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A NEW ALKALOID FROM THE ROOTS OF *RAUVOLFIA SERPENTINA*

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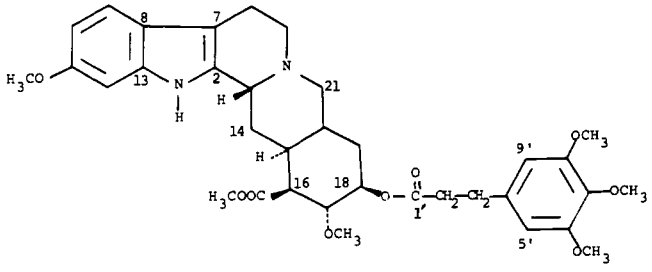
In the course of studies carried out in the 1930s on the alkaloidal constituents of *Rauvolfia serpentina* Benth. (Apocynaceae) roots collected from the Bihar province and Dehradun, Siddiqui (1-3) noted a marked difference in their character and yields. It was observed that the yellow oxidation bases of the white ajmaline group found in the plant growing in the hot swampy districts of Bihar do not occur in the temperate climatic conditions of the Dun valley. In light of these observations, studies were more recently undertaken by Siddiqui, *et al.* on the roots collected from Thailand and Nepal, as a result of which isolation and structure elucidation of two new indole alkaloids—sandwicoline (4) and sandwicolidine (5) from the Nepalian roots—and a new alkaloid—ajmalicidine (6) from the roots of Thai origin—have been reported earlier. The present paper deals with the isolation and structure elucidation of another new base named rescinnamidine [**1**] (2′,3′-dihydrorescinnamine) from the roots of *R. serpentina* collected from Thailand.

Rescinnamidine [**1**] has a molecular formula $C_{35}H_{44}N_2O_9$ (high resolution mass M^+ 636.7420). Its uv spectrum showed maxima at 210, 225, 270, and 295 nm characteristic of methoxyindole alkaloids. The ir spectrum showed peaks at 3450 (N-H), 1725, and 1740 cm^{-1} (carbonyl stretchings), in addition to other peaks at 3100-2890 (C-H stretching), 1530 (characteristic for methoxy indole), and 870-770 cm^{-1} (C-H bending of substituted benzene ring). The ^1H and ^{13}C nmr exhibited the presence of six methoxy groups while a fragment at *m/z* 397.2127 in the ms corresponding to $C_{23}H_{29}N_2O_4$ resulting from the loss of $C_{12}H_{15}O_5$ indicated the presence of a trimethoxy-dihydrocinnamoyl ester in the molecule. This was supported by the appearance of a two-proton singlet at δ 6.42 (H-5′ and H-9′), a six-proton singlet at δ 3.84 (6′-OCH_3 and 8′-OCH_3), a three-proton singlet at δ 3.81 (7′-OCH_3), and two two-proton triplets at δ 2.93 (J=7.5 Hz) and 2.63 (J=7.5 Hz) for H-2′ and H-3′. The spectral data of **1** showed a close relation with the reserpine series of alkaloids and a one-proton doublets of double doublets at δ 4.78 ($J_{17,18}$=9.2 Hz, $J_{18,19\alpha}$=4.8 Hz, and $J_{18,19\beta}$=14 Hz) led to the location of the dihydrocinnamoyl ester at C-18.

The ^1H- and ^{13}C-nmr spectra indicated that, in addition to the trimethoxydihydrocinnamoyl group, **1** contains a methoxy ester (δ ^1H=3.79, δ ^{13}C=51.81 and 172.97) and two methoxy functions (δ ^1H=3.81 and 3.42, δ ^{13}C=55.60 and 60.61). A one-proton singlet at δ 7.45 has been attributed to N-H, while two one-proton



