

## STUDIES ON THE CONSTITUENTS OF *HIBISCUS ROSA-SINENSIS*

M Amzad Hossain\* and SA Tarafdar

Chemistry Division, Atomic Energy Centre, Ramna, Dhaka-1000, Bangladesh

(Received 1 August 2000; accepted 3 October 2002)

A new flavone derivative, furanoflavone (**1**) was isolated along with ovalichalcone (**2**) from the leaves and stems of *Hibiscus rosa-sinensis* (Malvaceae). The structure of furanoflavone (**1**) was identified as 5-methoxy-3'-methyl-3',4'-methylenedioxyfurano[2'', 3'' : 7,8]flavone by spectroscopic and chemical analysis.

**Key words:** *Hibiscus rosa-sinensis*, Isolation, Characterization, Chalcone, Flavone.

### Introduction

The flavonoid constituents of the leaves and stems of *Hibiscus rosa-sinensis* continue to attract attention not only for their remarkable taste properties (Guadagni *et al* 1973 & 1976; Dubois *et al* 1982) but also for their therapeutic and pharmacological activities. They have been shown to possess biological activities such as anti-carcinogenic effect (Kato *et al* 1983; Nishino *et al* 1983; Harborne *et al* 1988; Venna *et al* 1988; Weitt *et al* 1990; Deschner *et al* 1991), anti-inflammatory properties (Busse *et al* 1984; Laudolfi *et al* 1984) and inhibitory activities against histamine release (Middleton *et al* 1984; Bronner *et al* 1985). The ethyl acetate extract of the leaves and stems of the plant yielded a novel furanoflavone (**1**) while the BuOH extract of the leaves and stems gave the ovalichalcone (**2**). The structures were identified on the basis of UV, IR, <sup>1</sup>H-NMR, mass and gas liquid chromatographic data. Compound **1** identified as 5-methoxy-3-methyl-3',4'-methylenedioxyfurano [2'', 3'' : 7, 8] flavone is a novel natural product.

### Experimental

Melting points were determined on an electrothermal melting point apparatus (Gallenkamp) which are uncorrected. IR spectra were recorded (KBr discs) on a Pye-Unicam SP3-300 IR spectrophotometer ( $\nu_{\max}$  in  $\text{cm}^{-1}$ ), <sup>1</sup>H-NMR spectra were recorded on a Perkin-Elmer R-32 (90 MHz) instrument in CDCl<sub>3</sub> with TMS as an internal standard (The chemical shifts are given in  $\delta$  values). UV spectra were recorded on a Shimadzu UV -1201 spectrophotometer "Ultraspek" in methanol. TLC were performed on silica gel plates. Mass spectra were recorded on VG 7070 E analytical mass spectrometer. A Varian gas chromatograph, Model 3300, equipped with Flame Ionization Detector (FID) was used for the GLC

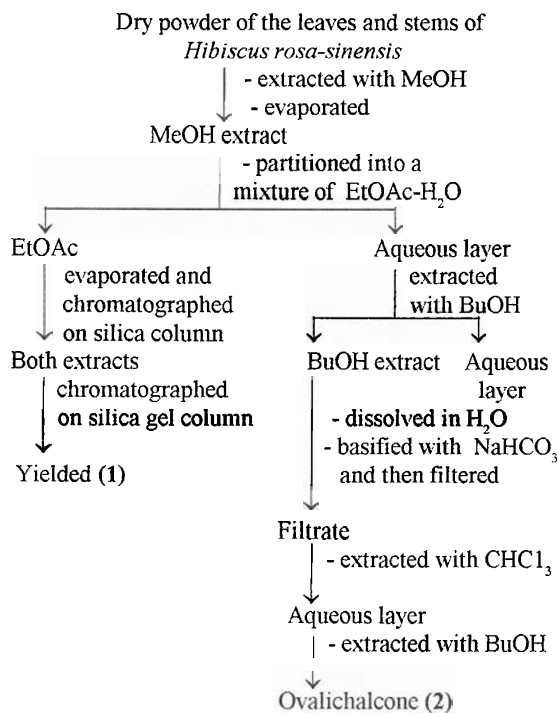
analysis. Samples were recorded on an Integrator, Varian model 4290. Hydrogen generator, model 7526, Canberra Industries Inc. was used for hydrogen production. A megabore column, SF 254 phase, 30 m long with 0.54 mm id., 1.2  $\mu$  film thickness was used for the chromatographic separation. Operation conditions:- Column temperature 90°C, detector temperature 300°C, injector temperature 250°C, carrier gas (N<sub>2</sub>), flow rate 18 $\pm$ 1 ml min<sup>-1</sup>.

