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Constituents of *Geranium thunbergii* SIEB. *et* ZUCC. XIII.[1] Isolation of Water-Soluble Tannins by Centrifugal Partition Chromatography, and Biomimetic Synthesis of Elaeocarpusin

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A highly water-soluble polyphenolic compound, geraniinic acid A, together with elaeocarpusin which is composed of equimolar geraniin and ascorbic acid, has been isolated from *Geranium thunbergii* SIEB. *et* ZUCC. by means of centrifugal partition chromatography and gel chromatography, and the structure was elucidated on the basis of degradative and spectroscopic studies. One of these compounds, elaeocarpusin, was synthesized by condensation of geraniin and ascorbic acid; this route is considered to be biomimetic. The distribution of elaeocarpusin in various plants including *Acer nikoense* MAXIM. was also investigated.

Keywords——*Geranium thunbergii*; Geraniaceae; *Acer nikoense*; Aceraceae; centrifugal partition chromatography; geraniinic acid A; elaeocarpusin; ascorbic acid; biomimetic synthesis; distribution

Geraniin (**1**),[1] which is the crystalline main tannin of the plants of *Geranium* species, and is widely distributed in other species of plants,[2] is almost insoluble in water after it has been isolated. However, it is present in a high concentration in the aqueous solution obtained upon concentration of the homogenate of the plant in acetone–water mixture, and is poorly extractable by ethyl acetate. Such a difference of solubility between the isolated compound and the same compound in the extract is often observed for various natural organic compounds, although the reason has not always been established. Interaction between the tannin and some substances present in the same extract presumably causes the solubilization of the tannin, but no compound with such ability has yet been isolated.

Upon investigation of the water-soluble fraction of the extract of *G. thunbergii* SIEB. *et* ZUCC., we have found two polyphenolic components, compound A and compound B, which are highly soluble in water. Compound A is extremely soluble in water but not in methanol when coexists with amino acids. The solubility in water was reduced to some extent when the amino acids were removed. Compound B is a condensation product of equimolar ascorbic acid and geraniin (**1**), and it was synthesized by a route considered to be biomimetic. Compound B, along with geraniin, was also isolated from *Acer nikoense* MAXIM. (Aceraceae) in a high yield. These findings suggest that some amino acids and ascorbic acid can induce solubilization of tannins in plant tissues, depending on the structures of the tannins.

This paper describes the effective isolation of these tannins by application of centrifugal partition chromatography (CPC),[3] and their structural elucidation based on unambiguous assignments of the nuclear magnetic resonance (NMR) peaks. The synthesis[4] of compound B,

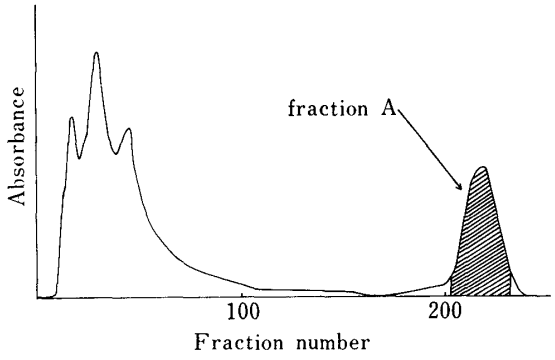


Fig. 1. CPC Chromatogram of the Methanol-insoluble Fraction from *Geranium thunbergii* Extract

Sample, 2.0 g; flow rate, 1 ml/min; fractionation, 5 g/fraction; detection, 254 nm.

which was identical with elaeocarpusin,[5,6] was also achieved through a route considered to be biomimetic.

The 70% aqueous acetone extract of *G. thunbergii* was extracted with ether, ethyl acetate and *n*-butanol, successively. The mother liquor of the ethyl acetate extract after separation of geraniin was sujected to CPC and also to column chromatography on Sephadex LH-20 and on Toyopearl HW-40F to give compound B. The aqueous layer after the extraction with *n*-butanol was evaporated and the residue was separated into methanol-soluble and methanol-insoluble portions. Separation of the latter by CPC gave compound A from fraction A in Fig. 1. A portion of the residue obtained by evaporation of this fraction, which was extremely soluble in water, showed on the thin-layer chromatogram (cellulose, 7% AcOH) positive color reactions for phenolic compound ($FeCl_3$) and amino acid (ninhydrin). The residue, after acidification with dil. HCl, was purified further on a column of Amberlite XAD-2 to give amino acids (mainly alanine and valine) from the water eluate, and compound A, a new tannin which we named geraniinic acid A, from the methanol eluate.

