[Chem. Pharm. Bull. 30(6)2061—2067(1982)]

Tannins and Related Compounds. IV.[1] Seven New Phenol Glucoside Gallates from *Quercus stenophylla* Makino (1)

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(Received December 5, 1981)

Along with gallic acid, ellagic acid, (+)-catechin and procyanidins B-1 and B-3, seven new gallates of phenol glucosides (compounds **1**—**7**) have been isolated from the bark of *Quercus stenophylla* Makino (Fagaceae). On the basis of chemical and spectral evidence, the structures of compounds **1**, **2**, **4**, **5** and **6** have been established respectively as 6″-*O*-gallate (**1**), 3″-*O*-gallate (**2**), 4′, 6″-di-*O*-gallate (**4**), 4″,6″-di-*O*-gallate (**5**) and 3″,4″,6″-tri-*O*-gallate (**6**) of salidroside (*p*-hydroxyphenethyl alcohol 1-*O*-*β*-D-glucopyranoside). Similarly, compounds **3** and **7** have been characterized as 3′,4′-dihydroxyphenethyl alcohol 1-*O*-*β*-D-(6″-*O*-galloyl)-glucopyranoside (**3**) and 2,4,6-trimethoxyphenol 1-*O*-*β*-D-(6′-*O*-galloyl)-glucopyranoside (**7**), respectively.

Keywords——*Quercus stenophylla*; Fagaceae; salidroside gallates; 3,4-dihydroxyphenethyl alcohol glucoside gallate; 2,4,6-trimethoxyphenol glucoside gallate; procyanidin B-1; procyanidin B-3

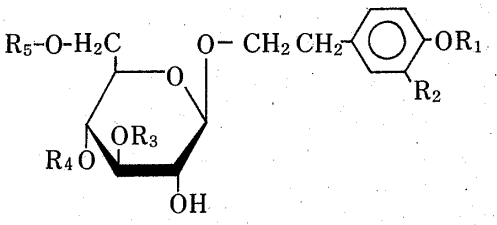
Although the members of the genus *Quercus* (Fagaceae) are known to produce a large amount of polyphenolic secondary metabolites, little phytochemical examination of these polyphenolic compounds has been done except for Schmidt and Mayer's notable work on ellagitannins; they proposed structures for pedunculagin,[2] castalin,[3] castalagin,[4] castavaloninic acid,[5] valolaginic acid[6] and isovalolaginic acid[7] isolated from *Quercus pedunculata*, *Q. sessiflora*, *Q. valonea*, *etc.* Wide distributions of low-molecular weight phenolics such as gallic acid, ellagic acid and catechin have also been recognized in this genus. As part of our continuing studies on tannins and related compounds, we have examined the polyphenolic constituents of the bark of *Quercus stenophylla* Makino (Japanese name: Urajirogashi), which has been used in Japan as a remedy for biliary and urinary calculi, and we isolated and characterized seven new phenol glucoside gallates, in addition to gallic acid,[5] ellagic acid,[5] (+)-catechin and procyanidins B-1[9] and B-3.[9a] This paper deals with the isolation and characterization of these compounds.

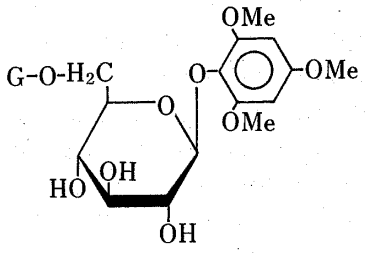
Initial separation of constituents in the ethyl acetate-soluble portion of the aqueous acetone extract was achieved through preparative column chromatography on Sephadex LH-20 using ethanol. Subsequent chromatography of each fraction on Sephadex LH-20 using acetone and 80% aqueous methanol afforded compounds **1**—**7**.

Compound **1** (**1**), after crystallization from water, yielded pale yellow needles, mp 116—117°C, $[\alpha]_D$ +15.1° (acetone), $C_{21}H_{24}O_{11}\cdot 1/2H_2O$, and was strongly positive (a dark blue color) to the ferric chloride reagent. The occurrence of a galloyl function in **1** was easily deduced from the proton nuclear magnetic resonance (^{1}H-NMR) [δ 7.17 (2H), s] and carbon-13 nuclear magnetic resonance (^{13}C-NMR) [δ 166.9, 145.6 (2C), 138.6, 121.0, 109.6 (2C)] spectra. On enzymatic hydrolysis with tannase in aqueous solution, **1** gave gallic acid and a colorless hydrolysate (**1a**), mp 162—163°C, $[\alpha]_D$ −28.2° (methanol). The ^{1}H-NMR spectrum of **1a** exhibited A_2B_2-type aromatic signals [δ 6.70 (2H), d, J=8 Hz; δ 7.08 (2H), d, J=8 Hz] and a two-proton triplet signal (δ 2.81, J=7 Hz) due to benzylic methylene protons. Furthermore, an anomeric proton signal (δ 4.32, d, J=8 Hz) and multiplet signals (δ 3.10—4.12) suggesting the existence of one mole of glucose residue in the molecule were observed. From these spectral

