_2O, 7:3:0.5), colourless needles (150 mg); mp >300°; $[\alpha]_D^{24}$ −83.4° (pyridine, *c* 0.50); IR ν_{max}^{KBr} cm^{-1}: 3350 (OH), 1610 (COO^-), 920, 900 (intensity 900 > 920), 865. FDMS *m/z*: 937 $[M+K]^+$.

Methanolysis of compound **2**. A soln of **2** (50 mg) in 2N HCl–MeOH (15 ml) was refluxed for 2 hr, diluted with H_2O and extracted with $CHCl_3$. The organic layer was evaporated to give a residue, which was crystallized from Me_2CO to afford an aglycone, colourless needles (6 mg), mp 205–207°, $[\alpha]_D^{23}$ −126.5° ($CHCl_3$; *c* 0.02), identical with diosgenin. IR ν_{max}^{KBr} cm^{-1}: 3350 (OH), 960, 918, 898, 863 (25*R* spiroketal). EIMS *m/z*: 415 $[M+1]^+$, 139 $[C_9H_{15}O]^+$. ^1H NMR ($CDCl_3$): δ0.77 (3H, *s*, 13-Me), 1.02 (3H, *s*, 10-Me), 3.12–3.76 (3H, *m*, 3-H, 26-H_2), 4.12–4.61 (1H, *m*, H-16), 5.31 (1H, *m*, H-6). The aq. layer was neutralized, concd and examined by TLC ($CHCl_3$–MeOH–H_2O, 7:3:0.5) to detect the respective methylsides of glucose (R_f 0.35), rhamnose (R_f 0.61) and glucuronic acid R_f 0.15).

Methylation of compound **2** *with* CH_2N_2. A soln of **2** (100 mg) in MeOH (50 ml) was treated with excess CH_2N_2 and left standing overnight in a refrigerator to give the methyl ester (**3**) R_f 0.47 ($CHCl_3$–MeOH–H_2O, 7:3:0.5), colourless needles

Table 1. ^{13}C NMR spectral data of compounds **2**, **3** and **4** (in pyridine-d_5)

	C	2	3	4
Aglycone	1	37.4	37.4	37.5
	2	29.9	29.9	30.0
	3	78.6	78.6	78.6
	4	38.6	38.6	38.7
	5	140.9	140.6	140.7
	6	121.8	121.9	121.8
	7	32.5	32.2	32.2
	8	31.8	31.7	31.7
	9	50.2	50.3	50.2
	10	37.2	37.1	37.1
	11	21.1	21.1	21.1
	12	39.9	39.9	39.9
	13	40.5	40.5	40.5
	14	56.6	56.6	56.6
	15	32.3	32.3	32.3
	16	81.2	81.1	81.1
	17	62.3	62.3	62.4
	18	16.4	16.3	16.4
	19	19.4	19.3	19.4
	20	42.0	42.0	41.9
	21	15.1	15.0	15.0
	22	109.2	109.2	109.2
	23	31.8	31.8	31.8
	24	29.3	29.3	29.2
	25	30.6	30.6	30.6
	26	66.9	66.9	66.8
	27	17.4	17.3	17.3
Sugar moiety	1′	99.6	100.4	99.9
	2′	76.6	76.3	77.8
	3′	87.0	88.2	89.5
	4′	71.2	71.2	69.5
	5′	75.6	76.4	77.0
	6′	178.0	169.8	62.4
	1″	106.1	104.5	104.5
	2″	72.6	72.7	72.7
	3″	72.2	72.3	72.4
	4″	73.9	74.0	74.0
	5″	69.5	69.6	69.6
	6″	18.6	18.6	18.7
	1‴	104.1	102.1	102.2
	2‴	74.5	74.8	74.9
	3‴	78.2	78.2	78.4
	4‴	71.6	71.4	71.4
	5‴	77.7	78.4	77.6
	6‴	62.9	62.9	62.9
	OMe		52.1	



1

2 R = COOK
3 R = COOMe
4 R = CH_2OH

(85 mg), mp 276–277°, $[\alpha]_D^{23}$ −83.4° (pyridine; *c* 0.50). IR ν_{max}^{KBr} cm^{-1}: 3400 (OH), 1725 (COOMe).

Reduction of compound **3** *with* $NaBH_4$. To a soln of **3** (80 mg) in MeOH (20 ml) $NaBH_4$ (30 mg) was added and the mixture left standing for 2 hr at room temp. Addition of AcOH (10 ml) and evaporation of the reaction mixture gave a residue, which was subjected to silica gel CC ($CHCl_3$–MeOH–H_2O, 7:3:0.3) to furnish compound **4**, R_f 0.29 ($CHCl_3$–MeOH–H_2O, 7:3:0.5), colourless needles (55 mg), mp 285–286°, $[\alpha]_D^{22}$ − 82.6° (pyridine; *c* 0.50).

Methylation of compound **4**. A soln of **4** (50 mg) in DMSO (3 ml) was methylated by Hakomori's method (NaH 300 mg, DMSO 5 ml, MeI 7 ml) and worked-up to yield the crude product, which was purified by silica gel CC using hexane–EtOAc, 3:1 → 1:1 as eluent to afford the permethyl ether (**5**), R_f 0.33 (hexane–Me_2CO, 2:1) as a white powder (35 mg).

Methanolysis of compound **5**. The methyl ether (5 mg) was refluxed with 2 N HCl–MeOH (3 ml) for 2 hr and the reaction mixture was examined by TLC (EtOAc–MeOH, 25:1) to detect methyl-4,6-di-*O*-methyl α-D-glucopyranoside (R_f 0.23), methyl-2,3,4,6-tetra-*O*-methyl α-D-glucopyranoside (R_f 0.62) and methyl-2,3,4-tri-*O*-methyl α-L-rhamnopyranoside (R_f 0.74) along with diosgenin.

Partial hydrolysis of compound **4**. After a soln of **5** (25 mg) in 0.5 N HCl–MeOH (4 ml) was refluxed for 1 hr, the reaction mixture was neutralized and evaporated *in vacuo* to dryness to give a residue, which was purified by silica gel CC using hexane–EtOAc (3:2 → 1:1) as the eluent to afford compound **6** (12 mg), R_f 0.13 (hexane–EtOAc, 1:1).

Methylation of compound **6**. A mixture of **6** (10 mg), Ag_2O (90 mg), MeI (4 ml) and DMF (2 ml) was stirred overnight at room temp. and worked-up to give the methyl ether **7** (6 mg), R_f 0.51 (hexane–EtOAc, 1:1).

Methanolysis of compound **7**. A soln of **7** (5 mg) in 1 N HCl–MeOH (3 ml) was refluxed for 2 hr and the reaction mixture was checked by TLC (EtOAc–MeOH, 25:1) to reveal the presence of methyl-2,3,4,6-tetra-*O*-methyl-α-D-glucopyranoside (R_f 0.64) and methyl-2,4,6-tri-*O*-methyl-α-D-glucopyranoside (R_f 0.38).

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