Geraniinic acid A (**2**), $C_{41}H_{38}O_{27}\cdot 7H_2O$, $[\alpha]_D$ $-13°$ (MeOH), was obtained as a pale yellow amorphous powder which gives a greyish blue color with ferric chloride, and the positive color reaction of ellagitannins with $NaNO_2$–AcOH reagent.[7] The proton nuclear magnetic resonance (^{1}H-NMR) spectrum of **2** shows the presence of a galloyl group and two hexahydroxydiphenoyl (HHDP) groups, as revealed by the aromatic signals at δ 7.15 (3H, s), 7.14, 6.84 and 6.71 (1H each, s). These groups were confirmed by methanolysis of the permethylated derivative (**2a**), prepared by methylation of **2** with diazomethane, to yield methyl tri-*O*-methylgallate and dimethyl hexamethoxydiphenate in a ratio of 1 : 2. The liberated sugar was characterized as glucose by gas-liquid chromatography (GLC) after trimethylsilylation. Upon partial hydrolysis with boiling water, **2** gave ellagic acid and corilagin (**3**), thus establishing the location of a galloyl group and an HHDP group at C-1, and C-3—C-6 of the glucose core in **2**, respectively.

The location of another HHDP group in **2** was determined as follows. The glucose proton signals in the 400 MHz ^{1}H-NMR spectrum of **2** were assigned on the basis of spin–spin decoupling experiments, as given in Table I. Comparison of these data with those for 1-*O*-galloyl-2,4;3,6-di-*O*-(*R*)-HHDP-β-D-glucose (**4**)[8] indicates that one of the carboxyl groups in the HHDP group is attached to O-4 of glucose, and the C-2 hydroxyl group is free, as the H-2 signal of **2** appears at significantly higher field (δ 4.24) than that of **4**, while the H-4 signal shows a chemical shift almost identical with the corresponding signal of **4**. This feature was also supported by the carbon-13 nuclear magnetic resonance (^{13}C-NMR) spectrum, in which a signal attributable to a free carboxyl group is observed at significantly lower field (δ 170.21) than the other four ester carbonyl carbon signals.

The proposed structure was finally substantiated by partial hydrolysis of **4** with aqueous ammonia, which yielded **2**. Based on the above evidence, the structure of geraniinic acid A is represented by **2**. As the solubility of isolated geraniinic acid A in water was moderate, the

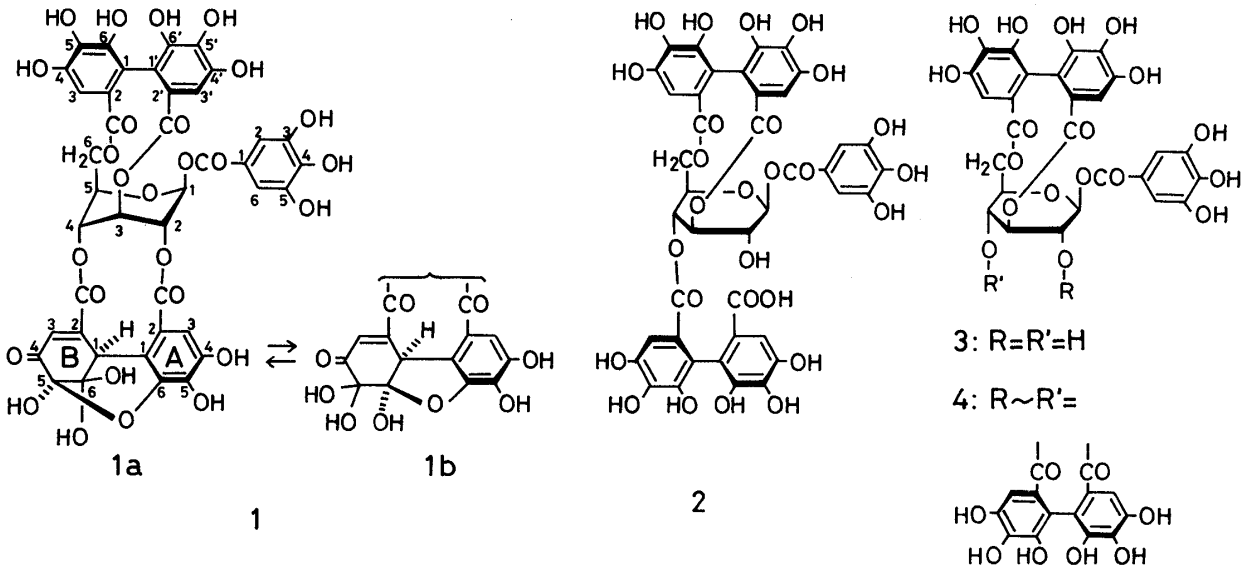


Chart 1

TABLE I. ^{1}H-NMR Data for **2** and **4** [Acetone-d_6, δ Values, J (Hz)]

	H-1	H-2	H-3	H-4	H-5 H-6	H-6′
2[a]	6.25 (d, J=3)	4.24 (br d, J=3)	4.86 (br s)	5.57 (br d, J=3)	4.75 (m)	4.29 (dd, J=6, 11)
4[b]	6.22 (d, J=4)	5.62 (d, J=4)	5.26 (d, J=2.8)	5.52 (d, J=2.8)	4.80 (m)	4.03 (dd, J=2, 9)

a) Measured at 400 MHz with TMS as an internal standard. *b*) Measured at 200 MHz.

