Medicinal Foodstuffs. V.[1] Moroheiya. (1): Absolute Stereostructures of Corchoionosides A, B, and C, Histamine Release Inhibitors from the Leaves of Vietnamese *Corchorus olitorius* L. (Tiliaceae)

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Three new ionone glucosides named corchoionosides A, B, and C were isolated from the leaves of *Corchorus olitorius*, commonly called "moroheiya" in Japanese, together with seven known compounds, an ionone glucoside (6*S*,9*R*)-roseoside, a monoterpene glucoside betulalbuside A, two flavonol glucosides astragalin and isoquercitrin, two coumarin glucosides scopolin and cichoriine, and chlorogenic acid. The absolute stereostructures of corchoionosides A, B, and C were determined by chemical and physicochemical evidence, which included the result of application of a modified Mosher's method, the CD helicity rule, and chemical correlation with (6*S*,9*R*)-roseoside. Corchoionosides A and B and (6*S*,9*R*)-roseoside were found to inhibit the histamine release from rat peritoneal exudate cells induced by antigen–antibody reaction.

Key words corchoionoside; *Corchorus olitorius*; ionone glucoside; histamine release inhibitor; (6*S*,9*R*)-roseoside; moroheiya

The leaves of *Corchorus olitorius* L. (Tiliaceae), which is commonly called "moroheiya" in Japanese, are used as a vegetable in Arabian countries. Recently, since "moroheiya" is considered to be rich in vitamins, carotenoids, calcium, and potassium, it has been consumed as a health food in Japan. As chemical constituents of *Corchorus olitorius* L., several cardiac glycosides and mucilages have been obtained from the seed,[2] while a triterpene and sterols were isolated from the root,[3] and phenolic acids were identified from the fresh bark and stem by HPLC.[4] Recently, known flavonoid glycosides, oleanolic acid, β-sitosterol, and its glucoside, were isolated from the leaves of Japanese *Corchorus olitorius* L.[5]

In the course of our studies on the bioactive principles of medicinal foodstuffs,[1,6] we have isolated three new ionone glucosides called corchoionosides A (**1**), B (**2**), and C (**3**) together with seven known compounds from the 1-butanol-soluble fraction of the leaves of Vietnamese *Corchorus olitorius* L., which is widely used by the health food industry in Japan. This paper describes the structure elucidation of corchoionosides (**1**—**3**) and the inhibitory activity of **1**, **2**, and (6*S*,9*R*)-roseoside (**4**) on the histamine release from rat peritoneal exudate cells induced by antigen–antibody reaction.[7]

The air-dried leaves of *Corchorus olitorius* L. cultivated in Vietnam were extracted with methanol under reflux. The methanol extract was partitioned into an ethyl acetate–water mixture to furnish the ethyl acetate-soluble portion and the water phase. The water phase was further extracted with 1-butanol to give the 1-butanol-soluble portion and the water-soluble portion. The 1-butanol-soluble portion was subjected to normal-phase silica-gel column chromatography to provide five fractions (Fr. 1—5). Fraction 2 was separated by normal and reversed-phase silica-gel column chromatography and then HPLC to give corchoionosides A (**1**, 0.0049%), B (**2**, 0.0004%), and C (**3**, 0.0003%) together with an ionone glucoside (6*S*,9*R*)-roseoside[8] (**4**, 0.0117%), a monoterpene glucoside betulalbuside A[9] (**5**, 0.0012%), a flavonol glucoside astragalin[5,10] (**6**, 0.0073%), and two coumarin glucosides cichoriine[11] (**8**, 0.0016%) and scopolin[12] (**9**, 0.0006%). Fraction 6 was also subjected to reversed-phase and normal-phase silica-gel column chromatography followed by HPLC to furnish a flavonol glucoside isoquercitrin[5,10] (**7**, 0.0022%) and chlorogenic acid[13] (**10**, 0.0028%).

