Phytochemistry, 1978, Vol 17, p 2138 © Pergamon Press Ltd Printed in England 0031-9422/78/1201-2138 $02.00/0

A NEW FLAVONOL GLYCOSIDE FROM THE FLOWERS OF *SOLANUM XANTHOCARPUM*

PRAMILA DUBEY and P. C. GUPTA

Department of Chemistry, University of Allahabad, Allahabad-211 002, India

(*Received* 14 *April* 1978)

Key Word Index—*Solanum xanthocarpum*; Solanaceae; quercetin 3-*O*-β-D-glucosyl-*O*-β-D-mannoside; apigenin; sitosterol

Although *Solanum xanthocarpum* is of some medicinal value there has been no previous chemical examination of the flowers. In the present work, apigenin [1] was identified in the petals and a new flavonol diglycoside and sitosterol were isolated from the stamens. The identity of apigenin and sitosterol was confirmed by standard procedures.

The new flavonol glycoside, $C_{27}H_{32}O_{19}$, mp 184°, gave the characteristic colour reactions of a flavonol [2, 3] and on hydrolysis with 10% ethanolic H_2SO_4 gave quercetin and a disaccharide, the component sugars of which were glucose and mannose. Glucose was confirmed by co-chromatography and by the preparation of its phenylosazone, mp 204° (lit. 205°); mannose was confirmed by mp 131°, $[\alpha]_D^{30}$ + 13.9° (water), co-chromatography and by the preparation of its phenylhydrazone, mp 196° (lit. 199-200°) and *N*-glycosyl aminobenzoic acid, mp 179° (lit. 181°). Periodate oxidation indicated that both sugars in the disaccharide had the pyranose configuration; 3.4 mol of periodate were consumed with the liberation of 1.3 mol of formic acid. On methylation of the glycoside [4] followed by hydrolysis, two methylated sugars were identified in the hydrolysate, viz. 2,3,6-tri-*O*-methyl-D-mannose and 2,3,4,6-tetra-*O*-methyl-D-glucose, indicating that C_1 of the mannose is linked with the C_3-OH of the aglycone and that C_4 of the mannose is attached to C_1 of the glucose.

Only glucose could be identified in the aqueous hydrolysate obtained after acid hydrolysis, indicating that glucose occupies the terminal position. The position of the disaccharide molecule at position −3 was confirmed by the ready H_2O_2 oxidation of the glycoside in dilute NH_4OH and was further confirmed by acid hydrolysis of the methylated glycoside. The methylated aglycone hydrolysate, mp 192–93°, was identified as 5,7,3',4'-tetra-*O*-methylquercetin by co-chromatography with an authentic compound obtained similarly from quercetin-3-glycoside. Since the glycoside failed to reduce Fehling's solution and also did not give a test with aniline hydrogen phthalate, the glucose molecule must be linked to mannose through its reducing group. Hydrolysis of the glycoside with emulsin gave glucose and mannose, indicating a β-linkage between the two sugar units in the disaccharide as well as between the aglycone and mannose.

Thus, the original glycoside is quercetin-3-*O*-β-D-glucopyranosyl-*O*-β-D-mannopyranoside.

REFERENCES

1. Nick, E. (1953) *Pharmazie* **8**, 940.
2. Asahina, Y. and Inubuse, M. (1928) *Chem. Ber.* **61B**, 1646.
3. Wilson, C. W. (1939) *J. Am. Chem. Soc.* **61**, 2303.
4. Parikh, V. M., Ingle, T. R. and Bhide, B. V. (1958) *J. Org. Chem.* **35**, 125.

