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NEW SPIROSTANE SAPONINS AND SAPOGENINS FROM *SOLANUM HISPIDUM* SEEDS*

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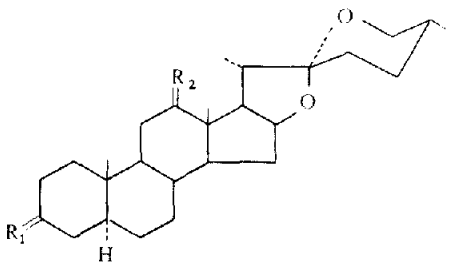
Key Word Index *Solanum hispidum*: Solanaceae; saponins; 6α-*O*-α-L-rhamnopyranosyl-(25*S*)-5α-spirostan-3-one; 6α-*O*-α-L-rhamnopyranosyl (1 → 3)-α-L-rhamnopyranosyl-(25*S*)-5α-spirostan-3-one; 6α-*O*-α-L-rhamnopyranosyl (1 → 3)-α-L-rhamnopyranosyl-(25*S*)-5α-spirostan-3β-ol; sapogenin; (25*S*)-6α-hydroxy-5α-spirostan-3-one.

We recently reported [1] the isolation of a novel spirostane sapogenin having an unusual 22β*O*-stereochemistry along with neochlorogenin and paniculogenin from the leaves of *Solanum hispidum*. The present paper deals with three new spirostane saponins and one new sapogenin isolated from the seeds of the same plant. We did not, however, encounter either hecogenin (**1**) or hispidogenin (**2**) in the seeds which were reported by the earlier workers [2].

The crushed dry seeds (3.5 kg) were extracted with methanol in a Soxhlet apparatus for 24 hr. The Dragendorff-negative extract was concentrated to 1.5 l. After removal of fats and oils by petrol, the methanol soluble part of the concentrate was diluted with water (2 l.) and thoroughly extracted with chloroform to yield the crude saponin mixture (20 g). On repeated chromatography over Si gel, it was resolved into three new saponins hispinin-A, $C_{33}H_{52}O_8$, mp 224–226°, $[\alpha]_D$ −44° (Py); hispinin-B, $C_{39}H_{62}O_{12}$, mp 258–260°, $[\alpha]_D$ −68.3° (Py) and hispinin-C, $C_{39}H_{64}O_{12}$, mp 285–288°, $[\alpha]_D$ −59° (Py).

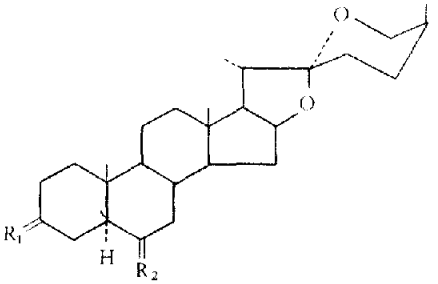
Hydrolysis (5% alcoholic HCl) of hispinin-A and hispinin-B gave the same hitherto unreported sapogenin, designated as solagenin (**3**), $C_{27}H_{42}O_4$ (M^+ at *m/e* 430), mp 218–220°, $[\alpha]_D$ −51.6° ($CHCl_3$). Its IR spectrum in KBr showed a broad band at 3400–3500 cm^{-1} for OH, a strong band at 1710 cm^{-1} for a six-membered ring C=O and the characteristic [3] bands at 848, 862, 892, 915 and 982 cm^{-1} for the spirostane ring system. It formed a monoacetate (**4**), mp 210–212°, $[\alpha]_D$ −49° ($CHCl_3$), ν_{max}^{KBr} 1730, 1710 and 1240 cm^{-1}, *m/e* 472 (M^+) and was oxidized by Jones' reagent to a diketone (**5**), mp 235–236°, *m/e* 428 (M^+). The carbonyl absorption ($\nu_{max}^{CHCl_3}$ 1705 cm^{-1}) of the latter clearly indicated that both the secondary OH and C=O groups of solagenin (**3**) must be located in six-membered rings of the molecule. That the two groups cannot be in the same ring became evident from the UV spectrum of **5**, which did not show the characteristic absorption for an α- or β-diketo chromophore. The MS of solagenin (**3**) exhibited prominent peaks at *m/e* 139 (base peak) and 115, besides the low intensity peaks at *m/e* 371 (M^+-59), 361 (M^+-69) and 358 (M^+-72) diagnostic [4a] of spirostane ring system.

The ^1H NMR data (Table 1) of solagenin acetate (**4**) showed that the carbinyl proton resonated at δ 4.75 as doublets of double doublets and the coupling constants (11, 11, 4.5 Hz) were in good agreement with the equatorial OAc group at either C-6 or C-11. The chemical shifts of C-18 (0.78) and C-19 (1.10) protons were found to be in excellent accord with those calculated [5] (C-18, 0.78 and C-19, 1.11) for 3-keto-6α-acetoxy-5α-spirostane. Furthermore, the resonance frequency of the C-27 protons coupled with the characteristic [6, 7] signals for



1 R_1 = 6α-OH, 6β-H; R_2 = O

2 R_1 = O; R_2 = 6α-OH, 6β-H



3 R_1 = O; R_2 = 6β-H, 6α-OH

4 R_1 = O; R_2 = 6β-H, 6α-OAc

5 R_1 = R_2 = O

6 R_1 = 6α-OH, 6β-H; R_2 = 6α-OH, 6β-H

7 R_1 = 6α-OH, 6β-H; R_2 = 6β-H, 6α-OH

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Table 1. ^{1}H NMR data* (δ) in $CDCl_3$ of **4** and **6**

