

## Isolation of 3,3'-Dihydroxychalcone from *Primula macrophylla*

Viqar Uddin Ahmad, Mohammad Ghani Shah,  
Mushtaq Noorwala, and Faryal Vali Mohammad

*J. Nat. Prod.*, **1992**, 55 (7), 956-958 • DOI:  
10.1021/np50085a018 • Publication Date (Web): 01 July 2004

Downloaded from <http://pubs.acs.org> on April 4, 2009

### More About This Article

---

The permalink <http://dx.doi.org/10.1021/np50085a018> provides access to:

- Links to articles and content related to this article
- Copyright permission to reproduce figures and/or text from this article



**ACS Publications**  
High quality. High impact.

Journal of Natural Products is published by the American  
Chemical Society, 1155 Sixteenth Street N.W., Washington,  
DC 20036

ISOLATION OF 3,3'-DIHYDROXYCHALCONE FROM  
*PRIMULA MACROPHYLLA*VIQAR UDDIN AHMAD,\* MOHAMMAD GHANI SHAH, MUSHTAQ NOORWALA,  
and FARYAL VALI MOHAMMAD

H.E.J. Research Institute of Chemistry, University of Karachi, Karachi 75270, Pakistan

ABSTRACT.—A new dihydroxy chalcone has been isolated from the EtOAc-soluble portion of the whole plant of *Primula macrophylla*, and its structure was determined by spectroscopic methods, including 2D nmr, as 3,3'-dihydroxychalcone [1]. 3'-Methoxyflavone and  $\beta$ -sitos-terol have also been isolated for the first time from this plant.

*Primula macrophylla* D. Don (Primulaceae), syn. *Primula stuartii* Wall., is a perennial herb found in the Karakorum and Kuram valleys in Pakistan at an elevation of 3600–4900 m (1). The plant (especially the farina on the leaves) is used locally in Pakistan as well as in Afghanistan for the treatment of eye diseases. A literature survey showed that only two chalcones have been reported from this family (2,3). This species has not been investigated chemically so far. In view of its medicinal importance, a phytochemical investigation was carried out which resulted in the isolation and structure elucidation of 3,3'-dihydroxychalcone [1]. This compound was synthesized previously (4) but has not been previously isolated from a natural source.

## RESULTS AND DISCUSSION

3,3'-Dihydroxychalcone [1] [1,3-bis(3-hydroxyphenyl)-2-propen-1-one] was isolated as a yellow crystalline solid, mp 155–156° (dec). Compound 1 gave uv absorption maxima at 252, 311, and 369 nm which are characteristic for chalcones (5). The ir spectrum displayed ab-

sorption bands at 1660  $\text{cm}^{-1}$ , indicating the presence of a conjugated carbonyl group. Other absorptions were observed at 3300 (OH), 1625 (C=C, ar.) and 1145 (C—O—C)  $\text{cm}^{-1}$ . The eims displayed a molecular ion peak at  $m/z$  [M]<sup>+</sup> 240 and prominent fragment ions at  $m/z$  239, 222, 221, 194, 147, 121, 93, 91, 65. The mass fragmentation pattern showed the loss of an H<sub>2</sub>O molecule and direct fission on both sides of the carbonyl group. The hreims of compound 1 displayed the molecular ion peak at  $m/z$  240.0795 (calcd 240.0786), consistent with the molecular formula C<sub>15</sub>H<sub>12</sub>O<sub>3</sub>.

The <sup>1</sup>H-nmr spectrum (Table 1) showed two characteristic broad doublets ( $J = 15.6$  Hz) of chalcone, each for 1H, due to the protons attached to C- $\alpha$  and C- $\beta$ . The upfield values of the protons at  $\delta$  6.89 and 6.98 supported the hydroxyl group at the C-3 and C-3' positions. The <sup>1</sup>H-nmr assignments were confirmed with the help of 2D  $J$ -resolved, COSY-45°, long range COSY, NOESY, and HOHAHA experiments.

