Available online at www.sciencedirect.com
SCIENCE DIRECT®
Phytochemistry 66 (2005) 589–597
PHYTOCHEMISTRY
www.elsevier.com/locate/phytochem

Further constituents from the bark of *Tabebuia impetiginosa*

Tsutomu Warashina [a,*], Yoshimi Nagatani [b], Tadataka Noro [a]

[a] *Institute for Environmental Sciences, University of Shizuoka, Yada 52-1, Shizuoka 422-8526, Japan*
[b] *Herbario Goro Hashimoto, Centro de Pesquisas de Historia Natural, Rua jaime Ribeiro Wright, 618 Sao Paulo/SP 08260-070, Brasil*

Received 27 July 2004; received in revised form 27 October 2004

Abstract

Further study on the constituents from the bark of *Tabebuia impetiginosa* (Mart. ex DC) Standley afforded twelve compounds, consisting of four iridoid glycosides, one phenylethanoid glycoside, five phenolic glycosides, and one lignan glycoside, along with seven known compounds. The structures of these compounds were determined based on the interpretation of their NMR and MS measurements and by chemical evidence.

Keywords: *Tabebuia impetiginosa* (Mart. ex DC) Standley; Bignoniaceae; Phenylethanoids; Phenolic glycosides; Lignans; Iridoids

1. Introduction

The bark of *Tabebuia impetiginosa* (Mart. ex DC) Standley has been used traditionally for treating diabetes, ulcers, and syphilis (Hashimoto, 1996). In a previous paper, we reported on some compounds from the bark of this plant (Warashina et al., 2004). In the present paper, we describe the isolation and structural determination of nineteen compounds from the polar fraction of the methanol extract of the bark.

2. Results and discussion

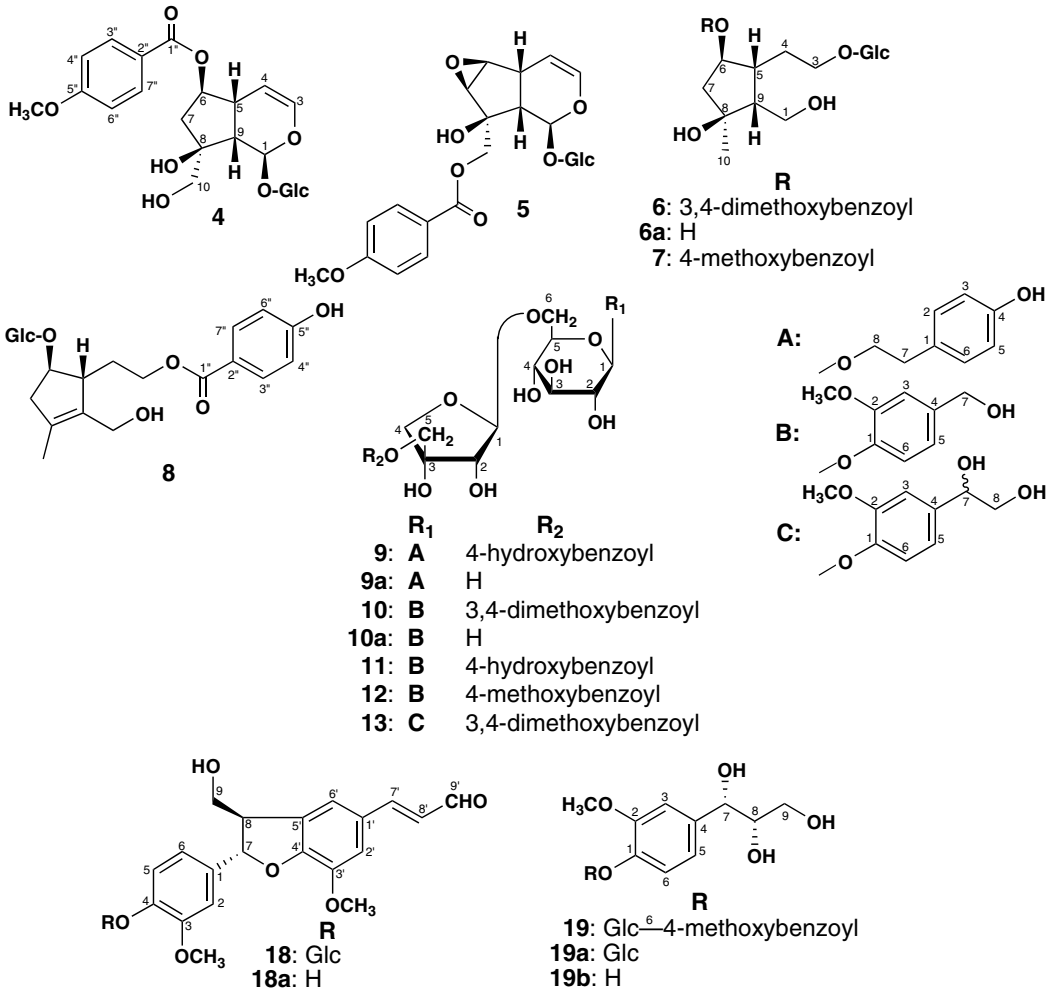
Details of the extraction of the bark of *T. impetiginosa* (Mart. ex DC) Standley was given in the previous paper (Warashina et al., 2004). The aqueous MeOH (1:1) eluate from the porous polymer gel "Mitsubishi Diaion HP-20" column was concentrated and the residue subjected to silica gel column chromatography followed by semi-preparative HPLC to give compounds **1**–**19**.

Compounds **1**–**3** were the known iridoid glucosides identified as 6-*O*-(4-hydroxybenzoyl)-ajugol (**1**) (Nakano et al., 1993; Nishimura et al., 1989), 6-*O*-vanilloyl-ajugol (**2**) (Nishimura et al., 1989), and 6-*O*-(4-hydroxybenzoyl)-6-epiaucubin (**3**) (Bianco et al., 1982). Compounds **14**–**17** were the known lignan glycosides identified as (+)-lyoniresinol 3a-*O*-β-D-glucopyranoside (**14**) (Achenbach et al., 1992), dihydrodehydrodiconiferyl alcohol 9-*O*-β-D-glucopyranoside (**15**) (Abe and Yamauchi, 1986; Otsuka et al., 2000), dihydrodehydrodiconiferyl alcohol 9′-*O*-β-D-glucopyranoside (**16**) (Takeda et al., 1998), and dihydrodehydrodiconiferyl alcohol 4-*O*-β-D-glucopyranoside (**17**) (Matsuda et al., 1996).

Compound **4** was proposed to have the molecular formula $C_{23}H_{30}O_{12}$ based on high resolution (HR)-FABMS [*m*/*z* 521.1606 $[M + Na]^+$]. Because the ^{13}C NMR spectrum of **4** showed nine carbon signals due to the aglycone, in addition to six carbon signals of the β-D-glucopyranosyl group and eight carbon signals of the acyl group (see Table 1), compound **4** was presumed to be an acylated-iridoid glucoside. The ^{1}H and ^{13}C NMR spectroscopic data of **4** were consistent with

* Corresponding author. Tel./fax: +81 54 264 5791.
E-mail address: warashin@sea.u-shizuoka-ken.ac.jp (T. Warashina).

0031-9422/$ - see front matter © 2005 Elsevier Ltd. All rights reserved.
doi:10.1016/j.phytochem.2005.01.005



those of 6-*O*-(4-hydroxybenzoyl)-5,7-bisdeoxycynanchoside (Iwagawa et al., 1991), except for the ester moiety; thus **4** was initially considered to be 6-*O*-acyl-5,7- bisdeoxycynanchoside. Since the ^{1}H NMR spectrum of **4** revealed the presence of an aromatic AA′XX′ system [δ 8.01 (2H, *d*, $J = 9.0$ Hz), 6.98 (2H, *d*, $J = 9.0$ Hz)] and a methoxyl group [δ 3.86 (3H, *s*)], the ester moiety of **4** was deduced to be a 4-methoxybenzoyl group. This was confirmed by alkaline hydrolysis of **4**. Based on the above evidence, the structure of **4** was determined to be 6-*O*-(4-methoxybenzoyl)-5,7-bisdeoxycynanchoside.