Absolute Stereostructures of Corchoionosides A (1), B (2), and C (3) Corchoionoside A (**1**) was isolated as a white powder with negative optical rotation ($[\alpha]_D^{28}$ $-24.8°$). The negative-mode and positive-mode FAB-MS of **1** showed quasimolecular ion peaks at *m/z* 387 $(M-H)^-$ and 411 $(M+Na)^+$, respectively, and high-resolution MS analysis of the quasimolecular ion peak $(M-H)^-$ revealed the molecular formula of **1** to be $C_{19}H_{32}O_8$. The IR spectrum of **1** showed absorption bands at 1655 and 1638 cm^{-1} ascribable to olefin and strong absorption bands at 3425 and 1076 cm^{-1} suggestive of a glycosidic structure. The ^{1}H-NMR (CD_3OD) and ^{13}C-NMR (Table 1) spectra of **1**, which was assigned by various NMR analytical methods,[14] showed three tertiary methyls [δ 0.96, 1.14, 1.19 (all s, 12, 13, 11-H_3)], a secondary methyl [δ 1.22 (d, $J=6.3$ Hz, 10-H_3)], two methines bearing a hydroxyl group [δ 3.86 (m, 3-H), 4.29 (dq, $J=5.6$, 6.3 Hz, 9-H)], a disubstituted olefin [δ 5.66 (dd, $J=5.6$, 15.5 Hz, 8-H), 5.91 (dd, $J=1.0$, 15.5 Hz, 7-H)], and a β-D-glucopyranosyl moiety [δ 4.33 (d, $J=7.9$ Hz, 1′-H)] together with two quaternary carbons bearing an epoxide function. In the heteronuclear multiple bond correlation (HMBC) experiment (Fig. 1) on **1**, a long-range correlation was observed between the anomeric proton and the 3-carbon. Furthermore, acetylation of **1** with acetic anhydride in pyridine yielded the penta-*O*-acetate (**1a**), whose ^{13}C-NMR spectrum showed a acetylation shift[15] around the 9-position of **1a**. These findings and comparisons of the ^{1}H-NMR and ^{13}C-NMR spectra of **1** and **1a** with those of known ionone glucosides[16] led us to formulate the 3-*O*-β-D-glucopyranosyl 5,6-epoxy-9-hydroxyionol struc-

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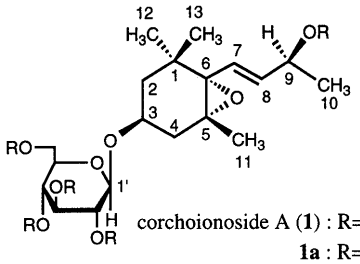
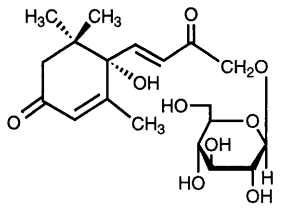
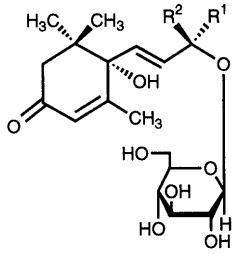


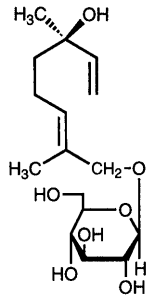
corchoionoside A (**1**) : R=H
1a : R=Ac



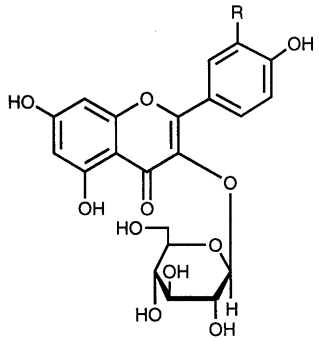
corchoionoside B (**2**)



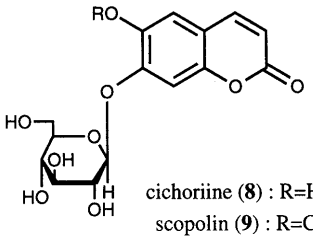
corchoionoside C (**3**) : R^1=CH_3, R^2=H
(6*S*, 9*R*)-roseoside (**4**) : R^1=H, R^2=CH_3



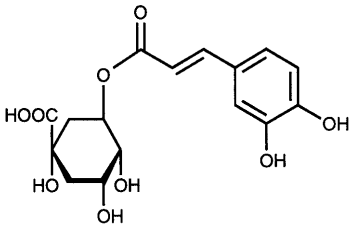
betulalbuside A (**5**)



astragalin (**6**) : R=H
isoquercitrin (**7**) : R=OH



cichoriine (**8**) : R=H
scopolin (**9**) : R=CH_3



chlorogenic acid (**10**)

