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NORTENUIPINE-2'β-N-OXIDE, A NEW BISBENZYLISOQUINOLINE ALKALOID FROM DAPHNANDRA DIELSII

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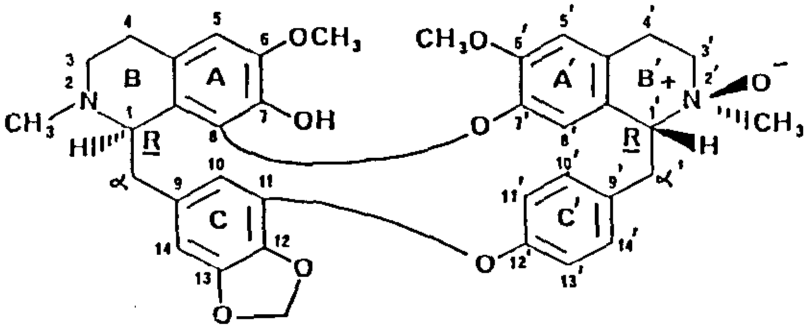
Abstract - Repeated chromatography of the nonquaternary alkaloid fraction of an extract of the bark of Daphnandra dielsii Perk. (Monimiaceae) over silica gel afforded the new bisbenzylisoquinoline alkaloid (-)-nortenuipine-2'β-N-oxide (1) which was characterized by a consideration of physicochemical data and reduction to (-)-nortenuipine. In addition, seven other bisbenzylisoquinoline alkaloids of known structure were isolated and identified.

Plants of the genus Daphnandra (Fam. Monimiaceae) are large trees indigenous to the rain forests of Australia.[1] This genus, some species of which have been used as timber,[1] is a rich source of bisbenzylisoquinoline alkaloids, with approximately twenty-five different compounds of this type having been isolated from among nine different species.[2-5] However, there appear to have been only two reports dealing with the alkaloids of Daphnandra dielsii Perk. The first appeared in 1947 and reported the isolation of repanduline,[6] while six years later a second paper detailed the isolation of O-methylrepandine, repandinine (racemic tenuipine), and tenuipine.[7] This present paper is to report the isolation and identification of a new bisbenzylisoquinoline alkaloid, (-)-nortenuipine-2'β-N-oxide (1), as well as that of seven other similar bisbenzylisoquinoline alkaloids of previously established structure.

Chromatography of a portion of the nonquaternary alkaloid fraction of an alcoholic extract of the bark of D. dielsii over silica gel and elution with petrol ether-$CHCl_3$ mixtures, followed by $CHCl_3$, failed to elute any alkaloids. However, elution with a $CHCl_3$-MeOH gradient successively afforded the bisbenzylisoquinoline alkaloids repanduline, pseudorepanduline[8], (-)-nortenuipine, and oxyacanthine. This is the first reported isolation of oxyacanthine from the genus Daphnandra. Elution of the column with $CHCl_3$-MeOH (96:4) afforded a fraction which, after repeated preparative tlc, gave additional quantities of oxyacanthine, plus the bisbenzylisoquinoline alkaloids N-methylapateline[9] and repandine. Continued elution of the column with a $CHCl_3$-MeOH gradient gave a complex mixture of alkaloids. Preparative tlc of this mixture afforded the bisbenzylisoquinoline alkaloid aromoline plus the new base (-)-nortenuipine-2' β -N-oxide (1).

