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Tannins and Related Compounds. LXIV.[1] Six New Phenol Glucoside Gallates from *Castanopsis cuspidata* var. *sieboldii* NAKAI. (2)

MASAYUKI AGETA, KANJI ISHIMARU, GEN-ICHIRO NONAKA, and ITSUO NISHIOKA*

Faculty of Pharmaceutical Sciences, Kyushu University, 62, 3-1-1 Maidashi, Higashi-ku, Fukuoka 812, Japan

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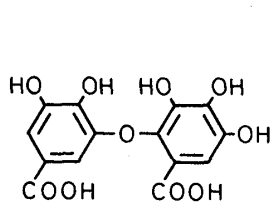
Six new phenol glucoside gallates (**3**—**5**, **7**, **9** and **10**) have been isolated from *Castanopsis cuspidata* var. *sieboldii* NAKAI (Fagaceae), together with the known compounds, dehydrodigallic acid (**1**), cretanin (**2**), chesnatin (**11**) and chestanin (**8**). On the basis of spectroscopic evidence and the result of tannase hydrolysis, the structures of compounds **3**—**5** have been established to be the mono- (**3**) and digallates (**4** and **5**) of crenatin (**6**). Similarly, compounds **7**, **9** and **10** have been characterized as the monogallate (**7**) of 3,4-dihydroxyphenethyl alcohol 4-*O*-*β*-D-glucopyranoside and the monogallates (**9** and **10**) of **8**. In addition, the structure of chesnatin (**11**), in which the orientation of the dehydrodigalloyl group had been unknown, was determined by detailed carbon-13 nuclear magnetic resonance examination.

Keywords——*Castanopsis cuspidata* var. *sieboldii*; Fagaceae; phenol glucoside gallate; 3,4,5-trihydroxybenzyl alcohol; 3,4-dihydroxyphenethyl alcohol; dehydrodigallic acid; chesnatin

Previously, we demonstrated that among many Fagaceous plants growing in Japan, *Quercus stenophylla* MAKINO, *Q. mongolica* FISHER *ex.* TURCZ. var. *grosseserrata* (BL.) REHD. *et* WILS. and *Q. acutissima* CARRUTH, contain simple phenol glucoside gallates,[2] accompanied with large quantities of hydrolyzable tannins.[3] In a continuing chemical examination of tannins and related compounds in the Fagaceous plants, we have now isolated six new phenol glucoside gallates (**3**—**5**, **7**, **9** and **10**), together with dehydrodigallic acid (**1**), cretanin (**2**), chesnatin (**11**) and chestanin (**8**), from the leaves of *Castanopsis cuspidata* var. *sieboldii* NAKAI. This paper deals with the isolation and structure determination of these compounds, and also describes the definitive structural characterization of **11**, in which the orientation of the dehydrodigalloyl group was not determined previously.

Extraction of the fresh leaves with aqueous acetone, followed by a combination of Sephadex LH-20, MCI-gel CHP-20P and Fuji-gel ODS-G3 chromatographies using a variety of solvent systems, afforded compounds **1**—**5** and **7**—**11**. Among these compounds, **1**, **2**, **8** and **11** were identified as dehydrodigallic acid (**1**), cretanin (**2**), chesnatin (**11**) and chestanin (**8**) by comparisons of their physical and spectral data with those of authentic samples.[4]

Compound **3**, colorless needles, mp 235—236 °C, $[\alpha]_D$ −32.5° (H_2O), $C_{20}H_{22}O_{13}$, showed a blue coloration with ferric chloride, suggestive of the presence of a pyrogallol ring in the molecule. The presence of a galloyl group was deduced from the observation of a characteristic two-proton singlet (δ 7.20) in the proton nuclear magnetic resonance (^{1}H-NMR) spectrum. On enzymatic hydrolysis with tannase, **3** yielded gallic acid and a hydrolysate (**6**). Ordinary phenol methylation of **6**, followed by acetylation, gave the dimethylpentaacetyl derivative (**6a**) as colorless needles, mp 87 °C, 556 $(M)^+$ [electron-impact mass spectrum (EI-MS)]. The ^{1}H-NMR showed, together with five acetyl [δ 2.03, 2.04 and