data, compound **1a** was likely to be a glucoside of *p*-hydroxyphenethyl alcohol, and was finally identified as salidroside, which had been isolated from *Salix triandra* L.[10] The location of the galloyl group in **1** was determined to be C(6) in the glucose moiety by comparison of the ^{13}C-NMR spectrum with that of salidroside; among six sugar carbon signals in **1** a triplet signal (in the off-resonance spectrum) easily assignable to the C(6)-carbon was shifted downfield (δ 64.4), while the C(5)-carbon signal showed an upfield shift (δ 74.5) as compared with those in salidroside [δ 62.6, C(6); δ 77.7, C(5)] (Table I). Accordingly, compound **1** was characterized as *p*-hydroxyphenethyl alcohol 1-*O*-β-D-(6″-*O*-galloyl)-glucopyranoside (**1**).

Compound **2** (**2**) was an off-white amorphous powder, $[\alpha]_D$ +4.5° (methanol), $C_{21}H_{24}O_{11}\cdot H_2O$, whose ^{1}H-NMR spectrum suggested the presence of a galloyl group [δ 7.12 (2H), s] and an aglycone moiety [δ 6.68 (2H), d, J=8 Hz; δ 7.05 (2H), d, J=8 Hz; δ 2.38 (2H), t, J=7 Hz] similar to those of compound **1**. This was also supported by the ^{13}C-NMR spectrum, which bore a close resemblance to that of compound **1** (Table I). Furthermore, the ^{13}C-NMR chemical shifts of sp^3-carbon signals indicated the existence of a glucose moiety in **2**. In the ^{1}H-NMR spectrum a triplet signal attributable to an ester-bearing proton was observed at lower filed (δ 5.12, J=9 Hz) and this signal could be assigned to either C(3)- or C(4)-hydrogen in the glucose moiety since this triplet signal remained unchanged upon irradiation of the anomeric proton (δ 4.42, d, J=8 Hz). The galloyl group could be conclusively placed on the C(3)-position in the glucose moiety based upon the fact that the C(3)-carbon signal was shifted downfield upon acylation of the hydroxy group, whereas the neighboring C(2)- and C(4)-carbon atoms resonated at higher filed than those of salidroside. Thus, the structure **2** was assigned to this compound.





7

1: $R_1=R_2=R_3=R_4=H$, $R_5=G$
2: $R_1=R_2=R_4=R_5=H$, $R_3=G$
3: $R_1=R_3=R_4=H$, $R_2=OH$, $R_5=G$
4: $R_2=R_3=R_4=H$, $R_1=R_5=G$
5: $R_1=R_2=R_3=H$, $R_4=R_5=G$
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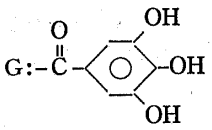

Chart 1

Compound **3** (**3**), an off-white amorphous powder, $[\alpha]_D$ −36.2° (acetone), $C_{21}H_{24}O_{12}\cdot H_2O$, gave the ^{1}H NMR spectrum almost identical with that of compound **1** except for ABX-type aromatic proton signals (δ 6.52, dd, J=2, 8 Hz; δ 6.69, d, J=8 Hz; δ 6.72, d, J=2 Hz). This fact coupled with the ^{13}C-NMR analysis which revealed the presence of a 3′,4′-dihydroxyphenethyl alcohol moiety (δ 116.0, 116.8, 121.6, 131.1, 144.1, 145.6) and six carbon signals due to a glucose residue analogous to that in compound **1** enabled us to establish the structure as 3′,4′-dihydroxyphenethyl alcohol 1-*O*-β-D-(6″-*O*-galloyl)-glucopyranoside (**3**).