Chart 1

Table 1. ^{13}C-NMR Data for **1**, **1a**, **11**, **2**, **3**, **4**, **14**

	1[a]	**1a**[a]	**11**[b]	**2**[a]	**3**[a]	**4**[a]	**14**[a]
C-1	35.9	35.6	34.9	42.7	42.4	42.4	42.5
C-2	45.7	44.8	47.0	50.6	50.8	50.8	50.8
C-3	73.0	73.8	64.2	200.4	201.2	201.1	201.8
C-4	38.5	38.3	40.8	128.1	127.1	127.1	127.2
C-5	67.7	67.1	66.4	164.5	167.1	167.2	167.5
C-6	71.3	71.1	69.4	80.2	80.0	79.9	80.0
C-7	125.8	128.9	124.9	148.5	133.8	135.2	136.9
C-8	139.1	134.6	137.8	127.5	133.7	131.5	130.1
C-9	68.7	71.9	68.7	198.6	74.6	77.3	68.8
C-10	23.8	21.1	23.7	74.1	22.2	21.2	23.9
C-11	20.2	20.3	19.8	19.2	19.5	19.5	19.6
C-12	25.2	25.5	24.7	23.5	23.5	23.4	23.5
C-13	29.8	29.4	29.5	24.8	24.7	24.7	24.5
C-1′	102.9	100.6		104.3	101.3	102.7	
C-2′	75.1	73.0		75.0	75.0	75.2	
C-3′	77.8	74.3		77.8	78.2	77.9	
C-4′	71.6	70.0		71.6	71.7	71.6	
C-5′	78.1	72.8		78.2	78.4	78.0	
C-6′	62.7	63.2		62.8	62.8	62.8	

The spectra were taken with CD_3OD[a] or $CDCl_3$.[b]

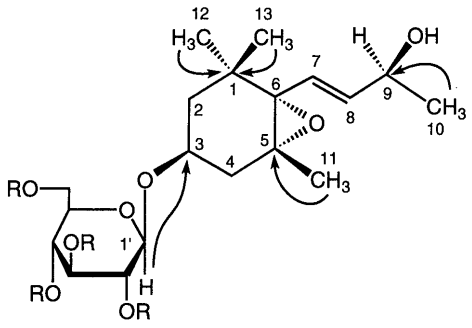


Fig. 1. Long-Range Correlations in the HMBC Spectrum of **1**

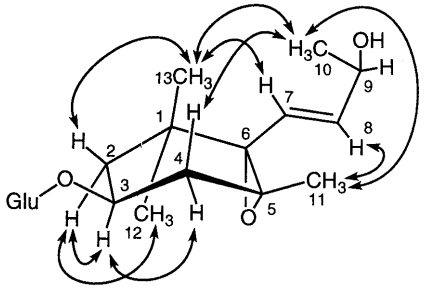
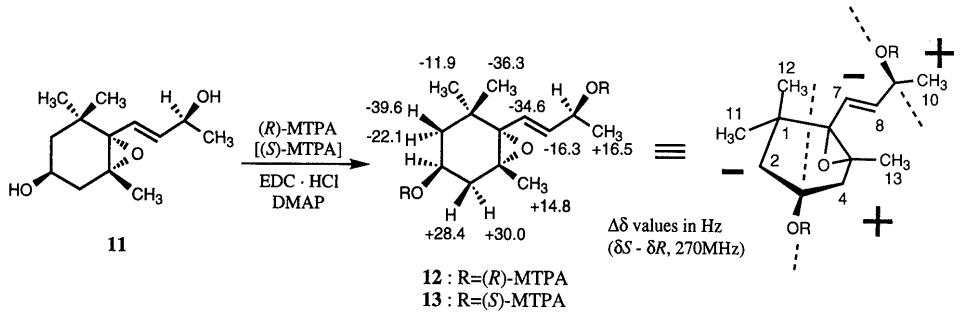


Fig. 2. NOEs Observed in the NOESY Spectrum of **1**



11

12 : R=(*R*)-MTPA
13 : R=(*S*)-MTPA

Chart 2

ture of **1**. The relative stereostructure of **1** was characterized by ^{1}H-NMR nuclear Overhauser and exchange spectroscopy (NOESY) as shown in Fig. 2; NOE correlations were observed between 2β-H and 13-H$_3$, 2α-H and 12-H$_3$, 2α-H and 3α-H, 3α-H and 4α-H, 4β-H and 7-H, 4β-H and 10-H$_3$, 7-H and 13-H$_3$, 8-H and 11-H$_3$, 10-H$_3$ and 11-H$_3$, and 10-H$_3$ and 13-H$_3$.

