HETEROCYCLES, Vol. 86, No. 1, 2012, pp. 381 - 389. © 2012 The Japan Institute of Heterocyclic Chemistry
Received, 9th June, 2012, Accepted, 20th July, 2012, Published online, 24th July, 2012
DOI: 10.3987/COM-12-S(N)29

ISOLATION OF ELLAGITANNIN MONOMER AND MACROCYCLIC DIMER FROM *CASTANOPSIS CARLESII* LEAVES

Yong-Lin Huang,[a,b] Takashi Tanaka,[*,a] Yosuke Matsuo,[a] Isao Kouno,[a] Dian-Peng Li,[b] and Gen-ichiro Nonaka[c]

[a]Graduate School of Biomedical Sciences, Nagasaki University, 1-14 Bunkyo-Machi, Nagasaki 852-8521, Japan; t-tanaka@nagasaki-u.ac.jp
[b]Guangxi Key Laboratory of Functional Phytochemicals Research and Utilization, Guangxi Institute of Botany, Guilin 541006, China
[c] Usaien Pharmaceutical Company, Ltd., 1-4-6 Zaimoku, Saga 840-0055, Japan

Abstract – In a phytochemical and chemotaxonomical investigation of *Castanopsis* species (Fagaceae), new monomeric and dimeric ellagitannins, named carlesiins A (**1**) and B (**2**), were isolated from fresh leaves of *Castanopsis carlesii* along with 55 known compounds. Carlesiin A was identified as 1-*O*-galloyl-4,6-(*S*)-tergalloyl-β-D-glucose. Carlesiin B is a macrocyclic ellagitannin dimer with a symmetrical structure composed of two tergalloyl and two glucopyranose moieties. Their structures were elucidated based on spectroscopic and chemical evidence.

INTRODUCTION

The species in the *Castanopsis* (Fagaceae) genus are evergreen trees that are found in East Asia, sometimes as the dominant species in a forest. These trees are often used as forestry or ornamental trees, and the wood is an important construction material. There are about 120 species in the genus, but the chemical compositions of only a few species have been studied. The leaves of Chinese and Japanese *Castanopsis* sp. are rich sources of polyphenols, and each species has a unique phenolic composition. We conducted a phytochemical and chemotaxonomical investigation of the *Castanopsis* sp. to determine how these plant resources could be effectively utilized. Our investigations of *C. cuspidata* var. *sieboldii*,[1,2] *C. hystrix*,[3,4] *C. fissa*,[5] and *C. sclerophylla*,[6,7] have revealed the presence of characteristic metabolites,

This paper is dedicated to Prof. Ei-ichi Negishi on the occasion of his 77th birthday.

including triterpene hexahydroxydiphenoyl (HHDP) esters, phenylpropanoid-substituted flavan-3-ols, ellagitannins, and galloyl esters of quinic acid. In the present study, we examined the leaves of *C. carlesii* collected in Southern China and isolated two new ellagitannins and 55 known compounds. The differences in the phenolic composition compared to other species are also discussed.

RESULTS AND DISCUSSION

The fresh leaves were extracted with 80% aqueous acetone, and the extract was subjected to a combination of column chromatography using Sephadex LH-20, Diaion HP20SS, MCI gel CHP 20P, Toyopearl Butyl 650C, Chromatorex ODS, Toyopearl HW F40, and Avicel cellulose to afford 57 compounds, including the two new ellagitannins **1** and **2** (Figure 1). The known compounds were identified by comparison of their spectroscopic data to data acquired from authentic samples and literature data (see Experimental).