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doublets at δ 7.31 ($J_{9,10}$=8.6 Hz) and 6.83 ($J_{10,12}$=2.0 Hz) and a one-proton doublet of doublets at δ 6.7 ($J_{9,10}$=8.6 Hz, $J_{10,12}$=2.0 Hz) have been assigned to the aromatic protons H-9, H-12, and H-10, respectively, indicating that one of the methoxy groups is present at C-11. A doublet of doublets at δ 3.73 ($J_{16,17}$=11.1 Hz, $J_{17,18}$=9.2 Hz) and a doublet of doublets at δ 2.6 ($J_{15,16}$= 4.6 Hz, $J_{16,17}$=11.1 Hz) attributed to H-17 and H-16, showed the position of the methoxy ester and methoxy function at C-16 and C-17, respectively. A broad singlet at δ 4.41 assigned to H-3 suggested its β-disposition (7). The structure of rescinnamidine [**1**] was finally substantiated by the ^{13}C-nmr (broad band and spin echo) spectral data and by the hydrolysis of **1** into methyl reserpate.

The values of chemical shifts and coupling constants showed that the stereochemistry of all the asymmetric centers of **1** is the same as reserpine and rescinnamine (8-11).

EXPERIMENTAL

GENERAL EXPERIMENTAL PROCEDURES.—Melting points were recorded in glass capillary tubes and are uncorrected; ir ($CHCl_3$) and uv (MeOH) spectra were measured on JASCO IRA-I and Pye-Unicam SP-800 spectrometers, respectively. Mass spectra were recorded on Finnigan MAT 112 and MAT 312 double focusing mass spectrometer connected to PDP 11/34 computer system. ^{1}H- and ^{13}C-nmr (broad band and gated spin echo) spectra were recorded in $CDCl_3$ with TMS as internal reference on 300 MHz instrument model Bruker aspect 3000. The ^{1}H- and ^{13}C-nmr spectral assignments have been made through a comparison of the chemical shifts with the published data for similar compounds (7,12). The purity of samples was checked on tlc (S.I.F.-254 precoated aluminium cards).

ISOLATION.—The viscous dark brownish residue from the alcoholic extract of the undried roots was partitioned between 5% HOAc and EtOAc. The residue, obtained on usual work-up and removal of the solvent from the EtOAc phase, was exhaustively treated with petroleum ether to remove the fatty constituents. The petroleum ether insoluble fraction was divided into C_6H_6-Et_2O (1:1) and EtOAc soluble fractions. The former was freed of the solvent and subjected to thick layer chromatography (Silica gel, $CHCl_3$-MeOH, 9.9:0.1) resulting in the isolation of rescinnamidine as a yellow crystalline solid which on recrystallization from $CHCl_3$-Et_2O (8:2) formed sharp needles mp 260-261°, $[\alpha]^{20}D$=207 ($CHCl_3$); eims *m/z* (rel. int. %) 636.7420 (M^+, calcd. for $C_{35}H_{44}N_2O_9$, 636.7423)$^+$ (100), 621.7056 ($C_{34}H_{41}N_2O_9$)$^+$ (16), 605.7063 ($C_{34}H_{41}N_2O_8$)$^+$ (18), 397.2127 ($C_{23}H_{29}N_2O_4$)$^+$ (34), 382.1832 ($C_{22}H_{26}N_2O_4$)$^+$ (8), 366.1940 ($C_{22}H_{26}N_2O_3$)$^+$ (18), 251.1172 ($C_{16}H_{15}N_2O$)$^+$ (16), 214.1182 ($C_{13}H_{14}N_2O$)$^+$ (18), 200.0929 ($C_{12}H_{12}N_2O$)$^+$ (20) and 181.0854 ($C_{10}H_{13}O_3$)$^+$ (36); uv λ max (nm) 210, 225, 270, 295; ir ν max (cm^{-1}) 3450 (-NH), 1740 and 1725 (C=O, stretching), 1530 (methoxy indole), 2890-3100 (C-H stretching), 1610 and 1465 (C=C aromatic vibration), 770-870 (C-H bending of substituted benzene ring).