In order to clarify the absolute stereostructure of **1**, the aglycone, corchoionol A (**11**),[17] which was obtained by enzymatic hydrolysis of **1** with β-glucosidase, was subjected to a modified Mosher's method.[18] Namely, **11** was treated with (*R*)- and (*S*)-α-methoxy-α-trifluoromethylphenyl acetate (MTPA) in the presence of 1-ethyl-3-(3-dimethylaminopropyl)carbodiimide hydrochloride (EDC · HCl) and 4-dimethylaminopyridine (DMAP) to give the 3,9-di-(*R*)-MTPA ester (**12**) and the 3,9-di-(*S*)-MTPA ester (**13**), respectively. As shown in Chart 2, the signals due to protons attached to the 4, 13 and 10-carbons in the 3,9-di-(*R*)-MTPA ester (**12**) were observed at higher fields ($\Delta\delta$: positive) as compared to those of the 3,9-di-(*S*)-MTPA ester (**13**), while the signals due to protons of the 2, 11, 12, 7, and 8 carbons in **12** were observed at lower fields ($\Delta\delta$: negative) as compared to those of **13**. Consequently, the absolute configurations at the 3 and 9-positions of **1** have been elucidated as 3*S* and 9*S*, and the absolute stereostructure of corchoionoside A (**1**) was determined to be as shown.

Corchoionoside B (**2**) was also isolated as a white powder with positive optical rotation ($[\alpha]_D^{23}$ +113.7°), and its IR spectrum showed absorption bands due to hydroxyl and enone functions at 3403, 1705, 1655, 1630, and 1076 cm^{-1}. In the negative-mode and positive-mode FAB-MS of **2**, quasimolecular ion peaks were observed at *m/z* 399 $(M-H)^-$, 401 $(M+H)^+$, and 423 $(M+Na)^+$, and the molecular formula, $C_{19}H_{28}O_9$, of **2** was determined by high-resolution MS measurement. The ^{1}H-NMR (CD_3OD) and ^{13}C-NMR (Table 1) spectra[14] of **2** showed two tertiary methyls [δ 1.01, 1.06 (both s, 13, 12-H$_3$)], a vinyl methyl [δ 1.90 (d, J=1.3 Hz, 11-H$_3$)], an oxymethylene [δ 4.50, 4.74 (ABq, J=17.2 Hz, 10-H$_2$)], two enones [δ 5.93 (d, J=1.3 Hz, 4-H), 6.66, 7.11 (both d, J=15.8 Hz, 8,7-H)] and a β-D-glucopyranosyl moiety [δ 4.33 (d, J=7.6 Hz, 1′-H)]. The connectivities of the quaternary carbons and the position of the glucosidic linkage were clarified by an HMBC experiment with **2**. Namely, HMBC correlations were observed between the following protons and carbons: 2-H$_2$ and 3-C, 11-H$_3$ and 4-C, 11-H$_3$ and 6-C, 7-H and 6-C, 8-H and 9-C, 10-H$_2$ and 9-C, 1′-H and 10-C (Fig. 3). In the NOESY experiment on **2**, NOE correlations were observed between the protons as shown in Fig. 4, so that the relative stereostructure of **2** was characterized. Finally, the absolute stereostructure of **2** was determined by application of the CD helicity rule.[19] Thus, since the CD spectrum of **2** showed a positive Cotton effect at 246 nm, the 6-position was determined to have *S* configuration (Fig. 5). On the basis of the above evidence, the absolute stereostructure of corchoionoside B (**2**) was elucidated.

