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 hypoglycemic²⁾ 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 ($J=2.0\text{ Hz}$ each 1H) represented H-8 and H-6, respectively. The downfield signal at δ 12.48, exchange-

able 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, $J=2.0\text{ Hz}$, 1H), 6.72 (d, $J=8.0\text{ Hz}$, 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, $J=7.9\text{ Hz}$) suggested β -configuration for sugar.⁸⁾ The other signals belonging to *trans*-cinnamoyl moiety at δ 6.65 (d, $J=16.0\text{ Hz}$, 1H), 7.67 (d, $J=16.0\text{ Hz}$, 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-

1

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. ^1H - and ^{13}C -NMR spectra were measured on a Bruker 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; ^1H -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 $[\text{M}+\text{Na}]^+$, 462, $[\text{M}+\text{H}-\text{cinnamoyl}]^+$ (5), 301 $[\text{M}+\text{H}-\text{cinnamoyl glucosyl}]^+$ (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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