Compound **4** (**4**), an off-white amorphous powder, $[\alpha]_D$ −21.0° (acetone), $C_{28}H_{28}O_{15}\cdot H_2O$, contained two galloyl groups as revealed by the ^{1}H-NMR spectrum (δ 7.17, 7.26, s). Close resemblance of aliphatic proton and carbon signals to those in compound **1** confirmed that one galloyl group was located at the C(6)-position in the glucose residue. The position of the other galloyl group was determined to be C(4′) in the aglycone moiety on the ground that A_2B_2-type aromatic proton signals were shifted downfield [δ 7.07 (2H), J=8 Hz; δ 7.19 (2H), J=8 Hz]

upon acylation of the phenolic hydroxyl group. Consequently, the structure of compound **4** is represented by the formula **4**.

Compound **5** (**5**), an off-white amorphous powder, $[\alpha]_D$ +20.9° (acetone), $C_{28}H_{28}O_{15}\cdot 1/2\text{-}H_2O$, showed two galloyl peaks (δ 7.14, 7.16, s) in the ^{1}H-NMR spectrum. The presence of a *p*-hydroxyphenethyl alcohol moiety and a glucose moiety in **5** was confirmed by analysis of the ^{13}C-NMR spectrum (Table I). The locations of the two galloyl groups were determined as follows. In the ^{1}H-NMR spectrum a triplet signal (δ 5.13, J=8 Hz) and a pair of double-doublet signals (δ 4.20, J=12, 4 Hz; δ 4.42, J=12, 3 Hz) due to protons attached to the galloyl-bearing carbon atoms were observed at relatively lower field. The latter two signals could be assigned to the C(6)-protons in the glucose moiety owing to a large geminal coupling constant and chemical shifts analogous to those of compounds **1**, **3** and **4**. The assignment of the triplet signal was achieved by the spin-decoupling technique (Fig. 1). Upon irradiation of the anomeric proton signal (δ 4.50, d, J=8 Hz) the triplet signal (δ 3.40, J=8 Hz) changed to a doublet, thus, permitting the assignment of this signal to the C(2)-proton. Since this C(2)-proton signal was shown not to be coupled with the triplet signal at δ 5.13 on irradiation of the C(2)-proton, the triplet signal in question was attributed to the C(4)-proton in the glucose moiety. On the basis of these results the structure of compound **5** was formulated as **5**.

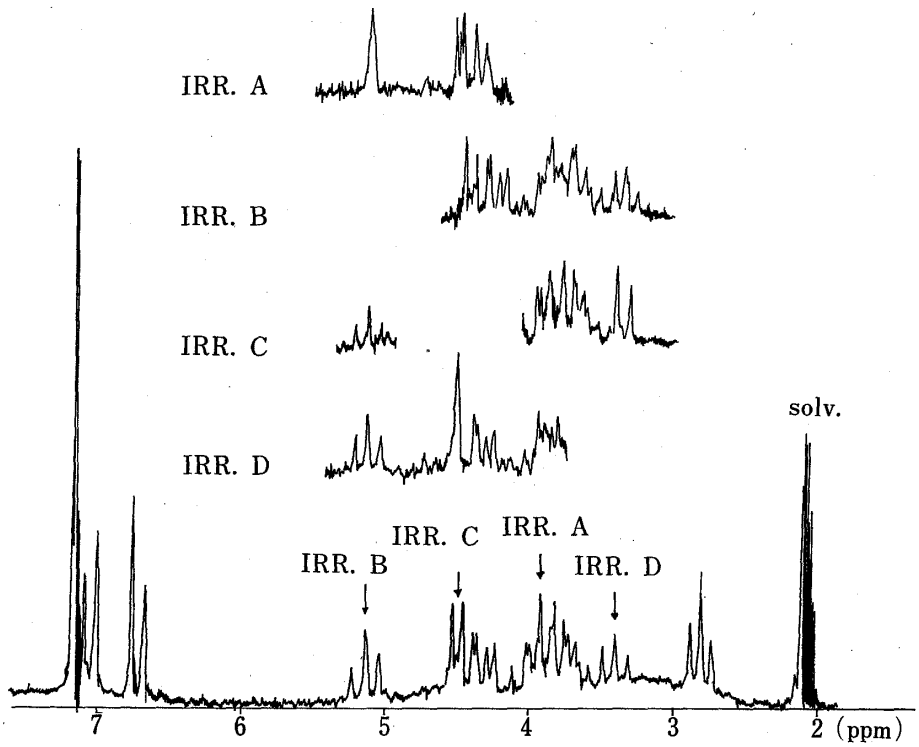

Fig. 1. ^{1}H-NMR Decoupling Experiments on Compound **5** (in Acetone-d_6)

IRR. stands for irradiation.

