

Chemical constituents of *Parkia javanica*, *Alocasia indica* and *Premna latifolia*

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Abstract : Isolation and characterization of one new phenyl propanoid, parkinol from *Parkia javanica* leaves; five known sterols, ergosterol, campesterol, stigmasterol, β -sitosterol and clionasterol from *Alocasia indica* tubers; and two known sterols, β -sitosterol and stigmasterol from *Premna latifolia* leaves are reported. The structure of parkinol was established as 4' (methylene nonanoyloxy)-3-phenyl propanol by spectroscopic studies.

Keywords : *Parkia javanica*, *Alocasia indica*, *Premna latifolia*, parkinol, sterols.

Introduction

Parkia javanica (Lamk.) Merr. Syn., *Parkia roxburghii* G. Don. and *Parkia timoriana* (DC) Merr. (local name : Safola, Long chak, Kuki-Tetai, Mimosaceae) is medium sized tree¹ and is widely distributed in different parts of North-East India. The tender fruits are eaten by the local people as vegetables. The methanol extracts of both the leaf and the stem-bark are found to possess significant anti-neoplastic activity against K 562 and EAC/Dox cell lines. Earlier we have reported² two iridoid glucosides, javanicosides A and B, ursolic acid and β -sitosterol. Here we report the isolation and characterization of one new phenyl propanoid, designated as parkinol from the leaf of the plant.

Air-dried fresh leaves of *P. javanica* (5 kg) (collected from the forest region of Ambassa, Dhalai District, Tripura in January 2005) were extracted with MeOH at room temperature by percolation process. The concentrated MeOH extract was fractionated into benzene, chloroform and *n*-butanol soluble fractions by partition with water. The CHCl_3 soluble fraction on column chromatography (CC) with silica gel afforded a solid, which on repeated CC gave a white amorphous solid (18 mg), $\text{C}_{19}\text{H}_{30}\text{O}_3$, m.p. 152 °C. It was homogeneous on TLC in different solvent systems. It showed UV absorption in MeOH at λ_{max} 270 nm (log ϵ , 3.17), suggesting its aromatic nature. It showed IR absorption band in KBr for ester (1730 cm^{-1}), aromatic (1605 cm^{-1}) and hydroxyl (3308 cm^{-1}) functions. The 400 MHz ^1H NMR spectrum of the com-

pound in $\text{C}_5\text{D}_5\text{N}$ displayed signals for four aromatic protons as AB doublets at δ 7.28 (2H, d, J 8.0 Hz, H-3', 5') and 7.41 (2H, d, J 8.0 Hz, H-2', 6'), a propanol side chain attached to benzyloxy group [δ 2.88 (2H, dd, J 7.0 and 7.0 Hz, H_2 -3), 1.77 (2H, quin, J 7.0 Hz, H_2 -2), 3.70 (2H, t, J 7.0 Hz, H_2 -1) and 2.0 (1H, s, HO-1)] and a benzylic methylene group at δ 3.58 (2H, s) attached to nonanoyloxy group [δ 2.31 (2H, t, J 11.0 Hz, H_2 -2''), 2.23 (2H, m, H_2 -3''), 1.53 (2H, quin, J 7.5 Hz, H-4''), 1.26 (6H, brs, H_2 -5''-8'') and 0.87 (3H, t, J 7.0 Hz, H_3 -9'') suggesting nonanoyloxybenzylpropanol structure³. The 100 MHz ^{13}C NMR spectrum in $\text{C}_5\text{D}_5\text{N}$ with DEPT experiments recorded signals at δ 62.2 (t, C-1), 34.4 (t, C-2), 29.7 (t, C-3), 136.3 (s, C-1'), 128.8 (d, C-2', 6'), 129.6 (d, C-3', 5'), 134.2 (s, C-4'), 63.6 (t, C-7'), 173.0 (s, C-1''), 33.9 (t, C-2''), 26.7 (t, C-3''), 32.2 (t, C-4''), 30.2-30.0 (t, C-5''-7''), 23.0 (t, C-8''), 14.4 (q, C-9'') supporting the said structure. The FAB-MS (positive mode) of the compound recorded mass ion peaks at m/z 307 $[\text{M}+\text{H}]^+$ (57), 289 $[\text{MH}-\text{H}_2\text{O}]^+$ (42), 166 $[\text{M}-\text{nonanoyl}+\text{H}]^+$ (21), 165 $[\text{M}-\text{nonanoyl}]^+$ (28), 159 (10), 141 (15), 122 (45), 113 $[\text{CH}_3-(\text{CH}_2)_7]^+$ (28), 107 $[\text{benzyl alcohol}-\text{H}]^+$ (100%), 91 (96), 78 (65), 77 (99), 55 (99) and 41 (70) (Fig. 1) corroborating the said structure. The compound on saponification with 2 *N* aqueous methanolic KOH afforded nonanoic acid, $\text{C}_9\text{H}_{18}\text{O}_2$ (2) (M^+ 158) and 4' (hydroxymethyl)-3-phenyl propanol, $\text{C}_{10}\text{H}_{14}\text{O}_2$ (3) (M^+ 166), characterized by Co-GC with authentic samples. Therefore, the structure of the compound was assigned as