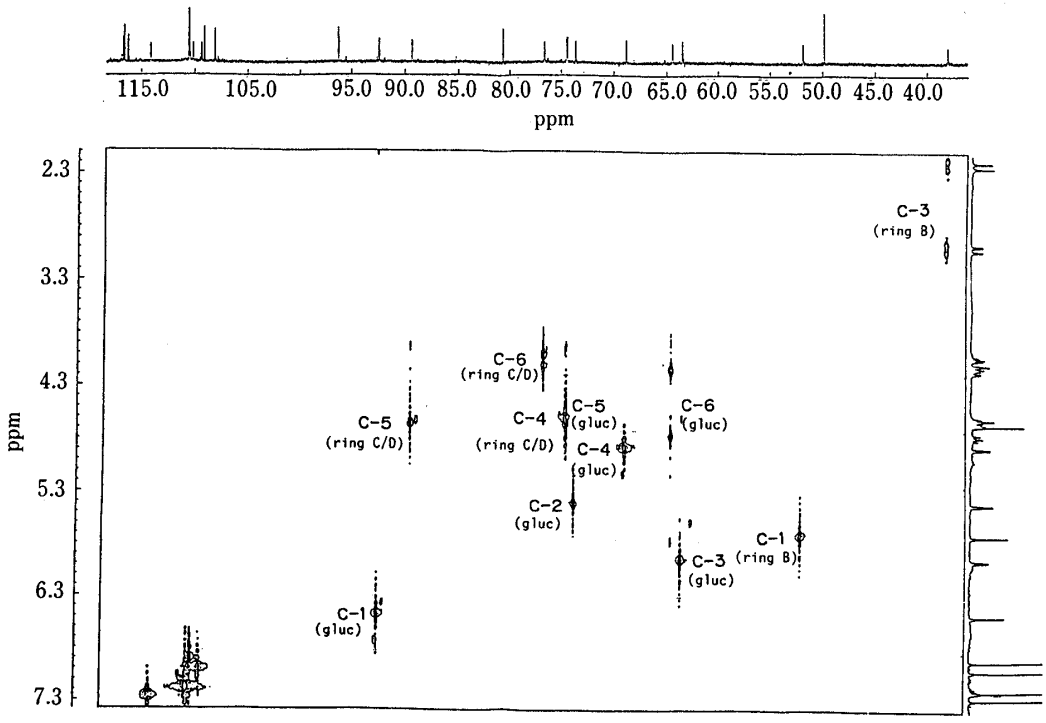


Fig. 2. Aliphatic Region of the 2D ^{1}H–^{13}C Shift Correlation Spectrum of **5** in Acetone-d_6

TABLE II. ^{1}H-NMR Data for the Aliphatic Moieties of **1**, **5** and **7** (400 MHz, Acetone-d_6)

		1[a]	**5**	**7**
Glucose	H-1	6.17 (dd, J=1.2, 1.6)	6.50 (d, J=3.7)	
	H-2	5.57 (dt, J=1.2, 2.4)	5.44 (dt, J=3.7, 1.5)	
	H-3	5.48 (ddd, J=1.6, 2.4, 4)	5.98 (dd, J=4, 1.5)	
	H-4	5.53 (br dd, J=1.2, 4)	4.89 (br d, J=4)	
	H-5	4.80 (br ddd, J=1.2, 8, 11)	4.62 (dd, J=8, 4.8)	
	H-6	4.32 (dd, J=8, 11)	4.82 (dd, J=12, 8)	
	H-6′	4.93 (t, J=11)	4.13 (dd, J=12, 4.8)	
Ring B	H-1	5.18 (s)	5.75 (d, J=2.4)	
	H-3	6.53 (s)	2.99 (dd, J=19.4, 2.4)	
			2.20 (d, J=19.4)	
Ring C/D	H-4		4.67 (s)	4.56 (d, J=4)
	H-5		4.67[b]	4.28 (ddd, J=5.4, 4, 3.2)
	H-6		4.10 (dd, J=9.7, 3.5)	4.10 (dd, J=13, 5.4)
	H-6′		4.05 (dd, J=9.7, 5.5)	4.02 (dd, J=13, 3.2)

a) Data for **1a**. *b*) Overlapped by the H-4 (ring C/D) signal.

