Phytochemistry, Vol. 27, No. 3, pp. 956–958, 1988.
Printed in Great Britain.

0031 9422/88 $3.00 + 0.00

7β-HYDROXY-*O*-METHYLSOLANOCAPSINE, A NEW 3-AMINO STEROIDAL ALKALOID FROM *SOLANUM CAPSICASTRUM**

AJIT K. CHAKRAVARTY and SATYESH C. PAKRASHI†

Indian Institute of Chemical Biology, Calcutta 700 032, India

(*Revised received* 15 *July* 1987)

Key Word Index—*Solanum capsicastrum*; steroidal alkaloid; 7β-hydroxy-*O*-methylsolanocapsine; ^{1}H NMR; ^{13}C NMR.

Abstract—A new minor 3-amino steroidal alkaloid has been isolated from the leaves of *Solanum capsicastrum* and characterised as 7β-hydroxy-*O*-methylsolanocapsine (**3**) on the basis of IR, MS, ^{1}H and ^{13}C NMR spectral analyses. The assignments of ^{13}C chemical shifts of the OMe and C-24 carbons of *O*-methylsolanocapsine (**6**) had to be revised on the basis of INEPT and selective proton decoupling experiments.

INTRODUCTION

We required solanocapsine (**1**) in quantities as starting material for the preparation of solanogantine (**2**) [1], a potent hypotensive and tranquilo-sedative (Ray Ghatak, B. J., unpublished data) steroidal alkaloid isolated in this laboratory from *Solanum giganteum* Jacq. In the course of its isolation from *S. capsicastrum* Link [2, 3], we encountered a new steroidal alkaloid characterized as 7β-hydroxy-*O*-methylsolanocapsine (**3**).

RESULT AND DISCUSSION

The alkaloids were isolated from the leaves of the plant by extraction with MeOH at room temperature followed by hydrolysis (10% methanolic HCl). The total crude base on chromatography over neutral alumina yielded, besides **1** (major, 0.2%), a mixture of other very minor components. Repeated column chromatography of the latter followed by preparative TLC [solvent system: $CHCl_3$–MeOH (98:2) saturated with NH_3] afforded the new alkaloid **3**, amorphous powder; $[\alpha]_D^{24^\circ} + 65.2^\circ$ ($CHCl_3$; *c* 0.46).

The mass spectrum of **3** exhibited the molecular ion peak at *m/z* 460 which is 30 mass units higher than that of the co-occurring solanocapsine (**1**). Its IR spectrum displayed a broad absorption band in the region 3340–3410 cm^{-1} for NH/OH group(s). On treatment with methanal–formic acid at 100° for 2 hr, it formed a trimethyl derivative (**4**). Acetylation of **3** with acetic anhydride–pyridine at room temperature yielded an *N,N',O*-triacetate (**5**).

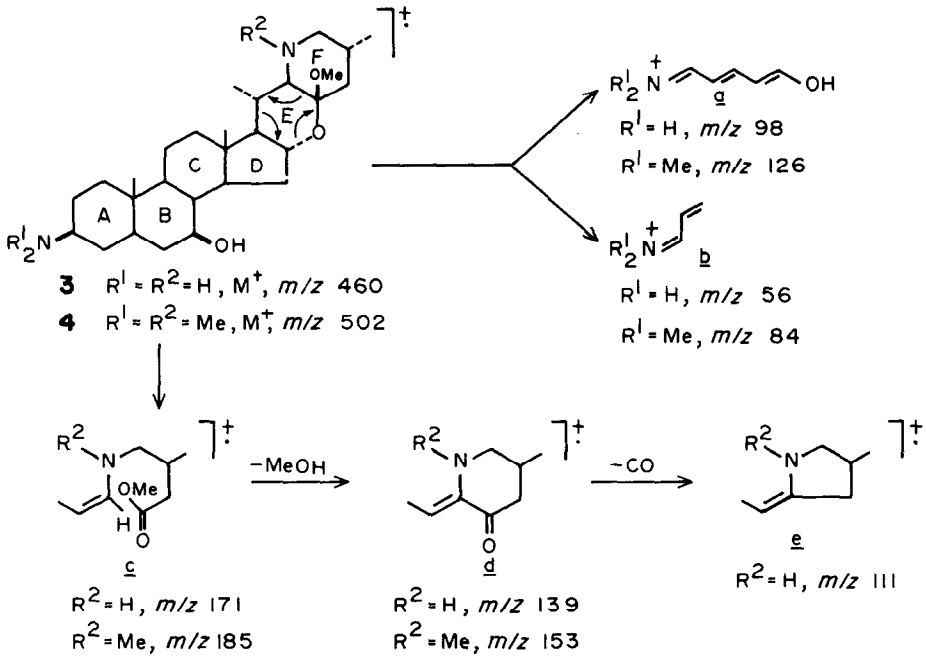
The above evidence clearly indicated the presence of NH_2, NH and OH functions in the alkaloid (one of each). In addition, the presence of one OMe group was apparent from the three-proton singlet at δ3.13 ppm in the ^{1}H NMR spectrum. The secondary nature and equatorial orientation of the OH group could also be inferred from the one-proton multiplet of the carbinyl proton at δ3.32 ppm ($w_{1/2} \approx 20$ Hz) shifted down-field to δ4.56 ppm in the spectrum of the *N,N',O*-triacetate (**5**). In addition, the spectrum of the alkaloid showed the signals for 10-Me, 13-Me, 20-Me, 25-Me and 16-H protons very close to those of *O*-methylsolanocapsine (**6**) [4].