Phytochemistry, 1978, Vol 17, pp 2138-2140 © Pergamon Press Ltd Printed in England 0031-9422/78/1201-2138 $02.00/0

GOMPHRENOL, A NEW METHYLENEDIOXYFLAVONOL FROM THE LEAVES OF *GOMPHRENA GLOBOSA* (AMARANTHACEAE)

M. L. BOUILLANT*, P. REDOLFI†, A. CANTISANI† and J. CHOPIN*

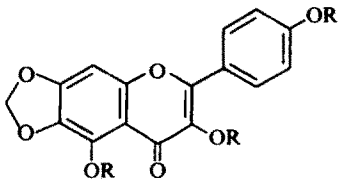
*Laboratoire de Chimie biologique, Université de Lyon I, 69621 Villeurbanne, France;
†Laboratorio di Fitovirologia Applicata, 10135 Torino, Italy

(*Received* 23 *June* 1978)

Key Word Index—*Gomphrena globosa*; Amaranthaceae; 3,5,4'-trihydroxy-6,7-methylenedioxyflavonol; gomphrenol.

INTRODUCTION

In a previous paper [1] two of us reported results on the role of phenolic compounds in the hypersensitive reaction of *Gomphrena globosa* infected with tomato bushy stunt virus. In the course of screening the phenolic constituents of the healthy plant, we have isolated, in addition to a number of common phenolics, a flavonol (**1a**), which was present in considerable amount and did not correspond to any of the known structures [2]. We describe here the identification of this new compound, the only flavonoid present in *G. globosa* leaves. Previous phytochemical knowledge on *G. globosa* is restricted to the identification of several betacyanins, from both flowers and leaves [3] [4].

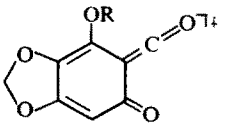


1a R = H gomphrenol
1b R = Me PM gomphrenol

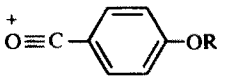
RESULTS

Healthy leaves of *G. globosa* were extracted with 50% MeOH. After acid hydrolysis and Sephadex LH 20 column chromatography in MeOH, a few mg of the pure flavonoid **1a** were isolated. From the yellow–brown color in UV light and the UV spectral data (as kaempferol), **1a** has a flavonol structure, with a free hydroxyl group at 3 and 5, but no free hydroxyl at 7. MS of **1a** (M^+ 314) and of its permethyl derivative **1b** (M^+ 356) showed the presence of three free hydroxyl groups in **1a**. High resolution MS established the molecular formula of **1b** as $C_{19}H_{16}O_7$ (M 356) and not $C_{20}H_{20}O_6$ (M 356) excluding a tetra-*O*-methyl *C*-methylkaempferol structure.

Both PMR spectra of the free compound **1a** and of the PM derivative **1b** exhibited two protons as a strong singlet near 6 ppm, typical of a methylenedioxy group, and two two proton doublets near 8 and 7 ppm, with *ortho*-coupling, corresponding to an aromatic A_2B_2 system. All these results agreed with a 3,5,4′-trihydroxyflavone structure bearing a methylenedioxy group on the A ring. This conclusion was also supported by the presence, in the MS of **1a** and **1b** of the corresponding RDA fragments:



R = H *m/e* 180
R = Me *m/e* 194



R = H *m/e* 121
R = Me *m/e* 135

The 6,7 position of the methylenedioxy group was deduced as follows. 5,6,7,4′-Tetrahydroxyflavonol was detected (MS and TLC) in the complex mixture obtained after heating **1a** with pyridinium hydrochloride; a singlet at 6.69 ppm, typical for a proton in 8 position, was observed in the PMR spectrum of the TMS derivative [5]. After removal of the OTMS in position 5 this signal was shifted upfield (0.07 ppm), whilst a proton in position 6 would be unaffected [5]; the A ring protons of the PM derivative **1b** exhibited the same chemical shifts as those given for 3,5-dimethoxy-6,7,3′,4′-bismethylenedioxyflavone (meliternatin) [6].

As compound **1a** is the first flavonol found in *G. globosa* we name it gomphrenol. Flavonoids bearing the methylenedioxy group are rare in nature. Some flavonols with such a group on the B ring have been described but only two with the 6,7-orientation are known: meliternatin from *Melicope* sp. [6–8] and the incompletely characterised 5,3′,4′-trihydroxy-3-methoxy-6,7-methylenedioxyflavone from *Spinacia oleracea* [9].

