Flavonoids of *Madhuca butyracea* Nut-Shell

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The flavonoid (m.p. 222–224° dec.) isolated from *Madhuca butyracea* nut-shell is a mixture of quercetin and the flavanonol dihydroquercetin, the latter being 90% of the crystallizate. Paper chromatography (*n*-butyl alcohol–acetic acid–water, 4:1:5) also revealed only two constituents.

The identity of quercetin was established by elemental analysis, coupled with comparison with an authentic sample in regard to its mixed melting point, ultraviolet spectra, and paper chromatography.

The *madhuca* flavanonol is soluble in common organic solvents and water, and insoluble in benzene. The characteristics are noted in the accompanying table; Wilson's boric acid test[1] with it is positive. The flavanonol obtained on crystallization from dilute alcohol (30%) is not solvated, whereas the dihydroquercetin, taxifolin,[2] was isolated with one or more molecules of water of hydration when crystallized from alcohol of various dilutions.

The ultraviolet spectra of the flavanonol is substantially different from those of common flavones and flavonols, though the bathochromic shift in alkaline solution is somewhat similar to to that in case of polyhydroxy flavones; the peak at 250–260 mμ (*cf.*, flavones[3]) is conspicuously absent and to that extent the curve resembles those of polyhydroxy isoflavones.[4] The combination peaks at 1620, 1591, 1456, and 1513 cm.$^{-1}$ in its infrared spectra show the absence of absorption in the lower region, 1640–1680 cm.$^{-1}$, reported as characteristic of flavonols and flavanones.[4]

When treated with dilute mineral acids the flavanonol is slowly converted into quercetin. While methylation with methyl iodide did not yield any well defined product, on prolonged treatment with dimethyl sulfate both penta- (m.p. 149–150°) and tetramethyl (m.p. 159–160°) ethers of quercetin were obtained in comparatively poor yields (*cf.*, ref. 5). The flavanonol pentaacetate, m.p. 130°, $[\alpha]^{31}{}^{\circ}{}_{D}$ + 60° (*c* 1.0, alcohol), is different from quercetin pentaacetate in contradistinction to the formation of the methyl ethers.

The alkaline hydrolysis of the flavanonol yielded protocatechuic acid, and veratric acid was obtained from the drastic alkali degradation products of the tetramethyl ether. In another experiment, the methylated product of the flavanonol on alkaline hydrolysis yielded a cream-white crystalline neutral product, m.p. 119°, which is being further investigated.

Reduction of *madhuca* flavanonol with zinc and hydrochloric acid yielded a colorless crystalline compound, m.p. 268–270° dec., apparently identical with eriodyctiol. Racemization of the flavanonol yielded the optically inactive isomer, m.p. and mixed m.p. 241° dec.

The characteristics of *madhuca* flavanonol, its derivatives, and degradation products pointedly show its similarity with dihydroquercetin and it is further substantiated by its iodine oxidation to quercetin in almost quantitative yield. The identity of the flavanonol with the optically active dihydroquercetin isolated from Douglas fir-wood (*Pseudotsuga douglasii*, Carr.; Coniferae) was confirmed by mixed melting point and their comparable infrared and ultraviolet spectra, very kindly made available by Dr. J. C. Pew to whom our grateful thanks are due. The following table shows the comparative data of the two natural flavanonols:

It is significant to note that dihydroquercetin has until now been isolated only from the stem (wood and bark) of certain temperate zone plants; it is for the first time isolated in appreciable yield along with quercetin from a nut-shell.

Experimental[6]

Isolation.—Coarsely powdered *M. butyracea* nut-shell (2 kg.) was percolated at room temperature (20–35°) with 95% alcohol (5 × 3 l.). The semisolid dark brown extractive (*c* 300 g.) obtained after removal of solvent under reduced pressure and finally *in vacuo*, was successively extracted with petroleum ether (2 l.), ether (2 l.), and ethyl acetate (500 ml.). The ether extract on concentration yielded a pale yellow crystallizate, m.p. 222–224° (*c* 40 g.) responding to the color tests for flavonoids, and on repeated fractional crystallizations from dilute alcohol (30%) it yielded quercetin, m.p. and mixed m.p. 310–312° dec., (4 g.) and the flavanonol as dull white needles, m.p. 241° dec. (35 g.). Ultraviolet (quercetin) $\lambda_{max}^{alc.}$ 257 mμ (log ε 4.32) and 375 mμ (log ε 4.30). *lit*[3]: 258 mμ (log ε 4.32) and 375 mμ (log ε 4.34).