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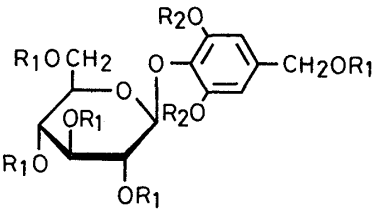
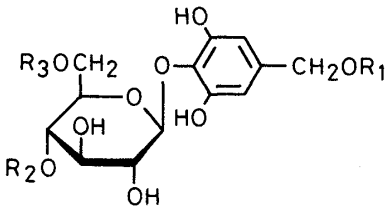
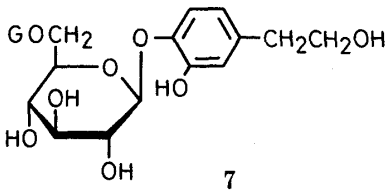


6 : $R_1=R_2=H$
6a : $R_1=Ac$, $R_2=Me$



2 : $R_1=G$, $R_2=R_3=H$
3 : $R_1=R_2=H$, $R_3=G$
4 : $R_1=R_3=G$, $R_2=H$
5 : $R_1=H$, $R_2=R_3=G$



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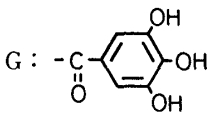
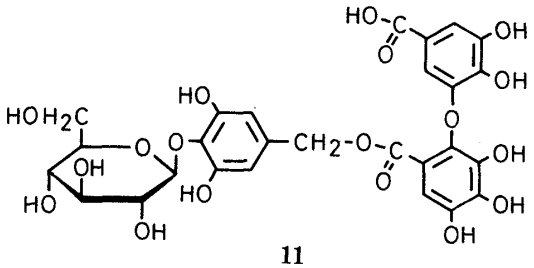


G :



11

2.12 (×3)] and two methoxyl singlets [δ 3.83 (×2)], the presence of a benzylic methylene [δ 5.01 (2H, s)], an aromatic ring with a symmetrical substitution system [δ 6.57 (2H, s)] and a sugar [δ 3.50—5.40 (7H)]. On the basis of these findings, **6** was concluded to be identical with crenatin, which had been obtained from the galls of *Castanea crenata* SIEB. *et* ZUCC. by Ozawa *et al.*[4a] The position of the galloyl group was concluded to be at the C(6)-position in the glucose moiety, since two proton signals [δ 4.38 (1H, dd, $J=6$, 13 Hz), δ 4.78 (1H, dd, $J=2$, 13 Hz)], assignable to the glucose C(6)-methylene protons, were shifted downfield. Consequently, **3** was characterized as 3,4,5-trihydroxybenzyl alcohol 4-*O*-β-D-(6″-*O*-galloyl)-glucopyranoside.

Compound **4**, a white powder, mp 235 °C (dec.), $[\alpha]_D$ −54.5° (H_2O), $C_{27}H_{26}O_{17}\cdot 5/2H_2O$, and **5**, an off-white amorphous powder, $[\alpha]_D$ −34.6° (H_2O), $C_{27}H_{26}O_{17}\cdot H_2O$, liberated gallic acid and crenatin (**6**) on enzymatic hydrolysis with tannase. The ^{1}H- and carbon-13 nuclear magnetic resonance (^{13}C-NMR) spectra of **4** and **5** revealed the presence of two galloyl groups [δ 7.16, 7.20 (each 2H, s) in **4**; δ 7.13, 7.19 (each 2H, s) in **5**] in each molecule. These observations were consistent with the field-desorption mass spectrum (FD-MS), which showed the same $[M+H]^+$ peak at m/z 623, corresponding to crenatin digallate. In the ^{1}H-NMR spectra of **4** and **5**, the lowfield shifts [δ 4.40 (dd, $J=6$, 12 Hz), δ 4.75 (dd, $J=2$, 12 Hz) in **4**; δ 4.20 (d, $J=16$ Hz), δ 4.64 (dd, $J=6$, 16 Hz) in **5**] of double doublet signals with large coupling constants, attributable to the glucose C(6)-methylene protons, indicated that one galloyl group is located at this position in both cases. The location of the other galloyl group in **4** and **5** was determined from the ^{1}H-NMR examinations as follows. In the case of **4**, the observation of the fairly lowfield shift [δ 5.13 (2H, s)] of benzylic methylene signals readily established the position of the galloyl group to be at the benzyl position. On the other hand, the spectrum of **5** exhibited a lowfield triplet (δ 5.24, $J=9$ Hz) due to the methine attached to the galloyl group. The assignment of this signal was achieved by spin-decoupling techniques. On irradiation at the anomeric proton signal [δ 4.78 (d, $J=8$ Hz)], an upfield triplet signal (δ 3.76, $J=9$ Hz) changed into a doublet, thus permitting the assignment of this signal to C(2)-H. Since this C(2)-H signal was shown by spin-decoupling not to be coupled with the triplet at δ 5.24, the lowfield signal was attributed to C(4)-H. On the basis of these results, the structures of **4** and **5** were established to be 1′-*O*-galloyl-3,4,5-

