Chemistry Research Journal, 2022, 7(2):43-45

Available online www.chemrj.org

Research Article

ISSN: 2455-8990
CODEN(USA): CRJHA5

Isoflavone from *Sida rhombifolia*

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Abstract A novel isoflavone has been isolated from the chloroform fraction of the root of the *Sida rhombifolia*. The structure of the newly isolated compound has been established as 6-hydroxy-7,3',4'-trimethoxy isoflavone based on the spectral (UV, IR, ^{1}H, ^{13}C NMR and mass) data.

Keywords Sida rhombifolia; Malvaceae; Isoflavone; 6-hydroxy-7,3',4'-trimethoxy isoflavone

Introduction

Metabolites are the transitional products of metabolism and restricted to small molecules. The plant produces a lot of chemicals that can be categorised into primary metabolites and secondary metabolites. Primary metabolites are necessary for cell function and they are present everywhere. Secondary metabolites are useful for human because of their diverse application [1]. *Sida rhombifolia* (family – Malvaceae) is a secondary metabolite found in marshy [2] place throught India. In Ayurveda, it is known as "Mahabala" and is use to cure fever, heart diseases, burning sensation, piles, urinary disorder and all kinds of inflamation.

The plant growth leads to the production of different chemical compound are called phytochemicals, phenols, terpenoids, flavonoid, steroid and phytosteroid [3].

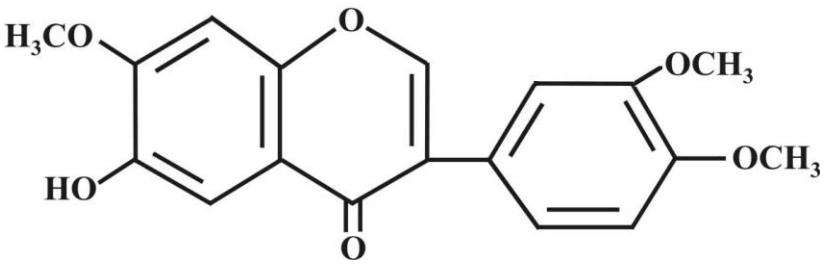
It is well known that these compound have medicinal application. The alkaloid have been recorded as potent poisons, for instance many alkaloid from medicinal plants indicate biological activity such as antispasmodic and pharmacological impact, antimalerial, antiinflamatory, antimicrobial [4].

The isolated steroids are known to have cardiac impact, and to have insecticidal and antibacterial properties [5].

Due to their biological activity they are commonly used in drugs. As a result of research, tannins have antibacterial action, a antitumor and antiviral action [6].

Other chemical constituents called respiratory glycone were handled for congestive heart failure and cardiac arrhythm [7].

Earlier co-workers have announce another various alkaloid from this plant are, ephedrine, ecdysterone etc. the present paper report the isolation and structure elucidation of novel isoflavone from *S. rhombifolia*.



6-hydroxy-7,3',4'-trimethoxy isoflavone

Result and Discussion

The $[M]^+$ of compound, m/z 328, corresponding to $C_{18}H_{16}O_6$ was determined by FABMS. UV λ_{max}^{MeOH} 269 nm, 296 sh, 348 nm and IR data υ_{max}^{KBr} 1655 cm^{-1} (= C = O) coupled with a low field singlet at δ 8.46 (H-2) in the NMR spectrum are suggestive of the presence of an isoflavone system [8].

The compound on acetylation with acetic anhydride and fused with sodium acetate there by accommodate the presence of only one hydroxyl group in the compound.

^{1}H-NMR spectrum of the compound showed the signal at δ 3.68 (3H, s), 3.78 (3H, s) and 3.92 (3H, s) relative to three methoxyl groups. This was also supported by ^{13}C NMR spectrum. Which showed the signals at δ 57.00, 57.82 and 58.02 respectively. On methylation, compound gave a methyl ether which analyse for four methoxyl group. Since three methoxyl group were already present in the compound, one phenolic hydroxyl group present in the compound must have got methylated. Compound gave negative test with Zirconium oxy-chloride [9] and vanillin-hydroxyl acid reagent [10] this is the suggestive of the absence of a free hydroxyl group at C-5 of ring A and absence of 1, 3-dihydoxyl system.