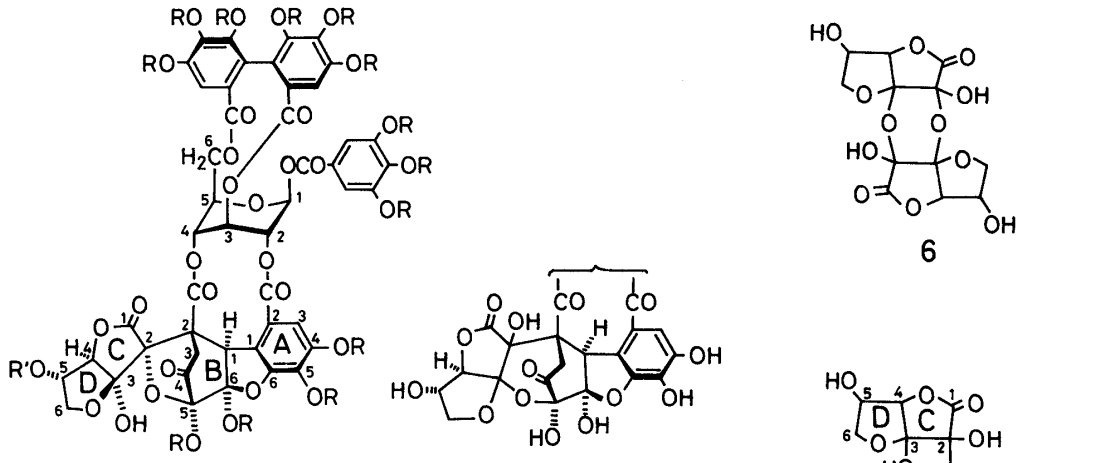
TABLE III. ^{13}C-NMR Resonances of the Aliphatic Carbons of **1**, **5** and **7** (δ Values, Acetone-d_6)

		1		**5**	**7**[a]
		1a	**1b**		
Glucose	C-1	90.81	91.76	92.37	
	C-2	69.89	70.37	73.58	
	C-3	63.27	62.28	63.44	
	C-4	65.85	66.78	68.79	
	C-5	72.55	73.14	74.44	
	C-6	63.64	63.80	64.40	
Ring B	C-1	46.05	51.89	51.87	
	C-2	154.32	149.09	49.83	
	C-3	128.59	125.01	38.00	
	C-4	191.73	194.80	197.59	
	C-5	96.14	92.37	96.23	
	C-6	92.37	108.00	108.09[b]	
Ring C/D	C-1			170.56	177.58
	C-2			80.67	80.62
	C-3			109.08[b]	109.15[c]
	C-4			74.35	75.75
	C-5			89.33	87.64
	C-6			76.59	75.97

a) Ascorbigen (**7**) was synthesized according to the method of Kiss and Neukom.[12] Assignments were made based on the off-resonance spectrum and a comparison of the signals with those of 3-methylindole and dimeric dehydroascorbic acid.[11] *b*) Assignments may be interchanged. *c*) Assignment may be reversed with that of the C-2 signal (δ 108.34).

high solubility of geraniinic acid A in the extract should be due to the coexistence of amino acids with this tannin.

Compound B (**5**), a pale brown amorphous powder, $[\alpha]_D$ +31° (MeOH), gives colorations with ferric chloride and $NaNO_2$–AcOH reagents which are similar to those obtained with **2**. The molecular formula, $C_{47}H_{34}O_{32}\cdot 5H_2O$, was determined from the elemental analysis data and the fast atom bombardment mass spectrum (FAB-MS) using glycerol and NaI or KI as a matrix [m/z 1133 $(M+Na)^+$ or m/z 1149 $(M+K)^+$]. The ^{1}H-



5: R=R'=H
5a: R=Me, R'=H
5b: R=Me, R'=Ac

5c

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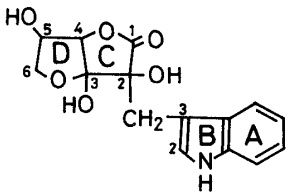

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Chart 2

NMR spectrum indicates the presence of a galloyl group (δ 7.22), and an HHDP group (δ 7.03 and 6.94), and a penta-substituted benzene ring (δ 7.22) in **5**. These assignments of the structural units were also supported by the ^{13}C-NMR spectrum, in which twenty-eight sp^2 carbon signals are almost identical with those of corilagin (**3**)[9] and of the aromatic part of dehydrohexahydroxydiphenyl (DHHDP) group (**1a**⇄**1b**) in geraniin (**1**).[1,9b] Among the ^{1}H- and ^{13}C-NMR peaks of the aliphatic portions, which were assigned by two-dimensional (2D) ^{1}H–^{1}H and ^{1}H–^{13}C shift correlation experiments as shown in Fig. 2, Table II and Table III, the signals due to the sugar residue showed a close resemblance to those of the glucose core of geraniin.