The <sup>13</sup>C-nmr assignments were carried out with the help of DEPT, 2D direct <sup>1</sup>H/<sup>13</sup>C chemical shift correlation (hetero-COSY) (Table 1), and HMBC experiments. The typical signals of C- $\beta'$ , C- $\alpha$ , and C- $\beta$  showed that compound 1 was of chalconoid nature (5). The fifteen signals are due to the unsymmetrically substituted pattern of the compound and disproved the positions of the hydroxyl groups at C-4, -4' and C-2, -2'

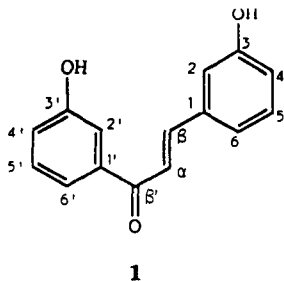


TABLE 1. Nmr Assignments and  $^1\text{H}/^{13}\text{C}$  Direct Correlation (hetero-COSY) of Compound 1.

Position	$^1\text{H}$ nmr (CDCl <sub>3</sub> , 300.13 MHz)			$^{13}\text{C}$ nmr (CDCl <sub>3</sub> , 75.43 MHz)		
	$\delta$	Multiplicity	$J$ (Hz)	$\delta$	DEPT	hetero-COSY
1'	—	—	—	123.10	Quaternary	No coupling
2'	6.89	m	—	117.22	CH	6.89 (H-2')
3'	—	—	—	164.48	Quaternary	No coupling
4'	6.98	m	—	120.13	CH	6.98 (H-4')
5'	7.51	dddd	1.6,7.3,8.1	137.22	CH	7.51 (H-5')
6'	8.05	dd	1.7,8.0	131.23	CH	8.05 (H-6')
$\beta'$	—	—	—	196.10	Quaternary	No coupling
$\alpha$	7.97	d	15.6	121.11	CH	7.97 (H- $\alpha$ )
$\beta$	8.21	d	15.6	142.93	CH	8.21 (H- $\beta$ )
1	—	—	—	121.60	Quaternary	No coupling
2	6.98	m	—	119.10	CH	6.98 (H-2)
3	—	—	—	159.24	Quaternary	No coupling
4	6.89	m	—	120.90	CH	6.89 (H-4)
5	7.26	dddd	1.7,7.3,8.1	133.32	CH	7.26 (H-5)
6	7.69	dddd	0.5,2.1,8.1	130.92	CH	7.69 (H-6)

(6,7). The position of the hydroxyl groups at C-3, -3', and not at C-2, -2', is further evidenced by the low field value of C- $\alpha$  and C- $\beta$  (6,7).

3'-Methoxyflavone, mp 126–127°, is a rare flavone; it was also isolated and was identified by comparison of its spectral data with those reported in the literature (8). The isolation of this flavone has been reported from *Pimelea decora* Domin. (Thymeliaceae) (8).  $\beta$ -Sitosterol, mp 137–138°, was also identified with the help of mass and other spectroscopic data reported in the literature (9–11).

## EXPERIMENTAL

**GENERAL EXPERIMENTAL PROCEDURES.**—Mp's were determined in an H<sub>2</sub>SO<sub>4</sub> bath (capillary) and are uncorrected. The uv spectrum was recorded in MeOH on a Shimadzu UV-240 spectrometer. Ir spectra were recorded in KBr discs on a JASCO IRA-1 ir spectrophotometer.  $^1\text{H}$ -nmr spectra were scanned on a Bruker AM-300 (300.13 MHz) spectrometer using CDCl<sub>3</sub> as solvent and TMS as an internal standard.  $^1\text{H}$  chemical shifts are reported from TMS and coupling constants are in Hz.  $^{13}\text{C}$ -nmr spectra were measured in CDCl<sub>3</sub> at 75.43 MHz with TMS as an internal standard using the Bruker AM-300 spectrometer. The DEPT experiments were carried out with  $\theta = 45^\circ, 90^\circ,$  and  $135^\circ$ ; the quaternary carbons were determined by subtraction of this spectrum from the broad band  $^{13}\text{C}$ -nmr spectrum. Eims were determined on a Finnigan MAT-312 Varian MAT-112 double focusing

mass spectrometer connected to a PDP 11/34 (DEC) computer system. Kieselgel 60 (70–230 mesh) was used for cc. Precoated Kieselgel 60, F<sub>254</sub> cards (thickness 0.25 mm, Riedel de Haën, Art. No. 37360) were used for tlc. The purity of the sample was checked on hptlc plates (E. Merck, Art. No. 5556). The chromatograms were sprayed with 0.1% Ce(SO<sub>4</sub>)<sub>2</sub> in 2 N H<sub>2</sub>SO<sub>4</sub> and heated at 80° for 5 min.

