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FLAVONOIDS FROM *IRYANTHERA SAGOTIANA*

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Key Word Index—*Iryanthera sagotiana*; Myristicaceae; dihydrochalcones; flavonols; flavanonols.

Abstract—Leaves and inflorescences of *Iryanthera sagotiana* were found to contain three known dihydrochalcones, two flavonol rhamnosides, four flavanonol rhamnosides, one dihydrocoumaric acid, besides the new 3,′3‴-bis-2′,4′,6′-trihydroxy-4-methoxydihydrochalcone and 4′,6′-dihydroxy-4-methoxydihydrochalcone-2′-*O*-β-D-glucopyranoside. © 1997 Elsevier Science Ltd

INTRODUCTION

There have been very few chemical studies of neotropical members of the Myristicaceae. Lignans, neolignans and flavones have been reported from *Virola surinamensis* Warb. [1] but most *Virola* species contain only lignans and neolignans in their leaf tissue. However, inflorescences of the neotropical tree *V. venosa* Warb. were found to produce only flavones [2]. The bark of *Iryanthera sagotiana* (Benth.) Warb., another member of the Myristicaceae collected at Belém was reported to contain dihydrochalcones **1** and **4** [3]. The present work describes the isolation and identification of four dihydrochalcones (**1–3** and **5**), two flavonol 3-rhamnosides (**10**, **11**), four flavanonol 3-rhamnosides (**12–15**) and dihydro-*p*-coumaric acid (**7**) from the inflorescence of *I. sagotiana*. Leaves of *I. sagotiana* afforded **1**, **3**, **6**, **10–13**. One dihydrochalcone glycoside (**5**) and a dihydrochalcone dimer (**6**) are new compounds.

RESULTS AND DISCUSSION

Dihydrochalcones **1–3** and dihydro-*p*-coumaric acid (**7**) were identified by comparison of ^{1}H and ^{13}C NMR data reported in the literature [1, 8, 9]. The location of hydroxyl and methoxyl groups of **1–3** was evidenced additionally through analysis of the MS ions.

Structural elucidation of **5** was based on ^{1}H and ^{13}C NMR and EIMS spectral data. Its ^{1}H NMR spectrum showed two triplets at 2.79 δ (2H, J = 7.4 Hz) and 3.24 δ (2H, J = 7.4 Hz), and two doublets at 6.78 δ (2H, J = 8.5 Hz) and 7.14 δ (2H, J = 8.5 Hz) of a dihydrochalcone with a *p*-substituted B-ring. A doublet at 4.87 δ (1H, J = 7 Hz) in addition to multiplets between 3.0 and 4.0 δ suggested the presence of a β-*O*-glucosyl unit linked to the dihydrochalcone. This was confirmed by PND and DEPT 135° ^{13}C NMR spectra which showed one signal for a methylene carbon (61.0 δ), four signals around 70 δ and one signal at 100.9 δ for an anomeric carbon. The mass spectrum showed peaks with m/z 121 (983) (**8**) and 134 (20) (**9**) which placed the methoxyl group in ring B. The ^{1}H NMR data also disclosed an unsymmetrical A ring ($\delta_{H3'}$ = 5.89 and $\delta_{H5'}$ = 5.66) which sited the glucosyl unit at C-2′. Thus, **5** is identified as 4′,6′-dihydroxy-4-methoxydihydrochalcone 2′-*O*-β-D-glucoside, a new compound.

The ^{1}H NMR spectrum of **6** showed two triplets at 2.8 and 3.5 δ, which are typical of a dihydrochalcone moiety. It also showed two doublets at 6.70 and 7.05 δ of a *p*-substituted aromatic ring and a singlet at 5.79 δ integrating for only one proton, which suggested the presence of two dihydrochalcone units linked through C-3′. This was confirmed by the ^{13}C NMR spectrum which showed only one signal around 90 δ for an aromatic carbon bearing one proton, which could be assigned to C-5′ (94.5 δ). The signal due to C-3′ was shifted downfield (115.0 δ) when compared to the dihydrochalcone monomer and appeared as a quaternary carbon as evidenced by a DEPT 135° experiment. These findings, allied to MS data, which established the methoxyl group position through observation of peaks with m/z 121 (100) and m/z 134 (40) for fragments **8** and **9**, respectively, confirmed the proposed structure for the new dihydrochalcone dimer as **6**. Measurement of specific rotation ($[\alpha_D^{21}] = -8°$) led to the assumption of a non-planar