Its ^{1}HNMR spectrum revealed an ABX pattern with two double doublet at δ 7.02 (2H, J = 8.5, 2 Hz) attributable to H-5' and H-2', and 7.36 (1H, J = 8.5, 2 Hz), attributable to H-6. The ^{1}HNMR spectra of the compound contained signals for the para coupled protons at δ 6.80 and 6.70 (1H, each, s), due to H-5 and H-8 respectively [11].

On the addition of NaOme with MeOH showed bathochromic shift of 26 and 20 nm in UV absorption bands I and II with reduced intensity, indicating the substitution at C–3' and C–4', where as the bathocrhomic shift of 72 nm with $AlCl_3$-HCl specify the hydroxyl substitution at C–5 [12].

The presence of a peak at m/e 162 for the retro-Diels-Alder fragment[13] confirms the presence of a B ring with two methoxyl substitutents.

The singlet at δ 12.35 (1H, s, OH) was ascribe to the proton of hydroxyl group at C – 6. Thus the position left for the substitution of three methoxyl groups were C-3', C-4' and C-7 respectively.

The molecular ion peaks at m/z 328 relative to $C_{18}H_{16}O_6$ and the other fragment at 327 (M-1) are typical of isoflavone moiety. Thus the structure of compound was thus explained as 6-hydroxy-7,3',4'-trimethoxyl isoflavone.

Plant Material

The root of *S. rhombifolia* were collected from NRIPT, Telearganj, Prayagraj and recognised by Dr. B.K. Shukla, Taxonomist, Botanical Survey of India (BSI) Prayagraj. It is widely spread through India and Nepal, specially in moist region arise to an altitude of 1800 cm in the Himalayas.

Extraction and Isolation

The shed dried and well ground roots (5 kg) were refluxed with (95%) ethanol and the extract was concentrated under reduced pressure through rotatory evaporator. It was partition between DCM and chloroform. The chloroform soluble fraction (60 g) was choromatographed over colum of silicagel and eluted with binary solution of Acetone: $CHCl_3$ in sequence of increasing polarity. The compound was isolated (hexane: chloroform) (7 : 3 v/v) fraction as pale yellow solid (20 mg). 6-hydroxy-7,3',4'-trimethoxyl isoflavone M.P. 215ºC.

Experimental Section

M.P. was measured in an open capillary tube and is incorrected. UV was recorded on a Beckmans – DK2 spectrophotometer. IR was recorded in KBr on a Perkin Elmer Spectrometer. ^{1}H-NMR of compound was recorded at 300 MHz. ^{13}C-NMR spectra at 100 MHz in $CdCl_3$ using TMS as an internal reference on a JEOLJNM – A500 spectrometer. Mass Spectra were recorded on a JEOLINSD 300 mass spectrometer.

UV λ_{max}^{MeOH}	:	269, 296 sh, 348 nm
IR υ_{max}^{KBr}	:	3440, 2930, 2810, 1640, 1250, 1185 cm^{-1}.
^{1}H-NMR	:	(300 MHz, $CdCl_3$) [δ 3.92 (3H s 4'-OCH_3), 3.98 (3H, s, 3'-OCH_3) 3.96 (3 H, s, 7-OCH_3); 7.02 (2H, dd, J =8.5 Hz 2.5 Hz, H-2', H-5'), 6.51 (1H, s, H-8), 6.74 (1H, s, H-5), 7.36 (1H, dd, J = 8.5 Hz, 2.5 Hz, H-6'), 7.56 (1H, s, H-2), 12.12 (1H, s, ─OH)-6].
^{13}C-NMR	:	(100 MHz, $CdCl_3$) [δ 154.2 (C-2, d), 165.0 (C-3, s), 180.2 (C-4, s), 129.2 (C-5, d), 143.8 (C-6, s), 147.2 (C-7, s), 130.2 (C-8, d), 140.2 (C-9, s), 100.31 (C-10, s), 120.3 (C-1', s), 147.2 (C-2', d), 106.00 (C-3', s), 144.47 (C-4', s), 109.37 (C-5', d), 114.91 (C-6', d), 57.00 (q, -OCH_3), 57.82 (q, –OCH_3), 58.02 (q, –OCH_3).
Mass spectra	:	M^+ 328, m/e 327, 313, 285, 166, 164, 162

Acknowledgement

The author thanks to UGC Delhi India for providing Teacher Research fellowship and also the author wish to express their gratitude to Dr. B.K. Shukla, Taxonomist, Botanical Survey of India (BSI) Prayagraj (U.P.) India for identification of plant.

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