Compound **6** (**6**), an off-white amorphous powder, $[\alpha]_D$ −20.3° (acetone), $C_{35}H_{32}O_{19}\cdot 3/2H_2O$, was shown to be a salidroside derivative having three galloyl groups by analyses of the ^{1}H-NMR (δ 7.02, 7.04, 7.18, s) and ^{13}C-NMR (Table I) spectra. The determination of the positions of the galloyl groups was achieved by spin-decoupling experiments of the ^{1}H-NMR spectrum as shown in Fig. 2. Two triplet signals (δ 5.36, J=9 Hz; δ 5.52, J=9 Hz) in the lower field due to methine protons bearing galloyl groups did not change on irradiation of the signal due to the anomeric proton (δ 4.69, d, J=8 Hz), thus indicating that these two triplets could be ascribed

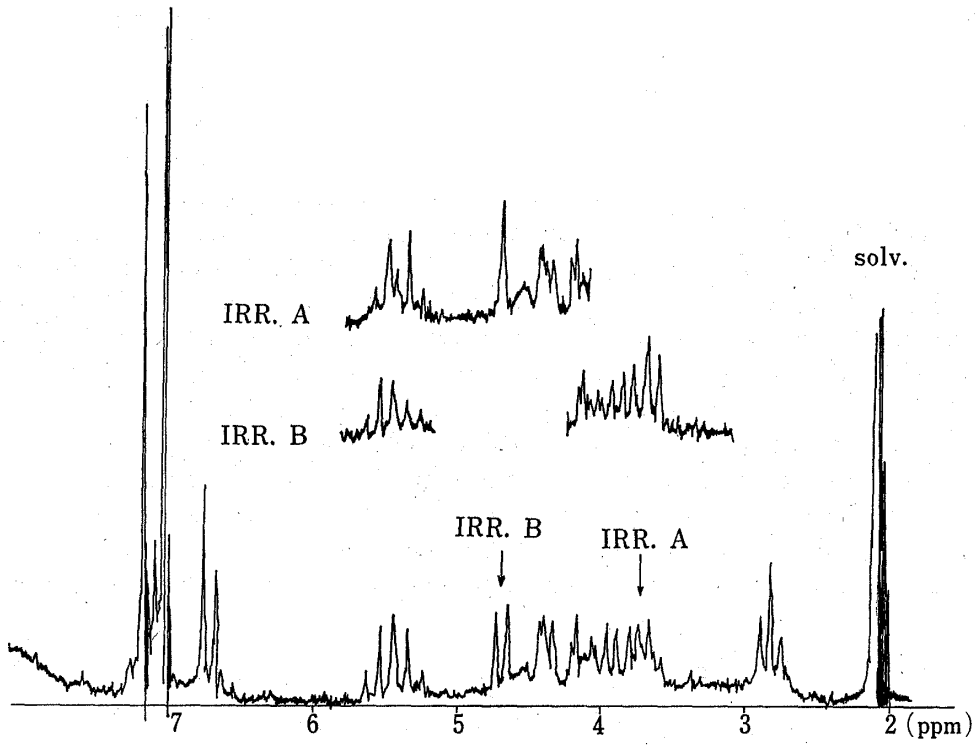

Fig. 2. ^{1}H-NMR Decoupling Experiments on Compound **6** (in Acetone-d_6)
IRR. stands for irradiation.

TABLE I. ^{13}C-NMR Chemical Shifts (δ-values)[a)]

	1a	**1**	**2**	**4**	**5**	**6**	**3**
Glucose							
1″	104.6	103.5	104.0	103.6	103.6	103.9	103.8
2″	74.9	74.2	73.4	74.2	74.5	73.1	74.7
3″	77.9	77.2	79.0	77.2	75.2	75.5	77.6
4″	71.4	71.4	69.7	71.0	72.0	69.9	71.2
5″	77.7	74.5	77.6	74.5	72.5	72.8	74.7
6″	62.6	64.4	62.3	64.5	63.5	63.3	64.5
Aglycone							
1	71.9	71.0	72.1	71.0	71.5	71.7	71.5
2	36.3	35.7	36.2	36.0	35.7	36.0	36.2
1′	130.4	129.7	130.5	136.8	129.6	129.8	131.1
2′	130.6	130.3	130.7	130.4	130.4	130.4	116.8
3′	115.8	115.6	115.9	121.1	115.7	115.7	144.1
4′	156.3	155.9	156.3	150.1	155.9	156.2	145.6
5′	115.8	115.6	115.9	121.1	115.7	115.7	116.0
6′	130.6	130.3	130.7	130.4	130.4	130.4	121.6
Galloyl							
1		121.0	121.5	120.2 121.0	120.5 120.7	120.5 121.2 121.3	121.0
2		109.6	110.2	109.6 110.0	109.7 109.9	109.8(6C)	110.0
3		145.6	146.0	145.7 145.8	145.7(4C)	145.5(6C)	145.6
4		138.6	139.4	138.7 139.3	138.8 139.0	138.5(2C) 138.7	138.9
–COO–		166.9	167.9	165.7 167.0	166.5 166.8	165.5 165.9	167.0

a) Measured in acetone-d_6+D_2O at 25.05 MHz with TMS as an internal standard.