trihydroxybenzyl alcohol 4-*O*-*β*-D-(6′′-*O*-galloyl)-glucopyranoside and 3,4,5-trihydroxybenzyl alcohol 4-*O*-*β*-D-(4′′,6′′-di-*O*-galloyl)-glucopyranoside, respectively.

Compound **7**, an off-white amorphous powder, $[\alpha]_D$ −44.1° (H_2O), $C_{21}H_{24}O_{12}\cdot H_2O$, showed, in the ^{1}H- and ^{13}C-NMR spectra, the presence of a galloyl group (δ 7.18, s), an aromatic ring with a 1,3,4-trisubstitution system [δ 6.73 (d, J=8 Hz), δ 6.82 (dd, J=2, 8 Hz), δ 7.08 (d, J=2 Hz)], adjacent methylenes [δ 2.66, 3.73 (each t, J=9 Hz)] and a hexose moiety (δ 63.8, 71.0, 74.1, 75.1, 76.7, 103.5). On treatment with tannase, **7** gave gallic acid and a hydrolysate, which was identified as 3,4-dihydroxyphenethyl alcohol 4-*O*-*β*-D-glucopyranoside by comparison of the ^{1}H-NMR data with those of an authentic sample.[5] Similar lowfield shifts of the glucose C(6)-signals [δ 4.33 (dd, J=6, 13 Hz), δ 4.71 (dd, J=2, 13 Hz), δ 63.8] in the ^{1}H- and ^{13}C-NMR spectra of **7** established the galloyl group to be at the C(6)-position. Thus, **7** was determined to be 3,4-dihydroxyphenethyl alcohol 4-*O*-*β*-D-(6′′-*O*-galloyl)-glucopyranoside.

Compound **9**, an off-white amorphous powder, $[\alpha]_D$ −21.5° (H_2O), $C_{47}H_{46}O_{30}\cdot 4H_2O$, and **10**, an off-white amorphous powder, $[\alpha]_D$ −18.6° (H_2O), $C_{47}H_{46}O_{30}\cdot 3H_2O$, gave almost the same ^{1}H-NMR spectra, showing two benzylic methylene signals [δ 4.96, 5.08 (each 2H, s)], two two-proton aromatic singlets [δ 6.39, 6.46 (each 2H, s) in **9**; δ 6.32, 6.48 (each 2H, s) in **10**], three aromatic signals [δ 6.77, 7.25 (each 1H, d, J=2 Hz), δ 7.13 (1H, s)] due to a dehydrodigalloyl group and a galloyl signal [δ 7.20 (2H, s)]. These signal patterns were similar

TABLE I. ^{1}H-NMR Spectral Data for Compounds **3**—**5**, **7**, **9** and **10** (in Acetone-d_6 + D_2O, δ Values)[a]