That the OH group of the alkaloid is located either at C-6 or at C-7 in ring B of a 3-amino steroid became evident from the mass spectra of **3** and its *N,N,N'*-trimethyl derivative (**4**). Thus, the mass spectra demonstrated peaks at *m/z* 98 in **3** and 126 in **4** (species **a**), 16 mass units higher than those expected of the fragmentation of otherwise unsubstituted A/B ring system [5] of 3-amino and 3-dimethyl amino steroids, apart from the diagnostic [4, 5] peaks at *m/z* 56 and 84 (species **b**) respectively. The other prominent ion peaks at *m/z* 428 [M − MeOH], 171 (species **c**), 139 (species **d**) and 111 (species **e**) in the spectrum of **3** and the corresponding ones in that of **4** were in complete agreement [6] with the fragmentation involving E/F ring system of an *O*-methylsolanocapsine structure (Scheme 1).

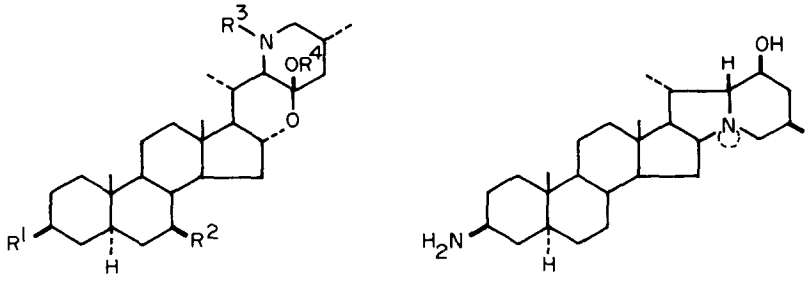
The location of the equatorial OH at C-7 rather than C-6 could, however, be deduced [7] from the deshielding of C-6 (β), C-7 (α), C-8 (β) and C-15 (δ) by 9.9, 42.8, 8.0 and 1.7 ppm, and shielding of C-5 (γ_t), C-9 (γ_t) and C-14 (γ_g) by 3.0, 2.1 and 0.8 ppm respectively in the ^{13}C NMR spectrum of the alkaloid compared to those of **6**. This was further corroborated by the expected [7] shift of the signals for C-6, C-7 and C-8 in the spectrum of the triacetate (**5**). That **3** possesses the same skeleton as well as stereochemistry at all the chiral centres as in *O*-methylsolanocapsine (**6**) received strong support from the very close ^{13}C chemical shifts of other carbons of **3** to those of **6** (Table 1).

*Part 85 in the series 'Studies on Indian Medicinal Plants'. For part 84 see Chakravarty, A. K., Das, B. and Pakrashi, S. C. *Phytochemistry* **26**, 2345.

†Author to whom correspondence should be addressed.