to C(3)- and C(4)-protons in the glucose moiety. A multiplet signal at around δ 4.40 might be assigned to the C(6)-methylene protons on the basis of the close similarities in the chemical shifts to those of compounds **1**, **3**, **4** and **5**. Further support for the existence of a galloyl group at the C(6)-position was provided by the ^{13}C-NMR chemical shift of the C(6)-carbon signal which, like that of compound **5**, was shifted downfield (+0.7 ppm) as compared with that of compound **1a**. These spectral data permitted the assignment of the structure (**5**) for this compound.

Compound **7** (**7**), colorless needles, mp 161—163°C, $[\alpha]_D$ −36.8° (methanol), $C_{22}H_{26}O_{13}\cdot 1/2H_2O$, showed a molecular ion peak at m/z 498 in the field-desporption mass spectrum (FD-MS). The ^{1}H-NMR spectra suggested the presence of a galloyl group (δ 7.10, s) and a sugar moiety (δ 64.6, 71.0, 74.4, 75.0, 77.3, 102.2). Enzymatic hydrolysis of **7** with tannase afforded gallic acid and a hydrolysate (**7a**), mp 210—212°C, $[\alpha]_D$ −86.2° (H_2O). The latter exhibited signals due to two aromatic protons (δ 6.39, s) and three methoxyl groups [δ 3.64 (3H), 3.71 (6H)] linked to the aglycone moiety in the ^{1}H-NMR spectrum. **7a**, when incubated with crude hesperidinase, yielded glucose and 2,4,6-trimethoxyphenol (**7b**), mp 145—146°C, thus establishing the structure of **7a** to be 2,4,6-trimethoxyphenol 1-*O*-glucoside. The mode of glucosidic linkage was determined to be β-form based on the coupling constant of the anomeric proton signal (δ 4.77, d, J=8 Hz) in the ^{1}H-NMR spectrum of **7a**. Furthermore, examination of the ^{1}H- and ^{13}C-NMR spectra of **7** allowed the galloyl group to be located on the C(6)-hydroxyl group in the glucose moiety [δ 4.39, dd, J=12, 6 Hz; δ 4.65, dd, J=12, 2 Hz, C(6)-H. δ 64,6, t, C(6)]. Based upon the results described above, the structure of **7** was characterized as 2,4,6-trimethoxyphenol 1-*O*-β-D-(6′-*O*-galloyl)-glucopyranoside (**7**).

Biological tests of these compounds and further chemical studies on the phenolic consti tuents of *Quercus stenophylla* MAKINO are in progress.

Experimental

All melting points were determined on a Yanagimoto micro-melting point apparatus and are uncorrected. Optical rotations were measured with a JASCO DIP-4 digital polarimeter (cell length: 0.5 dm). IR spectra were obtained with a JASCO IR-G spectrometer. FD-MS and EI-MS were taken with JEOL D-300 and JEOL-01SG spectrometers. ^{1}H-NMR and ^{13}C-NMR spectra were recorded on JEOL PS-100 and JEOL FX-100 spectrometers with tetramethylsilane (TMS) as an internal standard, and chemical shifts are expressed in δ-values (ppm). Thin-layer chromatography (TLC) was conducted on precoated Kieselgel 60 F_{254} plates (Merck, 0.20 mm thick) and precoated cellulose F_{254} plates (Merck, 0.10 mm thick), and the spots were detected by spraying 2% ethanolic ferric chloride, anisaldehyde–sulfuric acid and aniline–hydrogen phthalate reagents. Column chromatography was carried out with Sephadex LH-20 (Pharmacia Fine Chemicals, 25—100 mμ) and Kieselgel 60 (Merck, 70—230 mesh).

Extraction and Isolation——The air-dried bark (3.35 kg) of *Quercus stenophylla* MAKINO collected at Tokushima Prefecture (June, 1980) was powdered and extracted with 80% aqueous acetone at room temperature. The aqueous solution, after removal of the acetone under reduced pressure, afforded precipitates which were removed by filtration. The filtrate was extracted successively with Et_2O and AcOEt. The AcOEt-soluble portion (114.4 g) thus obtained was subjected to column chromatography on Sephadex LH-20. Elution with EtOH yielded three fractions; Fr. I (60.8 g), II (18.2 g) and III (17.7 g).