The presence of geraniin structure as a part of **5** was confirmed by the production of geraniin in 26% yield upon keeping a solution of **5** in water at 37 °C for 18 h. The hydrolysis of **5** in boiling water for 8 h gave corilagin. These results combined with the NMR data described above, indicate that **5** is a derivative of geraniin.

Among the NMR peaks other than those of the glucose residue, the peaks of six carbons and three protons (ring B) are assignable to the hydrated cyclohexenetrione portion of the DHHDP group[1] in which the double bond is saturated. The hemiacetal ring in this saturated DHHDP group should be five-membered, as indicated by the appearance of the C-6 (ring A) signal as a singlet at δ 148.96 in the deuterium-induced differential isotope shift measurement,[10] as in the case of geraniin,[1] and by comparison of the chemical shift of C-6 (ring B) in **5** (δ 108.09 or 109.08) with that in the **1a**- (δ 92.37) or **1b**-form (δ 108.00) of the DHHDP group in geraniin.[1]

When all of these signals are subtracted from the NMR spectra of **5**, there still remain signals due to six carbons (rings C and D) and four protons, which show a close similarity to those of the dimer (**6**)[11] of dehydroascorbic acid and also of the ascorbic acid portion of ascorbigen (**7**).[12] The assignments of the ^{1}H- and ^{13}C-NMR peaks of **7** are shown in Tables II and III. Compound B (**5**) can therefore be regarded as a product of condensation between geraniin and ascorbic acid, with a carbon-carbon bond between C-3 of ascorbic acid and C-2 (ring B) of geraniin, inducing saturation of the double bond in the DHHDP group. A comparison of the C-5 signal (ring B) in **5** with that in **1a** (δ 96.3) and **1b** (δ 92.3), taking into account the molecular weight of **5** determined from the FAB-MS, indicates that C-5 (ring B) in **5** forms another hemiacetal ring which involves a linkage to C-2 or C-3 of ascorbic acid, as

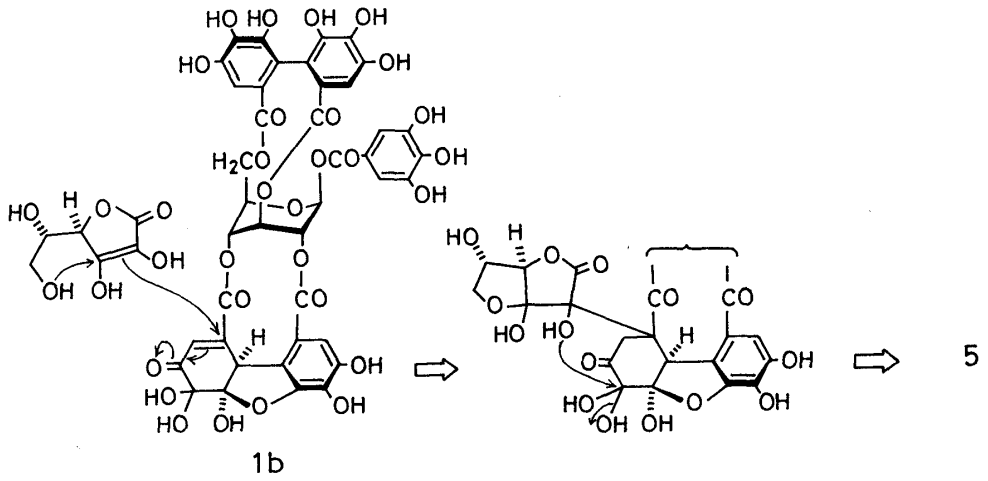

Chart 3

in **5** or **5c**. Methylation of **2** with diazomethane yielded a tridecamethyl derivative (**5a**), and acetylation of **5a** gave a monoacetate (**5b**).

These properties including the unambiguously assigned NMR data, indicated that compound B is identical with elaeocarpusin (**5**), whose structure was determined based on an X-ray analysis of the product obtained by hydrolysis of methylated **5**.[5a)]

The analogy to ascorbigen (**7**), which also contains combined ascorbic acid, and the presence of a large amount of **1** in the plant tissues, suggest that **5** may be biosynthetically formed by enzymatic or non-enzymatic condensation between geraniin and ascorbic acid.

