of tail just prior to and 1 and 3 h after the extract/standard drug administration. In sub-acute treatment, the administration of extract/standard drug was continued for 10 days, once daily. Blood samples were collected from the tip of the tail just prior to and on days 1, 3, 7 and 10 of the extract/standard drug administration. The blood glucose levels were determined for all the samples by the glucose-oxidase method (Varley et al. 1967). Statistical significance was determined by one-way analysis of variance (ANOVA) followed by Dunnet's t-test. $P < 0.05$ indicates significant differences between group means.

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A new homotriterpene from the roots of *Marsdenia tenacissima* Wight and Arn.

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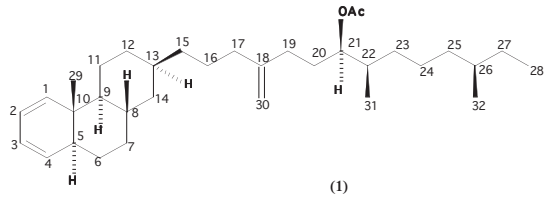
A new homotriterpene isolated from the roots of *Marsdenia tenacissima* Wight et Arn. has been characterized as 13-(31,32-dimethyl-30-methylene-21α-acetoxy-tetradecanyl)-29-methyl-perhydrophenanthr-1,3-diene on the basis of spectral analyses.

Marsdenia tenacissima Wight et Arn. (Asclepiadaceae), popularly known as Safed Nishoth, is a large and stout twinning shrub, with grey or pale brown bark. It is an important herbal drug used in the indigenous system of medicine as a purgative, antipyretic, antifilarial, bitter stomachic and uterine sedative (Anonymous 1966; Kirtikar and Basu 1994). It has also been found to contain antimutagenic factors (Nadkarni 1954; Lee H and Lin JY 1988). The presence of steroidal glycosides (Chen et al. 1999; Yang et al. 1981) and pregnane derivatives (Singhal and Khare 1980) from the various part of the plant has been reported.

Marsdemene (**1**) was isolated from a petroleum ether-chloroform (1 : 1) mixture as a colourless crystalline solid. It responded positively to the tetranitromethane and bromine water tests for unsaturation. Its IR spectrum exhibited absorption bands for ester group (1751 cm^{-1}) and unsaturation (1662 cm^{-1}). The FAB mass spectrum of **1** exhibited a molecular ion peak at m/z 496 corresponding to the molecular formula of tricyclic homotriterpene acetate, $C_{34}H_{56}O_2$, which was supported by its ^1H, ^{13}C and DEPT NMR spectra. The spectra indicated seven double bond equivalents; three of these were adjusted in a tricyclic carbon framework, three in olefinic linkages and the remaining one in the ester group.



(1)

The MS showed diagnostically important fragments at m/z 444 $[C_{1,10}–C_{4,5}$ fission$]^+$, 384 [444-AcOH]$^+$, 106, 390 $[C_{6,7}–C_{9,10}$ fission$]^+$, 120 $[C_{7,8}–C_{9,10}$ fission$]^+$, 146, 350 $[C_{8,14}–C_{9,11}$ fission$]^+$, 160 $[C_{8,14}–C_{11,12}$ fission$]^+$ and 174

$[C_{8,14}-C_{12,13}$ fission$]^+$, suggesting the location of the two vinylic linkages in ring A and saturated nature of rings B and C. The ion peaks at m/z 201, 295 $[C_{13}-C_{15}$ fission$]^+$, and generation of ion fragments at m/z 281, 267, 253 due to subsequent removal of methylene groups from the mass unit 295 and then m/z 227 $[253\text{-}C_2H_2]^+$, 213 $[227\text{-}CH_2]^+$, 199 $[213\text{-}CH_2]^+$ and 156 $[199\text{-Ac}]^+$, supported the location of unsaturated methylene function at C-18. The ion peaks appearing at m/z 397 $[C_{22}-C_{23}$ fission$]^+$, 243 $[397\text{-}(CH)_2(CH_2)_2CH(OAc)CH_2\ CH_3]^+$, 229 $[243\text{-}CH_2]^+$, 215 $[229\text{-}CH_2]^+$ and 201 $[215\text{-}CH_2]^+$ indicated the location of the acetoxy group at C-21.

The ^{1}H NMR spectrum of **1** displayed a two-proton broad signal at δ 5.30 assigned to vinylic H-1 and H-4 and five one-proton signals at δ 5.06 and 4.83 ascribed to H-2 and H-3, respectively, at δ 4.74, 4.63 attributed to vinylic C-30 methylene protons and at δ 4.57 with half-width of 16.5 Hz accounted to carbinol H-21α. A six-proton broad signal at δ 0.96 was associated with C-29 and C-31 methyl protons. A three-proton doublet at δ 0.84 (J = 6.2 Hz) was due to C-32 secondary methyl protons. A three-proton triplet at δ 0.54 (J = 6 Hz) and a three proton broad signal at δ 1.98 were assigned to C-28 primary methyl and to acetyl protons, respectively. The remaining methylene and methine protons appeared between δ 2.21 and 1.22. The appearance of all the methyl protons in the range δ 0.98–0.54 supported the location of these functionalities on the saturated carbons.