The molecular formula of compound **5** was $C_{23}H_{28}O_{12}$ based on HR-FABMS [*m*/*z* 519.1482 $[M + Na]^{+}$]. This compound was also considered to be an esterified iridoid glucoside, since the ^{13}C NMR spectrum revealed signals for the aglycone, the glucose moiety and a 4-methoxybenzoyl group (see Table 1). A comparison of the ^{13}C NMR spectroscopic data of **5** with those of 6β, 7β-epoxysplendoside peracetate (Damtoft et al., 1981) and consideration of its molecular formula indicated that **5** contained an epoxide ring. The HMQC spectrum showed the oxygenated methine carbon and proton signals of the epoxide ring at δ 58.9; 3.48 (1H, *d*, $J = 2.0$ Hz) and δ 60.8; 3.58 (1H, *d*, $J = 2.0$ Hz). These signals were assigned to the C-6/ H-6 and C-7/H-7 on the basis of the following HMBC correlations: δ 58.9 (C-6) and δ 4.86 (H-4), 3.12 (H-5); δ 60.8 (C-7) and δ 3.12 (H-5), 4.55 (H-10), 4.36 (H-10); and δ 3.58 (H-7) and δ 79.4 (C-8), 46.9 (C-9). The observation of NOEs between δ 3.58 (H-7) and 4.36 (H-10); and δ 4.55 (H-10) and 5.75 (H-1) indicated that H-7 was alpha and hence the epoxide ring was beta. The position of attachment of the ester group was deduced from HMBC correlations between the C-10 methylene protons and the 4-methoxybenzoyl carbonyl group. Thus, the structure of **5** was determined as shown, and named 10-*O*-(4-methoxybenzoyl)-impetiginoside A.

The molecular formulae of compounds **6** and **7** were indicated to be $C_{24}H_{36}O_{12}$ and $C_{23}H_{34}O_{11}$ from the results of HR-FABMS. The ^{13}C and ^{1}H NMR spectra of both compounds suggested that they were iridoid congeners with different ester functions. Alkaline hydrolysis of **6** and **7**, respectively, afforded 3,4-dimethoxy-

Table 1
^{13}C NMR Spectroscopic data of compounds **4**–**6**, **8**–**10**, **13**, **18**, and **19**

Carbon no.	**4**	**5**	**6**	**8**[A]	**9**	**10**	**13**	**18**	**19**
Aglycone moiety									
C-1(C-1′)	93.4	92.4	59.2	56.8	130.8	147.2	147.3	137.5 (129.8)	147.2
C-2(C-2′)	–	–	–	–	130.9	150.9	150.8	111.4 (114.5)	150.7
C-3(C-3′)	141.7	142.0	69.8	63.7	116.1	112.7	112.0	151.1 (146.1)	112.5
C-4(C-4′)	104.3	101.5	30.1	31.3	156.7	137.8	138.4	147.9 (152.9)	138.5
C-5(C-5′)	40.8	33.1	44.0	51.3	116.1	120.8	120.2	118.3 (131.1)	120.4
C-6(C-6′)	80.8	58.9	82.1	82.3	130.9	118.1	118.1	119.5 (120.0)	117.7
C-7(C-7′)	42.8	60.8	48.7	44.4	36.4	64.9	75.6	89.6 (155.9)	75.8
C-8(C-8′)	82.5	79.4	80.5	132.3[a]	72.1	–	68.6[a]	54.9 (127.2)	77.4
C-9(C-9′)	51.6	46.9	55.0	136.8[a]	–	–	–	64.7 (196.1)	64.2
C-10	68.0	68.3	25.2	13.9	–	–	–	–	–
C-OMe	–	–	–	–	–	56.7[a]	56.7[b]	56.8	56.7
	–	–	–	–	–	–	–	56.9	–
Sugar moiety									
Glc-1	99.6	99.5	104.4	103.8	104.4	103.0	103.0	102.8	102.7
Glc-2	74.8	74.7	75.1	75.1	75.1[a]	74.9[b]	74.9[c]	74.9	74.9
Glc-3	78.3[a]	78.2[a]	78.0[a]	78.5[b]	78.1	77.9	77.9	78.2[a]	77.8
Glc-4	71.7	71.7	71.6	71.8	71.8	71.7	71.6	71.4	72.0
Glc-5	78.0[a]	78.0[a]	77.8[a]	78.4[b]	76.9	77.0	77.1	77.9[a]	75.7
Glc-6	62.9	62.8	62.7	63.1	68.6	68.7	68.7[a]	62.6	65.1
Api-1	–	–	–	–	110.7	110.8	110.8	–	–
Api-2	–	–	–	–	78.5	78.7	78.7	–	–
Api-3	–	–	–	–	79.0	79.0	79.0	–	–
Api-4	–	–	–	–	75.0[a]	75.0[b]	75.0[c]	–	–
Api-5	–	–	–	–	67.5	67.6	67.7	–	–
Ester moiety									
C-1″	167.8	167.8	168.2	166.8	167.9	167.7	167.7	–	167.7
C-2″	123.9	123.5	124.3	122.0	122.0	123.5	123.5	–	123.5
C-3″	132.7	132.8	113.7	132.4	133.0	113.8	113.8	–	132.7
C-4″	114.8	114.9	150.2	116.1	116.3	150.2	150.2	–	114.9
C-5″	165.2	165.3	154.8	163.4	163.7	155.0	155.0	–	165.3
C-6″	114.8	114.9	112.0	116.1	116.3	112.1	112.1	–	114.9
C-7″	132.7	132.8	125.1	132.4	133.0	125.2	125.2	–	132.7
C-OMe	56.0	56.0	56.6	–	–	56.6[a]	56.6[b]	–	56.1
	–	–	56.5	–	–	56.5[a]	56.5[b]	–	–

Measured in CD_3OD solution at 35 °C.
[a–c] : Assignments may be interchangeable in each column.
[A] Measured in C_5D_5N solution at 35 °C.

benzoic acid and 4-methoxybenzoic acid, together with **6a**. The production of D-glucose by acid hydrolysis of **6a** suggested that it was an iridoid glucoside. The ^{13}C NMR spectral data of **6a** was similar to those of crescentin IV (Kaneko et al., 1997), with glycosylation shifts observed around the C-3 position (Kasai et al., 1979). Thus, **6a** was concluded to be crescentin IV 3-*O*-β-D-glucopyranoside. In its HMBC spectrum, **6** showed correlations between δ 168.2 (C-1″) and δ 5.21 (H-6); δ 69.8 (C-3) and δ 4.23 (H-1 of β-D-glucopyranose); and δ 104.4 (C-1 of β-D-glucopyranose) and δ 3.97 (H-3), 3.66 (H-3). On the basis of the above evidence, the structures of **6** and **7** were determined to be 6-*O*-(3,4-dimethoxybenzoyl)-crescentin IV 3-*O*-β-D-glucopyranoside and 6-*O*-(4-methoxybenzoyl)-crescentin IV 3-*O*-β-D-glucopyranoside, respectively.

The molecular formula of compound **8** was $C_{22}H_{30}O_{10}$ based on a HR-FABMS measurement. Signals due to the 4-hydroxybenzoyl and β-D-glucopyranosyl groups were exhibited in the ^{1}H and ^{13}C NMR spectra. On comparison of the ^{13}C NMR spectral data of the aglycone moiety in **8** with those of **6**, tetra-substituted double bond signals were seen at δ 136.8 and 132.3 in **8**, instead of an oxygenated quaternary carbon signal (C-8) and a methine carbon signal (C-9) in **6**. These observations suggested that the aglycone of **8** was 10-deoxyeucommiol (Bianco et al., 1981). The locations of the 4-hydroxybenzoyl and β-D-glucopyranosyl groups were established by correlations in the HMBC spectrum between δ 166.8 (C-1″) and δ 4.80 (H-3), 4.60 (H-3); and δ 82.3 (C-6) and δ 4.93 (H-1 of β-D-glucopyranose). Thus, the structure of **8** was proposed to be 3-*O*-(4-hydroxybenzoyl)-10-deoxyeucommiol 6-*O*-β-D-glucopyranoside.

Compound **9** was suggested to have the molecular formula $C_{26}H_{32}O_{13}$ based on HR-FABMS [*m*/*z*

575.1719 [M + Na]$^+$]. The ^{1}H NMR spectra of **9** showed the presence of methylene protons, a glucopyranosyl anomeric proton, an apiofuranosyl anomeric proton and aromatic protons. Alkaline hydrolysis of **9** afforded osmanthuside H (**9a**) (Sugiyama and Kikuchi, 1993) and 4-hydroxybenzoic acid, which were identified by the HPLC analysis with authentic samples. A comparison of the ^{1}H NMR spectroscopic data of **9** and **9a** showed downfield shifts of the C-5 methylene protons of the apiofuranose 6 moiety [δ 4.08 (1H, *d*, J = 10.0 Hz), 3.87 (1H, *d*, J = 10.0 Hz)]. This result suggested that the 4-hydroxybenzoyl group was located at the C-5–OH position of β-D-apiofuranose in **9a**, and the structure of **9** was determined to be 2-(4-hydroxyphenyl)ethyl 1-*O*-β-D-[5-*O*-(4-hydroxybenzoyl)]-apiofuranosyl-(1 → 6)-β-D-glucopyranoside.