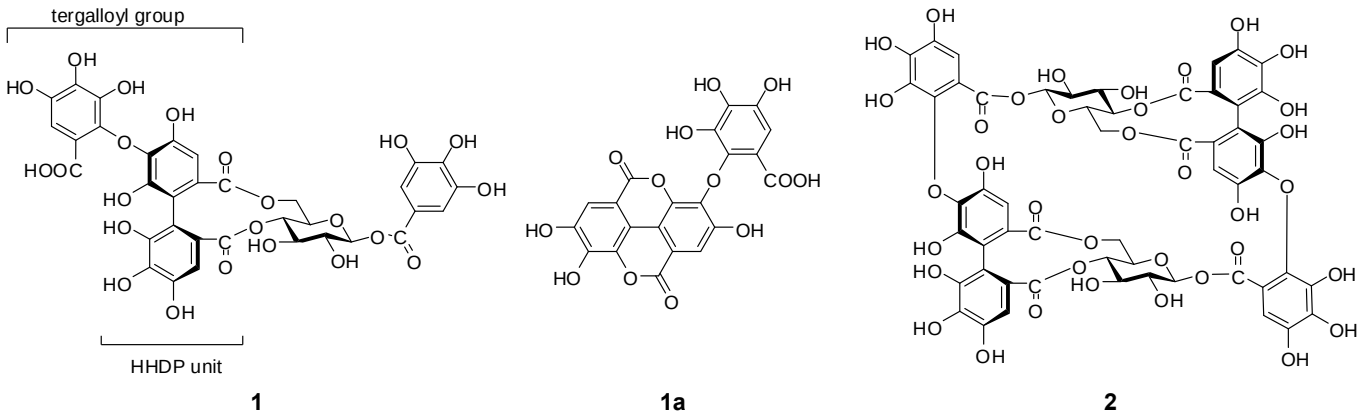


Figure 1. Structures of **1**, **2**, and **1a**

Carlesiin A (**1**) was obtained as a brown amorphous powder, which gave a dark blue color with ethanolic $FeCl_3$ reagent. The molecular formula $C_{34}H_{26}O_{23}$ was determined by high-resolution fast atom bombardment mass spectrometry (HR FAB-MS), which showed the $[M+H]^+$ ion peak at *m/z* 803.0962 (Calcd for $C_{34}H_{27}O_{23}$, 803.0943). The proton nuclear magnetic resonance (^{1}H-NMR) spectrum of **1** (Table 1) showed an anomeric proton signal at δ 5.72 (d, J = 8.2 Hz, H-1). The large coupling constants of the remaining sugar proton signals ($J_{2,3}$, $J_{3,4}$, and $J_{4,5}$ = 9–10 Hz) indicated that this sugar was a β-glucopyranose in the $^4C_1$ conformation. Downfield shifts of the glucose H-1, H-4 and H-6 indicated esterification of these positions. As for the acyl groups, the presence of a galloyl group was apparent from the ^{1}H [δ 7.17 (2H, s)] and ^{13}C NMR signals. Comparison among the chemical shifts of the remaining three aromatic protons [δ 6.55 (H-3'), 6.73 (H-3), 6.92 (H-6")] and 18 aromatic and three carboxyl carbons,

indicated the presence of a HHDP ester group with an additional gallic acid moiety. Among the aromatic carbon signals, the signal at δ 132.0 is characteristic of the C-2' of tergalloyl group.[8,9] This was confirmed by acid hydrolysis of **1**, which gave gallic acid and tergallic acid dilactone (**1a**). These compounds were also isolated from the same extract in the present study. The heteronuclear multiple bond connectivity (HMBC) correlations between the glucose H-1 and galloyl carboxyl carbon indicated that the galloyl group was attached to an anomeric hydroxyl group (Figure 2). The large difference in the ^{1}H NMR chemical shifts (Δδ 1.36) of the glucose C-6 methylene protons suggests that the HHDP unit of the tergalloyl moiety forms macrocyclic esters with O-4 and O-6 of the glucose. This was confirmed by the HMBC correlations of H-4 (δ 4.92) and H-6 (δ 3.79 and 5.15) with the tergalloyl C-7 (δ 168.2) and C-7' (δ 168.1) carboxyl carbons, respectively (Figure 2). The orientation of the tergalloyl esters, that is, the location of the additional gallic acid moiety, was determined by observation of the HMBC correlations of H-3' (δ 6.55) with C-4' (δ 149.4), C-5' (δ 136.3) and C-7'. Furthermore, atropisomerism of the tergalloyl biphenyl bond was revealed to be *S*, because the circular dichroism (CD) spectrum exhibited a strong positive Cotton effect at 236 nm ($[\theta]_{236}$ $+8.2\times10^5$) and a negative effect at 261 nm ($[\theta]_{261}$ -3.3×10^5).[10] The glucose core was deduced to be in the D configuration because coexisting known ellagitannins isolated from the same plant source have a D-glucopyranose core. Based on these spectroscopic and chemical results, the structure of compound **1** was confirmed to be 1-*O*-galloyl-4,6-*O*-(*S*)-tergalloyl-β-D-glucose and was named carlesiin A.