Subsequent separation of Fr. I on Sephadex LH-20 using acetone afforded three fractions; Fr. I-A (25.5 g), I-B (12.7 g) and I-C (5.3 g). Fr. I-A was crystallized from H_2O to give (+)-catechin (16.5 g) as colorless needles, mp 178—180°C, $[\alpha]_D^{20}$ +9.0° (c=1.07, acetone); this compound was identified by comparison of the IR and ^{1}H-NMR spectra with those of an authentic sample. The mother liquor, after the crystals of (+)-catechin had been filtered off, was separated by Sephadex LH-20 chromatography using acetone and MeOH to furnish gallic acid, mp 232—235°C, and compound **1** (2.5 g), the former being identified by comparison of the IR spectrum with that of an authentic sample. Fr. I-B in acetone deposited yellow fine needles (0.45 g), mp >300°C, which were shown to be identical with ellagic acid by IR spectral comparison. The mother liquor afforded a further crop of compound **1** (10.8 g) after purification by Sephadex LH-20 chromatography using acetone. Fr. I-C was rechromatographed over Sephadex LH-20 using acetone to give compound **7** (0.4 g).

Fr. II was further separated by Sephadex LH-20 chromatography using acetone into two fractions; Fr. II-A (9.0 g) and II-B (8.8 g). Rechromatography of Fr. II-A on Sephadex LH-20 using 80% aqueous MeOH furnished compounds **2** (0.7 g) and **3** (0.12 g). Similarly, repeated chromatography of Fr. II-B over

Sephadex LH-20 using acetone and 80% aqueous MeOH yielded compounds **4** (0.05 g), **5** (0.06 g) and **6** (0.2 g), along with procyanidins B-1 (0.05 g) and B-3 (0.2 g) which were identified by comparison with corresponding authentic samples (TLC, $[\alpha]_D$ and ^{1}H-NMR spectrum).

Compound 1 (**1**)——Pale yellow needles (H_2O), mp 116—117°C, $[\alpha]_D^{23}$ +15.1° (c=0.9, acetone). *Anal.* Calcd for $C_{21}H_{24}O_{11}\cdot 1/2H_2O$: C, 54.66; H, 5.46. Found: C, 54.92; H, 5.34. IR ν_{max}^{KBr} cm^{-1}: 3200—3600 (OH), 1710 (–COO–). ^{1}H-NMR (acetone-d_6): 2.78, 3.56 (each 2H, t, J=7 Hz, $-CH_2CH_2-$), 3.30—4.10 [4H, m, glu. C(2)–, C(3)–, C(4)–, C(5)–H], 4.40 (1H, d, J=8 Hz, anomeric H), 4.41 [1H, dd, J=12, 4 Hz, glu. C(6)–H], 4.58 [1H, dd, J=12, 2 Hz, glu. C(6)–H], 6.72, 7.03 (each 2H, d, J=8 Hz, arom. H), 7.17 (2H, s, gal. H). ^{13}C-NMR: Table I.

Hydrolysis of 1 with Tannase——A solution of **1** (103 mg) in H_2O was incubated with tannase at 37°C for 2 h. The reaction mixture was filtered and the filtrate was evaporated to dryness *in vacuo*. The residue was subjected to Sephadex LH-20 chromatography using EtOH to furnish gallic acid (12 mg), mp 240—241°C, and a hydrolysate (**1a**) (25 mg), colorless prisms (MeOH–AcOEt), mp 162—163°C, $[\alpha]_D^{21}$ −28.2° (c=0.13, MeOH). IR ν_{max}^{KBr} cm^{-1}: 3280 (OH), no ester absorption. ^{1}H-NMR (acetone-d_6+CD_3OD): 2.81, 3.30 (each 2H, t, J=7 Hz, $-CH_2CH_2-$), 3.10—4.12 (6H, m, glu. H), 4.32 (1H, d, J=8 Hz, anomeric H), 6.70, 7.08 (each 2H, d, J=8 Hz, arom. H). ^{13}C-NMR: Table I. This compound was shown to be identical with salidroside by comparison of its physical constants and chromatographic properties with those described in the literature.[10]