The molecular formulae of compounds **10**, **11**, and **12** were considered to be $C_{28}H_{36}O_{15}$, $C_{26}H_{32}O_{14}$, and $C_{27}H_{34}O_{15}$ on the basis of HR-FABMS analyses, respectively. The ^{1}H NMR spectra of these compounds commonly showed the presence of hydroxymethyl protons, a glucopyranosyl anomeric proton, an apiofuranosyl anomeric proton, and aromatic protons. Alkaline hydrolysis of **10** produced 3,4-dimethoxybenzoic acid and **10a**, $C_{19}H_{28}O_{12}$ (HR-FABMS). Comparison of the ^{1}H and ^{13}C NMR spectroscopic data of **10a** with those of osmanthuside H (**9a**) (Sugiyama and Kikuchi, 1993) indicated that the sugar moiety of **10a** was also β-D-apiofuranosyl-(1 → 6)-β-D-glucopyranoside. The ^{1}H NMR spectrum of **10a** showed an aromatic AMX systems [δ 7.13 (1H, *d*, J = 8.0 Hz), 7.03 (1H, *d*, J = 2.0 Hz), 6.90 (1H, *dd*, J = 8.0, 2.0 Hz)], a hydroxy methyl proton signal [δ 4.54 (2H, *s*)], and a methoxyl proton signal [δ 3.87 (3H, *s*)], arising from the aglycone. The following NOEs were observed for **10a**; δ 4.82 (H-1 of β-D-glucopyranose) and δ 7.13; δ 3.87 (–OMe) and δ 7.03; and δ 4.54 (–CH2OH) and δ 7.03, 6.90. These results showed that the aglycone was vanillyl alcohol. Acidic hydrolysis confirmed this conclusion (see section 3). Thus, compound **10a** was determined to be 4-hydroxymethyl-2-methoxyphenyl 1-*O*-β-D-apiofuranosyl-(1 → 6)-β-D-glucopyranoside. A comparison of the spectroscopic data of **10** and **9** established the position of acylation as C-5 of the apiofuranose moiety. Thus, the structure of **10** was established as 4-hydroxymethyl-2-methoxyphenyl 1-*O*-β-D-[5-*O*-(3,4-dimethoxybenzoyl)]-apiofuranosyl-(1 → 6)-β-D-glucopyranoside. Alkaline hydrolysis of **11** and **12**, respectively, produced 4-hydroxybenzoic acid and 4-methoxybenzoic acid together with **10a**, thus confirming their structures as shown.

Compound **13** had the molecular formula $C_{29}H_{38}O_{16}$ (HR-FABMS), an increase of CH_2O relative to **10**. Comparison of the ^{1}H and ^{13}C NMR spectroscopic data of **13** with those of **10** revealed that **13** also had a β-D-[5-*O*-(3,4-dimethoxybenzoyl)]-apiofuranosyl-(1 → 6)-β-D-glucopyranosyl group. In regard to the aglycone moiety of **13**, the ^{1}H and ^{13}C NMR spectra showed signals for an oxygenated methine [δ 75.6, 4.58 (1H, *dd*, J = 7.0, 4.5 Hz)] and a hydroxymethylene groups [δ 68.6, 3.59 (1H, *dd*, J = 11.5, 4.5 Hz), 3.55 (1H, *dd*, J = 11.5, 7.0 Hz)], in addition to the aromatic and methoxyl signals. Significant HMBC correlations were observed from this oxygenated methine proton to C-3 (δ 112.0), C-4 (δ 138.4), and C-5 (δ 120.2) of the aromatic ring as well as to the hydroxtmethylene carbon (δ 68.6). The methoxyl group (δ 3.84) showed a correlation to C-2 (δ 150.8). Moreover, NOEs were observed as follows: δ 4.58 (H-7) and δ 7.02 (H-3), 6.83 (H-5); δ 3.84 (–OMe) and δ 7.02 (H-3); δ 4.80 (H-1 of β-D-glucopyranose) and δ 7.08 (H-6). Thus, the aglycone of **13** was considered to be 1-(4-hydroxy-3-methoxyphenyl)ethane-1,2-diol. On the basis of the above arguments, the structure of **13** was established to be 4-(1,2-dihydroxyethyl)-2-methoxyphenyl 1-*O*-β-D-[5-*O*-(3,4-dimethoxybenzoyl)]-apiofuranosyl-(1 → 6)-β-D-glucopyranoside. The absolute configuration of the aglycone moiety was not determined.

Compound **18** was a lignan glucoside whose molecular formula was $C_{26}H_{30}O_{11}$. The ^{13}C and ^{1}H NMR spectra of **18** were similar to those of (−)-balanophonin (Haruna et al., 1982; Yuen et al., 1998) except for presence of a β-D-glucopyranosyl group. The anomeric proton signal of the β-D-glucopyranosyl group resonated at δ 4.89 (1H, *d*, J = 8.0 Hz), and showed a NOE interaction with H-5 [δ 7.16 (1H, *d*, J = 8.0 Hz)] in agreement with the attachment of the sugar at C-4 of the aglycone. Enzymatic hydrolysis of **18** produced (+)-balanophonin (**18a**). The CD spectrum of **18a** supported its 7*S*, 8*R* absolute configuration (Yuen et al., 1998). Accordingly, the structure of **18** was concluded to be (7*S*, 8*R*)-balanophonin 4-*O*-β-D-glucopyranoside.

The molecular formula of compound **19** was indicated as $C_{24}H_{30}O_{12}$ by HR-FABMS. Alkaline hydrolysis of **19** yielded 4-methoxybenzoic acid and **19a**. However, despite the correspondence of the ^{13}C NMR spectral data of **19a** with those of 2-methoxy-4-[(1*R*, 2*R*)-1,2,3-trihydroxypropyl]phenyl 1-*O*-β-D-glucopyranoside (Ishikawa and Kitajima, 2002), there was a difference in their optical rotations. Enzymatic hydrolysis of **19a** produced compound **19b**. The ^{13}C NMR spectral data of **19b** corresponded to those of (1*R*, 2*R*)-1-(4-hydroxy-3-methoxyphenyl)-1,2,3-propanetriol (Ishikawa and Kitajima, 2002). However, as compound **19b** showed a positive optical rotation value ($[\alpha]_D^{23}$ +25°) and (1*R*, 2*R*)-1-(4-hydroxy-3-methoxyphenyl)-1,2,3-propanetriol exhibited a negative optical rotation value ($[\alpha]_D^{25}$ −26°) (Greca et al., 1998; Ishikawa and Kitajima, 2002), the absolute stereochemistry of C-7 and C-8 in **19b** should be *S* and *S*. Therefore, **19b** was determined to be (1*S*, 2*S*)-1-(4-hydroxy-3-methoxyphenyl)-1,2,3-propanetriol. The HMBC spectrum indicated that the position of esterification was C-6 of glucose. Thus, com-

pound **19** was 2-methoxy-4-[(1*S*,2*S*)-1,2,3-trihydroxypropyl]phenyl 1-*O*-β-D-[6-*O*-(4-methoxybenzoyl)]-glucopyranoside.

It is known that Bignoniaceaus plants contain many kinds of iridoid glycosides. By our present investigation, some esterified-ajugols were afforded from the bark of *T. impetiginosa*. The previous literature (Nakano et al., 1993) reported that the same esterified-ajugols were obtained form the bark of *T. avellanedae*. Thus, esterified-ajugols are thus considered to be characteristic constituents of *Tabebuia* genus plant.

3. Experimental

Instrumental analysis was carried out as described previously (Warashina et al., 2004). ^{1}H and ^{13}C NMR were recorded in MeOH-d_4, pyridine-d_5 or D_2O solution, and chemical shifts are given on the δ (ppm) scale with tetramethylsilane (TMS) or dioxane as an internal standard. HPLC analyses employed a JASCO 800 system instrument and Shimamura YRD-880 RI detector.

3.1. Plant material

The bark of *T. impetiginosa* (Mart. ex DC) Standley was purchased on the herbal market in Sao Paulo, Brazil in March, 2000, and identified by Y. Nagatani (a staff of Herbario Goro Hashimoto, Centro de Pesquisas de Historia Natural). The voucher specimen is deposited in the herbarium of Herbario Goro Hashimoto, Centro de Pesquisas de Historia Natural.