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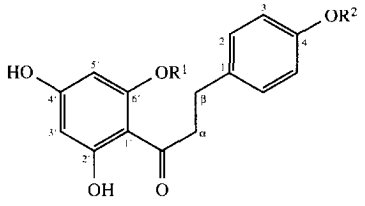

	R^1	R^2
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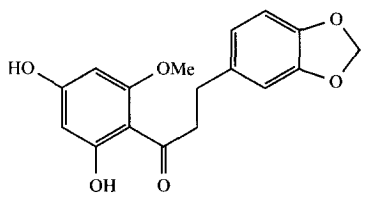
2	H	Me
3	Me	H



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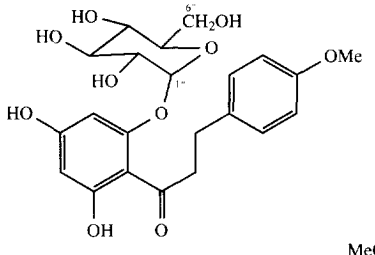

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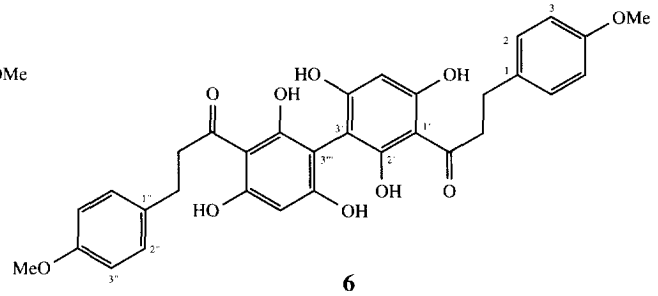

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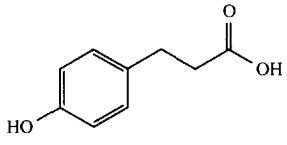

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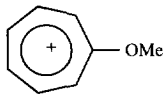

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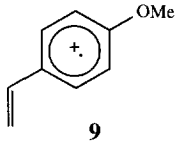

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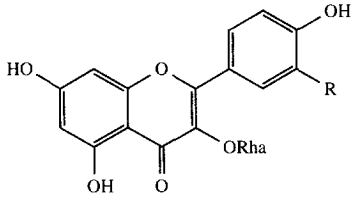
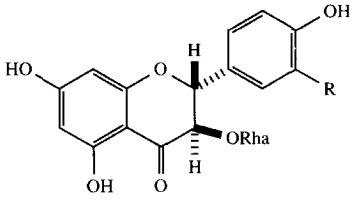
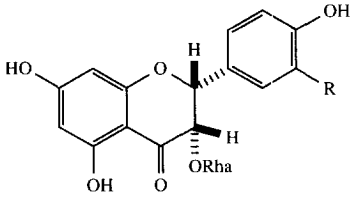


10	R = H	**12**	R = H
11	R = OH	**13**	R = OH





14	R = H
15	R = OH

and hence chiral molecule, probably due to the sterical crowding caused by the pentasubstituted aromatic rings that bear the 3′,3‴ linkage between the two dihydrochalcone moieties that form this dimer. The first example of a dimeric dihydrochalcone was described by Drews and Hudson from *Brackenridgea zanguebarica* (Ochnaceae) [10] and is composed of two C_α–C_α linked davidigenin units. Compound **6** is the second example of a dimeric dihydrochalcone and the first example of a biflavonoid in the Myristicaceae.

Compounds **10** (afzelin) and **11** (quercitrin) were identified through comparison of their ^1H and ^{13}C NMR spectra with literature data. The observed deshielding of C-2 (10.5 ppm for **10** and 10.0 ppm for **11**) and C-4 (1.9 ppm for **10** and 1.4 ppm for **11**) and shielding of C-3 (1.2 ppm for **10** and 1.4 ppm for **11**) of afzelin and quercitrin when compared with their aglycone moieties, kaempferol and quercetin, respectively, are in agreement with literature data [11].