Corchoionoside C (**3**), obtained as a white powder with

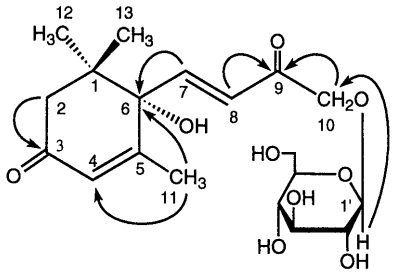


Fig. 3. Long-Range Correlations in the HMBC Spectrum of **2**

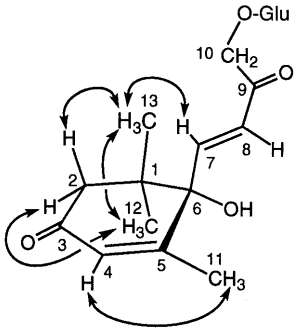


Fig. 4. NOEs Observed in the NOESY Spectrum of **2**

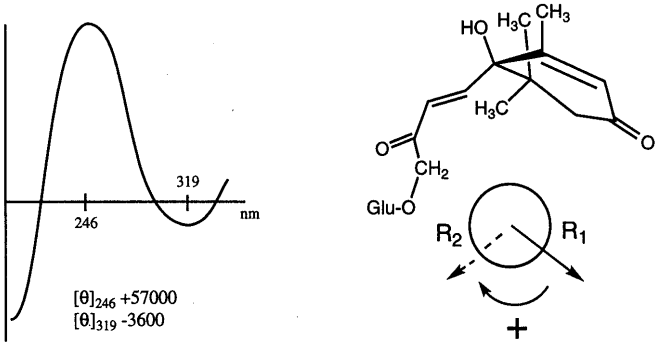


Fig. 5. The CD Curve of **2**

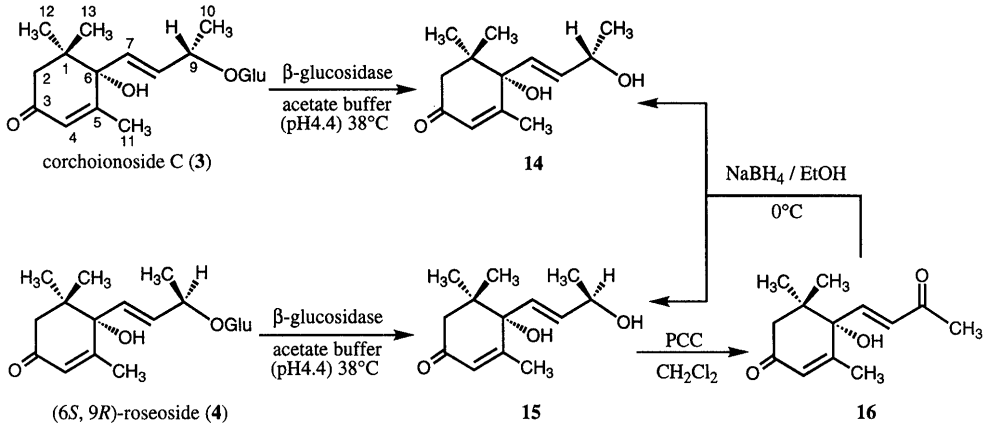


Chart 3

positive optical rotation ($[\alpha]_D^{29}$ +25.3°), showed absorption bands due to hydroxyl and enone functions in its IR spectrum. The molecular formula, $C_{19}H_{30}O_8$, of **3** was determined from the negative-mode and positive-mode FAB-MS [*m/z* 385 $(M-H)^-$ and 409 $(M+Na)^+$] and by high-resolution MS measurement. The ^{1}H-NMR (CD_3OD) and ^{13}C-NMR (Table 1) spectra[14] of **3** showed signals assignable to two tertiary methyls [δ 1.01, 1.03 (both s, 13,12-H_3)], a secondary methyl [δ 1.29 (d, J=6.3 Hz, 10-H_3)], an olefin [δ 5.73 (dd, J=7.3, 15.5 Hz, 8-H), 5.97 (d, J=15.5 Hz, 7-H)], an enone [δ 5.86 (br s, 4-H)], and a β-D-glucopyranosyl moiety [δ 4.27 (d, J=7.3 Hz, 1′-H)]. The proton and carbon signals in the ^{1}H-NMR and ^{13}C-NMR spectra[14] of **3** were superimposable on those of (6*S*,9*R*)-roseoside (**4**),[8] except for some signals around the 9-position. Thus, **3** was presumed to be the 9-stereoisomer of **4**. In order to verify this presumption, **3** was chemically correlated with **4**. Compounds **3** and **4** were subjected to enzymatic hydrolysis with β-glucosidase to yield the aglycone corchoionol C (**14**) and **15**,[8] respectively. Compound **15** was treated with pyridinium chlorochromate (PCC) in dichloromethane to give the 9-keto derivative (**16**),[8] followed by sodium borohydride ($NaBH_4$) reduction to furnish **14** and **15** in a *ca.* 2 : 1 ratio. On the basis of those findings, corchoionoside C was characterized as the 9-stereoisomer (**3**) of **4**.