		3	**4**	**5**	**7**	**9**	**10**
Sugar-H	H-1	4.68 (d, J=8)	4.70 (d, J=8)	4.78 (d, J=8)	4.80 (d, J=8)	4.60 (d, J=8)	4.58 (d, J=8)
	H-2	b)	b)	3.76 (t, J=9)	b)	b)	b)
	H-3	b)	b)	3.99 (t, J=9)	b)	b)	b)
	H-4	b)	b)	5.24 (t, J=9)	b)	b)	b)
	H-5	b)	b)	b)	b)	b)	b)
	H-6	4.38 (dd, J=6, 13)	4.40 (dd, J=6, 12)	4.20 (d, J=16)	4.33 (dd, J=6, 13)	4.38 (dd, J=6, 12)	4.39 (dd, J=5, 12)
		4.78 (dd, J=2, 13)	4.75 (dd, J=2, 12)	4.64 (dd, J=6, 16)	4.71 (dd, J=2, 13)	4.76 (dd, J=4, 12)	4.74 (dd, J=2, 12)
	$-CH_2O-$	4.47	5.13	4.46	3.73 (t, J=9)	4.96, 5.08	4.96, 5.08
	$-CH_2-$				2.66 (t, J=9)		
Aromatic-H		6.48 (2H, s)	6.54 (2H, s)	6.45 (2H, s)	6.73 (d, J=8)	6.39 (2H, s)	6.32 (2H, s)
					6.82 (dd, J=2, 8)	6.46 (2H, s)	6.48 (2H, s)
					7.08 (d, J=2)		
Galloyl-H		7.20	7.16, 7.20	7.13, 7.19	7.18	7.20	7.20
Dehydrodigalloyl-H						6.77, 7.25 (each d, J=2)	6.77, 7.25 (each d, J=2)
						7.13 (1H, s)	7.13 (1H, s)

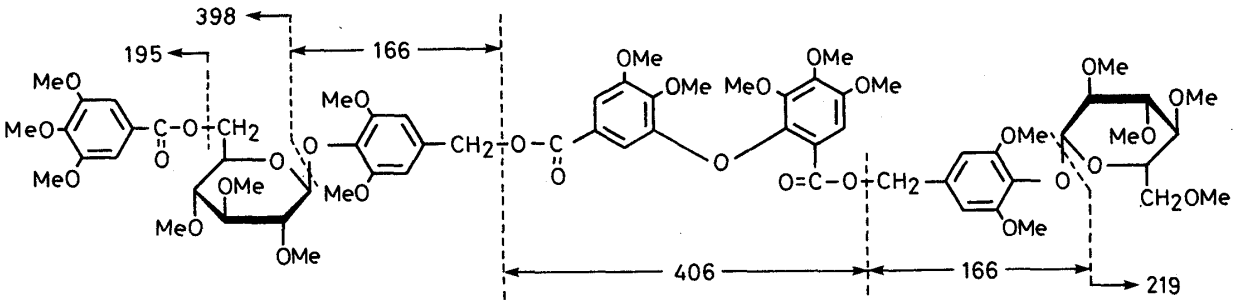
a) J values are expressed in Hz. b) Overlapped with solvent signals.

TABLE II. ^{13}C-NMR Spectral Data for Compounds **3**—**5**, **7**, **9** and **10** (δ Values)[a]

