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IRIDOID GLUCOSIDES FROM *AVICENNIA GERMINANS*

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Key Word Index—*Avicennia germinans*; Avicenniaceae/Verbenaceae; iridoid glucosides; 2′-cinnamoyl mussaenosidic acid; 2′-caffeoyl mussaenosidic acid; verbascoside.

Abstract—A new iridoid glucoside, namely 2′-caffeoyl mussaenosidic acid has been isolated from leaves of *Avicennia germinans* along with the known compounds 2′-cinnamoyl mussaenosidic acid and verbascoside. The structure of the new compound was established by spectroscopic methods.

INTRODUCTION

The systematics of the mangrove genus *Avicennia* has been controversial for a long time, mainly with regard to its relationship to other taxa. The genus is now better known [1], particularly with regard to the Australasian species [2], and some contemporary botanists consider *Avicennia*, with about 14 species, to be a member of the family Verbenaceae [3] although others have a different opinion [4].

The distribution of iridoids and compounds similar to verbascoside have been shown to be chemotaxonomically useful characters [5–7], and since iridoid glucosides have been reported from *A. officinalis* [4] and *A. marina* [8], we have now investigated *A. germinans* (L.) Stearn from Guyana for the presence of glucosides.

RESULTS AND DISCUSSION

The crude aqueous extract of the plant showed the presence of several compounds although in very small amounts. After extensive chromatographic separation (see Experimental) three compounds were obtained in the pure state. One compound was proved by comparison with an authentic specimen to be the known caffeic ester derivative verbascoside (**1**), earlier reported from *A. marina* [9]. The NMR spectra of the other compounds, **2** and **3**, showed signals characteristic of an iridoid glucoside esterified with an aromatic acid. The NMR spectra of **2** proved to be almost identical with those reported [8] for 2′-cinnamoyl-mussaenoside except for the presence of the methyl group present in the latter. Compound **2** is thus 2′-cinnamoyl mussaenosidic acid, and this compound has earlier been reported from *A. officinalis* [4] and *A. marina* [8].

The ^{1}H NMR spectrum of **3** was similar to that of **2**, except for a difference in the aromatic signals, which signified the presence of a caffeoyl moiety in **3**. This was confirmed by the ^{13}C NMR spectrum, where a caffeoyl moiety could be distinguished together with signals corresponding to the aglucone of mussaenosidic acid [7]. The remaining six signals from a substituted β-glucopyranosyl moiety were almost identical with those found for **2** with the diagnostic downfield shift of 1.3 ppm for C-2′ and upfield shifts of 2.0 and 3.2 ppm, respectively, for C-1′ and C-3′ when compared to the spectrum of mussaenoside [7]. This proves that the caffeic acid is esterified with the C-2′ oxygen of the mussaenosidic acid moiety in **3**. Characteristically, the H-2′ signal in both **2** and **3** is found at δ4.8, *ca* 1.6 ppm downfield from the usual position of this proton. To our knowledge, this is the first report of 2′-caffeoyl mussaenosidic acid.

Iridoids are very useful taxonomic markers and the chemical relationship between *Avicennia* and other taxa in Lamianae is confirmed by this character and by the presence of verbascoside in the genus.

EXPERIMENTAL

General. IR: KBr disks; MS: Nermag R-10-10 by DCI with NH_3 or FAB; ^{1}H and ^{13}C NMR: 250 and 62.5 MHz, respectively, in CD_3OD, using the solvent peaks at δ3.31 and 49.0 as standards. Leaves of *Avicennia germinans* were collected in the mangrove (Cayenne) in Guyana. A

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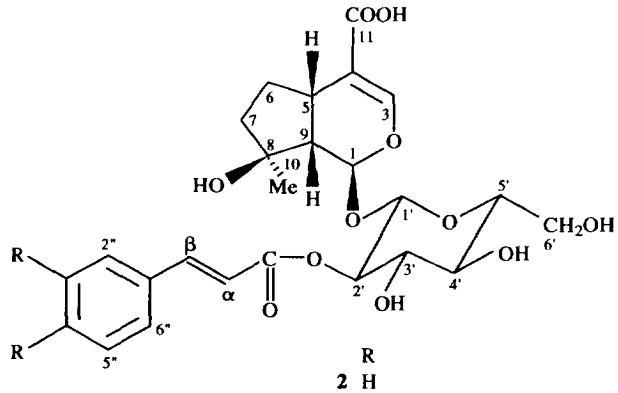

R
2 H
3 OH

voucher specimen (No. 2076) was identified by Dr F. Blasco and is deposited in the Herbarium of the Institute for the International Map of the Vegetation (France).