The ^{13}C NMR spectrum of **1** showed 34 Carbon signals in the molecule. The vinylic carbon signals appeared at δ 124.64 (C-1), 130.0 (C-2), 128.42 (C-3), 122.71 (C-4), 140.09 (C-18) and 109.72 (C-30). Signals at δ 172.76 and 19.31 were assigned to acetyl carbons. The degree of protonation of each carbon was determined by DEPT experiments, which indicated the presence of 5 methyl, 15 methylene, 11 methine and three quaternary carbons. On the basis of these spectral evidences, compound **1** was characterized as 13-(31,32-dimethyl-30-methylene-21α-acetoxytetradecanyl)-29-methyl-perhydrophenanthr-1,3-diene.

Experimental

1. General procedure

Melting point is uncorrected. IR spectra were recorded on Perkin Elmer 1710 FTIR spectrophotometer using KBr pellets. ^{1}H NMR (300 MHz), ^{13}C NMR (75 MHz) and 2D NMR spectra were screened by Brucker WM-400 spectrometer in $CDCl_3$ using TMS as standard. FAB-MS was run at 70 eV on a JEOL-JMS-D100 mass spectrophotometer.

Column chromatography was performed on silica gel (Merck, 60–120 mesh) and thin layer chromatography on silica gel G (Merck).

2. Plant material

Roots of *M. tenacissima* Wight et Arn. were purchased from the local market of Khari Baoli, Delhi, India in August 2002. The specimen were identified by Dr. MP Sharma (Taxonomist), in the Department of Botany, Jamia Hamdard. A voucher specimen is deposited in the herbarium of the Phytochemistry Research Laboratory, Jamia Hamdard.

3. Extraction

The roots of *M. tenacissima* were extracted with petroleum ether, chloroform and then finally with methanol. The chloroform fraction was concentrated and chromatographed on silica gel (60–120 mesh). The column was eluted with petroleum ether and chloroform in order of increasing polarity.

4. Isolation and characterization of 1

Elution of the column with petroleum ether-chloroform (1 : 1), furnished colourless crystals of **1**, which were recrystallized from chloroform-methanol (1 : 1) as colourless crystalline solid, 340 mg; m.p: 45–48 °C; R_f: 0.33 (hexane : benzene; 85 : 15); UV λ_{max} (MeOH): 223, 277 nm (log ε 6.3, 4.1); IR ν_{max} (KBr): 2926, 2860, 1751, 1662, 1592, 1374, 1218, 1079, 1027, 770 cm^{-1}; ^{1}H NMR ($CDCl_3$): δ 5.30 (2H, brs, H-1, H-4), 5.06 (1 H, brs, H-2), 4.83 (1 H, brs, H-3), 4.74 (1 H, brs, H_2-30a), 4.63 (1 H, brs, H_2-30b), 4.57 (1 H, brs, $w_{1/2} = 16.5$ Hz, H-21α), 2.21 (1 H, m, H-9), 1.98 (5 H, brs, H-8, H-13, H-26, CH_2-19), 1.98 (3 H, brs, $COCH_3$), 1.79 (4 H, brs, H_2-6, H_2-7), 1.63 (4H, brs, H_2-20, H_2-23), 1.56 (6 H, brs, $3 \times CH_2$), 1.22 (14 H, brs, $7 \times CH_2$), 0.96 (6 H, brs, Me-29, Me-31), 0.84 (3 H, d, J = 6.2 Hz, Me-32), 0.54 (3 H, t, J = 6.0 Hz, Me-28); ^{13}C NMR ($CDCl_3$): δ 124.64 (C-1), 130.00 (C-2), 128.42 (C-3), 122.71 (C-4), 56.50 (C-5), 39.14 (C-6), 39.59 (C-7), 51.40(C-8), 57.06 (C-9), 43.27 (C-10), 37.47 (C-11), 35.00 (C-12), 50.72 (C-13), 37.99 (C-14), 32.22 (C-15), 30.00 (C-16), 29.65 (C-17), 140.09 (C-18), 29.45 (C-19), 27.52 (C-20), 27.63 (C-21), 50.69 (C-22), 26.33 (C-23), 24.59 (C-24), 73.72 (C-25), 32.73 (C-26), 24.59 (C-27), 11.92 (C-28), 17.53 (C-29), 109.72 (C-30), 13.86 (C-31), 12.79 (C-32), 172.76 ($COCH_3$), 19.31 ($COCH_3$); + ve FAB-MS m/z (rel. int.): 496 $[M]^+$ ($C_{34}H_{56}O_2$) (1.1), 444 (11.3), 430 (8.7), 409 (8.5), 397 (10.3), 395 (11.2), 390 (3.6), 384 (11.6), 382 (6.7), 381 (18.3), 365 (65.7), 350 (16.3), 309 (2.3), 295 (10.6), 281 (9.7), 267 (7.5), 255 (31.7), 253 (21.3), 243 (11.3), 229 (21.5), 227 (14.1), 215 (20.7), 213 (33.1), 201 (21.8), 199 (23.6), 185 (22.6), 174 (25.4), 160 (60.1), 156 (23.6), 146 (81.6), 133 (61.9), 120 (67.6), 106 (100).

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