Among the few other flavonoids with this group in 6,7 position, three isoflavonoids have been isolated: tlatlancuayin and irisolone from *Iresine celosioides* (Amaranthaceae) [10] and betavulgarin from *Beta vulgaris* (Chenopodiaceae) [11], where a flavanone with the same A ring, betagarin, was also identified.

EXPERIMENTAL

MS spectra were recorded on an AEI MS 902 spectrograph to 70 eV. Temperatures (sample and source in the same order) varied between 175 and 210°. PMR spectra were recorded at 100 MHz on a Varian FT XL 100 apparatus and at 250 MHz on a CAMECA apparatus, both with TMS as internal standard.

Isolation of the compound **1a**. Fresh leaves of *G. globosa* were homogenized in 10 vol. 50% MeOH, the filtered extract made aq. and chlorophylls removed by repeated petrol extractions. The extract was then dried, the residue taken up in a small vol. of MeOH, layered on a Sephadex LH20 column and eluted with MeOH. The eluate was collected and hydrolysed in 2 N HCl at 100° for 30 min. The hydrolysate was extracted with Et_2O, evapd to dryness and the residue taken in MeOH, chromatographed as before and the main absorbing material collected. TLC, cellulose Merck, (R_f BAW 4:1:5; 0.80; BEW 5:4:2.2; 0.85; Forestal; 0.65; H_2O: 0.0; 15% HOAc; 0.05) (UV; brown; Na_2CO_3, yellow-green) UV λ_{max}^{MeOH} nm 262 sh, 272, 352; + $AlCl_3$ 279, 356 sh, 412; + $AlCl_3$ + HCl, 272, 308 sh, 384, 412; + NaOAc 262 sh, 272, 346, 375 sh, 408 sh; + NaOAc/H_3BO_3 262 sh, 272, 352; + NaOH 279, 356 sh, 412. PMR ($(CD_3)_2CO$)$\delta_{TMS}^{10^{-6}}$ 8.16 (*d*, 2H, *J* = 9 Hz) H-2′, 6′; 7.02 (*d*, 2H, *J* = 9 Hz) H-3′,5′; 6.77 (*s*, 1H) H-8; 6.14 (*s*, 2H), CH_2O_2-6,7. MS: *m/e* 314 (M^+, 100%); 286 (M − 28, 5%); 285 (M − 29, 6%); 181 (2%); 180 (2%); 157 (M/2*e*, 5%); 121 (18%). *Permethylation* was carried out using a stream of CH_2N_2 through a solution of **1a** in MeOH, during 30 min. PM derivative **1b** was obtained pure on TLC (Si gel, C_6H_6–Me_2CO 9:1) as a blue fluorescent spot. PMR ($CDCl_3$)$\delta_{TMS}^{10^{-6}}$ 8.02 (*d*, *J* = 9 Hz) H-2′,6′; 7.00 (*d*, *J* = 9 Hz) H-3′,5′; 6.66 (*s*) H-8; 6.06 (*s*) 6,7-CH_2O_2; 4.13 (*s*) OCH_3-5; 3.89 (*s*) and 3.86 (*s*) OCH_3-3 and 4′ (meliternatin [6] 6.64 (*s*) H-8; 6.02 (*s*) 6,7-CH_2O_2). High resolution MS *m/e* 356.08494 (M^+, calc for $C_{19}H_{16}O_7$ 356.08959, 100%); 355 (M − 1, 94%) 341 (M − 15, 11%), 340 (M − 16, 8%), 338 (M − 18, 7%); 337 (M − 19, 28%) 327 (M − 29, 39%), 313 (7%), 309 (10%), 297 (5%), 267 (3%), 195 (4%), 194.02152 (calc. for $C_9H_6O_5$ 194.0215, 8%) 166 (5%), 164 (13%), 135.044601 (calc. for $C_8H_7O_2$ 135.0442, 14%).