Compound	C-19	C-18	C-21	C-27	C-26	C-16	C-6	C-3	OAc
4	1.10	0.78	0.98*d* (7)	1.06*d* (7)	3.27*d* (12) 3.92*dd* (12, 3)	4.37*ddd* (7, 7, 7)	4.75*ddd* (11, 11, 4.5)	–	2.03
6	1.05	0.80	0.98*d* (7)	1.08*d* (7)	3.27*d* (11), 3.93*dd* (11, 3)	4.42*ddd* (7, 7, 7)	3.74*m* ($W_{\frac{1}{2}}$ = 9 Hz)	3.50*m*	—

* Spectra were recorded in a 90 MHz instrument, values in parentheses are the coupling constants in Hz.

the C-26 protons confirmed the 25*S*-stereochemistry.

The diketone **5** on $NaBH_4$ reduction yielded the diol **6**, mp 228–230°. Its ^{1}H NMR spectrum showed the presence of an expected [8] axial OH group (chemical shift of the carbinyl proton at δ 3.74, $W_{\frac{1}{2}}$ – 9 Hz) derived from reduction of the oxo group at C-6. The resonance frequencies of the C-18 and C-19 protons at δ 0.80 and 1.05 (calculated: 0.81 and 1.05) respectively are also in conformity with the 3β,6β-dihydroxy-5α-spirostane structure **6** for the diol. On the contrary, the introduction of a 11β-OH group would be expected to show a pronounced deshielding effect [5] on both C-18 and C-19 protons.

Finally, the structure of solagenin as (25*S*)-6α-hydroxy-5α-spirostan-3-one (**3**) was confirmed by its $NaBH_4$ reduction to neochlorogenin (**7**) [1] which was also obtained by acid hydrolysis of hispinin-C.

Hydrolysis of all the three saponins with Kiliani mixture (HOAc–HCl–H_2O, 1.5:3.5:5) and paper chromatography showed the presence of L-rhamnose. The MS of the peracetylated hispinin-A (M^+ at *m/e* 702) exhibited the characteristic [4b] peaks at *A/e* 273, 184, 171, 153 and 111 for the peracetylated rhamnose moiety. On the other hand, hispinin-B and hispinin-C peracetates showed an additional peak at *m/e* 503 for peracetylated dirhamnose unit. Evidently, hispinin-A is a monorhamnoside, and hispinin-B and hispinin-C are dirhamnosides.

Both hispinin-B and hispinin-C on Smith degradation [9] ($NaIO_4$ oxidation–$NaBH_4$ reduction–mild acid treatment) gave the same monorhamnoside, mp 188–190°, $[\alpha]_D$ –37.3° (Py), which on similar treatment furnished neochlorogenin (**7**). Therefore, the two rhamnose units in both the saponins must be coupled through a (1 → 3) linkage. Moreover, the signs and magnitudes of the rotation contribution [10] of the sugar component of all the saponins and the monorhamnoside mentioned above suggested an α-linkage of the rhamnose units.

On the other hand, hispinin-A (M^+ at *m/e* 576), as well as the monorhamnoside (M^+ at *m/e* 578) derived from the above two saponins, showed MS fragments due to the loss of 30, 59, 69 and 72 mass units from the parent ions besides peaks at *m/e* 139 and 115 for the intact spirostane ring system. They did not respond to Ehrlich reagent and also exhibited IR bands at 845, 890, 910 and 980 cm^{-1}. Thus, the sugar moieties in all the three saponins are linked through the 6α-OH group.

Considering all the above evidence, the complete structures and stereochemistry of the saponins were deduced to be 6α-*O*-α-L-rhamnopyranosyl-(25*S*)-5α-spirostan-3-one for hispinin-A, 6α-*O*-α-L-rhamnopyranosyl (1 → 3)-α-L-rhamnopyranosyl-(25*S*)-5α-spirostan-3-one for hispinin B and the corresponding 3β-ol for hispinin-C.

REFERENCES

1. Chakravarty, A. K., Dhar, T. K. and Pakrashi, S. C. (1978) *Tetrahedron Letters* **40**, 3875.
2. Maiti, P. C. and Mookherjea, S. (1965) *Chem. Ind.* 1653.
3. Jones, R. N., Katzenellenbogen, E. and Dobriner, K. (1953) *J. Am. Chem. Soc.* **75**, 158.
4. Budzikiewicz, H., Djerassi, C. and Williams, D. H. (1964) *Structure Elucidation of Natural Products by Mass Spectrometry*, Vol. 2, (a) p. 110, (b) p. 203. Holden-Day, San Francisco.
5. Tori, K. and Aono, K. (1964) *Annu. Rep. Shionogi Res. Lab.* **14**, 136.
6. Kutney, J. P. (1963) *Steroids* **2**, 225.
7. Williams, D. H. and Bhacca, N. S. (1965) *Tetrahedron* **21**, 1641.
8. Kirk, D. N. and Hartshorn, M. P. (1968) *Steroid Reaction Mechanisms*, p. 133. Elsevier, New York.
9. Smith, F. and Montgomery, R. (1959) *Chemistry of Plant Gums and Mucilages*, p. 194. Reinhold, New York.
10. Klyne, W. (1950) *Biochem. J.* **47**, XLI.