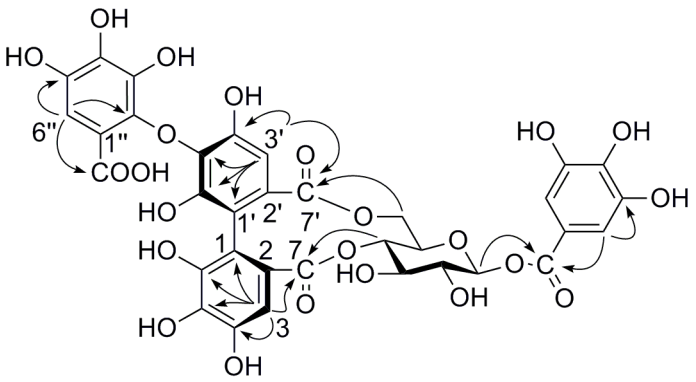


Figure 2. Selected HMBC correlations (H to C) of **1**

The ^{1}H NMR spectrum of carlesiin B (**2**) was closely related to that of **1**, showing three aromatic singlets at δ 6.46, 6.70, and 7.00, and a set of signals attributable to 1,4,6-*O*-acylated β-glucopyranose (Table 1). The presence of gallic acid trimer and glucopyranose residues was also apparent from the ^{13}C NMR signals. However, signals for the galloyl ester were not observed in the spectra of **2**, and the matrix assisted laser desorption/ionization time of flight mass spectrometry (MALDI-TOF-MS) $[M+Na]^+$ peak was at *m*/*z* 1287. The results indicate that **2** is a dimeric ellagitannin with a symmetrical structure composed of two glucopyranose and two triphenoyl groups. The C-2' of the triphenoyl group resonated at δ 127.9, which is different from that of tergalloyl group (δ 132.0).[8,9] The chemical shift was related to those of valoneoyl (C-2': about δ 126)[7] and macaranoyl groups (C-2': about δ 124),[9] which are positional isomers of the tergalloyl group (Figure 4).

Table 1. ^{1}H-(500 MHz) and ^{13}C-(125 MHz) NMR data for **1** and **2** (in acetone-d_6+D_2O)[a]

	1			**2**	
Position	^{1}H	^{13}C	Position	^{1}H	^{13}C
Anomer			Anomer		
1	5.72 (d, 8.2)	95.8	1	5.54 (d, 8.0)	96.5
2	3.71 (dd, 8.2, 9.8)	74.4	2	3.49 (dd, 8.0, 9.8)	74.1
3	3.84 (t, 9.8)	75.2	3	3.52 (t, 9.8)	75.2
4	4.92 (t, 9.8)	72.6	4	4.84 (t, 9.8)	73.1
5	4.11 (dd, 5.5, 9.8)	72.9	5	4.07 (dd, 6.3, 9.8)	72.6
6	5.15 (dd, 6.2, 13.3)	64.0	6	5.21 (dd, 6.3, 13.4)	63.5
	3.79 (d, 13.3)			3.76 (d, 13.4)	
Tergalloyl			Tergalloyl		
1		116.3	1		115.4
2		125.5	2		125.1
3	6.73 (s)	108.1	3	6.70 (s)	107.6
4		145.1	4		145.3
5		136.7	5		136.7
6		144.2	6		145.4
7		168.2	7		171.1
1'		115.6	1'		115.4
2'		132.0	2'		127.9
3'	6.55 (s)	108.0	3'	6.46 (s)	109.2
4'		149.4	4'		145.3
5'		136.3	5'		136.2
6'		149.4	6'		144.6
7'		168.1	7'		168.0
1"		114.7	1"		113.8
2"		139.7	2"		142.7
3"		139.2	3"		139.6
4"		140.2	4"		139.6
5"		142.2	5"		140.2
6"	6.92 (s)	108.3	6"	7.00 (s)	108.6
7"		170.9	7"		166.1
Galloyl					
1		120.4			
2,6	7.17 (2H, s)	110.1			
3,5		146.0			
4		139.4			
7		165.5			

[a] Chemical shifts are given in δ values; multiplicities and coupling constants (J in Hz) in parentheses.

