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THE CONSTITUTION OF BIFLAVONOIDS FROM GARCINIA PLANTS

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In the previous paper[1,2], we reported the structures of (±)-fukugetin (I), (+)-fukugetin (II), (±)-3'''-O-methyl fukugetin (III) and fukugiside (IV) which were isolated from the fresh bark of Garcinia spicata HooK. f. (Guttiferae). We have further studied on the constituents of this plant and isolated two new compounds, named dl-volkensiflavone (V) and spicataside (VI), a biflavonoid glycoside, along with GB-Ia (VII) and GB-2a (VIII)[3,4]. We have examined the wood and fresh leaves of G. xanthochymus HooK. f. and isolated I, IV, V, VII, VIII, GB-2 (IX)[3], and a new biflavanone glycoside, xanthochymusside (X), which is the first example of the glycoside of biflavanone compound. We also have isolated I, II, V, VII, VIII, IX and GB-1 (XI) from the bark of G. multiflora Champ. and I, VII, VIII, IX and XI from the bark of G. linii Chang.

Xanthochymusside (X), $C_{36}H_{32}O_{16}\cdot 2H_2O$, a colourless amorphous powder, m.p. 219°, showed the following ORD data: (c =0.31, MeOH) $[\alpha]^{20}$ (mμ) − 40 (589), − 110 (450) and − 140 (400). It gave blue-fluorescence under the ultraviolet light and gave deep red colour with Mg-HCl. Its UV absorption maxima in ethanol at 225 (shoulder), 290, and 330mμ were very similar to those of naringin and they showed the charactristic bathochromic shift of 5,7-dihydroxy systems[5] on addition of sodium acetate and $AlCl_3$. The IR absorption (nujol mull) appeared at 3300 (hydroxyl groups), 1630 (conjugated γ-pyrone), 1600 and 1510 cm^{-1} (benzene rings).

Hydrolysis of X with 5% sulfuric acid afforded, along with D-glucose, a colourless amorphous compound, which was identified with the authentic sample of GB-2a (VIII) isolated from G. eugeniifolia Wall.[4] by thin-layer chromatography (TLC) and IR spectral comparison. D-Glucose was identified by paper chromatography

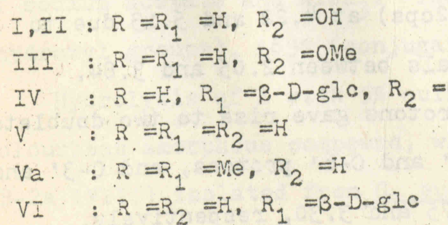
(PPC) and TLC (microcrystalline cellulose powder) of the sugar and its osazone together with the mixed m.p. of the latter. On acetylation X gave a decaacetate (Xa), which was negative for the ferric chloride test. NMR spectrum (60mc. in $CDCl_3$, τ scale) of Xa revealed the presence of ten acetoxyl groups (7.65 to 8.1) and C-1 to C-6 protons of the glucose moiety (multiplets 4.4 to 6.5). The NMR spectrum (acetone-d_6) of X showed several signals in the region between 2.50 and 7.50. Two hydrogen-bonded hydroxyl groups showed singlets at -2.18 and -2.06, other hydroxyl groups showed broad signals at 0.25, 1.50 and 1.88, one methylene groups (C-3" of ring F) showed multiplets at 7.30, and all the glucose protons showed broad signals at 4.50 - 6.7 (4.0 - 6.2 in pyridine-d_5). Aromatic protons showed signals between 2.50 and 4.18. Only three aromatic protons showed signals in the region of 3.6 - 4.2 of the spectrum. Two of them appeared as *meta*-coupled doublets (J =2cps) at 4.03 and 4.12, which were assigned to C-8 and C-6 protons respectively, whereas another proton showed a singlet at 3.69 due to C-6" proton. From these data X was suggested to be a GB-2a-7"-glucoside. In order to determine the position of glucose in the molecule, X was oxidized with sodium acetate and I_2 in ethanol[6,7] to give a yellow crystalline compound, m.p. 242-243° (decomp.), which was identified with fukugiside (IV) (mixed m.p., IR and NMR spectra). Hydrolysis of X with emulsin (Sigma Chemical Co.) gave GB-2a (VIII) and D-glucose indicating the β-glucoside linkage. From these data we propose structure X (=GB-2a-7"-β-glucoside) for xanthochymusside.

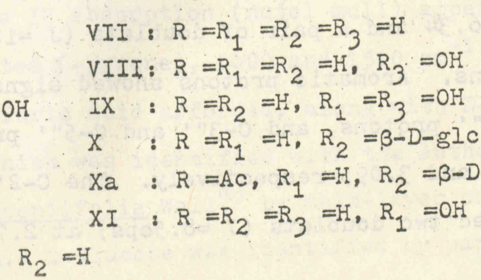
dl-Volkensiflavone (V), $C_{30}H_{20}O_{10}$, m.p. 290-293° (decomp.), $[\alpha]_D^{15}$ 0 (c =0.31, MeOH) (ORD), showed UV maxima (MeOH) at 225 (shoulder), 275, 289 and 330mμ, and IR absorptions (nujol mull) at 3100 (hydroxyl groups), 1640 and 1610 (conjugated γ-pyrone), 1570 and 1510 cm^{-1} (benzene rings). On methylation V gave hexamethyl ether (Va), m.p. 260-262°, $C_{36}H_{32}O_{10}$, which was negative for the ferric chloride test. NMR spectrum (acetone-d_6) of Va showed signals due to six methoxyl groups between 6.08 and 6.34 and a pair of doublets (J =12cps) at 4.22 and 5.13 due to C-2 and C-3 protons. Aromatic protons showed signals between 2.05 and 3.84. The C-2"' and C-6"' protons, and C-3"' and C-5"' protons gave rise to two doublets (J =9cps) at 2.19 and 3.09, respectively. The C-2' and C-6' protons, and C-3' and C-5' protons showed two doublets (J =8.5cps) at 2.75 and 3.30, respectively.