**Plant material:** The leaves and stems of *Hibiscus rosa-sinensis*, Malvaceae were collected from a 10 meter high tree in the vicinity of the Bangladesh Atomic Energy Commission building in Dhaka (Bangladesh) in March 1998.

**Isolation of Furanoflavone.** The dried leaves and stems of *Hibiscus rosa-sinensis* (1.4kg), were powdered and separately extracted four times with MeOH under refluxing conditions for 18 hr. The extracts were concentrated and partitioned into a mixture of EtOAc and H<sub>2</sub>O. The water layer was washed with EtOAc and extracted with BuOH. In this experiment, EtOAc-extracts (5 g from the leaves and 4.2 g from the stems) and BuOH-extracts (3.5 g from the leaves and 2.9 g from the stems) were obtained. The EtOAc extracts from the leaves and stems were combined, due to the identical TLC behaviour. This extract was chromatographed on silica gel column and the following eluates were obtained: hexane (1.8 g), hexane-benzene (4:1) (1.6 g), hexane-benzene (1:1) (0.8g) and benzene (1.2 g), CH<sub>2</sub>Cl<sub>2</sub> (3.1 g) ether (1.2 g) and EtOAc (0.2 g). The benzene and CH<sub>2</sub>Cl<sub>2</sub> eluates were combined and chromatographed repeatedly on a silica gel column, and the eluates with benzene: EtOAc (10: 1) were crystallized from petroleum ether (b.p. 40-80°C) to obtain yellow prisms of compound **1**, m.p. 161°C, yield (49 mg, yield 11.4%).

**5-Methoxy-3-methyl-3',4'-methylenedioxyfurano (2'',3'':7,8) flavone (1).** Prism from petroleum ether 161°C; (M<sup>+</sup>, m/z 350); UV: 232, 250, 364 nm; IR: (KBr, disc): 2840,

\*Author for correspondence  
e-mail:aced@ citechco.net

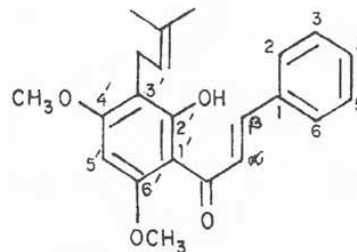
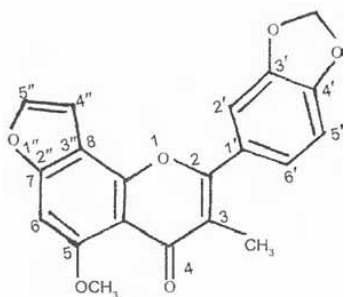


Scheme 1. Isolation of furanoflavone.

2210, 1642, 1605, 1595, 1475, 1363, 840, 740, 710 cm<sup>-1</sup>; <sup>1</sup>H-NMR: δ 2.14 (s, 3H, -CH<sub>3</sub>), 3.98 (s, 3H, -OCH<sub>3</sub>), 6.01 (s, 2H, -OCH<sub>2</sub>O-), 6.78 (s, 1H, H-6), 6.94 (s, 1H, H-2'), 7.12 (d, 1H, J=2.0 Hz, H-4''), 7.34 (d, 2H, J=9.0 Hz, H-5' and H-6'), 7.81 (d, 1H, J=2Hz, H-5''). (Found: C, 68.57; H, 4.00. C<sub>20</sub>H<sub>14</sub>O<sub>6</sub> requires: C, 68.66, H, 4.32%). It was identified as 5-methoxy-3-methyl-3',4'-methylenedioxyfurano (2'',3'': 7,8) flavone(1).

## Results and Discussion

The dried leaves and stems of *Hibiscus rosa-sinensis* were separately extracted with boiling methanol and the extracts were treated as shown on Scheme-1. Furanoflavone (1) was obtained in the part soluble in ethyl acetate from the leaves



and stems as yellow prism. Its molecular formula was obtained by the elemental analysis and the mass spectrum ( $M^+$ ,  $m/z$  350). The ethanolic solution of 1 showed a negative ferric chloride test indicating the absence of free -OH group in the structure.