3.2. Extraction and isolation

Extraction of the bark of *T. impetiginosa* is described in the previous paper (Warashina et al., 2004). The MeOH-H_2O (1:1) eluate from the porous polymer gel "Mitsubishi Diaion HP-20" column was concentrated and the residue (27.0 g) subjected to silica gel CC with a $CHCl_3$–MeOH–EtOAc–H_2O (46:15:35:2) as eluant to give six fractions (A (0.9 g), B (5.5 g), C (3.1 g), D (4.5 g), E (3.1 g), and F (10.5 g)). Using semi-preparative HPLC (Develosil-ODS-15/30, -C8 and YMC-ODS: 15–20% MeCN in water and 30–40% MeOH in water), fraction C (2.0 g) afforded compounds **1** (238 mg), **2** (66 mg), **3** (30 mg), **4** (4 mg), **5** (4 mg), **8** (5 mg), **9** (11 mg), **10** (16 mg), **11** (14 mg), **12** (10 mg), **15** (4 mg), **16** (7 mg), **17** (17 mg), **18** (9 mg), and **19** (11 mg). Similarly, fractions D (2.3 g) yielded compounds **6** (34 mg), **7** (18 mg), **13** (12 mg), **14** (6 mg), **17** (33 mg), and **19** (10 mg).

Compound ***4***: Amorphous powder. $[\alpha]_D^{23}$ −99° (MeOH; *c* 0.41). UV λ_{max}^{MeOH} nm (log ε): 201 (4.33), 256 (4.18). FABMS *m/z*: 521 $[M + Na]^+$, HR-FABMS *m/z*: 521.1606 (Calcd for $C_{23}H_{30}O_{12}Na$: 521.1635). For ^{13}C NMR spectrum, see Table 1. ^{1}H NMR (MeOH-d_4 at 35 °C): δ 8.01 (2H, *d*, *J* = 9.0 Hz, H-3″, -7″), 6.98 (2H, *d*, *J* = 9.0 Hz, H-4″, -6″), 6.28 (1H, *dd*, *J* = 6.0, 2.0 Hz, H-3), 5.57 (1H, *d*, *J* = 2.0 Hz, H-1), 5.08 (1H, *m*, H-6), 5.00 (1H, *dd*, *J* = 6.0, 3.0 Hz, H-4), 4.68 (1H, *d*, *J* = 8.0 Hz, H_{Glc}-1), 3.89 (1H, *dd*, *J* = 12.0, 2.0 Hz, H_{Glc}-6), 3.86 (3H, *s*, C-5″-OMe), 3.75 (1H, *d*, *J* = 11.5 Hz, H-10), 3.66 (1H, *dd*, *J* = 12.0, 6.0 Hz, H_{Glc}-6), 3.63 (1H, *d*, *J* = 11.5 Hz, H-10), 3.37 (1H, *t*, *J* = 8.0 Hz, H_{Glc}-3), 3.31 (overlapping, H_{Glc}-5), 3.27 (1H, *t*, *J* = 8.0 Hz, H_{Glc}-4), 3.20 (1H, *t*, *J* = 8.0 Hz, H_{Glc}-2), 3.09 (1H, *m*, H-5), 2.65 (1H, *dd*, *J* = 8.5, 4.5 Hz, H-9), 2.40 (1H, *dd*, *J* = 15.0, 6.5 Hz, H-7), 1.94 (1H, *dd*, *J* = 15.0, 2.5 Hz, H-7).

Compound ***5***: Amorphous powder. $[\alpha]_D^{22}$ −131° (MeOH; *c* 0.38). UV λ_{max}^{MeOH} nm (log ε): 202 (4.33), 256 (4.14). FABMS *m/z*: 519 $[M + Na]^+$, HR-FABMS *m/z*: 519.1482 (Calcd for $C_{23}H_{28}O_{12}Na$: 519.1478). For ^{13}C NMR spectrum, see Table 1. ^{1}H NMR (MeOH-d_4 at 35 °C): δ 8.04 (2H, *d*, *J* = 9.0 Hz, H-3″, -7″), 7.01 (2H, *d*, *J* = 9.0 Hz, H-4″, -6″), 6.26 (1H, *dd*, *J* = 6.5, 2.5 Hz, H-3), 5.75 (1H, *br s*, H-1), 4.86 (1H, *ddd*, *J* = 6.5, 2.0, 0.5 Hz, H-4), 4.59 (1H, *d*, *J* = 8.0 Hz, H_{Glc}-1), 4.55 (1H, *d*, *J* = 10.0 Hz, H-10), 4.36 (1H, *d*, *J* = 10.0 Hz, H-10), 3.88 (1H, *dd*, *J* = 11.5, 2.0 Hz, H_{Glc}-6), 3.87 (3H, *s*, C-5″-OMe), 3.67 (1H, *dd*, *J* = 11.5, 5.5 Hz, H_{Glc}-6), 3.58 (1H, *d*, *J* = 2.0 Hz, H-7), 3.48 (1H, *d*, *J* = 2.0 Hz, H-6), 3.35 (1H, *t*, *J* = 8.0 Hz, H_{Glc}-3), 3.29 (overlapping, H_{Glc}-4, $_{Glc}$-5), 3.18 (1H, *t*, *J* = 8.0 Hz, H_{Glc}-2), 3.12 (1H, *dt*, *J* = 8.5, 2.0 Hz, H-5), 2.29 (1H, *br d*, *J* = 8.5 Hz, H-9).

Compound ***6***: Amorphous powder. $[\alpha]_D^{23}$ −44.7° (MeOH; *c* 1.42) UV λ_{max}^{MeOH} nm (log ε): 203 (4.29), 218 (4.27), 260 (4.00), 290 (3.71). FABMS *m/z*: 539 $[M + Na]^+$, HR-FABMS *m/z*: 539.2101 (Calcd for $C_{24}H_{36}O_{12}Na$: 539.2104). For ^{13}C NMR spectrum, see Table 1. ^{1}H NMR (MeOH-d_4 at 35 °C): δ 7.72 (1H, *dd*, *J* = 8.5, 2.0 Hz, H-7″), 7.58 (1H, *d*, *J* = 2.0 Hz, H-3″), 7.02 (1H, *dd*, *J* = 8.5 Hz, H-6″), 5.21 (1H, *ddd*, *J* = 8.5, 7.5, 3.5 Hz, H-6), 4.23 (1H, *d*, *J* = 8.0 Hz, H_{Glc}-1), 3.97 (1H, *dt*, *J* = 10.0, 6.5 Hz, H-3), 3.89 (3H, *s*, C-5″-OMe), 3.88 (3H, *s*, C-4″-OMe), 3.78 (1H, *dd*, *J* = 12.0, 2.0 Hz, H_{Glc}-6), 3.73 (2H, overlapping, H-1), 3.66 (1H, *m*, H-3), 3.59 (1H, *dd*, *J* = 12.0, 5.5 Hz, H_{Glc}-6), 3.32 (1H, *t*, *J* = 8.0 Hz, H_{Glc}-3), 3.24 (1H, *t*, *J* = 8.0 Hz, H_{Glc}-4), 3.16 (1H, *t*, *J* = 8.0 Hz, H_{Glc}-2), 3.16 (1H, *m*, H_{Glc}-5), 3.00 (1H, *quint*, *J* = 7.5 Hz, H-5), 2.46 (1H, *dd*, *J* = 14.5, 8.5 Hz, H-7), 2.10 (1H, *m*, H-9), 1.96 (1H, *m*, H-4), 1.84 (1H, *m*, H-4), 1.75 (1H, *ddd*, *J* = 14.5, 3.5, 1.5 Hz, H-7), 1.36 (3H, *s*, H-10).

Compound ***7***: Amorphous powder. $[\alpha]_D^{22}$ −51.6° (MeOH; *c* 1.83) UV λ_{max}^{MeOH} nm (log ε): 204 (4.45), 256 (4.43). FABMS *m/z*: 509 $[M + Na]^+$, HR-FABMS *m/z*: 509.1979 (Calcd for $C_{23}H_{34}O_{11}Na$: 509.1999). The ^{13}C NMR and ^{1}H NMR spectra of the aglycone and sugar moieties were in good agreement with that of **6**. The ^{13}C NMR and ^{1}H NMR spectra of the ester moiety

(MeOH-d_4 at 35 °C): δ 168.2 (C-1″), 165.1 (C-5″), 132.7 × 2 (C-3″, -7″), 124.1 (C-2″), 114.7 × 2 (C-4″, -6″) and δ 8.01 (2H, *d*, *J* = 9.0 Hz, H-3″, -7″), 6.98 (2H, *d*, *J* = 9.0 Hz, H-4″, -6″), 3.86 (3H, *s*, C-5″-OMe).

