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8-*C*-METHYL-QUERCETIN-3-*O*-β-D-XYLOPYRANOSIDE, A NEW FLAVONE GLYCOSIDE FROM THE ROOTS OF *AMOORA ROHITUKA*

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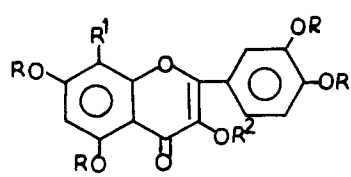
Amoora rohituka Wall (syn. *Aphanamixis polystachya*) (Meliaceae) has an astrigent action and has been used in the treatment of spleen and liver diseases, tumors, and abdominal complaints (1). We have recently reported the isolation and characterization of a new saponin, betulin-3-β-*O*-β-D-xylopyranoside (2), from its roots. Our further examination of the roots led to the isolation and characterization of a new flavone glycoside whose structure as 8-*C*-methyl-5,7,3′,4′-tetrahydroxyflavone-3-*O*-β-D-xylopyranoside (**1**) we now report.

The glycoside **1** was isolated by column chromatography from the MeOH extract of the stem bark of *A. rohituka*. Molecular weight determination by mass spectral and elemental analysis established the molecular formula as $C_{21}H_{20}O_{11}$ (M^+ 448); **1** gave all the positive color tests for a flavone glycoside. Acid hydrolysis of the glycoside afforded an aglycone (**2**) and a sugar identified as D-xylose by co-paper chromatography (co-pc) and osazone formation.

The aglycone (**2**), analyzed for $C_{16}H_{12}O_7$ (M^+ 316). The ir ν max (KBr) of **2** showed absorptions at 3440 (br, OH), 1650 and 1610 (α, β unsaturated >co), 2930 (Me) (3) and 1480, 1360, 1220, 1210, 1160, 910, 825, 725, and 712 cm^{-1} (complex aromatic substitution pattern). The ^{1}H-nmr spectrum of **2** exhibited signals at δ 12.40 (s, OH), 6.84, 7.30, 7.70 (3H, H-5′, 2′ and 6′), 2.10 (s, 3H, Me at C-8) (3,4), and 6.40 (s, 1H, H-6); **2** formed a pentaacetate and pentamethyl ether, both of which suggested the presence of five OH groups. The uv spectrum of **2** showed maxima at 260 and 360 nm in MeOH and gave positive bathochromic shifts with $AlCl_3$ (270 and 360 nm), $AlCl_3$+HCl [262, 300 (sh), 360 (sh), 410 nm], NaOAc (275 and 380 nm), and H_3BO_3+NaOAc (265 and 385 nm) (5,6), indicating the presence of free hydroxyls at the C-5, C-3, C-7, C-3′ and C-4′ positions, respectively; these were further confirmed by their color tests (7-11). KOH degradation (12) yielded 2,4,6 trihydroxytoluene (mmp and co-tlc) (3) and protocatechuic acid (mmp and co-tlc) (13), respectively. The $KMnO_4$ oxidation afforded protocatechuic acid (mmp and co-tlc) as one of the oxidation products. From the above data, it was clear that **2** was 8-C-methyl quercetin, which was in agreement with the mass spectral data [*m/z* 316 (M^+, 100), 315 (M^+-1, 76), 301 (M^+-15, 73), 298 (M^+-18, 10), 207 (M^+109, 22), 206 (M^+-110, 25), 180 (M^+-136, 28), 164 (M^+-152, 30), 150 (M^+-166, 16), and 149 (M^+-167, 80)].



1 R=H, R^1=CH_3, R^2= xylose
2 R=R^2=H, R^1=CH_3
3 R=R^1=CH_3, R^2=H

Compound **2** was identical (mmp, and co-tlc) to the iodine oxidation product of deodarin isolated from the stem bark of *Cedrus deodara* (14).

The glycoside **1** was methylated by Hakomori's method (15) followed by acid hydrolysis to afford another new flavone **3** and 2,3,4-tri-*O*-methyl-D-xylose (co-pc); **3** had a molecular formula of $C_{20}H_{20}O_7$ (M^+ 372). The ir spectrum (KBr) of **3** showed the absorption at 3450 (OH), 2935 (Me), 1645 and 1610 (CO), 2870 and 1170 (MeO), and 1485, 1310, 1210, 1155, 915, 895, 810, and 720 cm^{-1} (complex aromatic substitution pattern), while its pmr spectrum displayed signals at δ 2.12 (s, 1×Me), 3.90 (s, 2×OMe), 3.95 (s, 2×OMe), 6.40 (s, 1H, H-6), and 6.82, 7.35, 7.70 (3H, H-5′, 2′ and 6′). The uv spectrum of **3** exhibited maxima at 258 and 360 nm in MeOH. No bathochromic shifts of the longest wave length were observed with $AlCl_3$, NaOAc, and NaOAc+H_3BO_3 showing the blocked positions of C-5, C-7, C-3′, C-4′ by the methoxyl groups. Compound **3** gave a bathochromic shift with $AlCl_3$-HCl (260, 420 nm) and all the positive tests as described in references (7-9) for the presence of a free OH at C-3; the same was not produced by **1**, confirming the presence of xylose at the C-3 OH. KOH degradation of **3** yielded 1-methyl-2,4-dimethoxyphloroglucinol (mmp and co-tlc) (4) and veratric acid (mmp and co-tlc), respectively; while $KMnO_4$ oxidation gave veratric acid (mmp and co-tlc) as one of the oxidation products. Hence **3** was identified as 8-*C*-methyl-quercetin-5,7,3′,4′-tetramethyl ether; this structure was also supported by its mass spectrum (*m/z* 372, 371, 357, 354, 235, 234, 208, 192, 178, and 177). Compound **3** was also not reported previously by any other workers from nature.