Identification of the dihydroflavonol 3-*O*-α-L-rhamnosides: **12** (engeletin), **13** (astilbin), **14** (isoengeletin) and **15** (isoastilbin), including the chirality of the aglycones was made by comparison of their ^1H NMR shifts and coupling constants of H-2 and H-3 of the aglycone and H-1″ and H-6″ of the rhamnosyl moiety with literature data [4, 7]. Compounds **14** and **15** could not be isolated probably due to isomerization reactions taking place in the slightly acidic media used in HPLC isolation procedures followed by heating on a vacuum rota-evaporator [4]. However identification was possible from the chemical shifts obtained by spectra subtraction using spectral data for mixtures of **12** plus **14** and **13** plus **15** and that for pure **12** and **13**. The observed relative deshielding of C-3 (2.6 ppm)

and shielding of C-2 (3.2 ppm), C-4 (3.4 ppm) and C-1′ (2.6 ppm) of engeletin when compared to the aglycone moiety aromadendrin are in agreement with literature data [11].

It is of interest to note that the inflorescences of *I. sagotiana* have been found to contain only flavonoids, compounds, which absorb and emit strongly in the UV/VIS region of the electromagnetic spectrum and could possibly play an ecological role in the attraction of pollinators to the flowers.

EXPERIMENTAL

General. CC was carried out on Amberlite XAD-2 (100–250 μm) (Aldrich) and Sephadex LH-20 (25–100 μm) (Pharmacia). Flash chromatography was carried out on silica gel 60 (40–63 μm) (Merck). The ^{1}H NMR (200 MHz) and ^{13}C NMR (50 MHz) spectra were recorded on a Bruker-AC 200 in $CDCl_3$ and/or DMSO-d_6 with TMS as int. standard. EIMS was obtained at 70 eV on HP 5988-A.

Isolation of constituents. Inflorescences of *I. sagotiana* (Benth.) Warb. were collected near Belém, Pará State and identified by Dr William Rodrigues (voucher 133693, Herbarium João Murça Pires, Museu Paraense Emílio Goeldi, Belém, PA). They were dried, powdered and extracted with hexane and then EtOH. The ethanolic extract (8 g) was dissolved in 10% MeOH–H_2O and extracted successively with hexane, $CHCl_3$ and EtOAc. The $CHCl_3$ phase extract (5 g) was passed through an Amberlite XAD-2 column and eluted first with H_2O followed by MeOH–H_2O sols containing 20, 40, 60, 80 and finally 100% MeOH in order to remove sugars and other very polar compounds. Frs containing glycosylated flavonoids were submitted to flash chromatography over silica gel ($CHCl_3$–MeOH–H_2O, 79:18:3) followed by Sephadex LH-20 affording pure **7** (3 mg), **10** (30 mg) and **11** (38 mg) and two mixts of diastereoisomeric glycosylated dihydroflavonols: **12**+**14** and **13**+**15**. These mixts were purified by HPLC (RP-18 column 250 × 22 mm, 10 μm Perkin-Elmer; MeOH (HOAc 3%)–H_2O 53:47) to yield **12** (8 mg) and **13** (6 mg). Less polar frs from an XAD-2 column submitted to flash chromatography over silica gel (hexane–$CHCl_3$–MeOH, gradient) gave **1** (90 mg). Remaining frs submitted to HPLC (RP-18 column, 250 × 22 mm, 10 μm Perkin-Elmer; MeOH (HOAc 2%)–H_2O 65:35) gave **2** (92 mg) and **3** (3 mg) while a polar fr. submitted to HPLC (RP-18 column, 250 × 22 mm, 10 μm Perkin-Elmer; MeOH (HOAc 2%)–H_2O 6:4) gave **5** (3 mg).

Air-dried powdered leaves of *I. sagotiana* (220 g), collected from the same specimen, were submitted to the same procedure as described for inflorescences and yielded **1**, **3** and **10**–**13**. Fractionation through flash chromatography over silica gel (hexane–$CHCl_3$–MeOH, gradient) followed by purification by HPLC (RP-18 column, 250 × 22 mm, 10 μm Perkin-Elmer; MeOH (HOAc 2%)–H_2O 6:4) of a semi-polar fr. gave **6** (3 mg).