Compound 2 (**2**)——An off-white amorphous powder, $[\alpha]_D^{19}$ +4.5° (c=0.5, MeOH). *Anal.* Calcd for $C_{21}H_{24}O_{11}\cdot H_2O$: C, 53.62; H, 5.57. Found: C, 53.60; H, 5.61. ^{1}H-NMR (CD_3OD): 2.38 (2H, t, J=7 Hz, $-CH_2$–Ar), 3.30—4.20 [7H, m, glu. C(2)–, C(4)–, C(5)–, C(6)–H and $-OCH_2-$], 4.42 (1H, d, J=8 Hz, anomeric H), 5.12 [1H, t, J=9 Hz, glu. C(3)–H], 6.68, 7.05 (each 2H, d, J=8 Hz, arom. H), 7.12 (2H, s, gal. H). ^{13}C-NMR: Table I. FD-MS m/z: 453 (M+H)$^+$, 475 (M+Na)$^+$, 491 (M+K)$^+$.

Compound 3 (**3**)——An off-white amorphous powder, $[\alpha]_D^{23}$ −36.2° (c=0.24, acetone). *Anal.* Calcd for $C_{21}H_{24}O_{12}\cdot H_2O$: C, 51.85; H, 5.39. Found: C, 51.82; H, 5.38. ^{1}H-NMR (acetone-d_6): 2.75 (2H, t, J=7 Hz, $-CH_2$–Ar), 3.20—4.10 [6H, m, glu. C(2)–, C(3)–, C(4)–, C(5)–H and $-OCH_2-$]. 4.36 [1H, dd, J=12, 5 Hz, glu. C(6)–H], 4.36 (1H, d, J=8 Hz, anomeric H), 4.55 [1H, dd, J=12, 2 Hz, glu. C(6)–H, 6.52 1H, dd, J=8, 2 Hz, C(6′)–H], 6.69 [1H, d, J=8 Hz, C(5′)–H], 6.72 [1H, d, J=2 Hz, C(2′)–H], 7.15 (2H, s, gal. H). ^{13}C-NMR: Table I. FD-MS m/z: 469 (M+H)$^+$, 491 (M+Na)$^+$.

Compound 4 (**4**)——An off-white amorphous powder, $[\alpha]_D^{23}$ −21.0° (c=0.31, acetone). *Anal.* Calcd for $C_{28}H_{28}O_{15}\cdot H_2O$: C, 54.02; H, 4.86. Found: C, 53.95; H, 5.10. ^{1}H-NMR (acetone-d_6): 2.91 (2H, t, J=7 Hz, $-CH_2$–Ar), 3.10—4.12 [6H, m, glu. C(2)–, C(3)–, C(4)–, C(5)– and $-OCH_2-$], 4.36 [1H, dd, J=12, 6 Hz, glu. C(6)–H], 4.40 (1H, d, J=8 Hz, anomeric H), 4.58 [1H, dd, J=12, 2 Hz, glu. C(6)–H], 7.07, 7.19 (each 2H, d, J=8 Hz, arom. H), 7.26 (each 2H, s, gal. H). ^{13}C-NMR: Table I. FD–MS m/z: 605 (M+H)$^+$, 627 (M+Na)$^+$, 643 (M+K)$^+$.

Compound 5 (**5**)——An off-white amorphous powder, $[\alpha]_D^{26}$ +20.9° (c=0.31, acetone). *Anal.* Calcd for $C_{28}H_{28}O_{15}\cdot 1/2H_2O$: C, 54.81; H, 4.76. Found: C, 54.56; H, 5.11. ^{1}H-NMR (acetone-d_6): 2.87 (2H, t, J= 7 Hz, $-CH_2$–Ar), 3.40 [1H, t, J=8 Hz, glu. C(2)–H], 3.64—4.04 [4H, m, glu. C(3)–, C(5)–H and $-OCH_2-$], 4.20 [1H, dd, J=12, 4 Hz, glu. C(6)–H], 4.42 [1H, dd, J=12, 2 Hz, glu. C(6)–H], 4.50 (1H, d, J=8 Hz, anomeric H), 5.13 [1H, t, J=8 Hz, glu. C(4)–H], 6.71, 7.04 (each 2H, d, J=8 Hz, arom. H), 7.14, 7.16 (each 2H, s, gal. H). ^{13}C–NMR: Table I. FD-MS m/z: 605 (M+H)$^+$, 627 (M+Na)$^+$, 643 (M+K)$^+$.