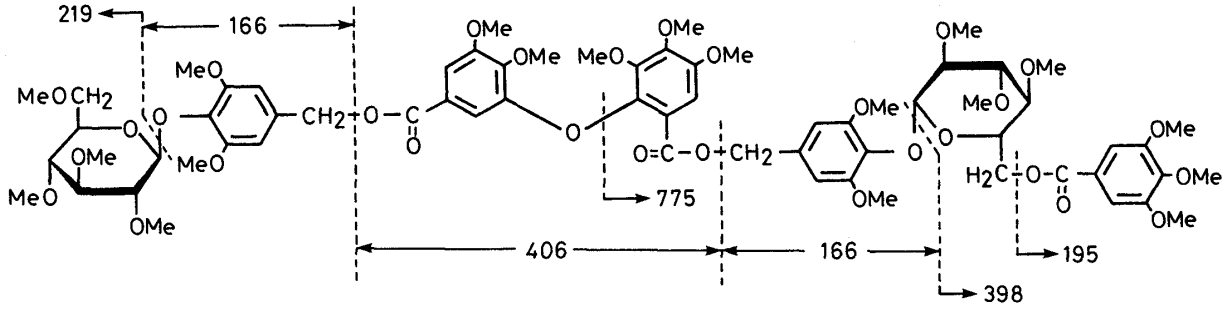
		3	**4**	**5**	**7**	**9**		**10**	
Aglycone	C-1	132.7	133.8	132.8	131.7	133.7	133.7	133.7	133.7
	C-2	106.9	108.0	106.6	116.6	107.8	107.2	107.1	108.1
	C-3	150.3	150.9	150.5	146.0	150.8	150.6	150.6	150.8
	C-4	140.5	135.6	141.1	146.0	135.3	135.0	134.8	135.4
	C-5	150.3	150.9	150.5	118.6	150.8	150.6	150.6	150.8
	C-6	106.9	108.0	106.6	124.8	107.8	107.2	107.1	108.1
	C-1′	63.9	66.3	63.0	38.8	66.3	66.3	66.4	66.4
	C-2′				64.8				
Glucose	C-1	106.9	107.4	107.3	103.5	107.5	107.5	107.5	107.5
	C-2	73.9	74.2	74.4	74.1	74.1[b]	74.2[b]	74.3[b]	74.1[b]
	C-3	76.5	76.8	74.7	76.7	76.7[b]	77.9	77.9	76.6[b]
	C-4	70.4	70.7	71.3	71.0	70.4	70.1	70.1	70.4
	C-5	75.6	76.0	73.9	75.1	75.8	76.8[b]	76.9[b]	75.8
	C-6	63.9	64.0	64.1	63.8	63.9	61.4	61.4	63.9
Galloyl	C-1	120.1	121.3 (2C)	120.6	121.1	121.1		121.0	
				120.9					
	C-2	110.2	110.0 (2C)	110.2 (4C)	110.0 (2C)	110.1 (2C)		110.1 (2C)	
			110.2 (2C)						
	C-3	145.8	146.0 (4C)	146.0 (4C)	146.0 (2C)	145.9 (2C)		145.9 (2C)	
	C-4	139.1	139.1 (2C)	139.1	139.3	139.1		139.1	
				139.3					
Dehydrodigalloyl	C-1					120.8		120.8	
	C-2					111.8		111.9	
	C-3					148.0		148.0	
	C-4					139.9		140.0	
	C-5					146.3		146.3	
	C-6					108.1		108.1	
	C-1′					115.0		115.1	
	C-2′					137.0		137.0	
	C-3′					140.2		140.2	
	C-4′					140.2		140.2	
	C-5′					143.2		143.3	
	C-6′					109.5		109.6	
	–COO–	167.6	167.1	166.6	167.4	165.4		165.6	
			167.3	166.9		166.7		166.7	
						167.3		167.4	

a) Measured in acetone-d_6+D_2O. b) Assignments may be interchanged.

to those of chestanin (**8**), except for the additional galloyl group. Enzymatic hydrolysis of **9** and **10** with tannase afforded gallic acid and **8**. The position of the galloyl group was determined from the similar lowfield shifts of the glucose C(6)-methylene signals [δ 4.38 (dd, J=6, 12 Hz), δ 4.76 (dd, J=4, 12 Hz) in **9**; δ 4.39 (dd, J=5, 12 Hz), δ 4.74 (dd, J=2, 12 Hz) in **10**] in each case. However, there are two possibilities for the location of the galloyl group in **9** and **10**, because of the unsymmetrical nature of the molecule. Thus, **9** and **10** were considered to be structural isomers in which the location of the galloyl group was different. Methylation of **9** and **10** with CH_2N_2, followed by Kuhn's methylation, furnished the respective nonadecamethylates (**9a** and **10a**), whose FD-MS showed the same molecular ion peak at m/z 1356. The EI-MS of **9a** and **10a** exhibited similar fragmentation patterns with prominent peaks at m/z 166, 195, 219, 398 and 406. In the spectrum of **10a**, the observation of the fragment peak at m/z 775, which resulted from fission of the ether linkage of the dehydrodigalloyl group, indicated clearly the position of the galloyl group as shown in the

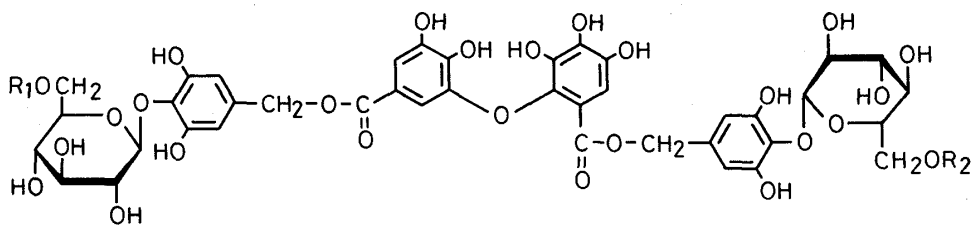


9a



10a

Chart 1. EI-MS Fragmentation Patterns of **9a** and **10a**



8 : $R_1=R_2=H$
9 : $R_1=G$, $R_2=H$
10 : $R_1=H$, $R_2=G$

G :