Anal. quercetin; Calcd. for $C_{15}H_{10}O_7$: C, 59.7; H, 3.3. Found: C, 59.9; H, 3.4.

Anal. flavanonol; Calcd. for $C_{15}H_{12}O_7$; C, 59.2; H, 3.95. Found: C, 59.1; H, 4.0.

Flavanonol pentaacetate was prepared by heating (8 hr., steam bath) the substance (500 mg.) in acetic anhydride (1.0 ml.) with fused sodium acetate (5 g.). After the usual work-up and crystallization (alcohol–charcoal) the acetate (150 mg.) was obtained as silky white needles, m.p. 130°. The ferric chloride test with the acetate was negative and the hydroxyl band was absent in its infrared spectra. Ultraviolet: $\lambda_{max}^{alc.}$ 280 mμ (log ε 3.47) and 330 mμ (log ε 3.80).

(1) C. W. Wilson, *J. Am. Chem. Soc.*, **61**, 2303 (1939).
(2) J. C. Pew, *ibid.*, **70**, 3031 (1948).
(3) L. H. Briggs and R. H. Locker, *J. Chem. Soc.*, 3136 (1951).
(4) K. Venkataraman (Review), *Fort. Chem. Org. Naturstoffe*, Springer-Verlag, **17**, 58 (1959).
(5) W. E. Hills, *Aust. J. Sci. Res.*, 379 (1952).
(6) Melting points uncorrected; petroleum ether, b.p. 40–60°; ultraviolet data recorded by Mr. B. P. Mittal; microanalyses by Mr. J. Saran, Central Drug Research Institute, Lucknow.

TABLE I

	Madhuca flavanonol	Dihydroquercetin from Douglas fir-wood
M.p.	241° dec.	240–242° dec.
Optical rotation	+36° (alc.)	+46° (acetone–water) +13 (absolute alc.)
Ultraviolet spectra:		
$\lambda_{max.}^{alc.}$ mμ	291 (log ε 4.18) and 330 (log ε 4.04)	290 and 330 (inflexion)
$\lambda_{max.}^{alc.}$ mμ (0.001 *N* alkali)	213 (log ε 4.40); 330 (log ε 4.38) and 410 (log ε 3.89)	
Infrared bands, μ (KBr)	2.98, 3.05, 6.17, 6.28, 6.61, 6.87, 7.33, 7.69, 7.93, 8.58, 8.81, 8.93, 9.24, 9.80, 10.28, 10.57, 11.65	2.96, 3.07, 6.10, 6.60, 6.80, 6.90, 7.37, 7.83, 7.95, 8.60, 8.81, 8.96, 9.25, 9.81, 10.04, 10.30, 10.62, 11.70
Mg or Zn and hydrochloric acid	Deep purple-red	Deep purple-red
Ferric chloride	Violet-brown-blueblack	Emerald green-black
Caustic alkali	Pink-yellow	Brown

Anal. Calcd. for $C_{25}H_{22}O_{12}$; C, 58.3; H, 4.3. Found: C, 57.92; H, 4.46.

Methyl Ethers.—Methylation was carried out by refluxing (36 hr.; steam bath) the flavanonol (1 g.) in acetone (100 ml.) with dimethyl sulfate (1.0 ml.) and anhydrous potassium carbonate (5 g.). After work-up and crystallization (alcohol), methylated products were obtained, one as a white crystalline product (50 mg.), m.p. 149–150°, and the other as pale-yellow needles, m.p. 159–160° (300 mg.). The former is identical with pentamethylquercetin (m.p. and mixed m.p. 149–150°) while the latter was found to be the tetramethyl ether of quercetin (*lit.*, m.p. 159–160° of 3,7,3′,4′-tetramethylquercetin). A ferric chloride test with both methyl ethers was negative but the tetramethyl ether showed the OH band (2.98 μ) in its infrared spectra.