Thus, we attempted a synthesis of elaeocarpusin (**5**) from geraniin and ascorbic acid, and achieved it by keeping a solution of **1** and L-ascorbic acid in McIlvaine's buffer (pH 4.0) or in a mixture of water and methanol at 37 °C for 5 h. The product, $[\alpha]_D$ +30° (MeOH) (60% yield),[13)] which was isolated by Toyopearl column chromatography, was identical with **5**. This facile biomimetic synthesis therefore indicates that the biogenesis of **5** most likely occurs as shown in Chart 3, in spite of the suggestion[5b)] that the possible precursors are dehydroascorbic acid and **4** which has not yet been isolated from natural sources.

We have also examined the distribution of elaeocarpusin in the plant kingdom, as this compound should be present whenever geraniin and ascorbic acid coexist in plant cells which are weakly acidic. High performance liquid chromatography (HPLC) analysis of the extracts of fresh plants of Geraniaceae (*Erodium stephanianum* WILLD., *Geranium carolinianum* L., *G. robertianum* L., *G. yoshinoi* MAKINO) revealed the presence of elaeocarpusin as a peak distinct from that of geraniin. Therefore, elaeocarpusin can be regarded as being generally present in geraniin-containing species of plants. This assumption was supported by the results of HPLC analysis on several species from other families which showed that geraniin in their extracts is accompanied with elaeocarpusin: *Acer buergerianum* MIQ., *A. saccharum* MARSH., *A. palmatum* THUNB. (Aceraceae); *Rhus trichocarpa* MIQ., *R. succedanea* L. (Anacardiaceae); *Cercidiphyllum japonicum* SIEB. *et* ZUCC. (Cercidiphyllaceae).

Experimental

Optical rotations were measured on a JASCO DIP-4 polarimeter, infrared (IR) spectra on a JASCO A-102 spectrometer and ultraviolet (UV) spectra on a Hitachi 200-10 spectrometer. NMR spectra were recorded on a Bruker AM-400 (400 MHz for ^1H and 100.2 MHz for ^{13}C) with tetramethylsilane (TMS) as an internal standard, and the chemical shifts were given in δ values (ppm). Normal-phase HPLC was run on a column of Nomura Develosil 60-5 (4 i.d. × 150 mm) with *n*-hexane–MeOH–THF–formic acid (55 : 33 : 11 : 1, v/v) containing oxalic acid (450 mg/l) as an eluant. Reversed-phase HPLC was performed on a column of YMC-Pack A312 (ODS) (6 i.d. × 150 mm),

developing with 0.05 M H_3PO_4–0.05 M KH_2PO_4–EtOH–EtOAc (42.5 : 42.5 : 10 : 5, v/v). Detection was effected with a Shimadzu SPD-6A (280 nm) and a multi channel photo detector MCPD-350PC system II (Union Giken) coupled with a Shimadzu LC-6A system. GLC was run on a Hitachi 163 gas chromatograph. CPC was carried out on a Sanki Engineering CPC, model L-90 (12 cartridges; 1000 rpm). Sephadex LH-20 (100 μm) (Pharmacia Fine Chemicals), and Toyopearl HW-40F (Toyo Soda) were used for column chromatography. A CS-900 dual wavelength TLC scanner (Shimadzu) was used for quantitation on TLC plates. Solvents were removed by evaporation under reduced pressure below 40 °C.

Isolation of Tannins——a) Dried leaves (500 g) of *G. thunbergii* collected in August at the herbarium of the Faculty of Pharmaceutical Sciences, Okayama University, were homogenized in 70% aqueous acetone (6 l), and the homogenate was filtered. The filtrate was concentrated to *ca.* 500 ml and extracted successively with ether (500 ml × 5), EtOAc (500 ml × 40) and *n*-BuOH (500 ml × 20) to afford the EtOAc extract (52 g) and the BuOH extract (9.2 g).

The EtOAc extract was crystallized from a mixture of water and a small amount of MeOH to give geraniin (11.8 g). The mother liquor was evaporated to give a brown residue (36.6 g), a part (2 g) of which was separated by ascending CPC using *n*-BuOH–*n*-PrOH–water (4 : 1 : 5, v/v) and finally purified on a Sephadex LH-20 column with EtOH and then EtOH–MeOH (5 : 1) to yield elaeocarpusin (**5**, 26 mg).

The BuOH extract (9.2 g) was also chromatographed over Sephadex LH-20 with EtOH containing increasing amounts of MeOH (up to 100%). The eluate with MeOH was rechromatographed on Toyopearl HW-40F using 50% aqueous EtOH to give **5** (26 mg).