The periodate oxidation (16) of **1** showed the consumption of 2.00 mol of periodate with the liberation of 1.00 mol of HCO_2H per mol of glycoside, suggesting the presence of one unit of D-xylose in pyranose form. Enzymatic hydrolysis of **1** gave **2** (mmp and co-tlc) and D-xylose, confirming the β-linkage between **2** and D-xylose. On the basis of the above data, structure **1** was assigned to the glycoside.

EXPERIMENTAL

PLANT MATERIAL.—Plant material of *A. rohituka* was procured from United Chemicals and Allied Products, Calcutta, India.

EXTRACTION AND ISOLATION.—The air dried and powdered roots (4 kg) were extracted with ethanol under reflux for 120 h. The ethanolic extract (40 liters) was concentrated (500 ml) under reduced pressure and poured into H_2O (1 liter) with continuous stirring. The H_2O soluble fraction was concentrated to a syrupy mass that was then extracted with MeOH. The methanolic extract yielded the reported glycoside **1**, mp 360-365° (dec). Compound **1** was purified over a column of magnesol (hydrated magnesium silicate) developed with MeOH and crystallized (MeOH-$CHCl_3$) as a dark brown amorphous solid (yield 1.3 g), (found; C, 56.22; H, 4.40; $C_{21}H_{20}O_{11}$ required; C, 56.25; H, 4.46); tlc, silica gel, Rf 0.45 (Me_2CO-MeOH, 8:2) and 0.28 (MeOH-$CHCl_3$, 5:5); pc, Rf 0.84 (*n*-BuOH-HOAc-H_2O, 4:1:5).

HYDROLYSIS OF **1**.—Compound **1** (800 mg) was hydrolyzed with 7% H_2SO_4 (40 ml) for 4 h under reflux to yield the aglycone **2** and the sugar D-xylose [Rf 0.28 in *n*-BuOH-HOAc-H_2O, 4:1:5, co-pc and osazone formation, mp 157-158° (lit mp 159°)].

CHARACTERIZATION OF **2**.—Compound **2** was purified over a column of magnesol (Me_2CO-MeOH, 5:5) and crystallized (MeOH-EtOAc) as light brown needles, mp 295-300° (dec) (found; C, 60.72; H, 3.75; $C_{16}H_{12}O_7$ required; C, 60.75, H, 3.79); tlc, silica gel, Rf 0.48 ($CHCl_3$-MeOH, 6:4) and 0.56 (Me_2CO-MeOH, 5:5); pc, Rf 0.90 (*n*-BuOH-HOAc-H_2O, 4:1:5); pentaacetate (100 mg of **2**+6 ml Ac_2O+5 ml C_5H_5N, yield 85 mg) (found; C, 58.25; H, 4.08; $C_{26}H_{22}O_{12}$ required; C, 59.31; H, 4.18); pentamethyl ether (80 mg of **2**+5 ml Me_2SO_4+2 g K_2CO_3, yield 60 mg); (found; C, 65.23; H, 5.70; $C_{21}H_{22}O_7$ required; C, 65.28, H, 5.69).

METHYLATION OF **1**.—Compound **1** (400 mg) was methylated by Hakomori's method (15) followed by acid hydrolysis to afford 2,3,4-tri-*O*-methyl-D-xylose (Rg 0.94 in *n*-BuOH-EtOH-H_2O, 5:1:4 and co-pc) and **3** mp 340-342° (found; C, 64.48; H, 5.35; $C_{20}H_{20}O_7$ required; C, 64.51; H, 5.37%); tlc, silica gel, Rf 0.68

(Me_2CO-MeOH, 8:2) and 0.42 ($CHCl_3$-MeOH, 6:4); pc, Rf 0.92 (*n*-BuOH-HOAc-H_2O, 4:1:5).

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LITERATURE CITED

1. "The Wealth of India, Vol. I," New Delhi: CSIR publication, 1956, p. 86.
2. A. Jain and S.K. Srivastava, *Indian J. Pharm. Sci.*, (in press).
3. S.K. Srivastava and H.O. Gupta, *Planta Med.*, **48**, 58 (1983).
4. Z.A. Rebeza and B. Viorin, *Tetrahedron Lett.*, 2149 (1978).
5. T.J. Mabry and H. Mabry, "The Flavonoids," London: Chapman and Hall, 1970, p. 46.
6. L. Jurd, K. Stevens and G. Manners, *Tetrahedron Lett.*, 2149 (1972).
7. R.D. Hill and G.D. Meakins, *J. Chem. Soc.*, 760 (1958).
8. H. Pacheco and M.C.R. Chandenson, *Acad. Sci. Paris*, **242**, 1621 (1956).
9. R. Neu, *Arch. Pharm. Berl.*, **292**, 431 (1959).
10. W.E. Hillis and G. Urbach, *Nature*, **182**, 637 (1958).
11. R.M. Horowitz and B. Gentilli, *J. Am. Chem. Soc.*, **82**, 2803 (1960).
12. L.H. Briggs and R.H. Locker, *J. Chem. Soc.*, 2157 (1949).
13. I. Heilbron and H.M. Bunburry, "Dictionary of Organic Compounds," London: Eyre and Spottiswoode, 1953, p. 260.
14. K. Raghunathan, S. Rangaswami and T.R. Seshadri, *Indian J. Chem.*, **12**, 1126 (1974).
15. S. Hakomori, *J. Biochem.*, **55**, 205 (1964).
16. E.L. Hirst and J.K.N. Jones, *J. Chem. Soc.*, 1959 (1949).

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