Compound ***8***: Amorphous powder. $[\alpha]_D^{24}$ −40° (MeOH; *c* 0.47) UV λ_{max}^{MeOH} nm (log ε): 202 (4.39), 256 (4.19). FABMS *m/z*: 477 $[M + Na]^+$, HR-FABMS *m/z*: 477.1737 (Calcd for $C_{22}H_{30}O_{10}Na$: 477.1737). For ^{13}C NMR spectrum, see Table 1. ^1H NMR (pyridine-d_5 at 35 °C): δ 8.20 (1H, *d*, *J* = 8.5 Hz, H-3″, -7″), 7.15 (1H, *d*, *J* = 8.5 Hz, H-4″, -6″), 4.93 (1H, *d*, *J* = 8.0 Hz, H_{Glc}-1), 4.80 (1H, *ddd*, *J* = 11.0, 8.5, 5.5 Hz, H-3), 4.71 (1H, *quint*, *J* = 3.5 Hz, H-6), 4.60 (1H, *ddd*, *J* = 11.0, 8.5, 7.0 Hz, H-3), 4.55 (1H, *dd*, *J* = 12.0, 2.0 Hz, H_{Glc}-6), 4.50 (1H, *br d*, *J* = 12.5 Hz, H-1), 4.40 (1H, *br d*, *J* = 12.5 Hz, H-1), 4.35 (1H, *dd*, *J* = 12.0, 5.5 Hz, H_{Glc}-6), 4.24 (1H, *t*, *J* = 8.0 Hz, H_{Glc}-3), 4.20 (1H, *t*, *J* = 8.0 Hz, H_{Glc}-4), 3.97 (overlapping, H_{Glc}-2, Glc-5), 3.58 (1H, *br s*, H-5), 2.73 (1H, *dd*, *J* = 17.0, 6.5 Hz, H-7), 2.62 (1H, *br d*, *J* = 17.0 Hz, H-7), 2.44 (1H, *m*, H-4), 1.95 (1H, *m*, H-4), 1.57 (3H, *br s*, H-10).

Compound ***9***: Amorphous powder. $[\alpha]_D^{24}$ −57.2° (MeOH; *c* 1.11) UV λ_{max}^{MeOH} nm (log ε): 202 (4.42), 208 (sh), 223 (sh), 257 (4.17). FABMS *m/z*: 575 $[M + Na]^+$, HR-FABMS *m/z*: 575.1719 (Calcd for $C_{26}H_{32}O_{13}Na$: 575.1741). For ^{13}C NMR spectrum, see Table 1. ^1H NMR (MeOH-d_4 at 35 °C): δ 7.91 (2H, *d*, *J* = 9.0 Hz, H-3″, -7″), 7.02 (2H, *d*, *J* = 9.0 Hz, H-2, -6), 6.83 (2H, *d*, *J* = 9.0 Hz, H-4″, -6″), 6.67 (2H, *d*, *J* = 9.0 Hz, H-3, -5), 5.05 (1H, *d*, *J* = 2.0 Hz, H_{Api}-1), 4.34 (1H, *d*, *J* = 11.5 Hz, H_{Api}-5), 4.33 (1H, *d*, *J* = 11.5 Hz, H_{Api}-5), 4.27 (1H, *d*, *J* = 8.0 Hz, H_{Glc}-1), 4.08 (1H, *d*, *J* = 10.0 Hz, H_{Api}-4), 4.01 (1H, *dd*, *J* = 11.5, 2.0 Hz, H_{Glc} -6), 3.99 (1H, *d*, *J* = 2.0 Hz, H_{Api}-2), 3.95 (1H, *ddd*, *J* = 9.5, 8.0, 7.0 Hz, H-8), 3.87 (1H, *d*, *J* = 10.0 Hz, H_{Api}-4), 3.67 (1H, *ddd*, *J* = 9.5, 8.0, 7.0 Hz, H-8), 3.62 (1H, *dd*, *J* = 11.5, 6.0 Hz, H_{Glc}-6), 3.41 (1H, *m*, H_{Glc}-5), 3.34 (1H, *t*, *J* = 8.0 Hz, H_{Glc}-3), 3.27 (1H, *t*, *J* = 8.0 Hz, H_{Glc}-4), 3.18 (1H, *t*, *J* = 8.0 Hz, H_{Glc}-2), 2.78 (2H, *m*, H-7).

Compound ***10***: Amorphous powder. $[\alpha]_D^{24}$ −66.8° (MeOH; *c* 1.16) UV λ_{max}^{MeOH} nm (log ε): 202 (4.74), 219 (4.52), 261 (4.18), 281 (sh). FABMS *m/z*: 635 $[M + Na]^+$, HR-FABMS *m/z*: 635.1932 (Calcd for $C_{28}H_{36}O_{15}Na$: 635.1952). For ^{13}C NMR spectrum, see Table 1. ^1H NMR (MeOH-d_4 at 35 °C): δ 7.70 (1H, *dd*, *J* = 8.0, 2.0 Hz, H-7″), 7.51 (1H, *d*, *J* = 2.0 Hz, H-3″), 7.07 (1H, *d*, *J* = 8.0 Hz, H-6), 7.01 (1H, *d*, *J* = 8.0 Hz, H-6″), 6.98 (1H, *d*, *J* = 2.0 Hz, H-3), 6.78 (1H, *dd*, *J* = 8.0, 2.0 Hz, H-5), 5.00 (1H, *d*, *J* = 2.0 Hz, H_{Api}-1), 4.80 (1H, *d*, *J* = 8.0 Hz, H_{Glc}-1), 4.46 (2H, *s*, H-7), 4.38 (1H, *d*, *J* = 11.0 Hz, H_{Api}-5), 4.34 (1H, *d*, *J* = 11.0 Hz, H_{Api}-5), 4.07 (1H, *d*, *J* = 9.5 Hz, H_{Api}-4), 4.02 (1H, *dd*, *J* = 11.0, 2.0 Hz, H_{Glc}-6), 4.00 (1H, *d*, *J* = 2.0 Hz, H_{Api}-2), 3.86 (1H, *d*, *J* = 9.5 Hz, H_{Api}-4), 3.88 (3H, *s*, C-5″-OMe), 3.84 (3H, *s*, C-4″-OMe or C-2-OMe), 3.83 (3H, *s*, C-2-OMe or C-4″-OMe), 3.53 (1H, *m*, H_{Glc}-5), 3.49 (1H, *t*, *J* = 8.0 Hz, H_{Glc}-2), 3.44 (1H, *t*, *J* = 8.0 Hz, H_{Glc}-3), 3.34 (1H, *t*, *J* = 8.0 Hz, H_{Glc}-4).

Compound ***11***: Amorphous powder. $[\alpha]_D^{24}$ −70.6° (MeOH; *c* 1.45) UV λ_{max}^{MeOH} nm (log ε): 201 (4.66), 226 (sh), 258 (4.13). FABMS *m/z*: 591 $[M + Na]^+$, HR-FABMS *m/z*: 591.1664 (Calcd for $C_{26}H_{32}O_{14}Na$: 591.1690). The ^{13}C and ^1H NMR spectra of the aglycone and sugar moieties were in good agreement with those in **10**. The ^{13}C and ^1H NMR spectra of the ester moiety (MeOH-d_4 at 35 °C): δ 167.9 (C-1″), 163.6 (C-5″), 133.0 × 2 (C-3″, -7″), 122.0 (C-1″), 116.2 × 2 (C-4″, -6″) and δ 7.91 (2H, *d*, *J* = 8.5 Hz, H-3″, -7″), 6.82 (2H, *d*, *J* = 8.5 Hz, H-4″, -6″).

Compound ***12***: Amorphous powder. $[\alpha]_D^{24}$ −69° (MeOH; *c* 0.97) UV λ_{max}^{MeOH} nm (log ε): 202 (4.59), 226 (3.98) 257 (4.18). FABMS *m/z*: 605 $[M + Na]^+$, HR-FABMS *m/z*: 605.1846 (Calcd for $C_{27}H_{34}O_{14}Na$: 605.1846). The ^{13}C and ^1H NMR spectra of the aglycone and sugar moieties were in good agreement with those in **10**. The ^{13}C and ^1H NMR spectra of the ester moiety (MeOH-d_4 at 35 °C): δ 167.7 (C-1″), 165.3 (C-5″), 132.8 × 2 (C-3″, -7″), 123.1 (C-1″), 114.9 × 2 (C-4″, -6″) 56.1 (C-5″-OMe) and δ 8.00 (2H, *d*, *J* = 8.0 Hz, H-3″, -7″), 6.98 (2H, *d*, *J* = 8.0 Hz, H-4″, -6″), 3.85 (3H, *s*, C-5″-OMe).