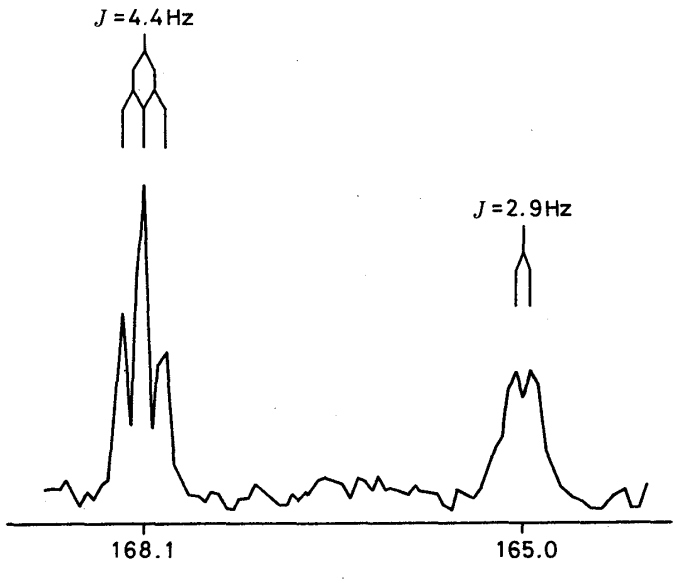


Fig. 1. Non-decoupled ^{13}C-NMR Spectrum of Chesnatin (**11**)

chart, thus permitting the assignments of the structures of **9** and **10** for these compounds.

Although chesnatin (**11**) was formerly isolated from chestnut galls and structurally examined, the orientation of the dehydrodigalloyl group still remained to be solved. Therefore, we attempted to elucidate this point. The ^{13}C-NMR spectrum of **11** showed two signals assignable to an ester carbon (δ 165.0) and a carboxylic acid carbon (δ 168.1).[6] In a nondecoupled ^{13}C-NMR experiment (Fig. 1) these two signals appeared as a triplet (δ 168.1, $J=4.4$ Hz) and a doublet (δ 165.0, $J=2.9$ Hz), respectively, through three-bond long-range couplings between carboxyl carbons and aromatic protons. From these observations, the structure of chesnatin is determined unequivocally to be **11**.

3,4,5-Trihydroxybenzyl alcohol derivatives, which are considered to be biosynthetically interesting from the viewpoint of gallic acid metabolism, were previously known to occur only in the galls of *Castanea crenata* SIEB. *et* ZUCC. The presence of similar metabolites in *Castanopsis cuspidata* var. *sieboldii* NAKAI suggests a close taxonomical relationship between these plants.

Experimental

Melting points were determined on a Yanagimoto micromelting point apparatus and are uncorrected. Optical rotations were measured with a JASCO DIP-4 digital polarimeter. EI- and FD-MS were taken with JEOL JMS D-300 and DX-300 instruments. ^{1}H- and ^{13}C-NMR spectra were recorded on JEOL PS-100 and JEOL FX-100 spectrometers, respectively, with tetramethylsilane as an internal standard, and chemical shifts are given in δ (ppm). Column chromatography was carried out with Sephadex LH-20 (25—100 μm, Pharmacia Fine Chemical Co., Ltd.), MCI-gel CHP-20P (75—150 μm, Mitsubishi Chemical Industries, Ltd.), Fuji-gel ODS-G3 (43—65 μm, Fuji Gel Hanbai Col., Ltd.) and Kieselgel 60 (70—230 mesh, Merck). Thin-layer chromatography (TLC) was conducted on precoated Kieselgel 60 F_{254} plates (Merck, 0.2 mm thick) with benzene–ethyl formate–formic acid (1 : 7 : 1 or 1 : 5 : 1.5), and spots were located by spraying 2% ethanolic ferric chloride and 10% H_2SO_4 reagents.