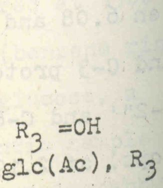
The remaining three aromatic protons showed signals in the region of 3.55 - 3.84 of the spectrum. Two of them appeared as <u>meta</u>-coupled doublets (J =2.5cps) at 3.65 and 3.77, which were assigned to C-8 and C-6 protons respectively, whereas another proton showed a singlet at 3.63 due to C-6" proton. A singlet at 3.47 can be assigned to C-3" proton. These NMR spectral data are in agreement with the structure Va and these data together with other experimental results described in this paper support structure V for <u>dl</u>-volkensiflavone.

Recently, Scheinmann et al[8] have obtained volkensiflavone, m.p. 245-250°, to which they assigned the structure V. Volkensiflavone was thought to be an optically active compound[9]. The TLC and UV spectrum of <u>dl</u>-volkensiflavone (V) were identical with those of volkensiflavone[9].

Spicataside (VI), $C_{36}H_{30}O_{15}\cdot 2H_2O$, m.p. 232-233° (decomp.), showed the following ORD data: (c =0.31, MeOH) $[\alpha]^{14}$ (mμ) + 20 (589), + 100 (450) and + 360 (400). Its UV absorption maxima in ethanol at 225 (shoulder), 275, 292 and 330mμ were very similar to those of V and the spectrum showed the charactristic bathochromic shift of 5,7-dihydroxy system on addition of $AlCl_3$ and sodium acetate. The IR absorptions (nujol mull) appeared at 3300 (hydroxyl groups), 1640 (conjugated γ-pyrone), 1600 and 1560 cm^{-1} (benzene rings). Hydrolysis of VI with 5% sulfuric acid afforded D-glucose and a yellow crystalline compoumd, m.p. 290-293°, which was identified with <u>dl</u>-volkensiflavone (V) (mixed m.p., IR and NMR spectra). D-Glucose was identified by PPC and TLC. The NMR spectrum (acetone-d_6) of VI







β-D-glc (R =H, Ac, Me)

I,II : R =R_1 =H, R_2 =OH
III : R =R_1 =H, R_2 =OMe
IV : R =H, R_1 =β-D-glc, R_2 =OH
V : R =R_1 =R_2 =H
Va : R =R_1 =Me, R_2 =H
VI : R =R_2 =H, R_1 =β-D-glc
VIa : R =Me, R_1 =β-D-glc(Me), R_2 =H

VII : R =R_1 =R_2 =R_3 =H
VIII: R =R_1 =R_2 =H, R_3 =OH
IX : R =R_2 =H, R_1 =R_3 =OH
X : R =R_1 =H, R_2 =β-D-glc, R_3 =OH
Xa : R =Ac, R_1 =H, R_2 =β-D-glc(Ac), R_3 =OAc
XI : R =R_2 =R_3 =H, R_1 =OH

showed several signals in the region between 1.85 and 6.70, two broad singlets at -3.15 and -2.23 due to hydrogen-bonded hydroxyl groups, broad signals at 0.5 and 1.5 due to other hydroxyl groups, and all the glucose protons showed broad signals between 4.5 - 6.7. Methylation of VI with methyl iodide and silver oxide in dimethylformamide (Kuhn's method)$^{10)}$ gave a permethyl ether (VIa) as a colourless syrup, which was negative for the ferric chloride test. Hydrolysis of VIa with boiling ethanolic potassium hydroxide gave *p*-methoxybenzoic acid and anisaldehyde. The NMR spectrum (acetone-d_6) of VIa showed signals due to nine methoxyl groups between 6.08 and 6.70 and multiplets at 4.90 and 5.25 - 7.0 due to C-1 and C-2 to C-6 protons of the glucose moiety. Aromatic protons of the ring E and the ring B were identical with those of hexamethyl volkensiflavone (Va). The remaining three aromatic protons showed signals in the region of 3.40 - 3.84 of the spectrum. Two of them appeared as *meta*-coupled doublets (J =2.5cps) at 3.65 and 3.75, which were assigned to C-8 and C-6 protons respectively, whereas another proton at C-6" showed a singlet at 3.61. A singlet at 3.45 can be assigned to C-3" proton. Thus, the NMR spectrum is in agreement with the structure VIa$^{2)}$. These NMR data together with other experimental results described in this paper indicate the structure of spicataside (VI) to be 7"-β-glucoside of V.

The occurrence of the biflavanones and the flavanone-flavones in *Garcinia* plants is interesting from the chemotaxonomic and biogenetic points of view.

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