The IR spectrum of 1 showed the presence of carbonyl group at 1642 cm<sup>-1</sup> characteristic of flavone (Mukerjee *et al* 1969) carbonyl and the absorptions peaks at 1605 and 1595 cm<sup>-1</sup> indicated the presence of unsymmetric ethylenic double bond and the aromatic rings, respectively (Hossain *et al* 1998) The <sup>1</sup>H-NMR spectrum of the furanoflavone (1) showed three sharp singlets at δ 2.14 (3H), δ 3.98 (3H) and δ 6.02 (2H) indicating the presence of one methyl, one methoxy and one methylenedioxy groups. Two doublets at δ 7.12 and δ 7.81 indicated the presence of vinylic H-4'' and H-5'' protons on the furano ring (Hossain 1999). The remaining aromatic protons have usual chemical shift values. The known compound ovalichalcone (Islam *et al* 1993) was identified by comparison of its spectral properties with those reported in the literatures (Islam *et al* 1993). This compound was earlier isolated from *Milletia ovalifolia*. In the GLC chromatogram, the compound (2) had the retention time 9.56 min. which is same as the retention time of the standard ovalichalcone (2). The elemental analysis also showed complete agreement (within ± 0.4%).

## Acknowledgement

Authors are grateful to Dr. Giasuddin Ahmed, Department of Chemistry, Dhaka University, Bangladesh for <sup>1</sup>H-NMR, IR and UV spectroscopy. Authors are also grateful to Dr. J. Palige, Department Number-6, Institute of Nuclear Chemistry and Technology, Warsaw, Poland for the mass and elemental analysis. Thanks are due to Mr. Md. Zakaria Mondol for the collection of plant materials.

## References

- Bronner C, Landry Y 1985 Synthesis of Desmodol. *Agents Actions* 16 147.
- Busse W W, Kopp D E, Middleton E 1984 Synthesis of anti-invasive activity of novel 1,3-diarylpro-penones. *J Al-*

- lergy Clin Immunol* **73** 801.
- Deschner E E, Reperto J, Wong G, Newmark H L 1991 New type chalcone from licorice root. *Carcinogenesis* **7** 1193.
- Dubois G E, Stephenson R A 1982 Synthesis of furanoflavone. *J Agric Food Chem* **30** 676.
- Guadagni D, Maier V P, Turnbaugh T G 1973 Flavonoids of *Chromolaena odorata*. *J Sci Food Agric* **24** 1277.
- Guadagni D, Maier V P, Turnbaugh T G 1976 Flavonoids of *Chromolaena odorata*. *J Sci Food Sci* **41** 681.
- Harborne J B 1988 *Plant Flavonoids in Biology and Medicine II*. Alam R Liss, New York pp 235.
- Hossain M A, Islam A 1998 New synthesis of triangularin. *Bangladesh J Sci Ind Res* **33** 94.
- Hossain M A 1999 New synthesis of pongaglabrone, a naturally occurring furanoflavone: *J Bang Chem Soc* **23** 9.
- Islam A, Hossain M A, Synthesis of some derivatives of ovalichalcone. *J Bang Chem Soc* **6** 199.
- Kato R, Nakadate T, Yamaiuoto S, Sugiyama T 1983 Synthesis of flavonoids. *Carcinogenesis* **4** 1301.
- Laudolfi R, Mower R L, Steiner M 1984 Isolation of chalcone from bioactive plants. *Biochem. Pharmacol* **33** 1525.
- Middleton E, Drzowieck G 1984 Chemotaxonomy of the genus *Pleiotaxis* *Curr. Biochem. Pharmaco* **21** 3333.
- Mukerjee S K, Sarker S C, Seshadri T R 1969 Synthesis of ovalichalcone from the seeds of *Milletia ovalifolia*. *Tetrahedron* **25** 1063.
- Nishino H, Nagao M, Fujiki H, Sugiyama T 1983 Synthesis of antifungal. *Cancer Lett* **21** 1.
- Venna A K, Johnson J A, Gould M N, Tanner M A, 1988 Prenylated flavonoids from seeds of *Calopogonium mucunoids*. *Cancer Res* **48** 5754.
- Weitt T, Bresnick E, Brit D F 1990 *The Biochemistry of Plants*. Academic Press, New York Vol. 7 pp-499.