Compound 6 (**6**)——An off-white amorphous powder, $[\alpha]_D^{27}$ −20.3° (c=1.03, acetone). *Anal.* Calcd for $C_{35}H_{32}O_{19}\cdot 3/2H_2O$: C, 54.27; H, 4.42. Found: C, 54.36; H, 4.44. ^{1}H-NMR (acetone-d_6): 2.83 (2H, t, J= 7 Hz, $-CH_2$–Ar), 3.67 [1H, t, J=8 Hz, glu. C(2)–H], 3.64—4.20 [3H, m, glu. C(5)–H and $-OCH_2-$], 4.69 (1H, d, J=8 Hz, arom. H), 7.02, 7.04, 7.18 (each 2H, s, gal. H). ^{13}C-NMR: Table I. FD-MS m/z: 757 (M+H)$^+$, 779 (M+Na)$^+$, 795 (M+K)$^+$.

Compound 7 (**7**)——Colorless needles (H_2O), mp 161—163°C, $[\alpha]_D^{18}$ −36.8° (c=0.62, MeOH). *Anal.* Calcd for $C_{22}H_{26}O_{13}\cdot 1/2H_2O$: C, 52.07; H, 5.36. Found: C, 52.39; H, 5.33. IR ν_{max}^{KBr} cm^{-1}: 3400 (OH), 1680 (–COO–). ^{1}H-NMR (acetone-d_6+CD_3OD): 3.64 (3H, s, OMe), 3.71 (6H, s, 2 OMe), 3.40—4.00 [4H, m, glu. C(2)–, C(3)–, C(4)–, C(5)–H], 4.39 [1H, dd, J=12, 6 Hz, glu. C(6)–H], 4.65 [1H, dd, J=12, 2 Hz, glu. C(6)–H], 4.98 (1H, d, J=8 Hz, anomeric H), 6.41 (2H, s, arom. H), 7.10 (2H, s, gal. H). ^{13}C-NMR (acetone-d_6+CD_3OD): 56.2 (q, 2 OMe), 60.6 (q, OMe), 64.6 [t, glu. C(6)], 71.0 [d, glu. C(4)], 74.4 [d, glu. C(2)], 75.5 [d, glu. C(5)], 77.3 [d, C(3)], 95.6 [d, C(3) and C(5)], 102.2 [d, glu. C(1)], 109.6 [d, gal. C(2) and C(6)], 121.1 [s, gal. C(1)], 134.1 [s, C(1)], 138.8 [s, gal. C(4)], 145.8 [s, gal. C(3) and C(5)], 154.1 [s, C(2) and C(6)], 154.9 [s, C(4)], 166.9 (s, –COO–). FD-MS m/z: 498 (M)$^+$.

Hydrolysis of 7 with Tannase——A suspension of **7** (105 mg) in H_2O was incubated with tannase at 37°C for 5.5 h. The reaction mixture was treated in the same way as described above to give a mixture of products which was separated by chromatography over Sephadex LH-20. Elution with EtOH followed by crystallization furnished gallic acid (15 mg) and **7a** (26 mg), colorless needles, mp 210—212°C, $[\alpha]_D^{19}$ −86.2° (c=0.1, H_2O). ^{1}H-NMR (DMSO-d_6): 3.56 (3H, s, OMe), 3.72 (6H, s, 2 OMe), 4.77 (1H, d, J=8 Hz, anomeric H), 6.37 (2H, s, arom. H). FD–MS: m/z 346 (M)$^+$.

Hydrolysis of 7a with Crude Hesperidinase——A solution of **7a** (15 mg) in H_2O was incubated at 37°C for 30 min. After filtration of the reaction mixture, the filtrate was shaken with AcOEt. The AcOEt layer,

after removal of the solvent by evaporation, yielded colorless prisms (**7b**) (4 mg), mp 145—146°C. ^{1}H-NMR ($CDCl_3$): 3.76 (3H, s, OMe), 3.80 (6H, s, 2 OMe), 6.06 (2H, s, arom. H). EI-MS: *m/e* 184 $(M)^+$, 169 $(M-Me)^+$, 139 $(M-3\ Me)^+$. The aqueous layer was concentrated under reduced pressure. The residue was subjected to TLC analysis on a cellulose plate using *n*-BuOH–pyridine–H_2O (6: 4: 3), and glucose was identified by co-chromatography (*Rf*: 0.38) with an authentic specimen.

Acknowledgement The authors thank Dr. T. Nohara and Mr. K. Murakami of Tokushima University for the kind supply of the plant materials. They are also indebted to Mr. Y. Tanaka, Mr. I. Maetani and Miss K. Soeda for the measurements of EI-MS, FD-MS, ^{1}H- and ^{13}C-NMR spectra. This work was supported in part by a grant from the Ministry of Education, Science and Culture, Japan, which is gratefully acknowledged.

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