Anal. Calcd. for $C_{15}H_5O_2(OCH_3)_5$: C, 64.6; H, 5.4. Found: C, 64.9; H, 6.0.

Anal. Calcd. for $C_{15}H_6O_3(OCH_3)_4$: C, 63.7; H, 5.3. Found: C, 63.67; H, 5.8.

Conversion of the Flavanonol to Quercetin.—(a) The flavanonol (300 mg.) was heated (24 hr.; steambath) with dilute sulfuric acid (7%) and the reaction mixture on fractional crystallization with dilute alcohol (30%) yielded quercetin (24 mg.), m.p. and mixed m.p. 310–312° dec., while the major fraction of the flavanonol (250 mg.) was recovered unchanged.

(b) The flavanonol (300 mg.) was heated (1 hr.; steam bath) with iodine (100 mg.) in glacial acetic acid (3 ml.) and fused sodium acetate (500 mg.). The yellow precipitate obtained on dilution and cooling was worked up and crystallized (glacial acetic acid) to give yellow needles of quercetin (270 mg.), m.p. and mixed m.p. 310–312° dec.

Reduction of the Flavanonol with Zinc and Hydrochloric Acid.—The flavanonol (200 mg.) was treated in methanolic solution (10 ml.) with zinc powder (2 g.) and concd. hydrochloric acid (4 ml. added dropwise) with vigorous shaking (1.5 hr.; room temperature). The reaction mixture was kept overnight and the clear filtrate after separation of the unchanged zinc was diluted with ice-cold water (20 ml.) and the resultant white precipitate collected and dried. On repeated crystallizations (water) it yielded crystalline needles (50 mg.), m.p. 268–270° (eriodictyol, lit.,[2] m.p. 272°). Its characteristic color reactions are same as those reported for eriodictyol.

Anal. Calcd. for $C_{15}H_{12}O_6$: C, 62.5; H, 4.2. Found: C, 62.3; H, 4.5.

Alkali Degradation.—(a) The flavanonol (300 mg.) was refluxed (1 hr.) with caustic potash (2.5 g. in 25 ml. water and 25 ml. alcohol) and worked up as usual to give a cream-white acidic product (30 mg.), m.p. 190° after purification through sublimation (150°/3 mm.). There was no depression in its melting point in admixture with authentic sample of protocatechuic acid.

(b) The total methylated product of the flavanonol (m.p. 150–155°; 1 g.) was boiled (3 hr.) with a potassium hydroxide solution (aqueous, 5%; 50 ml.) and the neutral precipitate obtained on cooling and dilution yielded after crystallization (alcohol) rhombic plates (250 mg.), m.p. 119°, $C_{17}H_{18}O_6$.

Anal. Calcd. for $C_{17}H_{18}O_6$: C, 64.1; H, 5.6. Found: C, 63.7; H, 5.76.

The filtrate on acidification (hydrochloric acid, 5 ml.) yielded a solid residue (100 mg.) which on sublimation (130°/3 mm.) gave veratric acid, m.p. and mixed m.p. with authentic sample, 179–180°.

Anal. Calcd. for $C_9H_{10}O_4$: C, 59.3; H, 5.49. Found: C, 59.5; H, 5.7.

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Crystalline Complexes of 1,3,5-Trinitrobenzene and Alkali Sulfites

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Muraour[1] reported that aqueous solutions of sodium sulfite (3–5%) would dissolve 1,3,5-trinitrobenzene (TNB) and lesser quantities of 2,4,6-trinitrotoluene (TNT) through the formation of red-colored addition compounds but would not dissolve 2,4,6-trinitro-*m*-xylene. From these aqueous solutions the TNB or TNT could be recovered by acidification or in the case of TNT by dilution with water. Although Muraour stated that this complex of TNB and sodium sulfite would be studied further, no other information has appeared.

Dark red crystalline compounds, in which the mole ratio of TNB to sulfite is 1:2, can be isolated if TNB is dissolved in warm aqueous solutions of sodium or potassium sulfite (20% by weight) and the resulting solutions quickly cooled to ambient temperature. If these solutions stand for twenty-four hours, the complex disappears completely and the TNB is destroyed (see below). When dry, these compounds are very stable; samples have

(1) H. Muraour, *Bull. soc. chim. France*, **35**, 367 (1924).