^{1}H nmr ($CDCl_3$) δ 7.45 (1H, s, NH), 7.31 (1H, d, $J_{9,10}$=8.6 Hz, H-9), 6.83 (1H, d, $J_{10,12}$=2.0 Hz, H-12), 6.7 (1H, dd, $J_{9,10}$=8.6 Hz $J_{10,12}$=2.0 Hz, H-10), 6.42 (2H, s, H-5′, H-9′), 4.78 (1H, ddd, $J_{17,18}$=9.2 Hz, $J_{18,19\alpha}$=4.8 Hz, $J_{18,19\beta}$=14 Hz, H-18), 4.41 (1H, br. s, H-3β), 3.84 [6H, s, C-6′, 8′ $(OCH_3)_2$], 3.81 (6H, s, 2×OCH_3), 3.79 (3H, s, $COOCH_3$), 3.73 (1H, dd, $J_{16,17}$=11.1 Hz, $J_{17,18}$=9.2 Hz, H-17), 3.42 (3H, s, C-17 OCH_3), 3.20 (2H, m, H-5), 3.05 (1H, dd, $J_{20,21\beta}$=4.0 Hz, $J_{21\alpha,21\beta}$=11.9 Hz, H-21β), 2.93 (2H, t, J=7.5 Hz, H-3′), 2.63 (2H, t, J=7.5 Hz, H-2′), 2.6 (1H, dd, $J_{15,16}$=4.6 Hz, $J_{16,17}$=11.1 Hz, H-16), 2.18 (1H, m, $J_{18,19\beta}$=14 Hz, $J_{19\alpha,19\beta}$=13.3 Hz, $J_{19\beta,20}$=11.7 Hz, H-19β), 1.93 (1H, ddd, $J_{14\alpha,15}$=3.9 Hz, $J_{14\beta,15}$=14.8 Hz, $J_{15,16}$=4.6 Hz, $J_{15,20}$=3 Hz, H-15) and 1.86 (1H, ddd, $J_{19\alpha,20}$=4 Hz, $J_{19\beta,20}$= 11.7 Hz, $J_{20,21\alpha}$=2 Hz, $J_{20,21\beta}$=4 Hz, H-20).

^{13}C nmr ($CDCl_3$) δ 172.79[a] (C=O), 172.22[a] (C-1′), 156.35 (C-11), 153.37 (C-6′, 8′), 136.54 (C-7′), 136.26 (C-13), 130.41 (C-2), 122.29 (C-4′), 118.58 (C-9), 109.15 (C-10), 108.10 (C-7), 105.56 (C-5′,9′), 95.42 (C-12), 77.77[b] (C-17), 77.60[b] (C-18), 60.85[c] (7′-OCH_3), 60.61[c] (17-OCH_3), 56.21[d] (6′,8′-OCH_3), 55.60[d] (11-OCH_3), 53.81 (C-3), 52.09 (C-16), 51.81 ($COOCH_3$), 51.26 (C-5), 44.09 (C-21), 36.56 (C-2′), 34.07 (C-20), 32.33 (C-15), 31.45 (C-3′), 29.67 (C-19), 24.25 (C-14), 16.65 (C-6). (Assignments with superscripts a,b,c,d may be interchanged).

HYDROLYSIS OF **1** TO METHYL RESERPATE.—Rescinnamidine (6 mg) was heated at 70° with 5% NaOH for 30 min. The reaction mixture was then shaken out with EtOAc, and on usual work up, methyl reserpate was obtained as crystalline solid, mp 241-243°; eims *m/z* (rel. int. %) 414.2153 (M^+, calcd. for $C_{23}H_{30}N_2O_5$ 414.2155) (30); ir ν max 1735 cm^{-1}; ^{1}H nmr ($CDCl_3$) δ 7.57 (1H, s, NH), 4.41 (1H, br. s, H-

3β), 3.83 (3H, s, OCH_3), 3.79 (3H, s, OCH_3), 3.56 (3H, s, OCH_3), 3.50 (1H, m, H-17β), 3.54 (1H, m, H-18α).

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