The uv spectrum of (-)-nortenuipine-2' β -N-oxide (1) showed a single maximum at 282 nm (log 3.81). The ms of the alkaloid showed the M^+ at m/z 638 (5%) and other significant fragment ions at m/z 623 (8), 622 (24), 592 (22), 431 (3), 430 (3), 382 (10), 381 (38), 367 (13), and 191 (61). These spectral data were characteristic of a bisbenzylisoquinoline mono-N-oxide alkaloid[2-5,10] which was confirmed by reduction of the alkaloid with H_2SO_3 to afford (-)-nortenuipine (2), thereby establishing the oxygenation pattern, the structure and stereochemistry of the skeleton. The placement of the oxide-nitrogen atom at N-2 or N-2' and the spatial relationship of that oxygen atom to its neighboring H-1 or H-1' proton remained to be established. The ^{1}H-nmr spectrum (300 MHz, $CDCl_3$, TMS, δ in ppm) of the N-oxide indicated the presence of one N-methyl group at 2.40, one N-oxide methyl group at 2.98, two methoxyl groups at 3.39 (C-6') and 3.79 (C-6), one methylenedioxy group at 6.00 (C-12 + C-13) and a cluster of nine aromatic protons from 6.11 to 7.40. These protons could be assigned the following chemical shift values by analogy with limacine-2' β -N-oxide[11] (3) and nortenuipine (2): 6.12 (1H,s)(H-8'), 6.23 (1H,dd,J=2.5, 9 Hz)(H-10'), 6.32 (1H,s)(H-5 or H-14), 6.35 (1H,s)(H-14 or H-5), 6.58 (1H,s)(H-5' or H-10), 6.60 (1H,s)(H-10 or H-5'), 6.80 (1H,dd,J=2.5, 9 Hz)(H-11'), 7.02 (1H,d,J=9 Hz)(H-13'), and 7.40 (1H,d,J=9 Hz)(H-14'). The methine proton at H-1' is found at 4.65 (dd) but the corresponding proton for H-1 is difficult to observe since it is hidden beneath the methoxyl peak at 3.79.[11] Since it has been established that the chemical shift of the 2-N-methyl group in nortenuipine, as well as other tail-to-tail linked bisbenzylisoquinoline alkaloids containing two diaryl ether bridges (subgroup C [8-7', 11-12' linked]), is at approximately 2.30-2.35 while that of the 2'-N-methyl group is at about 2.60-2.65,[10] the presence of a three proton singlet at 2.40 in the isolated alkaloid plus the absence of a signal at about 2.6 fixes the N-oxide at the N-2' position. It has also been determined that the chemical shift of a 2-N-methyl (or 2'-N-methyl) of this same subgroup as nortenuipine (R,R) undergoes a downfield shift of 0.3-0.4 ppm when H-1 (or H-1') is on the same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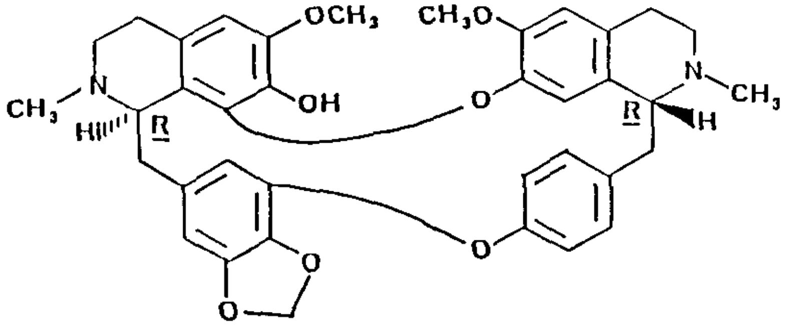
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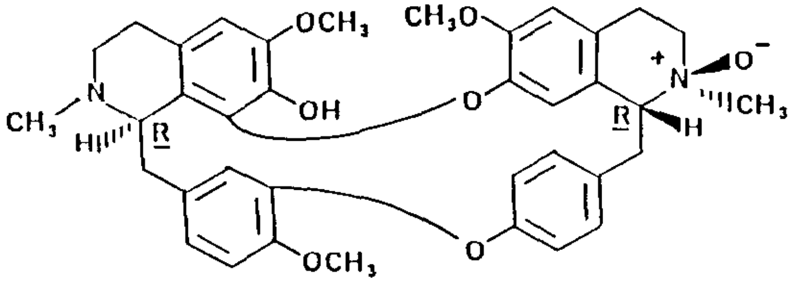
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side as the N-oxide oxygen atom, with a larger downfield shift (0.7-0.9 ppm) being operative when the H-1 (or H-1') is on the side opposite to the N-oxide oxygen atom.[10-12] Since the chemical shift of the 2'-N-methyl signal of nortenuipine is at 2.65 while that of the naturally occurring 2'-N-oxide is at 2.98, the downfield shift of 0.33 ppm suggests a β-N-oxide configuration in which the H-1' proton and the N-oxide oxygen atom bear a *syn*-relationship to one another. Although this is the first reported isolation of a bisbenzylisoquinoline-N-oxide from the genus *Daphnandra* and from the family Monimiaceae, there are at least sixteen additional bisbenzylisoquinoline mono-N-oxide alkaloids which have been previously isolated from higher plants.[2-5,13,14] Twelve of these bases have been found in nine different genera of the family Menispermaceae. Of the remaining four alkaloids, three were isolated from two different species of the genus *Berberis* (Fam. Berberidaceae), while the last was isolated from *Thalictrum sultanabadense* Stapf. (Fam. Ranunculaceae). Six of these twelve bases are of the same linkage (8-7',11-12')(family subgroup C)[10](type VIII)[2-4] as nortenuipine-2' β-N-oxide and are found in four genera of the family Menispermaceae. Finally, four of these six alkaloids have identical stereochemistry (*R*,*R*).

