A New Acylated Isoflavone Glucoside from *Pterocarpus santalinus*

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Phytochemical investigation on the constituents of heartwood of *Pterocarpus santalinus* resulted in the isolation of a new acylated isoflavone glucoside. The structure of the new compound was elucidated on the basis of spectral studies as 4′,5-dihydroxy-7-*O*-methyl isoflavone 3′-*O*-β-D-(3″-*E*-cinnamoyl)glucoside.

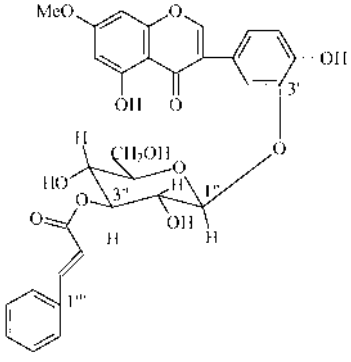
Key words *Pterocarpus santalinus*; Fabaceae; acylated isoflavone glucoside

The genus *Pterocarpus* is known to be a rich source of flavonoids and related phenolic compounds. Extracts of a few species of this genus are known for their hypoglycemic[2] and hypolipidemic activities.[3] The extracts of *Pterocarpus santalinus* L. (Fabaceae) have been used medicinally in the treatment of inflammations, mental aberrations and ulcers, while an infusion of the wood has long been regarded as useful in the treatment of diabetes.[4] As a part of our ongoing phytochemical investigation on *P. santalinus*[5—7] we report herein the isolation and characterization of a new acylated isoflavone glucoside (**1**) from the heartwood of *P. santalinus.*

Compound **1** was isolated as yellow amorphous powder whose molecular formula $C_{31}H_{28}O_{12}$ was determined by FAB-MS (*m*/*z* 615 $[M+Na]^+$). It gave positive visualization with ferric chloride and Molish reagent indicating that **1** is a glycoside. The IR spectrum suggested the hydroxyl group (br 3410 cm^{-1}), carbonyl groups (1690 and 1630 cm^{-1}) and ether function (1060 cm^{-1}). The broad band decoupled ^{13}C-NMR spectrum showed 29 signals corresponding to all 31 carbons (two carbons have the same chemical shift) of the molecule. Multiplicities of carbon signals were determined by DEPT spectrum: 11 quaternary carbons which includes two carbonyl carbons (181.9 and 165.2 ppm), 18 CH carbons, one CH_2 and CH_3 carbon. The ^1H-NMR spectrum of **1** exhibited a flavonoid pattern and showed a signal at δ 8.14 (1H, s) typical of a proton of C-2 of an isoflavone skeleton.[8,9] This was supported by UV spectrum, which exhibited absorptions at 265 nm and 322 (sh) nm[10,11] and this was confirmed by ^{13}C-NMR resonances at 154.8, 124.5 and 181.9 ppm, respectively for 2, 3 and 4 carbons.[12] Two *meta* coupled doublets at δ 6.54 and 6.38 (J=2.0 Hz each 1H) represented H-8 and H-6, respectively. The downfield signal at δ 12.48, exchangeable with D_2O, indicated the presence of free hydroxyl at C-5, which is in chelation with carbonyl function.[8] The signals at δ 6.96 (d, J=2.0 Hz, 1H), 6.72 (d, J=8.0 Hz, 1H) and 7.10 (dd, J=8.0 and 2.0 Hz, 1H) established the presence of three aromatic protons in ring B. The signal at δ 3.84, integrating for 3 protons, indicated the presence of a methoxy group and was assigned to C-7 on the basis of HMBC correlations (Fig. 1).

The ^1H-NMR resonances at δ 3.8—3.5 and signals in the ^{13}C-NMR spectrum just below δ 70 and a signal at 61.30 ppm showed by DEPT to represent CH_2 group, indicated the presence of sugar moiety. The signal for an anomeric proton at δ 5.35 (d, J=7.9 Hz) suggested β-configuration for sugar.[8] The other signals belonging to *trans*-cinnamoyl moiety at δ 6.65 (d, J=16.0 Hz, 1H), 7.67 (d, J=16.0 Hz, 1H), 7.40 (m, 3H) and 7.71 (m, 2H) were observed. A loss of 130 and 162 mass units from the molecular ion in the FAB-MS indicated the presence of cinnamic acid and glucose moieties in **1**. This was confirmed by the formation of *trans*-cinnamic acid and D-glucose upon hydrolysis. Alkaline hydrolysis of **1** gave *trans*-cinnamic acid and 4′,5-dihydroxy-7-*O*-methyl isoflavone 3′-*O*-β-D-glucoside,[7] indicating that the cinnamoyl moiety was attached to the glucosyl residue. The cross peak between H-3″ (δ 5.10) of the glucose and the carbonyl ester carbon (165.2 ppm) of the cinnamoyl residue (HMBC spectrum, Fig. 1), confirmed the attachment of cinnamoyl moiety at C-3″ hydroxyl of glucose.