Dry leaves (500 g) were successively extracted with H_2O and MeOH (70%) and the extracts combined and lyophilized followed by chromatography on a XAD_4 column. The fr. (6 g) eluted with H_2O–MeOH was re-chromatographed on silica gel eluting successively with CH_2Cl_2–MeOH (4:1), CH_2Cl_2–MeOH–H_2O (40:10:1) and CH_2Cl_2–MeOH–H_2O (80:20:3). The first fr. contained **2** (162 mg), the following two frs were mixts of verbascoside (**1**) and **3** (213 mg). Rechromatography on a polyamide column with a gradient of H_2O–MeOH (0–100% MeOH) gave 2′-cinnamoyl mussaenosidic acid (**2**) (31 mg) in the (1:1) fractions, while the mixt. of **1** and **3** was chromatographed with water on Sephadex LH 20 and yielded **1** (15 mg), 2′-caffeoyl mussaenosidic acid **3** (18 mg) was eluted with H_2O–MeOH (7:3).A final purification on a RP_{18} Bond Elut cartridge was performed with a gradient of H_2O–MeOH.

2′-Cinnamoyl mussaenosidic acid (**2**) Syrup. IR ν_{max}^{neat} cm^{-1}: 1628 (CO_2H) and 1698 (ester C=O); MS, positive FAB, glycerol matrix (*m/z*): 539 $[M + Na]^+$. ^1H NMR (only significant H's): δ7.85 (*d*, J = 16 Hz, b-H), 7.78 (2H, arom.), 7.39 (3H, arom.), 7.28 (*s*, H-3), 6.45 (*d*, J = 16 Hz, a-H), 5.49 (*d*, J = 3 Hz, H-1), 4.9 (*d*, partly covered by HDO, H-1′), 4.81 (*t*, J = 8 Hz, H-2′), 1.27 (*s*, Me-10), close to the values reported for the methyl ester [8], except for the value given for H-3 (*d*, 6.3), which is apparently a misprint. ^{13}C NMR: δ95.2 (C-1), 148.6 (C-3), 114.2 (C-4), 31.4 (C-5), 30.3 (C-6), 41.4 (C-7), 79.9 (C-8), 52.6 (C-9), 24.4 (C-10), 170.2 (C-11), 97.8 (C-1′), 76.0 (C-2′), 75.0 (C-3′), 71.8 (C-4′), 78.6 (C-5′), 62.8 (C-6′), 167.6 (CO″), 118.8 (C-a″), 148.6 (C-b″), 136.0 (C-1″), 130.0 (C-2″, C-6″), 129.4 (C-3″, C-5″), 131.4 (C-4″). Methyl ester: MS *m/z*, DCI, NH_3: 538 $[M + NH_4]^+$.

2′-Caffeoyl mussaenosidic acid (**3**). Syrup. UV λ_{max}^{MeOH} nm: 329; IR ν_{max}^{neat} cm^{-1}: 1628 (CO_2H) and 1698 (ester C=O); MS, DCI NH_3 (*m/z*): 556 $[M + NH_4]^+$, 521 $[M - H_2O]^+$. ^1H NMR: δ7.51 (*d*, J = 16 Hz, β-H), 7.27 (*s*, H-3), 7.03 (*d*, J = 2 Hz, H-2″), 6.93 (*dd*, J = 2 and 8 Hz, H-6″), 6.77 (*d*, J = 8 Hz, H-5″), 6.20 (*d*, J = 16 Hz, a-H), 5.47 (*d*, J = 3 Hz, H-1), 4.9 (covered by HDO, H-1′), 4.83 (*t*, J = 9 Hz, H-2′), 3.3–4.0 (5H, C-3′–C-6′), 3.0 (*m*, H-5), 2.2 (2H, H-9 and H-7), 1.3–1.7 (3H, H-7 and 2 × H-6), 1.27 (*s*, Me-10); ^{13}C NMR: δ95.1 (C-1), 151.2 (C-3), 114.2 (C-4), 31.5 (C-5), 30.3 (C-6), 41.3 (C-7), 80.0 (C-8), 52.6 (C-9), 24.4 (C-10), 170.4 (C-11), 97.8 (C-1′), 76.1 (C-2′), 74.8 (C-3′), 71.8 (C-4′), 78.6 (C-5′), 62.8 (C-6′), 168.2 (CO″), 115.1 (C-a″), 147.2 (C-b″), 128.0 (C-1″), 115.4 (C-2″), 146.7 (C-3″), 148.5 (C-4″), 116.5 (C-5″), 123.0 (C-6″).

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