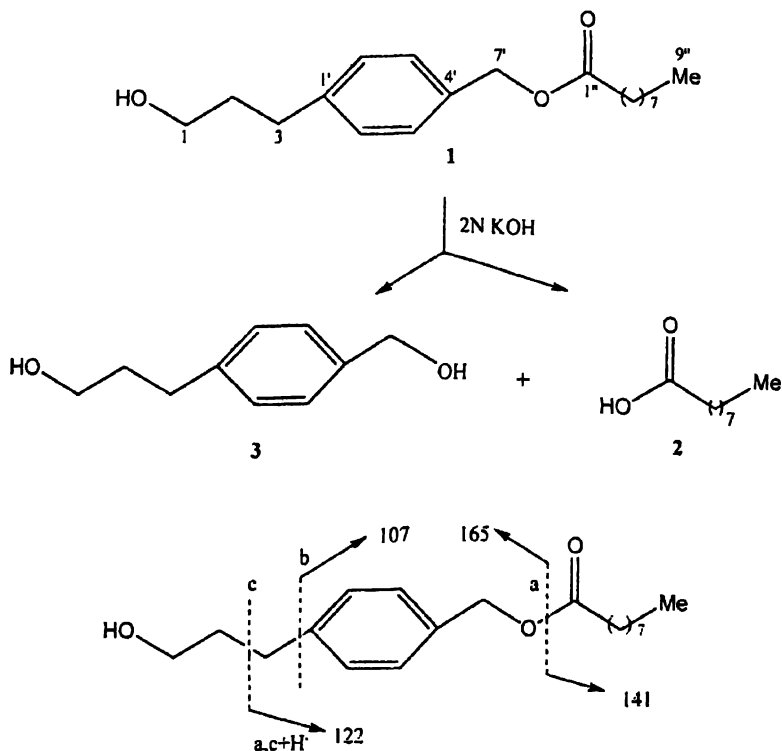


Fig. 1. Mass fragmentation of compound 1.

4' (methylene-nonanoyloxy)-3-phenyl propanol (1). It is a new natural product to the best of our knowledge.

Alocasia indica Schott.⁴ (local name : Mankachu; Araceae) is cultivated in North-East region and West Bengal of India for use as delicious vegetable as well as for use of the tuber/rhizome in treatment of piles, constipation and anasarca in traditional medicine. Earlier investigation on the tubers of the plant reported⁵, the presence of free amino acids, organic acids and sugars. We reinvestigated the tubers and report here the isolation and characterization of five known sterols.

Air dried fresh tubers (2 kg) (collected from Agartala in January 2006) were extracted with MeOH at room temperature by percolation process. The concentrated MeOH extract was fractionated in benzene, chloroform and *n*-butanol. The CHCl₃ soluble fraction on silica gel column chromatography afforded a colourless crystalline solid, m.p. 132–134 °C (MeOH-CHCl₃). It showed positive Liebermann-Burchard test for steroids and IR absorption in KBr at 3433, 2960, 2937, 2889, 2866, 1640,