The acyl group was chemically confirmed by hydrolysis of **2**, which gave tergalloyl acid dilactone (**1a**). Furthermore, methylation and subsequent methanolysis of **2** afforded a trimethyl ester **2a**, which was identified as (*S*)-trimethyl octamethyltergallate ($[\alpha]_D^{23}$ -20.5) (Figure 4). The *S*-biphenyl bond was also

confirmed by observation of a positive Cotton effect at 235 nm ($[\theta]_{235}$ $+1.5\times10^6$) and negative Cotton effect at 261 nm ($[\theta]_{261}$ -3.0×10^5) in the CD spectrum of **2**.[10] Similar glucose proton signals for **2** and **1** suggested that the HHDP unit of the triphenoyl group was located at the O-4 and O-6 positions. This was confirmed by the HMBC correlations of ester carboxyl carbons C-7 and C-7' with glucose H-4 and H-6, respectively (Figure 3). The HMBC spectrum also revealed the remaining gallic acid unit of the triphenoyl group was attached to the glucose anomeric position. Thus, the structure of carlesiin B was determined (**2** in Figure 1). To the best of our knowledge, this compound is the first macrocyclic ellagitannin dimer with tergalloyl esters. The upfield shift of the tergalloyl C-2' was probably caused by steric strain, and the upfield sifts of the glucose protons compared to those of **1** could be explained by anisotropic effect of the aromatic rings.

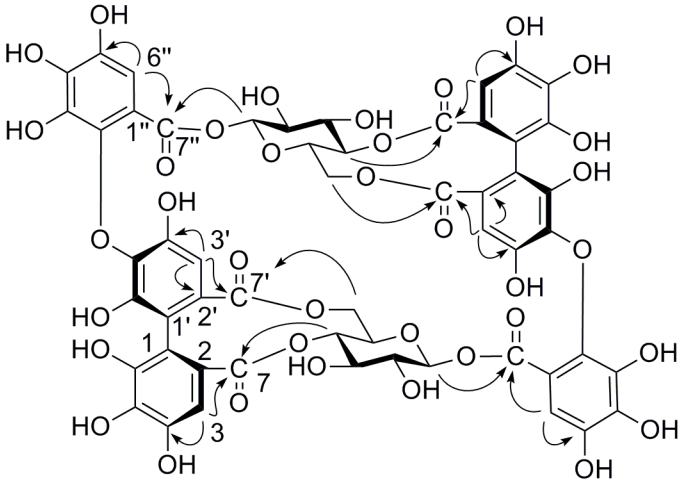
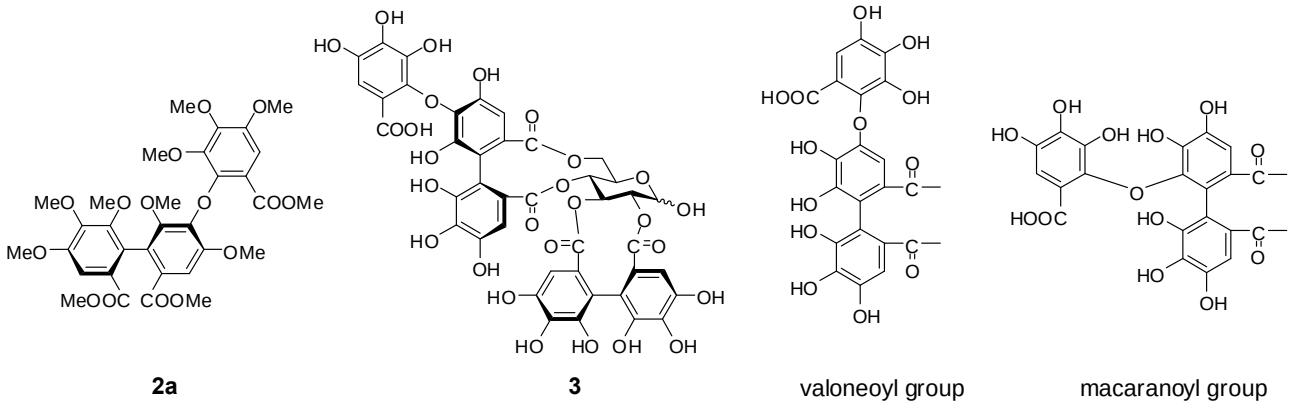


Figure 3. Selected HMBC correlations (H to C) of **2**.