Compound ***13***: Amorphous powder. $[\alpha]_D^{23}$ −66.7° (MeOH, *c* 1.15). UV λ_{max}^{MeOH} nm (log ε): 262 (4.98), 286 (sh). FABMS *m/z*: 665 $[M + Na]^+$, HR-FABMS *m/z*: 665.2057 (Calcd for $C_{29}H_{38}O_{16}Na$: 665.2058). The ^1H NMR spectra of the sugar and ester moieties were in good agreement with those in **10**. The ^1H NMR spectrum of the aglycone moiety (MeOH-d_4 at 35 °C): δ 7.08 (^1H, *d*, *J* = 8.0 Hz, H-6), 7.02 (1H, *d*, *J* = 2.0 Hz, H-3), 6.83 (1H, *dd*, *J* = 8.0, 2.0 Hz, H-5), 4.58 (1H, *dd*, *J* = 7.0, 4.5 Hz, H-7), 3.84 (3H, *s*, C-2-OMe), 3.59 (1H, *dd*, *J* = 11.5, 4.5 Hz, H-8), 3.55 (1H, *dd*, *J* = 11.5, 7.0 Hz, H-8).

Compound ***18***: Amorphous powder. $[\alpha]_D^{24}$ +7.5° (MeOH; *c* 0.94) UV λ_{max}^{MeOH} nm (log ε): 203 (4.67), 227 (4.30), 240 (sh), 286 (sh), 340 (4.32). FABMS *m/z*: 541 $[M + Na]^+$, HR-FABMS *m/z*: 541.1704 (Calcd for $C_{26}H_{30}O_{11}Na$: 541.1686). CD. $\Delta\varepsilon$ (nm): −4.66 (233), +0.38 (258), −1.32 (277), + 1.85 (338) (MeOH; *c* 0.0671). For ^{13}C NMR spectrum, see Table 1. ^1H NMR (MeOH-d_4 at 35 °C): δ 9.58 (1H, *d*, *J* = 8.0 Hz, H-9′), 7.61 (1H, *d*, *J* = 16.0 Hz, H-7′), 7.27 (1H, *br s*, H-6′), 7.23 (1H, *d*, *J* = 1.5 Hz, H-2′), 7.16 (1H, *d*, *J* = 8.0 Hz, H-5), 7.03 (1H, *d*, *J* = 2.0 Hz, H-2), 6.94 (1H, *dd*, *J* = 8.0, 2.0 Hz, H-6), 6.67 (1H, *dd*, *J* = 16.0, 8.0 Hz, H-8′), 5.66 (1H, *d*, *J* = 6.0 Hz, H-7), 4.89 (1H, *d*, *J* = 8.0 Hz, H_{Glc}-1), 3.92 (3H, *s*, C-3′-OMe), 3.91 ∼ 3.86 (2H, overlapping, H-9), 3.84 (overlapping, H_{Glc}-6), 3.83 (3H, *s*, C-3-OMe), 3.68 (1H, *dd*, *J* = 12.0, 4.0 Hz, H_{Glc}-6), 3.54 (1H, *q*, *J* = 6.0 Hz, H-8), 3.49

(1H, *t*, *J* = 8.0 Hz, H_{Glc}-2), 3.45 (1H, *t*, *J* = 8.0 Hz, H_{Glc}-3), 3.39 (overlapping, H_{Glc}-4, Glc-5).

Compound ***19***: Amorphous powder. $[\alpha]_D^{23}$ −18.5° (MeOH; *c* 1.04) UV λ_{max}^{MeOH} nm (log ε): 203 (4.68), 227 (4.05), 256 (4.23). FABMS *m/z*: 533 $[M + Na]^+$, HR-FABMS *m/z*: 533.1646 (Calcd for $C_{24}H_{30}O_{12}Na$: 533.1635). For ^{13}C NMR spectrum, see Table 1. ^1H NMR (MeOH-d_4 at 35 °C): δ 7.97 (2H, *d*, *J* = 9.0 Hz, H-3″,-7″), 7.07 (1H, *d*, *J* = 8.0 Hz, H-6), 7.06 (1H, *d*, *J* = 2.0 Hz, H-3), 7.01 (2H, *d*, *J* = 9.0 Hz, H-4″, -6″), 6.74 (1H, *dd*, *J* = 8.0, 2.0 Hz, H-5), 4.89 (1H, *d*, *J* = 8.0 Hz, H_{Glc}-1), 4.68 (1H, *dd*, *J* = 12.0, 2.5 Hz, H_{Glc}-6), 4.55 (1H, *d*, *J* = 6.0 Hz, H-7), 4.36 (1H, *dd*, *J* = 12.0, 7.5 Hz, H_{Glc}-6), 3.88 (3H, *s*, C-5″-OMe), 3.85 (3H, *s*, C-2-OMe), 3.74 (1H, *m*, H_{Glc}-5), 3.64 (1H, *td*, *J* = 6.0, 4.0 Hz, H-8), 3.55 (1H, *t*, *J* = 8.0 Hz, H_{Glc}-2), 3.51 (1H, *t*, *J* = 8.0 Hz, H_{Glc}-3), 3.48 (1H, *dd*, *J* = 12.0, 4.0 Hz, H-9), 3.44 (1H, *t*, *J* = 9.0 Hz, H_{Glc}-4), 3.35 (1H, *dd*, *J* = 12.0, 6.0 Hz, H-9).

3.3. Alkaline and acid hydrolysis of compounds *6*, *10* and *19*

Compounds **6** (14 mg), **10** (6 mg), and **19** (10 mg) were individually dissolved in 0.05 M NaOH (1 ml) and stirred for 2 h to 4 h at room temperature under a N_2 gas atmosphere. Each reaction mixture was neutralized with an Amberlite IR-120B column and the eluate was concentrated to dryness. The residue was partitioned between EtOAc and H_2O. Both layers were concentrated to dryness, and HPLC analysis of the residue from each EtOAc layer suggested that 3,4-dimethoxybenzoic acid was produced from **6** and **10**, and 4-methoxybenzoic acid was obtained from **19**. HPLC conditions: column, YMC-ODS 4.6 mm × 25 cm; flow rate, 1.0 ml/min; 20% MeCN + 0.05% trifluoroacetic acid (TFA); R_t, 3,4-dimethoxybenzoic acid 15.8 min; 25% MeCN + 0.05% TFA; R_t, 4-methoxybenzoic acid 16.2 min. Purification of the residue from each H_2O layer using HPLC afforded **6a** (6 mg), **10a** (3 mg) and **19a** (5 mg). HPLC conditions: column; YMC-ODS 10 mm × 25 cm, flow rate; 3.0 ml/min; **6a**, 2% MeCN in water; **10a**, 5% MeCN in water; **19a**, 2% MeOH in water.

Compound ***6a***: Amorphous powder. $[\alpha]_D^{22}$ −23° (MeOH; *c* 0.58). FABMS *m/z*: 375 $[M + Na]^+$, HR-FABMS *m/z*: 375.1611 (Calcd for $C_{15}H_{28}O_9Na$: 375.1631). ^{13}C NMR (MeOH-d_4 at 35 °C): δ 104.4 (C_{Glc}-1), 80.1 (C-8), 78.1 × 2, 78.0 (C-6, $_{Glc}$-3, $_{Glc}$-5), 75.2 (C_{Glc}-2), 71.7 (C_{Glc}-4), 70.3 (C-3), 62.8 (C_{Glc}-6), 59.6 (C-1), 55.2 (C-9), 50.6 (C-7), 47.1 (C-5), 30.3 (C-4), 25.2 (C-10). (D_2O at 35 °C): δ 102.9 (C_{Glc}-1), 79.9 (C-8), 76.8, 76.7, 76.5 (C-6, $_{Glc}$-3, $_{Glc}$-5), 73.9 (C_{Glc}-2), 70.5 (C_{Glc}-4), 70.1 (C-1), 61.6 (C_{Glc}-6), 59.0 (C-1), 53.5 (C-9), 49.2 (C-7), 45.3 (C-5), 29.0 (C-4), 23.5 (C-10). ^1H NMR (MeOH-d_4 at 35 °C): δ 4.29 (1H, *d*, *J* = 8.0 Hz, H_{Glc}-1), 4.01 (1H, *dt*, *J* = 10.0, 4.5 Hz, H-3), 3.98 (1H, *ddd*, *J* = 8.5, 7.0, 5.0 Hz, H-6), 3.87 (1H, *dd*, *J* = 12.0, 2.0 Hz, H_{Glc}-6), 3.71 (1H, *dt*, *J* = 10.0, 6.5 Hz, H-1), 3.66 (overlapping, H_{Glc}-6), 3.66 (1H, *dd*, *J* = 11.5, 4.5 Hz, H-1), 3.62 (1H, *dd*, *J* = 11.5, 4.5 Hz, H-1), 3.35 (overlapping, H_{Glc}-3), 3.27 (overlapping, H_{Glc}-4, $_{Glc}$-5), 3.17 (1H, *t*, *J* = 8.0 Hz, H_{Glc}-2), 2.44 (1H, *m*, H-5), 2.17 (1H, *dd*, *J* = 14.0, 8.5 Hz, H-7), 2.09 (1H, *m*, H-9), 1.89 (1H, *m*, H-4), 1.76 (1H, *m*, H-4), 1.68 (1H, *ddd*, *J* = 14.0, 5.0, 1.5 Hz, H-7), 1.25 (3H, *s*, H-10).