1464, 1380, 1053, 1022, 970, 958, 800 cm⁻¹. The 400 MHz ¹H NMR spectrum in CDCl₃ shows tertiary methyl signals at δ 0.63, 0.68, 0.69, 0.94, 1.01, 1.02 and 1.04 a couple of secondary methyl signals in the region δ 0.81–0.92, two carbiny methine protons at δ 3.52 and 3.63 and six olefinic protons at δ 5.02 (1H, dd, *J* 14.5 and 8.5 Hz), 5.15 (1H, dd, *J* 14.5 and 8.5 Hz), 5.18 (1H, dd, *J* 14.5 and 6.5 Hz), 5.34 (1H, m), 5.38 (1H, m) and 5.56 (1H, dd, *J* 4.5 and 2.0 Hz) suggesting it to be a mixture of Δ^5 , $\Delta^{5,22}$ and $\Delta^{5,7,22}$ sterols⁶. The 100 MHz ¹³C NMR spectrum in CDCl₃ recorded methyl signals at δ 11.85, 11.97, 12.04, 12.24, 15.36, 16.27, 17.59 and 18.25 and olefinic carbon signals at δ 121.70 and 140.75 (Δ^5 carbons), 129.27 and 138.31 (Δ^{22} carbons), 119.58 and 139.78 (Δ^5 carbons for ergosterol), 116.29 and 141.3 (Δ^7 carbons for ergosterol), and 131.97 and 135.56 (Δ^{22} carbons for ergosterol), and carbiny carbons at δ 70.45 and 71.80 supporting it to be a mixture of sterols⁷ also. EI-MS of the solid recorded mass peaks at *m/z* 414, 412, 400, 399, 397, 396, 273, 271, 255, 213 suggesting it to be a mixture of sterols. On GC analysis on a glass col-

umn packed with OV-17 using N₂ gas, isothermal column temperature of 240 °C and injector and FID temperatures at 270 °C, the solid showed five peaks corresponding to campesterol (RR_t : 1.31, content : 10.2%), ergosterol (RR_t : 1.35, 21.3%), stigmasterol (RR_t : 1.43, 17.3%), β -sitosterol (RR_t : 1.63, 31.0%) and clionasterol (RR_t : 1.65, 20.2%). All these sterols were identified by comparison of RR_t (relative to cholesterol) (R_t : 1.00) and Co-GC with those of standard compounds⁸ under identical conditions. This report of phytosterols is for the first time from this plant.

Premna latifolia Roxb. (local name : Gohara; Verbenaceae) a medium sized tree is cultivated as ornamental tree in different regions of India. Its leaves are spasmolytic and are applied externally in dropsy⁹. Earlier investigation on the leaves reported the isolation of furanoid, premnalatin and flavone glycosides¹⁰. We reinvestigated the leaves of this plant and isolated β -sitosterol and stigmasterol so far.

Air-dried fresh leaves of *Premna latifolia* (2.5 kg) (collected from the M.B.B. College campus area, Agartala on July 2006) were extracted with MeOH at room temperature by percolation process. The concentrated MeOH extract was fractionated into benzene, chloroform and *n*-butanol soluble fractions. The benzene fraction on silica gel column chromatography afforded a colourless needle shaped shiny crystals, m.p. 151–153 °C. It showed positive Liebermann-Burchard test for steroids. Its IR spectrum in KBr showed bands for hydroxyl (3410 cm⁻¹) and olefinic double bonds (1640 and 970 cm⁻¹) functions. The 400 MHz ¹H NMR spectrum in CDCl₃ recorded signals for four tertiary methyls at δ 0.68, 0.69, 1.01, and 1.03, three olefinic protons at δ 5.01 (1H dd, *J* 12.5 and 8.0 Hz), 5.15 (1H dd, *J* 12.5 and 9.5 Hz) and 5.35 (1H, m), one carbinyl methine proton at δ 3.52 (1H, m) and secondary methyl signals in the region δ 0.79–0.92 suggesting it was to be a mixture C₂₉ Δ^5 -sterols. The EI-MS of the solid showed mass ion peaks at *m/z* 414 (base peak), 412, 399, 397, 396, 381, 329, 303, 273, 271, 255, 231 and 213 supporting it to be a mixture of sterols. The solid on GC analysis on a glass column packed with OV-17 using N₂ gas, isothermal column temperature of 240 °C and injector and FID temperatures at 270 °C showed two peaks corresponding to stigmasterol (RR_t : 1.43, 64.8%) and β -sitosterol (RR_t : 1.63, 35.2%) relative to cholesterol of R_t, 1.00. Both these sterols were

identified by Co-GC with standard samples under identical conditions.

Experimental

All organic solvents were distilled before use. Electronic spectra were recorded on a Spectronic 21 spectrometer, IR spectra on a Perkin-Elmer 781 spectrophotometer, ¹H and ¹³C NMR spectra on a Varian XL 400 spectrometer using TMS as internal reference. GC was carried out on Pye Unicam 104 gas chromatograph and EI-MS and FAB-MS on a JEOL JMS-AX505H, JEOL JMS-700 Mstaion or JEOL X102/DA-6000 mass spectrometer. Extracts were column chromatographed on silica gel (Qualigen, 60–120 mesh) and thin layer chromatographed on silica gel G (Merck).

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