The (proton carbon chemical shift) correlations for all the carbons directly bonded to protons were established by HETCOR experiment and ^1H–^1H COSY and HMBC (Fig. 1) confirmed the assignment of the proton and carbon frequencies. Thus, compound **1** was characterized as 4′,5-dihydroxy-7-*O*-



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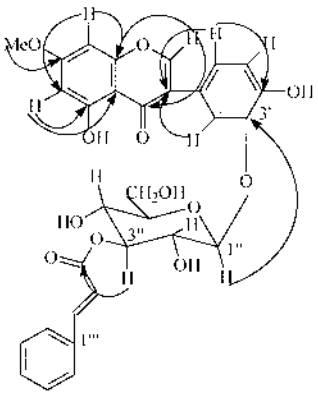

Fig. 1. HMBC Correlations of **1**

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methyl isoflavone 3′-*O*-*β*-D-[3″-*E*-cinnamoyl]glucoside

Experimental

General Procedures Melting points are uncorrected. UV spectra were taken in MeOH on a Beckman 25 spectrophotometer. IR spectra were run in KBr. FAB-MS was obtained using a glycerol matrix on a VG Micro Mass Zab-HF mass spectrometer. ^{1}H- and ^{13}C-NMR spectra were measured on a Brucker AC 300 spectrometer at 300 and 75 MHz, respectively. Samples were run in DMSO-d_6 TMS as internal standard.

Plant Material The heartwood of *P. santalinus* was collected from the Tirumala Hills, Tirupati and a voucher specimen has been deposited in the Herbarium of the Botany Department, Sri Venkateswara University, Tirupati.

Extraction and Isolation The air-dried and powdered heartwood (600 g) of *P. santalinus* was defatted and exhaustively extracted with methanol at room temperature. The MeOH extract was concentrated under reduced pressure and the resulting residue was suspended in water and extracted with CH_2Cl_2. The CH_2Cl_2 soluble portion was subjected to column chromatography over silica gel ,eluted with varying portions of a mixture of $CHCl_3$–EtOAc and the fractions were combined on the basis of TLC analysis leading to four series. Compound **1** (35 mg) was obtained from series C, eluted with $CHCl_3$–EtOAc (7 : 3) upon removal of solvent followed by recrystallization.

4′,5-Dihydroxy 7-*O*-methylisoflavone 3′-*O*-*β*-D-(3″-*E*-cinnamoyl) Glucoside (**1**): Light yellow amorphous powder from MeOH, UV (MeOH); λ_{max} (log ε) 265 (4.43), 322 (3.73) nm; IR (KBr) ν_{max} cm^{-1} 3410, 3300, 1690, 1630, 1060; ^{1}H-NMR (DMSO-d_6) δ 12.48 (1H, s, OH-5), 8.14 (1H, s, H-2), 7.71 (2H, m, H-2‴, 6‴), 7.67 (1H, d, J=16.0 Hz, H-7‴), 7.40 (3H, m, H-3‴, 4‴, 5‴), 7.10 (1H, dd, J=8.0, 2.0 Hz, H-6′), 6.96 (1H, d, J=2.0 Hz, H-2′), 6.72 (1H, d, J=8.0 Hz, H-5′), 6.65 (1H, d, J=16.0 Hz, H-8‴), 6.54 (1H, d, J=2.0 Hz, H-8), 6.38 (1H, d, J=2.0 Hz, H-6), 5.35 (1H, d, J=7.9, H-1″), 5.10 (1H, dd, J=9.2, 9.2 Hz, H-3″), 3.84 (3H, s, OMe-7), 3.80 (1H, m, H-6″a), 3.5 (4H, m, H-2″, 4″, 5″, 6″b); ^{13}C-NMR 181.9 (C-4), 165.2 (C-9‴), 164.6 (C-7), 159.7 (C-9), 157.3 (C-5), 154.8 (C-2), 151.9 (C-3′), 144.8 (C-7‴), 139.8 (C-4′), 133.9 (C-1‴), 130.1 (C-4‴), 129.2 (C-3‴, 5‴), 128.9 (C-6′), 128.0 (C-2‴, 6‴), 126.2 (C-1′), 124.5 (C-3), 119.2 (C-8‴), 115.8 (C-5′), 114.9 (C-2′), 112.2 (C-10), 101.9 (C-1″), 98.4 (C-6), 95.7 (C-8), 78.8 (C-3″), 77.2 (C-5″), 73.9 (C-2″), 71.0 (C-4″), 61.3 (C-6″), 58.4 (OMe); FAB-MS (m/z) 615 $[M+Na]^+$, 462, $[M+H-cinnamoyl]^+$ (5), 301 $[M+H-cinnamoyl\ glucosyl]^+$ (51).

Acid hydrolysis of 1 Compound **1** (15 mg) on acid hydrolysis with 5 N HCl at 90 °C for 3 h after usual workup gave 3′,4′,5-trihydroxy-7-*O*-methyl isoflavone,[6] cinnamic acid and glucose (identified by co-paper chromatography in BAW).

Alkaline hydrolysis of 1 Ten mg of compound **1** was refluxed in 1% KOH for 2 h. The reaction mixture was processed further in the usual way which yielded 4′,5-dihydroxy-7-*O*-methyl isoflavone-3′-*O*-*β*-D-glucoside and cinnamic acid.

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References and Notes

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