The aqueous layer after extraction with *n*-BuOH was evaporated to a syrup, to which MeOH (300 ml) was added. A part (2 g) of the MeOH-insoluble portion (43.7 g) was subjected to CPC by the ascending method using *n*-BuOH–AcOH–H_2O (4 : 1 : 5, v/v). Fraction A (Fig. 1) containing **2** was then purified on a Toyopearl HW-40F column using H_2O–EtOH (95 : 5, v/v). The fraction which gave a positive coloration with both $FeCl_3$ and ninhydrin reagents was adjusted to pH 2.0 with dil. HCl, and subjected to column chromatography over Amberlite XAD-2 developed with H_2O and MeOH. The H_2O eluate was shown by the amino acid analyzer to contain alanine and valine, accompanied with a small amount of aspartic acid and serine. The MeOH eluate gave **2** (8 mg), which is negative to ninhydrin. Total yields of **2** and **5** from the dried leaves were 0.04% and 0.05%, respectively.

b) Dried leaves (200 g) of *A. nikoense* were extracted in essentially the same manner as described in a) to give the EtOAc extract (16.44 g) and the BuOH extract (15.64 g). A part (1 g) of the EtOAc extract was subjected to column chromatography on Sephadex LH-20 to yield geraniin (395 mg). A part (3 g) of the BuOH extract was separated by successive chromatography on columns of Sephadex LH-20 (EtOH–H_2O, 7 : 3) and Toyopearl HW-40F (EtOH–H_2O, 4 : 6) to give elaeocarpusin (**5**, 130 mg). Total yields of **1** and **5** from the dried leaves were 3.25% and 0.34%, respectively.

Geraniinic Acid A (2)——A pale yellow amorphous powder, TLC (cellulose, 7% AcOH), *Rf* 0.45. $[\alpha]_D$ −13 °(c = 0.8, MeOH). UV λ_{max}^{MeOH} nm (log ε): 216 (4.84), 271 (4.48). IR ν_{max}^{KBr} cm^{-1}: 3430, 1720, 1615, 1520, 1450, 1350—1310, 1220—1180, 1100. ^{1}H-NMR, see Table I. ^{13}C-NMR (acetone-d_6) δ: 170.21 (carboxyl), 168.32, 167.90, 167.31, 166.76 (ester carbonyl), 95.37 (glucose C-1), 74.06, 71.06, 69.31, 64.99 (glucose C-2—6). *Anal.* Calcd for $C_{41}H_{30}O_{27}\cdot 7H_2O$: C, 45.56; H, 4.10. Found: C, 45.74; H, 4.39.

Partial Hydrolysis of Geraniinic Acid A (2)——A solution of **2** (1.1 mg) in H_2O (1 ml) was heated on a boiling water bath for 8 h. Analysis of the residue by HPLC (normal-phase and reversed-phase) demonstrated the production of ellagic acid and corilagin (**3**).

Methylation of 2——A solution of **2** (10 mg) in abs. EtOH (0.2 ml) was treated with excess ethereal CH_2N_2 at 5 °C for 1 h. After removal of the solvent, the residue was purified by preparative TLC (silica gel, light petroleum–$CHCl_3$–acetone, 4 : 6 : 3) to yield the hexadecamethyl derivative (**2a**) (4.4 mg) as pale yellow needles, mp 134 °C, $[\alpha]_D$ −6.4 ° (c = 0.5, acetone). ^{1}H-NMR (acetone-d_6) δ: 7.32 (2H, s, galloyl), 7.37, 7.21, 6.96, 6.93 (1H each, s, HHDP), 3.95—3.32 (16 × OMe). Upon methanolysis with 5% NaOMe for 4 h at room temperature, **2a** gave methyl tri-*O*-methylgallate and dimethyl hexamethoxydiphenate, in a ratio of 1 : 2 [by densitometry on TLC (silica gel, benzene–acetone, 14 : 1)], and glucose (TMS ether, GLC).

Formation of 2 from 1-*O*-Galloyl-2,4;3,6-di-*O*-(*R*)-hexahydroxydiphenoyl-β-D-glucose (4)——A solution of **4** (240 mg) in H_2O (24 ml) was hydrolyzed with 3% aq. NH_4OH (0.24 ml) for 2 h at room temperature. The reaction mixture was acidified with AcOH and evaporated. The residue was chromatographed on Toyopearl HW-40F (EtOH–H_2O, 4 : 6), and then on Sephadex LH-20 (EtOH–H_2O, 7 : 3) to give geraniinic acid A (**2**, 36.7 mg) which was shown to be identical with the natural compound by ^{1}H-NMR and HPLC (normal-phase and reversed-phase).