5,6,7,4′-TetraOH flavonol from **1a**. About 1 mg **1a** was mixed with 10 mg Py HCl and heated, *in vacuo*, in a sealed tube at 180° during 1 hr. After usual work-up, TLC of the ether extract on silanized Si gel in hexane–EtOAc–Me_2CO (6:3:1) showed a complex mixture in which one spot corresponded to the 5,6,7,4′-tetrahydroxyflavonol prepared in the same way from a sample of synthetic 5,7,4′-trihydroxy-3,6-dimethoxyflavone (from Dr. Goudard, Lyon). MS 302 (M^+ 100%), 301 (15%), 273 (8%), 262 (8%), 250 (9%), 245 (7%), 193 (75%), 186 (27%), 169 (7%), 168 (10%), 153 (19%), 151 (15%), 121 (46%), 94 (58%). *MS of synthetic 5,6,7,4′-tetraOH flavonol* 302 (M^+ 100%), 301 (15%), 273 (10%), 262 (6%), 250 (5%), 245 (7%), 229 (6%), 193 (12%), 186 (18%), 168 (15%), 153 (21%), 151 (9%), 121 (28%), 94 (32%).

Trimethylsilylation of **1a** was carried out by usual method [5]. PMR ($CDCl_3$) 6.69 (*s*) H-8 in the 5-*O*-TMS derivative; 6.62 (*s*) H-8 in the 5-OH derivative.

Acknowledgements—The authors express their sincere thanks to Dr. J. Martin (Laboratoire de Physiologie végétale—INRA—Dijon, France) for her help in this work.

REFERENCES

1. Redolfi, P., Cantisani, A. and Pennazio, S. (1978) *Phytopath. Z.* (in press).
2. Pennazio, S., Redolfi, P., Cantisani, A. and Vecchiati, M. (1978) *Phytopath. Z.* (in press).

3. Piattelli, M. and Minale, L. (1964) *Phytochemistry* **3**, 574.
4. Minale, L., Piattelli, M. and De Stefano, S. (1967) *Phytochemistry* **6**, 703.
5. Mabry, T. J., Markham, K. R. and Thomas, M. B. (1970) *The Systematic Identification of Flavonoids*. Springer, Berlin.
6. Ritchie, E., Taylor, W. C. and Vautin, S. T. K. (1965) *Aust. J. Chem.* **18**, 2021.
7. Geissman, T. A. (1962) *The Chemistry of Flavonoid Compounds*. Pergamon Press, Oxford.
8. Fukui, K. and Matsumoto, T. (1965) *Bull. Chem. Soc Jpn* **38**, 887.
9. Ottmeier, W. and Hempel A. (1972) *Z. Naturf.* **B27**, 177.
10. Fukui, K., Matsumoto, T. and Nakamura, S. (1965) *Bull. Chem. Soc. Jpn* **38**, 1168.
11. Geigert, J., Stermitz, F. R., Johnson, G., Maag, D. D. and Johnson, D. K.(1973) *Tetrahedron* **29**, 2703.

Phytochemistry, 1978, Vol 17, pp 2140 2141 © Pergamon Press Ltd Printed in England 0031-9422/78 1201-2140 $02 00/0

BIFLAVONOIDS FROM FRUITS OF POISON IVY *TOXICODENDRON RADICANS*

MAHMOUD A. ELSOHLY*, JOHN C. CRAIG†, COY W. WALLER and CARLTON E. TURNER*
Research Institute of Pharmaceutical Sciences, School of Pharmacy, University of Mississippi, University, MS 38677, U.S.A.

(*Received* 20 *April* 1978)

Key Word Index—*Toxicodendron radicans*; Anacardiaceae; poison ivy; biflavonoids; 3′,8″-binaringenin, amentoflavone; tetrahydroamentoflavone.

During the course of our analytical work [1] with poison ivy (*Toxicodendron radicans*), an ethanolic extract of the fruits of the plant was prepared. Upon partitioning of the ethanol extract between chloroform and water, a brown colored amorphous residue was obtained in the interface. TLC on silica gel plates using 20% methanol in chloroform showed the presence of two major phenolic spots (R_f 0.55 and 0.62). Repeated chromatography of the residue obtained above results in the isolation of two compounds A and B. The structure determination of these two phenolic compounds is the subject of this note.