Scheme 1



2

1 $R^1 = NH_2$, $R^2 = R^3 = R^4 = H$
3 $R^1 = NH_2$, $R^2 = OH$, $R^3 = H$, $R^4 = Me$
4 $R^1 = NMe_2$, $R^2 = OH$, $R^3 = R^4 = Me$
5 $R^1 = NHAc$, $R^2 = OAc$, $R^3 = Ac$, $R^4 = Me$
6 $R^1 = NH_2$, $R^2 = R^3 = H$, $R^4 = Me$

Since the alkaloid **3** was isolated after hydrolysis with methanolic HCl under reflux, its possibility of being an artefact could not altogether be ruled out. Unfortunately, paucity of the plant material prevented us from settling the issue. However, the corresponding 23-*O*-desmethyl derivative, *viz.* 7β-hydroxy-solanocapsine is a new alkaloid hitherto not encountered in nature.

During this investigation, we had to re-examine our earlier ^{13}C chemical shift assignments [4] of **6** which were based on T_1 data. We are now obliged to conclude on the basis of INEPT and selective proton decoupling experiments that the chemical shifts of δ54.5 and 46.0 ppm previously ascribed to OMe and C-24 carbons should be changed to δ46.4 and 39.5 ppm respectively. The significant up-field shift of C-24 signal (by ~6.5 ppm) compared to that of **1** [4] and the OMe carbon may then be ascribed to their mutual γ-interaction.

EXPERIMENTAL

Mps: uncorr. ^1H and ^{13}C NMR spectra were taken at 100 and 25.05 MHz respectively. EIMS were recorded on a direct insertion probe at 80 eV.

Plant material. The plant was collected for us by M/s United

Table 1. ^{13}C chemical shifts (δ ppm, $CDCl_3$) of **3** and its derivatives

C	3	4	5	6	C	3	4	5	6
1	37.3	37.5	36.9	37.5	18	13.5	13.7	13.6	13.6
2	32.4	24.4	28.6	31.8	19	12.4	12.4	12.1	12.4
3	50.9	64.1	48.6	51.0	20	33.0	23.2	33.0	33.0
4	38.8	30.8	34.6	39.0	21	15.3	15.2	17.0	15.3
5	42.7	43.2	42.4	45.7	22	68.8	70.5	73.8	68.8
6	38.8	38.9	34.0	28.9	23	98.4	100.1	99.2	98.5
7	75.0	75.2	76.6	32.2	24	39.4	40.1	40.5	39.5
8	43.0	43.0	38.9	35.0	25	30.6	30.6	29.4	30.6
9	52.8	52.8	52.4	54.9	26	54.8	62.8	58.3	54.9
10	35.1	35.3	34.9	35.0	27	18.6	18.5	18.0	18.7
11	20.4	20.5	20.3	20.4	OMe	46.3	46.1	47.0	46.4
12	39.1	39.3	38.8	39.3	NMe_2	—	41.6	—	—
13	42.4	42.5	43.0	42.0	N'Me	—	36.0	—	—
14	54.1	54.2	53.5	54.9	NCOMe	—	—	21.7, 23.4	—
15	30.6	30.6	29.6	28.3	NCOMe	—	—	169.1, 169.8	—
16	74.1	74.0	74.3	74.0	OCOMe	—	—	24.1	—
17	60.6	61.1	61.1	61.4	OCOMe	—	—	170.1	—

Chemical and Allied Products, 10, Clive Row, Calcutta 700001 (who possess a voucher specimen) during November to December from the Nilgiri Hills.

Extraction and isolation. Defatted crushed leaf (6 kg) was extracted for 48 hr with MeOH (3 × 20 l.) at room temp. The extract was concd under red. pres. to 1 l. The concentrate was poured into 4M HOAc (5 l.) with constant stirring and the HOAc acid insoluble part was removed by $CHCl_3$ extraction. The clear aq. part was basified with NH_3 and the separated gummy ppt. was collected by filtration. It was hydrolysed with 10% methanolic HCl (1 l) under reflux for 15 hr. The reaction mixture was concentrated (0.5 l) under red. pres., diluted with H_2O (3 l), basified with NH_3 and extracted with $CHCl_3$ to give the crude alkaloid (80 g) as a gum.

