A New Xanthone from the Fern Cystopterys fragilis

Filippo Imperato

J. Nat. Prod., **1991**, 54 (2), 603-605• DOI:
10.1021/np50074a043 • Publication Date (Web): 01 July 2004

Journal of Natural Products
Vol. 54, No. 2, pp. 603-605, Mar-Apr 1991

A NEW XANTHONE FROM THE FERN *CYSTOPTERYS FRAGILIS*

FILIPPO IMPERATO

Department of Chemical Sciences, University of Catania, I-95125 Catania, Italy

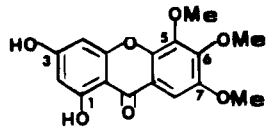
ABSTRACT.—A new pentaoxygenated xanthone has been isolated from the fern *Cystopteris fragilis*. Its structure has been determined as 1,3-dihydroxy-5,6,7-trimethoxyxanthone [**1**] by spectroscopic methods and chemical reactions.

The *Cystopteris fragilis* complex has recently been described as "perhaps the most formidable biosystematic problem in ferns" (1). Xanthone analysis may be of interest in the study of relationships of the various taxa of this complex, as it is well known that the taxonomic value of xanthones in ferns is in the identification of allopolyploidy (2). *Cystopteris fragilis* Bernh. (Aspleniaceae) has a worldwide distribution and occurs in tetraploid, hexaploid, and octoploid forms. Previous work on the chemical constituents of this fern has led to the identification of two *C*-glycosylxanthones (mangiferin and isomangiferin) (3) and four flavonol glycosides (the 3-*O*-glucoside, the 3,4′-bis-glucoside, the 3-*O*-(3″-sulfate)glucoside, and the 3-*O*-(6″-sulfate)glucoside of kaempferol) (4). In addition, a new xanthone (1,6-dihydroxy-3,5,7-trimethoxyxanthone) has recently been isolated from *Cy. fragilis* (5), and it is of interest that this compound is the first pentaoxygenated xanthone found in ferns. In the present work another new pentaoxygenated xanthone has been found in *Cy. fragilis* and identified as 1,3-dihydroxy-5,6,7-trimethoxyxanthone.

RESULTS AND DISCUSSION

Color reactions (yellow to orange in uv + NH_3) and uv spectral analysis in the presence of the usual shift reagents (6) suggest that the isolated compound is a xanthone with a free hydroxyl group at position 3 and/or 6 (shift with NaOAc) and a free hydroxyl group at position 1 and/or 8 (shift with $AlCl_3$ and $AlCl_3$/HCl). In addition, a 1,3-dihydroxy system must be in the A ring of this xanthone, as the uv spectra in the presence of NaOMe and NaOAc are not superimposable (6). The ^1H nmr shows the presence of three -OMe (three singlets at δ 3.83, 3.95, and 3.99) and two -OH (two singlets at δ 12.98 and 5.05), one of which (δ 12.98) must be in the peri position (7). In addition, the ^1H nmr shows two meta split doublets (δ 6.51 and 6.36, $J = 2.3$ Hz), which are consistent with this xanthone having a 1,3-dioxygenated A ring (7). A singlet at δ 7.41 has been assigned to H-8 because the signal of this proton [generally between δ 7.70 and 8.05 (8)] is shifted to higher fields by the presence of three substituents (7) in the B ring of this xanthone as in the case of synthetic 1,3,5,6,7-pentamethoxyxanthone which shows a singlet at δ ($CDCl_3$) 7.38 (H-8) (9). The mass spectrum gives a molecular ion at *m/z* 318 for $C_{16}H_{14}O_7$; this result, taking into account the ^1H-nmr data, is in accordance with a xanthone containing two hydroxyl groups and three methoxyl groups. The above observations are in agreement with the isolated compound being a 1,3,5,6,7-pentaoxygenated xanthone; this result was confirmed by demethylation with HI, which gave 1,3,5,6,7-pentahydroxyxanthone identified by ms and direct comparison with an authentic sample from *Canscora decussata* (10).

The above data show that the isolated xanthone must be 1,3-dihydroxy-5,6,7-trimethoxyxanthone [**1**], which is a new natural product; in addition this substance has not previously been described in the course of snythetic or partial degradation work. The structure of **1** was confirmed in the following way. Selec-



1

tive demethylation (11) of 1,3,5,6,7-pentamethoxyxanthone, prepared according to Quillinan and Scheinmann (9), gave 1,6-dihydroxy-3,5,7-trimethoxyxanthone which was different from the isolated xanthone **1** in paper chromatography [solvent system H_2O-HOAc (85:15)]. R_f values were: xanthone **1** 0.10; 1,6-dihydroxy-3,5,7-trimethoxyxanthone 0.17. In addition the mp of xanthone **1**, 240–243°, differs by a significant amount from that of 1,6-dihydroxy-3,5,7-trimethoxyxanthone, 251–254°.

In 1969 Carpenter *et al.* (12) predicted that 1,3,5,6,7-pentaoxygenated xanthones must occur in plants if the phenol oxidative coupling biosynthetic mechanism is operative. This suggestion was confirmed in 1974 by Ghosal *et al.* (10), who isolated a number of such xanthones from *Ca. decussata*. As mentioned above, recently (5) a pentaoxygenated xanthone has been isolated for the first time from *Cy. fragilis* and has been identified as 1,6-dihydroxy-3,5,7-trimethoxyxanthone; hence the isolation of xanthone **1** confirms that 1,3,5,6,7-pentaoxygenated xanthones occur in ferns.