Note

A preliminary unpublished communication has combined *Daphnandra dielsii* Perk. with *Daphnandra repandula* F. Muell.[15]

EXPERIMENTAL

Melting points were determined on a Fisher-Johns apparatus and are uncorrected. The uv spectra were taken on a Perkin-Elmer Model 552A spectrophotometer in MeOH while the optical rotations were measured on a Perkin-Elmer Model 241 Automatic Polarimeter. The ^{1}H-nmr spectra were recorded on a Bruker Model AF 300 - 300MHz Fourier Transform Spectrometer in $CDCl_3$. The mass spectra were obtained on a Finnigan EI Mass Spectrometer, Spectral Electronics, interfaced with a Finnigan INCOS data system. Column chromatography, tlc, and plc utilized silicic acid or silica gel.

Isolation of Nortenuipine-2' β-N-Oxide (**1**): - Extraction of the bark (1 kg) of *D. dielsii* with EtOH[7] and systematic partitioning in a classical manner[7] afforded a nonquaternary alkaloid fraction (8.1 g). The nonquaternary alkaloid fraction (8.1 g) was adsorbed on silica gel (16 g) and chromatographed over silicic acid (300 g) in petrol ether-$CHCl_3$ (8:1). Elution with petrol ether-$CHCl_3$ mixtures (500 ml each), followed by $CHCl_3$ (500 ml), failed to elute alkaloids. Elution with a $CHCl_3$-MeOH gradient ([99:1] to [97:3]) (14 x 500 ml) afforded repanduline (419 mg)(mp,uv,ir,^{1}H-nmr,ms,$[\alpha]_D$), pseudorepanduline (102 mg)(mp,uv,ir,^{1}H-nmr,ms,$[\alpha]_D$), (-)-nortenuipine (30 mg)(uv,ir,ms,$[\alpha]_D$)(^{1}H-nmr: δ 2.35 (N-2CH_3,3H,s), 2.62 (N-2'CH_3,3H,s), 3.36 (C-6'OCH_3,3H,s), 3.73 (H-1,1H,d,J=9.4 Hz), 3.77 (C-6OCH_3,3H,s), 3.90 (H-1',1H,dd,J=13.0,6.2 Hz), 5.99 (CH_2O_2,2H,s), 6.02 (H-8',1H,s), 6.16 (H-14,1H,s), 6.28 (H-5,1H,s), 6.31 (H-10',1H,d,J=8.3 Hz), 6.53 (H-5'or H-10, 1H,s), 6.55 (H-10 or H-5',1H,s), 6.81 (H-11',1H,d,J=8.2 Hz), 7.15 (H-13',1H,d,J=8.1 Hz), and 7.34 (H-14',1H,d,J=8.2 Hz) and oxyacanthine (32 mg)(uv,ir,^{1}H-nmr,ms,[α]$_D$). Elution with $CHCl_3$-MeOH (96:4)(7 x 500 ml) afforded a fraction which on repeated preparative tlc ([C_6H_6-Me_2CO-NH_4OH (28%)][6:24:0.65]) gave additional oxyacanthine, plus N-methylapateline (3 mg) (uv,ir,^{1}H-nmr,ms, [α]$_D$) and repandine (10 mg)(uv,ir,^{1}H-nmr,ms,[α]$_D$). Continued elution with the $CHCl_3$-MeOH gradient ([95:5] to [80:20]) gave a complex mixture of alkaloids. Preparative tlc ([C_6H_6-Me_2CO-MeOH-NH_4OH (28%)][20:20:0.5:1]) of this mixture afforded aromoline (3 mg) and (-)-nortenuipine-2'β-N-oxide (1)(9 mg), the latter as a pale-yellow amorphous residue; $[\alpha]_D^{27}$ -71° (c 0.24, $CHCl_3$).

Reduction of (-)-Nortenuipine-2' β-N-Oxide (1): - Alkaloid 1 (9 mg) was dissolved in H_2SO_3 (7.9%)(1 ml) and allowed to stand overnight at room temperature. The solution was basified with NH_4OH (28%) and extracted with Et_2O (3 x 20 ml), then $CHCl_3$ (3 x 20 ml). The Et_2O and $CHCl_3$ solutions were combined, dried (anhydrous Na_2SO_4), filtered, and evaporated to afford (-)-nortenuipine (2) (4 mg)(uv,^{1}H-nmr,ms,[α]$_D$) as a colorless, amorphous residue.

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