2a **3** valoneoyl group macaranoyl group

Figure 4. Structures of **2a**, platycaryanin B (**3**), and valoneoyl and macaranoyl groups.

CONCLUSION

Although dimeric and oligomeric ellagitannins have been isolated from many plant families,[11] only two *Castanopsis* plants have been shown to contain the ellagitannin dimers. One is *C. sclerophylla*, which contains rugosin E with a valoneoyl group (Figure 4) as one of the major polyphenolic constituents (0.50% from fresh leaves).[7] The other is *C. carlesii*, which was examined in this study, although the yield of the dimeric ellagitannin **2** is very low (isolation yield 0.0007% from fresh leaves). *C. carlesii* contains monomeric ellagitannins as the major constituents, pedunculagin [2,3;4,6-bis-(*S*)-HHDP-D-glucose;

0.14%], 1(β)-*O*-galloyl pedunculagin (0.42% from fresh leaves), and platycaryanin B [2,3-(*S*)-HHDP-4,6-(*S*)-tergalloyl-D-glucose (**3**); 0.18%]. These were accompanied by their degradation products, ellagic acid (0.49%) and tergallic acid dilactone (**1a**) (0.08%). Unlike *C. sclerophylla*, *C. carlesii* contains ellagitannins with a tergalloyl group, such as platycaryanin B (**3**). This is an important characteristic feature of this plant from the viewpoint of chemotaxonomy of the genus *Castanopsis*. Carlesiins A (**1**) and B (**2**) are biogenetically related to platycaryanin B, and it should be noted that the symmetrical macrocyclic structure with tergalloyl ester groups of **2** is the first example in the gallic acid metabolites. Triterpene HHDP esters isolated from *C. cuspidata* var. *sieboldii*, *C. hystrix*, and *C. fissa*, and phenylpropanoid-substituted flavan-3-ols isolated from *C. hystrix*, and *C. sclerophylla* were not detected in the leaves of *C. carlesii*. Further phytochemical and chemotaxonomical studies on *Castanopsis* species are now underway.

EXPERIMENTAL

IR spectra were obtained with a JASCO FT/IR-410 spectrophotometer, UV spectra were obtained using a JASCO V-560 UV/Vis spectrophotometer, and optical rotations were measured with a JASCO DIP-370 digital polarimeter (Jasco Co., Tokyo, Japan). ^{1}H- and ^{13}C-NMR spectra were measured in acteone-d_6 or CD_3OD at 27 °C using a Varian Unity plus 500 spectrometer (500 MHz for ^{1}H and 125 MHz for ^{13}C) (Varian, Palo Alto, CA) or a JEOL JNM-AL 400 spectrometer (400 MHz for ^{1}H and 100 MHz for ^{13}C) (JEOL Ltd., Tokyo, Japan). Coupling constants are expressed in Hz and chemical shifts are given in δ (ppm). MS were recorded on a Voyager DE-PRO (Applied Biosystems, Foster City, CA) and a JEOL JMS-700N spectrometer (JEOL Ltd.). 2,5-Dihydroxybenzoic acid and glycerol were used as the matrices for MALDI-TOF-MS and FAB-MS measurements, respectively. Column chromatography was performed using a Diaion HP20SS (Mitsubishi Chemical, Tokyo, Japan), MCI gel CHP 20P (75–150 μm; Mitsubishi Chemical), Sephadex LH-20 (25–100 μm; GE Healthcare Bio-Science AB, Uppsala, Sweden), Toyopearl Butyl 650C (Tosoh Corp., Tokyo, Japan), Toyopearl HW F40 (Toshoh Corp.), Avicel Cellulose (Funakoshi Co., Tokyo, Japan), and Chromatorex ODS (100–200 mesh; Fuji Silysia Chemical, Tokyo, Japan) columns. Thin-layer chromatography was performed on precoated Kieselgel 60 F_{254} plates (0.2 mm thick; Merck, Darmstadt, Germany) and Cellulose plates (0.1 mm thickness; Merck), using solvent systems of toluene-ethyl formate-formic acid (1:7:1, v/v/v) and 2% acetic acid, respectively. Spots were detected by UV illumination (254 nm) and by spraying with a 2% ethanolic $FeCl_3$ and 10% sulfuric acid reagent, followed by heating.