Elaeocarpusin (5)——A pale brown amorphous powder, easily soluble in H_2O (more than 1 g dissolves in 1 ml of H_2O at 15 °C). TLC (cellulose, 7% AcOH), *Rf* 0.25. $[\alpha]_D$ +31 ° (c = 1.0, MeOH). UV λ_{max}^{MeOH} nm (log ε): 223 (4.89), 282 (4.50). IR ν_{max}^{KBr} cm^{-1}: 3430, 1800, 1750—1730,1625, 1530—1510, 1450, 1360—1310, 1230—1210, 1030. ^{13}C-NMR (acetone-d_6) δ: 120.47 (galloyl, C-1), 110.51 (2C) (galloyl, C-2,6), 145.99 (galloyl, C-3,5), 139.58 (galloyl, C-4), 116.71, 116.58 (HHDP, C-1,1′), 125.14, 123.14 (HHDP, C-2,2′), 110.12, 109.36 (HHDP, C3,3′), 145.14, 145.07, 144.85, 144.65 (HHDP, C-4,4′,6,6′), 116.21 (ring A, C-1), 118.65 (ring A, C-2), 114.08 (ring A, C-3), 147.53 (ring A, C-4), 136.08 (ring A, C-5), 148.59 (ring A, C-6), 167.99, 167.93, 166.35, 165.25, 164.63 (ester carbonyl), aliphatic potion, see

Table III. *Anal.* Calcd for $C_{47}H_{34}O_{32} \cdot 5H_2O$: C, 47.01; H, 3.69. Found: C, 47.17; H, 3.45.

Partial Hydrolysis of Elaeocarpusin (5)——(1) A solution of **5** (90 mg) in H_2O (20 ml) was kept at 37 °C for 18 h. The reaction mixture was then concentrated and chromatographed on Sephadex LH-20 using EtOH–H_2O (7 : 3) as an eluant to give geraniin (**1**, 23.2 mg), which was found to be identical with an authentic sample by ^{1}H-NMR and HPLC (normal-phase and reversed-phase).

(2) A solution of **5** (1 mg) in water (1 ml) was kept in a boiling water bath for 8 h. Upon analysis of the products by normal-phase and reversed-phase HPLC, corilagin (**3**), gallic acid and ellagic acid were identified.

Methylation of Elaeocarpusin (5)——A solution of **5** (30 mg) in absolute EtOH (0.75 ml) was treated with excess ethereal CH_2N_2 for 30 min at room temperature. After evaporation of the solvent, the residue was further methylated in an analogous way. The crude methyl ether was purified by preparative TLC (SiO_2, light petroleum–$CHCl_3$–acetone, 4 : 6 : 3, v/v) to yield trideca-*O*-methylelaeocarpusin (**5a**, 6.6 mg) as pale yellow needles, mp 221 °C. $[\alpha]_D$ +37° (c=1.0, acetone). IR ν_{max}^{KBr} cm^{-1}: 3420, 2950, 2850, 1800, 1750, 1590, 1460, 1340. ^{1}H-NMR (acetone-d_6) δ: 7.43 (1H, s), 7.30 (2H, s, galloyl), 7.12, 6.97 (each 1H, s, HHDP), 3.93—3.41 (13 × OMe). *Anal.* Calcd for $C_{60}H_{60}O_{32} \cdot H_2O$: C, 54.96; H, 4.79. Found: C, 55.23; H, 4.65.

Acetylation of **5a** (8.3 mg) with Ac_2O and pyridine followed by preparative TLC (SiO_2, light petroleum–$CHCl_3$–acetone), gave a monoacetate (**5b**, 5.2 mg) as white needles, mp 199 °C. $[\alpha]_D$ +30° (c=1.0, acetone). IR ν_{max}^{KBr} cm^{-1}: 3430, 2950, 2850, 1800, 1750, 1590, 1460, 1330. ^{1}H-NMR (acetone-d_6) δ: 7.53 (1H, s), 7.23 (2H, s, galloyl), 6.78, 6.69 (each 1H, s, HHDP), 3.98, 3.97, 3.96, 3.95, 3.90, 3.85, 3.73, 3.72, 3.71, 3.69, 3.55, 3.36, 3.28 (each 1 × OMe), 2.16 (2 × OAc). *Anal.* Calcd for $C_{62}H_{62}O_{33} \cdot H_2O$: C, 55.03; H, 4.67. Found: C, 55.23; H, 4.65.

Condensation of Geraniin (1) with L-Ascorbic Acid——A solution of ascorbic acid (200 mg) in H_2O (100 ml) was added to a solution of geraniin (**1**, 200 mg) in MeOH–H_2O (9 : 1), and the reaction mixture was kept for 15 h at room temperature. The solvent was evaporated off, and the residue was chromatographed over Toyopearl HW-40F using H_2O–EtOH–AcOH (65 : 35 : 1) to afford recovered geraniin (120 mg) and elaeocarpusin (**5**, 49.6 mg) which was identical with the natural product.

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