Extraction and Isolation——The fresh leaves (15 kg) of *Castanopsis cuspidata* var. *sieboldii* were extracted three times at room temperature with 80% aqueous acetone. The extract was concentrated under reduced pressure, and the resulting precipitates, consisting mainly of chlorophylls and waxes, were filtered off. The filtrate was chromatographed over Sephadex LH-20. Elution with H_2O containing increasing amounts of MeOH furnished seven fractions, 1 (500 g), 2 (150 g), 3 (150 g), 4 (65 g), 5 (140 g), 6 (21 g) and 7 (24 g). Fr. 2 was rechromatographed over MCI-gel CHP-20P (H_2O–MeOH), followed by Fuji-gel ODS-G3 (H_2O) chromatography, to give dehydrodigallic acid (**1**) (450 mg), chesnatin (**11**) (*ca.* 30 g), and compounds **3** (510 mg) and **7** (220 mg). Repeated chromatography of fr. 5 over Sephadex LH-20 (EtOH, 60% aqueous MeOH), MCI-gel CHP-20P (30% aqueous MeOH) and Fuji-gel ODS-G3 (30% aqueous MeOH) afforded cretanin (**2**) (350 mg), chestanin (**8**) (*ca.* 7 g) and compounds **4** (*ca.* 1 g), **5** (74 mg), **9** (200 mg) and **10** (180 mg).

Compound 3——Colorless needles (H_2O), mp 235—236 °C, $[\alpha]_D^{23}$ −32.5° ($c=0.90$, H_2O). *Anal.* Calcd for $C_{20}H_{22}O_{13}$: C, 51.07; H, 4.71. Found: C, 50.77; H, 4.55. FD-MS *m/z*: 471 $[M+H]^+$.

Hydrolysis of 3 with Tannase——An aqueous solution of **3** (100 mg) was incubated with tannase at room temperature for 30 min. The reaction mixture was concentrated to dryness under reduced pressure, and the residue was treated with EtOH. The EtOH-soluble portion was subjected to chromatography over Sephadex LH-20. Elution with EtOH yielded gallic acid (29 mg) and 3,4,5-trihydroxybenzyl alcohol 4-*O*-β-D-glucopyranoside (**6**) (61 mg) as an off-white amorphous powder, $[\alpha]_D^{20}$ −43.5° ($c=0.86$, H_2O). ^{1}H-NMR (acetone-d_6 + D_2O): 3.40—4.10 (6H, m, sugar-H), 4.48 (2H, s, –CH_2O–), 4.60 (1H, d, $J=8$ Hz, anomeric-H), 6.47 (2H, s, aromatic-H).

Methylation of 6, Followed by Acetylation——An ice-cooled solution of **6** (50 mg) in MeOH (2 ml) was treated with ethereal CH_2N_2. The mixture was allowed to stand at room temperature for 2 h, then the solvent was evaporated *in vacuo* to give the methyl ether, which was treated with pyridine (0.5 ml) and acetic anhydride (0.5 ml) at room temperature overnight. Usual work-up gave the product, which was chromatographed over silica gel. Elution with benzene–acetone (3 : 1) furnished the dimethyl pentaacetate (**6a**) (30 mg) as colorless needles, mp 87 °C, $[\alpha]_D^{19}$ −16.1° ($c=0.83$, MeOH). ^{1}H-NMR ($CDCl_3$): 2.03, 2.04, 2.12 (15H in total, s, $OCOCH_3$), 3.83 (6H, s, OMe), 5.01 (2H, s, –CH_2O–), 3.50—5.40 (7H, m, sugar-H), 6.57 (2H, s, aromatic-H). EI-MS *m/z*: 556 $(M)^+$; this product was identified as 3,5-dimethyl-3,4,5-trihydroxybenzyl alcohol 4-*O*-β-D-glucopyranoside pentaacetate by comparison of its physical and ^{1}H-NMR data with those reported in the literature.[4a]

Compound 4——A white powder (H_2O), mp 235 °C (dec.), $[\alpha]_D^{20}$ −54.5° ($c=0.77$, H_2O). *Anal.* Calcd for $C_{27}H_{26}O_{17}\cdot 5/2H_2O$: C, 48.58; H, 4.68. Found: C, 48.76; H, 4.48. FD-MS *m/z*: 623 $[M+H]^+$.

Hydrolysis of 4 with Tannase——An aqueous solution of **4** (50 mg) was shaken with tannase at 37 °C for 1 h.

Work-up as described before yielded gallic acid (10 mg) and **6** (14 mg).

Compound 5——An off-white amorphous powder, $[\alpha]_D^{21}$ −34.6° (c=1.03, H_2O). *Anal.* Calcd for $C_{27}H_{26}O_{17}\cdot H_2O$: C, 51.35; H, 4.31. Found: C, 51.41; H, 4.40. FD-MS m/z: 623 $[M+H]^+$.