EXPERIMENTAL

GENERAL EXPERIMENTAL PROCEDURES.—Mp's were determined on a Kofler hotstage microscope and are uncorrected. ^{1}H nmr were taken in $CDCl_3$; chemical shifts are given in ppm downfield from TMS; coupling constants are given in Hz. The ^{1}H-nmr spectra were obtained with a Brucker AC-250 instrument operating at 250 MHz. Eims were run on a Kratos MS 50S instrument at 70 eV. Ir spectra were measured on a Perkin-Elmer spectrophotometer 521 and uv spectra on a Beckman DU-GT spectrophotometer.

PLANT MATERIAL.—Aerial parts of *Cy. fragilis* were collected on Mount Etna, Sicily in 1982. The fern was identified by Dr. A. D'Urso (Botanic Institute, University of Catania); a voucher specimen has been deposited in the Department of Chemical Sciences, University of Catania.

EXTRACTION AND ISOLATION.—Air-dried aerial parts (50 g) of *Cy. fragilis* were homogenized and extracted 3 times with 95% EtOH; evaporation of the solvent in vacuo gave 14 g of residue. The new xanthone **1** was isolated by preparative paper chromatography on Whatman no. 3 MM paper in *n*-BuOH–HOAc–H_2O (4:1:5), upper phase) (R_f = 0.82). The bands, as observed under uv light with NH_3 vapor, were cut out, eluted with EtOH, concentrated and rechromatographed in 15% HOAc (R_f = 0.10) and in BuOH-EtOH-H_2O (4:1:2.2) (R_f = 0.79). Further purification was carried out over Si gel cc (eluting with C_6H_6), which gave 7 mg of the xanthone **1**.

1,3-DIHYDROXY-5,6,7-TRIMETHOXYXANTHONE [**1**].—Mp 240–243°; uv (MeOH) λ max 358 (ε = 5650), 313 (10700), 253 (33970) nm; +$AlCl_3$ 403, 342, 267 nm; +$AlCl_3$/HCl 402, 340, 267 nm; +NaOAc 374, 267, 261 nm; +NaOMe 378, 263 nm. Ir (Nujol) 3388 br, 1661, 1610, 1584, 1321, 1291, 1217, 1200, 1162, 1100, 1061, 1033, 963, 884, 812, 800 cm^{-1}; ^{1}H nmr ($CDCl_3$) δ 3.83, 3.95, 3.99 (3H each, s, OMe), 5.05 (1H, s, OH), 6.36, 6.51 (1H each, d, J = 2.3 Hz, H-4 and H-2), 7.41 (1H, s, H-8), 12.98 (1H, s, 1-OH); eims (70 eV) *m/z* (rel. int. %) $[M]^+$ 318 (100), 303 (8), 289 (12), 275 (10), 257 (11), 229 (4), 229 (4), 159 (6), 145 (7), 129 (5). Calcd for $C_{16}H_{14}O_7$: C 60.37, H 4.43; found C 60.49, H 4.47.

DEMETHYLATION OF 1,3-DIHYDROXY-5,6,7-TRIMETHOXYXANTHONE.—The xanthone (2 mg) and HI (4 ml; d = 1.7) were refluxed for 6 h in the dark. Removal of HI under high vacuum gave a residue which was identified as 1,3,5,6,7-pentahydroxyxanthone by paper co-chromatography [solvent system HOAc-37% HCl-H_2O (30:3:10), R_f 0.37] and co-tlc on Si gel [solvent system C_6H_6-MeOH (4:1), R_f 0.30] by using an authentic sample from *Ca. decussata* (10). The identification of 1,3,5,6,7-pentahydroxyxanthone was confirmed by eims (70 eV), which revealed a molecular ion at *m/z* 276 $[M]^+$ (23%) and significant ions at 248 $[M-CO]^+$ (18), 220 $[M-2CO]^+$ (28), and 192 $[M-3CO]^+$ (40).

ACKNOWLEDGMENTS

The author thanks the Board of Education (Rome) for financial support.

LITERATURE CITED

1. J.D. Lovis, *Adv. Bot. Res.*, **4**, 229 (1977).
2. P.M. Richardson, *Biochem. Syst. Ecol.*, **12**, 1 (1984).

3. P.M. Richardson and E. Thaddeus, *J. Nat. Prod.*, **46**, 747 (1983).
4. F. Imperato, *Chem. Ind. (London)*, 204 (1983).
5. F. Imperato, *Z. Naturforsch*, **45b**, 1603 (1990).
6. A.A. Lins Mesquita, D. De Barros Correa, O.R. Gottlieb, and M. Taveira Magalhaes, *Anal. Chim. Acta*, **42**, 311 (1968).
7. D. Barraclough, H.D. Locksley, F. Scheinmann, M. Taveira Magalhaes, and O.R. Gottlieb, *J. Chem. Soc. B*, 603 (1970).
8. K. Hostettmann and H. Wagner, *Phytochemistry*, **16**, 821 (1977).
9. A.J. Quillinan and F. Scheinmann, *J. Chem. Soc., Perkin Trans. 1*, 1329 (1973).
10. S. Ghosal, R.K. Chauduri, and K.R. Markham, *J. Chem. Soc., Perkin Trans. 1*, 2538 (1974).
11. K.R. Chauduri, F. Zymalkowski, and S. Ghosal, *J. Pharm. Sci.*, **67**, 1321 (1978).
12. I. Carpenter, H.D. Locksley, and F. Scheinmann, *Phytochemistry*, **8**, 2013 (1969).

Received 28 June 1990