Plant material

The leaves of *C. carlesii* were collected at Guangxi Institute of Botany, Guangxi, China, in August 2009,

and identified by Prof. Wei Fa-nan. The voucher specimen (CC20090826) was deposited in the Herbarium of Guangxi Institute of Botany, China.

Extraction and separation

The fresh leaves of *C. carlesii* (7.60 kg) were cut into small pieces and extracted with acetone-H_2O (8:2, v/v) at room temperature. After filtration, the plant debris remaining on the filter paper was extracted with the same solvent twice. The filtrate was concentrated under reduced pressure to give a dark green precipitate consisting mainly of chlorophylls and waxes, which were removed by filtration. The filtrate was then concentrated to give an extract (610 g). The extract was partitioned between Et_2O and water. The aqueous layer was subjected to Sephadex LH-20 column chromatography (10 cm i. d. × 40 cm) with water containing increasing proportions of methanol (MeOH) (0–100%, 10% stepwise elution) and finally 60% acetone to give the following eight fractions: 1 (153.3 g), 2 (35.5 g), 3 (33.9 g), 4 (82.0 g), 5 (122.5 g), 6 (17.9 g), 7 (18.9 g), and 8 (42.8 g). Fraction 5 gave a precipitate in aqueous MeOH, which was collected by filtration and identified as ellagic acid (37.2 g). The soluble part of the fraction was separated by a Diaion HP20SS column (6 cm i. d. × 40 cm) with 0–100% MeOH (10% stepwise elution) to give 9 subfractions. Subfraction 5-4 (15.7 g) was further separated by column chromatography using Sephadex LH-20 (0–100% MeOH, 10% stepwise elution), Toyopearl Butyl 650C (0–100% MeOH, 10% stepwise elution), Avicel Cellulose (2% AcOH), and MCI-gel CHP-20P (10–100% MeOH, 10% stepwise elution) to yield **1** (159 mg), **2** (56 mg), (–)-epicatechin (122 mg), pedunculagin (10.5 g), platycaryanin B (13.5 g), 1-*O*-galloyl-4,6-*O*-(*S*)-valoneoyl-β-D-glucose (38 mg), flavogallonic acid (24 mg), valoneic acid dilactone (3.31 g), tergallic acid dilactone (6.13 g), and isoquercitrin (34 mg).

The known compounds isolated from other fractions by similar chromatographic separation were as follows: 5-*O*-(β-apiosyl-(1→2)-*O*-β-xylopyranosyl)gentisic acid (192 mg), gentisic acid 5-*O*-β-D-glucoside (46 mg), and benzyl-α-L-rhamnopyranosyl(1→6)-β-D-glucoside (172 mg) from fraction 1; 1-*O*-galloyl-β-D-glucose (77 mg), 4,6-(*S*)-HHDP-D-glucose (494 mg), gentisic acid 5-*O*-β-D-xylopyranoside (160 mg), 1,6-di-*O*-galloyl-β-D-glucose (484 mg), brevifolin carboxylic acid (131 mg), and 6,7-dihydroxycoumarin (10 mg) from fraction 2; 4,6-*O*-(*S*)-valoneoyl-D-glucose (600 mg), gallic acid (725 mg), 6-*O*-galloyl-glucose (26 mg), (2*R*,3*R*)-taxifolin (191 mg), gentisic acid 5-*O*-β-D-(6'-*O*-galloyl)glucopyranoside (9 mg), 4-hydroxy-3-methoxyphenol l-*O*-β-D-(6'-*O*-galloyl)-glucoside (161 mg), 4-hydroxy-2-methoxyphenol 1-*O*-β-D-(6'-*O*-galloyl)glucoside (133 mg), 3,4-dihydroxyphenethyl alcohol 4-*O*-β-D-(6''-*O*-galloyl)glucopyranoside (13 mg), 4-methoxycatechol (2 mg), 4-*O*-(6'-*O*-galloyl-β-glucopyranosyl)-*cis*-*p*-coumaric acid (9 mg), and 2*R*,3*R*-dihydromyricetin 3-*O*-β-L-arabinopyranoside (25 mg) from fraction 3; sanguiin H-5 (29 mg), casuariin (1.78 g), platycariin