*4′,6′-Dihydroxy-4-methoxydihydrochalcone 2′-O-β-*D*-glucopyranoside* (**5**). ^{1}H NMR (200 MHz, DMSO-d_6): δ 7.14 (2H, *d*, *J* = 8.5 Hz, H-2 and H-6), 6.78 (2H, *d*, *J* = 8.5 Hz, H-3 and H-5), 5.89 (1H, *sl*, H-3′), 5.66 (1H, *sl*, H-5′), 4.87 (2H, *d*, 7.0 Hz, H-1″), 3.8–3.0 (5H, *m*), 3.24 (2H-α, *m*), 2.79 (2 H-β, *m*). ^{13}C NMR (50 MHz, DMSO-d_6): δ 134.1 (C-1), 129.7 (C-2, C-6), 114.0 (C-3, C-5), 158.0 (C-4), 104.0 (C-1′), 161.2 (C-2′), 98.0 (C-3′), 166.3 (C-4′), 96.2 (C-5′), 157.0 (C-6′), 44.8 (C-a), 29.7 (C-b), 100.9 (C-1″), 69.9, 73.7, 77.2, 77.5 (C-2″, C-3″, C-4″, C-5″), 61.0 (C-6″). EIMS (70 eV) *m/z* (rel. int.): $[M]^+$-Glc+1 288 (100%), $[M]^+$-Glc-1 286 (33), 270 (19), 256 (21), 180 (17), 153 (5), 134 (19), 121 (93).

3′,3‴-bis-2′,4′,6′-Trihydroxy-4-methoxydihydrochalcone (**6**) $[\alpha]_D^{21} = -8°$ (MeOH, *c* = 0.16). ^{1}H NMR (200 MHz, DMSO-d_6): δ 7.05 (2H, *d*, *J* = 8.4 Hz, H-2 and H-6), 6.70 (2H, *d*, *J* = 8.4 Hz, H-3 and H-5), 5.79 (*s*, H-5), 3.5–3.6 (2H-α, *m*), 2.9–2.7 (2H-β, *m*). ^{13}C NMR (50 MHz, DMSO-d_6): δ 133.4 (C-1), 128.8 (C-2, C-6), 113.1 (C-3, C-5), 157.1 (C-4), 103.7 (C-1′), 163.9 (C-2′), 115.0 (C-3′), 164.0 (C-4′), 94.5 (C-5′), 163.9 (C-6′), 45.0 (C-α), 29.2 (C-β), 203.9 (C=O), 54.6 (CH_3O). EIMS (70 eV) *m/z* (rel. int.): 180 (10), 163 (14), 153 (8), 135 (18), 134 (40), 121 (100). FAB MS: $[M]^+$ + 1 575 (14%).

Engeletin (**10**). ^{13}C NMR (50 MHz, DMSO-d_6): δ 81.8 (C-2), 76.3 (C-3), 195.4 (C-4), 163.7 (C-5), 96.4 (C-6), 167.4 (C-7), 95.4 (C-8), 162.5 (C-9), 101.3 (C-10), 126.8 (C-1′), 129.4 (C-2′, C-6′), 115.5 (C-3′, C-5′), 158.2 (C-4′), 100.6 (C-1″), 69.3, 70.4, 70.7, 71.9 (C-2″, C-3″, C-4″, C-5″), 18.0 (C-6″).

Isoengeletin (**12**). ^{13}C NMR (50 MHz, DMSO-d_6): δ 80.3 (C-2), 75.6 (C-3), 193.0 (C-4), 164.3 (C-5), 96.6 (C-6), 167.7 (C-7), 95.5 (C-8), 162.9 (C-9), 100.4 (C-10), 126.1 (C-1′), 128.1 (C-2′, C-6′), 115.1 (C-3′, C-5′), 157.5 (C-4′), 98.8 (C-1″), 70.4, 70.5, 71.4, 73.4 (C-2″, C-3″, C-4″, C-5″), 17.8 (C-6″).

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