Compound ***10a***: Amorphous powder. $[\alpha]_D^{24}$ −91° (MeOH; *c* 0.34). UV λ_{max}^{MeOH} nm (log ε): 201 (4.49), 225 (3.86), 275 (3.31). FABMS *m/z*: 471 $[M + Na]^+$, HR-FABMS *m/z*: 471.1456 (Calcd for $C_{19}H_{28}O_{12}Na$: 471.1478). ^{13}C NMR (MeOH-d_4 at 35 °C): δ 151.0 (C-2), 147.3 (C-1), 137.9 (C-4), 120.9 (C-5), 118.4 (C-6), 112.8 (C-3), 111.0 (C_{Api}-1), 103.1 (C_{Glc}-1), 80.5 (C_{Api}-3), 78.1, 77.9 (C_{Glc}-3, $_{Api}$-2), 77.1 (C_{Glc}-5), 75.0 × 2 (C_{Glc}-2, $_{Api}$-4), 71.7 (C_{Glc}-4), 68.7 (C_{Glc}-6), 65.7 (C_{Api}-5), 65.0 (C-7), 56.8 (C-2-OMe). ^1H NMR (MeOH-d_4 at 35 °C): δ 7.13 (1H, *d*, *J* = 8.0 Hz, H-6), 7.03 (1H, *d*, *J* = 2.0 Hz, H-3), 6.90 (1H, *dd*, *J* = 8.0, 2.0 Hz, H-5), 4.95 (1H, *d*, *J* = 2.0 Hz, H_{Api}-1), 4.82 (1H, *d*, *J* = 8.0 Hz, H_{Glc}-1), 4.54 (2H, *s*, H-7), 3.98 (1H, *dd*, *J* = 11.5, 2.0 Hz, H_{Glc}-6), 3.93 (1H, *d*, *J* = 10.0 Hz, H_{Api}-4), 3.87 (overlapping, H_{Api}-2), 3.87 (3H, *s*, C-2-OMe), 3.73 (1H, *d*, *J* = 10.0 Hz, H_{Api}-4), 3.61 (1H, *dd*, *J* = 11.5, 6.5 Hz, H_{Glc}-6), 3.56 (2H, *s*, H_{Api}-5), 3.53 (1H, *m*, H_{Glc}-5), 3.48 (1H, *t*, *J* = 8.0 Hz, H_{Glc}-2), 3.44 (1H, *t*, *J* = 8.0 Hz, H_{Glc}-3), 3.34 (overlapping, H_{Glc}-4).

Compound ***19a***: Amorphous powder. $[\alpha]_D^{22}$ −31° (MeOH; *c* 0.45). UV λ_{max}^{MeOH} nm (log ε): 202 (4.54), 225 (3.91), 275 (3.37). FABMS *m/z*: 399 $[M + Na]^+$, HR-FABMS *m/z*: 399.1241 (Calcd for $C_{16}H_{24}O_{10}Na$: 399.1267). ^{13}C NMR (pyridine-d_5 at 35 °C): δ 150.0 (C-2), 147.2 (C-1), 138.6 (C-4), 120.0 (C-5), 116.2 (C-6), 112.4 (C-3), 102.6 (C_{Glc}-1), 78.8, 78.6 (C_{Glc}-3, $_{Glc}$-5), 77.6 (C-8), 75.0 (C_{Glc}-2), 74.6 (C-7), 71.3 (C_{Glc}-4), 64.3 (C-9), 62.4 (C_{Glc}-6), 56.0 (C-2-OMe). ^1H NMR (pyridine-d_5 at 35 °C): δ 7.58 (1H, *d*, *J* = 8.0 Hz, H-6), 7.49 (1H, *d*, *J* = 2.0 Hz, H-3), 7.31 (1H, *dd*, *J* = 8.0, 2.0 Hz, H-5), 5.62 (1H, *d*, *J* = 8.0 Hz, H_{Glc}-1), 5.29 (1H, *d*, *J* = 5.5 Hz, H-7), 4.19 (1H, *dd*, *J* = 11.0, 4.5 Hz, H-9), 4.04 (1H, *dd*, *J* = 11.0, 6.0 Hz, H-9) 3.67 (3H, *s*, C-2-OMe).

Compounds **6a**, **10a**, and **19a** (ca. 0.5 mg) were individually dissolved in dioxane and 2 M HCl (50 μl each). Each solution was heated at 100 °C for 1 h, and the reaction mixture was partitioned between EtOAc and H_2O. By HPLC analysis, vanillyl alcohol was detected from the EtOAc layer of compound **10a**. HPLC conditions: column, YMC-ODS 4.6 mm × 25 cm; flow rate, 1.0 ml/min; 10% MeCN in water; R_t, vanillyl alcohol 10.0 min. The H_2O layer was passed through an Amberlite IRA-60E column. The eluate was concentrated to

dryness and the residue was stirred with D-cysteine methyl ester hydrochloride, hexamethyldisilazane and trimethylsilylchloride in pyridine using the same procedures as in previous reports (Hara et al., 1987; Zhang et al., 1996). After the reactions, the supernatant was subjected to the GC analysis. GC conditions: column; GL capillary column TC-1 (GL Science, Inc.) 0.25 mm × 30 m, Carrier gas N_2, column temperature 230 °C; R_t, D-glucose 21.9 min, L-glucose 20.9 min. D-Glucose was detected from compounds **6a**, **10a**, and **19a**.

Additionally, **10a** was dissolved in dioxane and 2 M HCl (50 μl each) and heated at 100 °C for 5 min. The procedures following hydrolysis were the same as described above. The neutralized H_2O layer was reduced with $NaBH_4$ (ca. 1 mg) for 1 h at room temperature. Methods for the preparation of alditol acetates are described in the literature (Nagatani et al., 2001). Apiitol acetate was detected from **10a** by the GC analysis. GC conditions: column; Supelco SP-2380™ capillary column 0.25 mm × 30 m, Carrier gas N_2, column temperature 250 °C; R_t, apiitol acetate 8.1 min.

3.4. Alkaline and/or acid hydrolysis of compounds *4*, *5*, *7–9*, *11–13* and *18*

Compounds **4**, **5**, **7–9**, **11–13** (ca. 0.5 mg) were dissolved in 0.05 M NaOH, and stirred for 2.5 h to 1 day at room temperature under a N_2 gas atmosphere. The procedures after alkaline hydrolysis were carried out as described above. 4-Hydroxybenzoic acid was detected from the residue of the EtOAc layer of compounds **8**, **9** and **11** by HPLC analysis. HPLC conditions: column, YMC-ODS 4.6 mm × 25 cm; flow rate, 1.0 ml/min; 20% MeCN + 0.05% TFA; R_t, 4-hydroxybenzoic acid 6.6 min. Similarly, HPLC analysis detected 4-methoxybenzoic acid from **4**, **5**, **7**, and **12**, and 3,4-dimethoxybenzoic acid from **13**. (Conditions of HPLC analysis were described above.) Additionally, crescentin IV 3-*O*-β-D-glucopyranoside (**6a**) was identified from the residue of the H_2O layer of compound **7** by HPLC analysis. As the same way, 2-(4-hydroxyphenyl)ethyl 1-*O*-β-D-apiofuranosyl-(1 → 6)-β-D-glucopyranoside (osmanthuside H (**9a**)) was detected from compound **9**, and 4-hydroxymethyl-2-methoxyphenyl 1-*O*-β-D-apiofuranosyl- (1 → 6)-β-D-glucopyranoside (**10a**) was confirmed from **11** and **12**, respectively. HPLC conditions: column, YMC-ODS 4.6 mm × 25 cm; flow rate, 1.0 ml/min; 2% MeCN in water; R_t, **6a** 14.2 min, 10% MeCN in water; R_t, **9a** 10.6 min, 5% MeCN in water; R_t, **10a** 19.6 min.