(210 mg), tamarixetin 3-*O*-α-rhamnopyranoside (525 mg), myricitrin (568 mg), mearnsitrin (3.43 g), gemin D (2.57 g), 1-*O*-galloyl-4,6-(*S*)-HHDP-β-D-glucose (880 mg), (+)-catechin (608 mg), and (+)-gallocatechin (8 mg) from fraction 4; flosin A (1.11 g), 1,2-di-galloyl-4,6-*O*-(S)-HHDP-β-D-glucose (20 mg), alnusnin B (156 mg), (–)-epigallocatechin 3-*O*-gallate (597 mg), (–)-epicatechin 3-*O*-gallate (314 mg), eschweilenol A (12 mg), and myricetin 3-*O*-(3"-*O*-galloyl)-α-rhamnopyranoside (69 mg) from fraction 6; 1,2,3,6-tetra-*O*-galloyl-β-D-glucose (48 mg), and nobotanin D (53 mg) from fraction 7; and 1(β)-*O*-galloyl-pedunculagin (32.1 g), platycaryanin A (311 mg), 1,2,3,4,6-penta-*O*-galloyl-β-D-glucose (12 mg), 1-desgalloyl eugeniin (17 mg), eugeniin (466 mg), cuspinin (305 mg), and rugosin C (16 mg) from fraction 8.

Carlesiin A (1). Brown amorphous powder, $[\alpha]_D^{24}$ –123.3 (*c* 0.14, MeOH); HR FAB-MS *m*/*z*: 803.0962 $[M+H]^+$ (Calcd for $C_{34}H_{27}O_{23}$: 803.0943); IR ν_{max} cm^{-1}: 3348, 1711, 1563, 1228; UV λ_{max} (MeOH) nm (log ε): 261 (4.53), 212 (5.07); CD (MeOH) [θ] (nm): $+8.2\times10^5$ (236), 0 (251), $–3.3\times10^5$ (261), 0 (275), $+8.6\times10^4$ (283), 0 (292); MALDI-TOF-MS *m*/*z*: 825 $[M+Na]^+$; ^{1}H-NMR (500 MHz, acetone-d_6+D_2O) and ^{13}C-NMR (125 MHz, acetone-d_6+D_2O). See Table 2.

Carlesiin B (2). Brown amorphous powder, $[\alpha]_D^{24}$ +12.0 (*c* 0.13, MeOH); MALDI-TOF-MS *m*/*z*: 1287 $[M+Na]^+$; HR FAB-MS *m*/*z*: 1265.1334 $[M+H]^+$ (Calcd for $C_{54}H_{41}O_{36}$: 1265.1378); IR ν_{max} cm^{-1}: 3350, 1702, 1560, 1214; UV λ_{max} (MeOH) nm (log ε): 263 (4.58), 212 (5.10); CD (MeOH) [θ] (nm): $+1.5\times10^6$ (235), 0 (253), $–3.0\times10^5$ (261), 0 (271), $+2.2\times10^5$ (279), 0 (292); ^{1}H-NMR (500 MHz, acetone-d_6+D_2O) and ^{13}C-NMR (125 MHz, acetone-d_6+D_2O). See Table 2.