Hydrolysis of 5 with Tannase——An aqueous solution of **5** (30 mg) was treated with tannase at 37 °C for 1 h. Work-up in the same way as for **3** gave gallic acid (7 mg) and **6** (14 mg).

Compound 7——An off-white amorphous powder, $[\alpha]_D^{20}$ −44.1° (c=1.05, H_2O). *Anal.* Calcd for $C_{21}H_{24}O_{12}\cdot H_2O$: C, 51.85; H, 5.39. Found: C, 51.49; H, 5.44. FD-MS m/z: 469 $[M+H]^+$.

Hydrolysis of 7 with Tannase——An aqueous solution of **7** (16 mg) was incubated with tannase at room temperature for 2 h. The reaction mixture was concentrated to dryness under reduced pressure. The EtOH-soluble portion was applied to a column of Sephadex LH-20 with EtOH to give gallic acid and crude **7a**, which was purified by chromatography over silica gel with $CHCl_3$–MeOH–H_2O (7 : 3 : 0.3) to furnish **7a** as an off-white amorphous powder (9 mg), $[\alpha]_D^{19}$ −46.3° (c=0.50, MeOH). ^{1}H-NMR (acetone-d_6 + D_2O): 2.73 (2H, t, J=7 Hz, 2′-H), 3.70 (2H, t, J=7 Hz, 1′-H), 3.30—4.10 (6H, m, sugar-H), 4.80 (1H, d, J=8 Hz, anomeric-H), 6.74 (1H, dd, J=2, 8 Hz, 6′-H), 6.82 (1H, d, J=8 Hz, 5′-H), 7.09 (1H, d, J=2 Hz, 2′-H).

Compound 9——An off-white amorphous powder, $[\alpha]_D^{20}$ −21.5° (c=1.20, H_2O). *Anal.* Calcd for $C_{47}H_{46}O_{30}\cdot 4H_2O$: C, 48.62; H, 4.52. Found: C, 48.28; H, 4.74. FD-MS m/z: 1091 $[M+H]^+$.

Hydrolysis of 9 with Tannase——An aqueous solution of **9** (30 mg) was treated with tannase at 37 °C for 1 h. The reaction mixture was worked up in the same way as described above to furnish gallic acid and chestanin (20 mg) (**8**).

Permethylation of 9——A solution of **9** (30 mg) in MeOH was treated with an ethereal solution of CH_2N_2 under cooling for 1 h. The solvent was evaporated off under reduced pressure to give the crude methylate, which was further methylated with silver oxide (0.3 g) and methyl iodide (0.5 ml) in dimethylformamide (0.5 ml) at room temperature for 4 h. After removal of inorganic salts by filtration, the solvent was evaporated off *in vacuo* to give a yellow oily residue, which was chromatographed over silica gel with benzene–acetone (5 : 1) to furnish **9a** (14 mg) as a white amorphous powder, $[\alpha]_D^{20}$ −13.0° (c=0.64, $CHCl_3$). FD-MS m/z: 1356 $(M)^+$. EI-MS m/z (%): 406 (42), 398 (72), 219 (100), 195 (90), 166 (98).

Compound 10——An off-white amorphous powder, $[\alpha]_D^{20}$ −18.6° (c=0.90, H_2O). *Anal.* Calcd for $C_{47}H_{46}O_{30}\cdot 3H_2O$: C, 49.39; H, 4.41. Found: C, 49.12; H, 4.77. FD-MS m/z: 1091 $[M+H]^+$.

Hydrolysis of 10 with Tannase——An aqueous solution of **10** (30 mg) was shaken with tannase at 37 °C for 3 h. Treatment of the reaction mixture as before yielded gallic acid and **8** (19 mg).

Permethylation of 10——**10** (30 mg) was treated with an ethereal solution of CH_2N_2 and was further methylated by Kuhn's method (dimethylformamide 0.5 ml, silver oxide 0.3 g, methyl iodide 0.5 ml) to yield **10a** (21 mg) as a white amorphous powder, $[\alpha]_D^{19}$ −15.8° (c=0.44, $CHCl_3$). FD-MS m/z: 1356 $(M)^+$. EI-MS m/z (%): 775 (4), 406 (58), 398 (72), 219 (98), 195 (100), 166 (99).

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