The crude base was dissolved in $CHCl_3$ (250 ml) and repeatedly extracted with 2 MAcOH (15 × 200 ml). The regenerated base (55 g) on CC over neutral Al_2O_3 with $CHCl_3$ (4 l) and $CHCl_3$–MeOH (98:2, 1 l) afforded **1** (12 g, 0.2%), mp 208°. Further elution with $CHCl_3$–MeOH (97:3, 1 l; 9:1, 1 l; 85:15, 500 ml) furnished a gum (10 g) which on repeated chromatography and prep. TLC (solvent system: $CHCl_3$–MeOH, 98:2 saturated with NH_3) yielded **3** (0.1 g) as amorphous powder, $[\alpha]_D^{24°} + 65.2°$ ($CHCl_3$; *c* 0.46). IR ν_{max} (KBr) cm^{-1}: 3340–3410 (*br*). EIMS *m/z* (rel. int.): 460(M^+, 0.5), 445 (1), 428 (36), 424 (15), 171 (31), 139 (72), 111 (96), 98 (25), 56 (100). ^1H NMR ($CDCl_3$): δ0.76 (3H, *s*, 10-Me), 0.81 (3H, *s*, 13-Me), 0.83 (3H, *d*, J = 6 Hz, 25-Me), 0.98 (3H, *d*, J = 6 Hz, 20-Me), 2.66 (1H, *br*, 3-H), 3.04 (1H, *dd*, J = 12, 4 Hz, 26-H_{eq}), 3.13 (3H, *s*, OMe), 3.32 (1H, *m*, 7-H_{ax}), 4.08 (1H, *ddd*, J = 10, 10, 5 Hz, 16-H).

Methylation of **3** to **4**. Compound **3** (30 mg) was dissolved in 85% HCO_2H (1 ml) and 40% HCHO soln (1 ml) was added to it. The mixture was heated on a steam bath for 2 hr. Usual work-up gave an oil which on CC over neutral Al_2O_3 yielded **4** (24 mg) as viscous oil. EIMS *m/z* (rel. int.): 502 (M^+, 2.5), 487 (6), 470 (45), 452 (8), 185 (18), 184 (12), 153 (63), 126 (44), 84 (100).

Acetylation of **3** to **5**. Compound **3** (30 mg) was acetylated with Ac_2O–Py at room temp. for 12 hr to give **5** (25 mg), mp 204–206° (aq. acetone), $[\alpha]_D^{24°} - 90.8°$ ($CHCl_3$; *c* 1.0). IR ν_{max}^{KBr} cm^{-1}: 1730 (OAc), 1645 and 1655 (NAc). EIMS *m/z* (rel. int.): 554 [M − MeOH], 494 [M − MeOH − AcOH]$^+$. ^1H NMR ($CDCl_3$): δ0.80 (3H, *s*, 10-Me), 0.83 (3H, *s*, 13-Me), 0.88 (3H, *d*, J = 6 Hz, 25-Me), 0.94 (3H, *d*, J = 6 Hz, 20-Me), 1.94 (3H, *s*, NAc), 2.00 (3H, *s*, NAc), 2.06 (3H, *s*, OAc), 3.11 (3H, *s*, OMe), 3.72 (1H, *br. d*, J = 12 Hz, 26-H_{eq}), 4.14 (1H, *ddd*, J = 10, 10, 4 Hz, 16-H), 4.56 (1H, *m*, $W_{1/2}$ = 20 Hz, 7-H_{ax}), 5.30 (1H, *d*, J = 8 Hz, -C(3)H-NH-CO-).

REFERENCES

1. Pakrashi, S. C., Chakravarty, A. K. and Ali, E. (1977) *Tetrahedron Letters* 645.
2. Schreiber, K. and Ripperger, H. (1962) *Z. Naturforsch.* **17b**, 217.
3. Lin, C.-N., Chung, M.-I. and Lin, S.-Y. (1987) *Phytochemistry* **26**, 305.
4. Chakravarty, A. K., Das, B., Ali, E. and Pakrashi, S. C. (1984) *J. Chem. Soc. Perkin Trans. 1* 467.
5. Budzikiewicz, H. (1964) *Tetrahedron* **20**, 2267.
6. Ali, E., Chakravarty, A. K., Pakrashi, S. C., Biemann, K. and Hignite, C. E. (1977) *Tetrahedron* **33**, 1371.
7. Eggert, H., VanAntwerp, C. L., Bhacca, N. S. and Djerassi, C. (1976) *J. Org. Chem.* **41**, 71.