Acid hydrolysis of 1 and 2

A solution of **1** (1.6 mg) in 0.5 mol/L H_2SO_4 (0.2 mL) was kept at 90–100 °C for 2 h. An equal volume of MeOH was added to dissolve the resulting precipitates, and the products were analyzed by HPLC under the following conditions: column, Cosmosil 5C_{18} AR II (250×4.6 mm i.d., Nacalai Tesque Inc., Kyoto, Japan); solvent, MeCN in 50 mmol/L H_3PO_4; solvent gradient, 4–30% MeCN (39 min) and 30–75% MeCN (15 min); flow rate, 0.8 mL/min; and detector, Jasco photodiode array detector MD-910. The peaks at 8.69 min and 24.57 min corresponded to the derivatives prepared from gallic acid and tergallic acid dilactone, respectively. Hydrolysis of **2** (1.0 mg) was also achieved in a similar manner, and the HPLC showed a peak for the tergallic acid dilactone.

Production of 2a from 2

A solution of **2** (5.2 mg) in MeOH (0.5 mL) was treated with an ether solution of CH_2N_2 at 0 °C for 2 h.

After concentration under reduced pressure, the residue was dissolved in 1% NaOH in 50% MeOH (1.0 mL) and heated at 80 °C for 1 h. The reaction mixture was acidified with 0.5 mol/L HCl (1.5 mL) and partitioned between ethanol and water. The organic layer was dried with anhydrous Na_2SO_4, and then treated with an ether solution of CH_2N_2 at 0 °C for 2 h. The reaction mixture was concentrated, and the product was purified by silica gel chromatography with toluene-acetone (100:1–92:8, 1% stepwise elution) to yield (*S*)-trimethyl octa-*O*-methyl tergallate (**2a**) (2.5 mg) as a colorless syrup, $[\alpha]_D^{23}$ –20.5 (*c* 0.13, $CHCl_3$); MALDI-TOF-MS *m*/*z*: 683 $[M+Na]^+$; ^{1}H-NMR (400 MHz, $CDCl_3$) δ: 3.35, 3.59, 3.60, 3.61 (×2), 3.74, 3.79, 3.86 (×2), 3.89, 3.92 (each 3H, s, OMe), 7.16, 7.32, 7.37 (each 1H, s).

ACKNOWLEDGEMENTS

The authors are grateful to Mr. K. Inada, Mr. N. Yamaguchi and Mr. N. Tsuda (Nagasaki University Joint Research Center) for NMR and MS measurements.

REFERENCES AND NOTES

1. M. Ageta, K. Ishimaru, G. Nonaka, and I. Nishioka, *Chem. Pharm. Bull.*, 1988, **36**, 870.
2. M. Ageta, G. Nonaka, and I. Nishioka, *Chem. Pharm. Bull.*, 1988, **36**, 1646.
3. H. F. Chen, T. Tanaka, G. Nonaka, T. Fujioka, and K. Mihashi, *Phytochemistry*, 1993, **32**, 1457.
4. H. F. Chen, T. Tanaka, G. Nonaka, T. Fujioka, and K. Mihashi, *Phytochemistry*, 1993, **33**, 183.
5. Y. L. Huang, T. Tsujita, T. Tanaka, Y. Matsuo, I. Kouno, D. P. Li, and G. Nonaka, *Phytochemistry*, 2011, **72**, 2006.
6. Y. L. Huang, Y. Matsuo, T. Tanaka, I. Kouno, D. P. Li, and G. Nonaka, *Heterocycles*, 2011, **83**, 2321.
7. Y. L. Huang, T. Tanaka, Y. Matsuo, I. Kouno, D. P. Li, and G. Nonaka, *Phytochemistry Letters*, 2012, **5**, 158.
8. T. Tanaka, S. Kirihara, G. Nonaka, and I. Nishioka, *Chem. Pharm. Bull.*, 1993, **41**, 1708.
9. J. H. Lin, M. Ishimatsu, T. Tanaka, G. Nonaka, and I. Nishioka, *Chem. Pharm. Bull.*, 1990, **38**, 1844.
10. T. Okuda, T. Yoshida, T. Hatano, T. Koga, N. Toh, and K. Kuriyama, *Tetrahedron Lett.*, 1982, **23**, 3937.
11. T. Okuda, T. Yoshida, and T. Hatano, *Prog. Chem. Org. Nat. Prod.*, 1995, **66**, 1.