**PLANT MATERIAL.**—The whole plant of *P. macrophylla* was collected from Sora Lasht near the Pakistan Afghanistan border at Chitral (N.W.F.P.) Pakistan in the month of August; identification was kindly carried out by Yasin J. Nasir of the National Herbarium (Stewart Collection) Pakistan Agricultural Research Council, Islamabad, Pakistan. A voucher specimen is available for inspection in the herbarium.

**EXTRACTION AND ISOLATION.**—Dried material (15 kg) of the whole plant was extracted three times with MeOH after percolation for 15 days. The combined MeOH extracts were evaporated under reduced pressure, yielding a greenish syrupy residue which was then partitioned between EtOAc and H<sub>2</sub>O. The EtOAc layer was evaporated to dryness. The crude extract was chromatographed using Si gel 60 (70–230 mesh) (1 kg). Elution with *n*-hexane–EtOAc (96:4) afforded compound 1 (38 mg), a yellow crystalline solid. A spot of this substance on filter paper turned deep orange after exposure to NH<sub>3</sub> vapors. The compound was recrystallized from MeOH: mp 155–156° (dec); uv  $\lambda$  max (MeOH) nm 252, 311, 369; ir  $\nu$  max (KBr) cm<sup>-1</sup> 3300, 1660, 1625, 1460, 1225, 1145, 750;  $^1\text{H}$  nmr and  $^{13}\text{C}$  nmr see Table 1; ireims  $m/z$  (rel. int.) 240 (24), 239 (8), 222 (100), 221 (92), 119 (8), 107 (4),

147 (20), 121 (72), 93 (16), 91 (20), 65 (29); hreims  $m/z$   $[M]^+$  240.0795 (calcd 240.0786) for  $C_{15}H_{12}O_3$ .

3'-Methoxyflavone (40 mg) was isolated by eluting the column with *n*-hexane-EtOAc (98:2); it formed colorless crystals in  $CHCl_3$ , mp 125–126° [lit. (8) mp 129–131°].

$\beta$ -Sitosterol (30 mg) was also obtained by eluting the main column with *n*-hexane-EtOAc (98:2) and was purified by crystallization from  $CHCl_3$ , mp 137–138° [lit. (9) mp 136–137°]. The compound was identified by comparison of its spectroscopic data with those in the literature (10,11).

#### ACKNOWLEDGMENTS

The authors are thankful to the University Grants Commission for financial support. Thanks are also due to Yasin J. Nasir of the National Herbarium (Stewart Collection) Pakistan Agricultural Research Council, Islamabad, Pakistan, and Dr. A.R. Beg and Mr. Saeed of Pakistan Forest Institute Peshawar, Pakistan, for the identification of the plant material.

#### LITERATURE CITED

1. S.I. Ali and E. Nasir, "Flora of West Pakistan," Fakhri Printing Press, Karachi, Pakistan, 1972, p. 539.

2. E. Wollenweber and K. Mann, *Biochem. Physiol. Pflanz.*, **181**, 665 (1986).
3. E. Wollenweber, K. Mann, M. Iinuma, T. Tanaka, and M. Mizuno, *Phytochemistry*, **28**, 295 (1989).
4. Y. Kazutaka, S. Youjiro, and K. Masateru, *Chem Abstr.*, **78**, 97330d (1973).
5. T.J. Mabry, K.R. Markham, and M.B. Thomas, "The Systematic Identification of the Flavonoids," Springer-Verlag, New York, 1970, p. 227.
6. P.K. Agrawal, "Carbon-13 NMR of Flavonoids," Academic Press, London, 1982, pp. 30, 31, 368.
7. K.R. Markham and B. Ternai, *Tetrahedron*, **32**, 2607 (1976).
8. P.W. Freeman, S.T. Murphy, J.E. Nemoirin, and W.C. Taylor, *Aust. J. Chem.*, **34**, 1779 (1981).
9. "Dictionary of Organic Compounds," 5th ed., Chapman and Hall, New York, 1982, Vol. 5, p. 5009.
10. I. Rubinstein, L.J. Goad, A.D.H. Clague, and L.J. Mulheirn, *Phytochemistry*, **15**, 195 (1976).
11. H.L. Holland, P.R.P. Diakow, and G.J. Taylor, *Can. J. Chem.*, **56**, 3121 (1978).

Received 16 August 1991