Compound **18** (ca. 0.5 mg) and the residues of the H_2O layer of compounds **4**, **5**, **8**, and **13** were hydrolyzed individually with 2 M HCl and dioxane (50 μl each). Each solution was heated at 100 °C for 1 h, and the following procedures were described above. D-Glucose was detected from compounds **4**, **5**, **8**, **13**, and **18**. (Conditions of the GC analysis were described above.)

Moreover, the solution of the residue of H_2O layer of **13** in 2 M HCl and dioxane (50 μl each) was heated at 100 °C for 5 min. The following procedures were the same as described above. Apiitol acetate was detected from **13**. (Conditions of the GC analysis were described above.)

3.5. Enzymatic hydrolysis of compounds *18* and *19a*

Compounds **18** (5 mg) and **19a** (3 mg) were dissolved in EtOH (50 μl) and H_2O (0.70 ml), respectively, and then cellulase (Sigma Chem. Co.) (ca. 20 mg) was added into each solution. The mixture was stirred at 40 °C for 1 day. After hydrolysis, the reaction mixture of **18** was diluted with H_2O and extracted with EtOAc. (+)-(7*S*, 8*R*)-Balanophonin (**18a**, 2 mg) was purified from the residue of the EtOAc layer. (Conditions: YMC-ODS 10 mm × 25 cm, 27.5% MeCN in water.) The reaction mixture of **19a** was concentrated to dryness, and (1*S*, 2*S*)-1-(4-hydroxy-3-methoxyphenyl)-1,2,3-propanetriol (**19b**, 1 mg) was purified from above residue using HPLC. (Conditions: YMC-ODS 10 mm × 25 cm, 2% MeCN in water.)

*Compound **18a***: $[\alpha]_D^{22}$ +67° ($CHCl_3$; *c* 0.23) (lit: $[\alpha]_D^{23}$ + 108° ($CHCl_3$; *c* 0.41) (Yuen et al., 1998)). CD [θ] (nm): −12,336 (235), +3391(256), −1587 (279), +5540 (335) (MeOH; *c* 0.0494) (lit: CD [θ] (nm): +5956 (213), −20,615 (235), +5447(256), −1732 (279), +10,442 (335) (EtOH) (Yuen et al., 1998)).

*Compound **19b***: $[\alpha]_D^{23}$ +25° (MeOH *c* 0.11). ^{13}C NMR (pyridine-d_5 at 35 °C): δ 148.5 (C-2), 147.3 (C-1), 120.6 (C-5), 116.1 (C-6), 111.9 (C-3), 77.7 (C-8), 74.9 (C-7), 64.3 (C-9), 56.0 (C-2-OMe). The C-4 signal was overlapping with the solvent signal. ^1H NMR (pyridine-d_5 at 35 °C): δ 7.48 (1H, *d*, *J* = 1.5 Hz, H-3), 7.31 (1H, *dd*, *J* = 8.0, 1.5 Hz, H-5), 7.23 (1H, *d*, *J* = 8.0 Hz, H-6), 5.29 (1H, *d*, *J* = 6.0 Hz, H-7), 4.39 (1H, *m*, H-8), 4.21 (1H, *dd*, *J* = 11.0, 4.0 Hz, H-9), 4.07 (1H, *dd*, *J* = 11.0, 6.0 Hz, H-9), 3.70 (3H, *s*, C-2-OMe).

References

Abe, F., Yamauchi, T., 1986. Lignans from *Trachelospermum asiaticum* (Tracheolospermum. II). Chem. Pharm. Bull. 34, 4340–4345.

Achenbach, H., Lowel, M., Waibel, R., Gupta, M., Solis, P., 1992. New lignan glucosides from *Stemmadenia minima*. Planta Med. 58, 270–272.

Bianco, A., Bonini, C., Guiso, M., Iavarone, C., Trogolo, C., 1981. ^{13}C NMR spectroscopy of cyclopentenpolyols. Tetrahedron 37, 1773–1777.

Bianco, A., Passacantilli, P., Nicoletti, M., Alves de Lima, R., 1982. Iridoids in equatorial and tropical flora. V. A new glucosidic iridoid from *Tecoma chrysantha* Jacq. Gazz. Chim. Ital. 112, 227–229.

Damtoft, S., Jensen, S.R., Nielsen, B.J., 1981. ^{13}C and ^1H NMR spectroscopy as a tool in the configurational analysis of iridoid glucosides. Phytochemistry 20, 2717–2732.

Greca, M.D., Ferrara, M., Fiorentino, A., Monaco, P., Previtera, L., 1998. Antialgal compounds from *Zantedeschia aethiopica*. Phytochemistry 49, 1299–1304.

Hara, S., Okabe, H., Mihashi, K., 1987. Gas–liquid chromatographic separation of aldose enontiomers as trimethylsilyl ethers of methyl 2-(polyhydroxyalkyl)-thiazolidine-4(*R*)-carboxylate. Chem. Pharm. Bull. 35, 501–506.

Haruna, M., Koube, T., Ito, K., Murata, H., 1982. Balanophonin, a new neo-lignan from *Balanophora japonica* MAKINO. Chem. Pharm. Bull. 30, 1525–1527.

Hashimoto, G., 1996. Illustrated Cyclopedia of Brazilian Medical Plants. Aboc-sha, Kamakura, p. 158.

Ishikawa, T., Kitajima, J., 2002. Water-soluble constituents of anise: new glucosides of anethole glycol and related compounds. Chem. Pharm. Bull. 50, 1460–1466.

Iwagawa, T., Hamada, T., Kuroki, S., Hase, T., Okubo, T., Kim, M., 1991. Iridoids from *Catalpa bignonioides*. Phytochemistry 30, 4057–4060.

Kaneko, T., Ohtani, K., Kasai, R., Yamasaki, K., Duc, N.M., 1997. Iridoids and iridoid glucosides from fruits of *Crescentia cujete*. Phytochemistry 46, 907–910.

Kasai, R., Okihara, M., Asakawa, J., Mizutani, K., Tanaka, O., 1979. ^{13}C NMR study of α- and β-anomeric parts of D-mannopyranosides and L-rhamnopyranosides. Tetrahedron 35, 1427–1432.

Matsuda, N., Sato, H., Yaoita, Y., Kikuchi, M., 1996. Isolation and absolute structures of neolignan glycosides with enantiomeric aglycones from leaves of *Viburnum awabuki* K. KOCH. Chem. Pharm. Bull. 44, 1122–1123.

Nagatani, Y., Warashina, T., Noro, T., 2001. Studies on the constituents from the aerial part of *Baccharis dracunculifolia* DC. Chem. Pharm. Bull. 49, 1388–1394.

Nakano, K., Maruyama, K., Murakami, K., Takaishi, Y., Tomimatsu, T., 1993. Iridoids from *Tabebuia avellanedae*. Phytochemistry 32, 371–373.

Nishimura, H., Sasaki, H., Morita, T., Chin, M., Mitsuhashi, H., 1989. Six iridoid glycosides from *Rehmannia Glutinosa*. Phytochemistry 28, 2705–2709.

Otsuka, H., Hirata, E., Shinzato, T., Takeda, Y., 2000. Isolation of lignan glucosides and neolignan sulfate from the leaves of *Glochidion zeylanicum* (Gaertn) A. JUSS. Chem. Pharm. Bull. 48, 1084–1086.

Sugiyama, M., Kikuchi, M., 1993. Phenylethanoid glycosides from *Osmanthus asiaticus*. Phytochemistry 32, 1553–1555.

Takeda, Y., Mima, C., Masuda, T., Hirata, E., Takushi, A., Otsuka, H., 1998. Glochidioboside, a glucoside of (7*S*, 8*R*)-dihydrodehydrodiconiferyl alcohol from leaves of *Glochidion obovatum*. Phytochemistry 49, 2137–2139.

Warashina, T., Nagatani, Y., Noro, T., 2004. Constituents from the bark of *Tabebuia impetiginosa*. Phytochemistry 65, 2003–2011.

Yuen, M.S.M., Xue, F., Mak, T.C.W., Wong, H.N.C., 1998. On the absolute structure of optically active neolignans containing a dihydrobenzo[*b*]furan skeleton. Tetrahedron 54, 12429–12444.

Zhang, D., Miyase, T., Kuroyanagi, M., Umehara, K., Ueno, A., 1996. Studies on the constituents of *Polygala japonica* HOUTT. III. Structures of polygarasaponins XX–XXVII. Chem. Pharm. Bull. 44, 173–179.