Compound A, mp 250–257° was optically inactive; gave orange color with Mg–HCl test and green color with $FeCl_3$. It formed a hexaacetate, mp 245–248° and a hexamethyl ether, MW 622 (Mass). This compound was characterized as amentoflavone (**1**) by comparing the spectral data with those reported in the literature [2]. Particular attention was drawn to the ^{13}C NMR, which was identical to that reported [3] for amentoflavone. In addition direct comparison was made with an authentic sample (mp, IR and ^1H NMR).

Compound B was isolated as light tan powder, which gave purple color with Mg–HCl and purple blue with $FeCl_3$; mp 234–239° dec.; $[\alpha]_D^{23}$ −19° (*c* 0.68, MeOH); IR, ν_{max}^{KBr} 3460 (br, OH) and 1650 (C=O) cm^{-1}. The UV spectrum [λ_{max}^{MeOH} 321 nm (log ε 4.43), 290 (4.52) and 228 (4.64)] with bathochromic shift in basic medium [$\lambda_{max}^{MeOH-NaOMe}$ 322 nm (log ε 4.72) and 227 (4.57)] was similar to that of naringenin; which also underwent bathochromic shift in the presence of NaOAc or $AlCl_3$ indicating the presence of OH groups at 5 and 7 positions [$\lambda_{max}^{MeOH-NaOAc}$ 322 nm (log ε 4.74) and 230 (5.12); $\lambda_{max}^{MeOH\ AlCl_3}$ 375 nm (log ε 3.99), 311 (4.69) and 227 (4.74)].

The ^1H NMR (60 MHz, DMSO-d_6) of compound B showed peaks at δ 7.25 (*m*, 4H), 6.89 (*d*, *J* = 8 Hz, 1H), 6.73 (*d*, *J* = 8 Hz, 2H), 6.12 (*s*, 1H), 5.95 (*s*, 2H), 5.47 (*br d*, *J* = 12 Hz, 2H), 3.18 (*br m*, 4H) and two D_2O exchangeable protons at 11.13 (*s*, 1H) and 11.25 (*s*, 1H) (two bonded phenolic OH groups). The mass spectrum showed a molecular ion at *m/e* 542, for $C_{30}H_{22}O_{10}$, indicating a biflavanone with 6 phenolic hydroxy groups. The fact that compound B had 6 OH groups and 10 aromatic protons along with UV similarities with naringenin indicated a binaringenin structure with a C-C linkage. The structure was proven to be 3′,8″-binaringenin (tetrahydroamentoflavone) as follows.

The ^1H NMR spectrum showed 7 aromatic protons as part of the A_2B_2 system, four of which were further downfield indicating that C-3′ position of one naringenin unit was substituted. Since it is known that, in the flavonoids, the C-6 proton appears up field from C-8 proton [4], the two protons singlet at δ 5.92 was assigned to the C-6 and C-6″ protons and the one proton singlet at δ 6.12 was assigned to H-8. Comparison of the ^{13}C NMR of compound B with that of rhusflavanone (6,8″-binaringenin) and naringenin (Table 1) clearly indicated a 3′,8″-linkage [3]. The chemical shift of C-3′ in the two model compounds was 115.4 and 115.2 ppm, respectively. The ^{13}C NMR spectrum of B showed a peak at 120.1 ppm which shows a shift of ~ 5 ppm units indicating a C-C linkage at C-3′. Also, the ^{13}C NMR spectrum of compound B showed 3 signals at 95.8, 95.7 and 95.1 ppm assigned to carbons 6,6″ and 8, respectively. The signal for C-8″ was found at 105.9 ppm again indicating a C-C linkage at that position since resonances for carbons 6 and 8, in 5,7-dihydroxyflavoids absorb between 90 and 100 ppm with C-6 about 0.9 ppm downfield from C-8 [3].

Finally, dehydrogenation of B (I_2/HOAc/KOAc) [5] resulted in the formation of a biflavone which was purified by preparative TLC, the hexaacetate of which was

* To whom correspondence should be addressed.
† Present address: Department of Chemistry, Western Kentucky University, Bowling Green, KY 42101, U.S.A.