Inhibitory Effect of Corchoionosides A (1) and B (2) and (6*S*,9*R*) Roseoside (4) on Histamine Release from Rat Peritoneal Exudate Cells As part of our characterization studies on antiallergic constituents of foodstuffs, we recently reported several antiallergic constituents such as isocoumarins (thunberginols A, B),[20] benzylidenephthalide (thunberginol F),[20] dihydroisocoumarins (thunberginols C, D, E, G),[21] phthalides (hydramacrophyllols A, B),[22] secoiridoid glucoside complexes (hydramacrosides A, B),[23] ent-isopimarane type diterpene ketones (trifoliones A, B, C, D),[6b] pungent principles (6-gingeol, 6-shogaol),[24] methyl-migrated *seco*-dammarane triterpene glycosides (hovenidulciosides A1, A2, B1, B2),[25] and cyanoglucosides (rhodiocyanosides A, B).[26] In the course of our continuing survey on antiallergic compounds from medicinal foodstuffs, we have examined the inhibitory activity of corchoionosides A (**1**) and B (**2**) and (6*S*,9*R*)-roseoside (**4**), the principal ionone glucosides of "moroheiya", on the histamine release from rat peritoneal exudate cells induced by antigen–antibody reaction. As shown in Table 2, **2** and **4**, both of which contain an enone function, were found to inhibit the histamine release, while **1** showed weak activity. It is interesting as a new bioactive function of health vegetable "moroheiya" that the ionone glucosides (**2**, **4**) from "moroheiya" show inhibitory effect on the histamine release.

Table 2. Inhibitory Effect of Corchoionoside A (**1**) and B (**2**) and (6*S*,9*R*)-Roseoside (**4**) on Histamine Release from Rat Peritoneal Exudate Cells Induced by Antigen–Antibody Reaction

Sample	Conc. (M)	*n*	Inhibition of histamine release (%)
Corchoionoside A (**1**)	10^{-5}	4	8.7±3.0
	10^{-4}	4	11.9±2.7
Corchoionoside B (**2**)	10^{-5}	4	4.6±5.3
	10^{-4}	4	29.4±3.9
(6*S*,9*R*)-Roseoside (**4**)	10^{-6}	4	22.2±2.7
	10^{-5}	4	31.7±5.8
	10^{-4}	4	49.7±1.7
Amlexanox	10^{-6}	4	−4.2±1.6
	10^{-5}	4	11.8±3.4
	10^{-4}	4	61.2±3.1

Each value represents the mean±S.E.

Experimental

The following instruments were used to obtain physical data: specific rotations, Horiba SEPA-300 digital polarimeter (l=5 cm); UV spectra, Shimadzu UV-1200 spectrometer; CD spectra, Jasco J 500C spectropolarimeter; IR spectra, Shimadzu FTIR-8100 spectrometer; FAB-MS and high-resolution FAB-MS, JMS-SX 102A mass spectrometer; ^{1}H-NMR spectra, JEOL EX-270 (270 MHz) spectrometer; ^{13}C-NMR spectra, JEOL EX-270 (68 MHz) spectrometer with tertramethylsilane as an internal standard; HPLC, Shimadzu LC-10AS chromatograph.

The following experimental conditions were used for chromatography: ordinary-phase column chromatography, Silica-gel BW-200 (Fuji Silysia Chemical Ltd., 150—300 mesh); reversed-phase column chromatography, Chromatorex DM1020T (Fuji Silysia Chemical Ltd., 100—200 mesh); TLC, pre-coated TLC plates with Silica-gel $60F_{254}$ (Merck, 0.25 mm) (ordinary-phase) and Silica-gel RP-18 $60F_{254}$ (Merck, 0.25 mm) (reversed-phase). Detection was done by spraying 1% $Ce(SO_4)_2$–10% aqueous H_2SO_4 followed by heating.

Extraction and Isolation The air-dried leaves of *Corcholus olitorius* L. (5 kg, cultivated in Vietnam and purchased from Honso Pharmaceutical Co., Ltd., Nagoya) were minced and extracted three times with MeOH under reflux. Removal of the solvent from the MeOH extract solution under reduced pressure gave a residue (926 g) and 463 g of the residue was partitioned into AcOEt–H_2O (1 : 1) mixture. The aqueous

layer was further extracted with 1-butanol. Removal of the solvent under reduced pressure from the AcOEt-soluble portion and 1-butanol-soluble portion yielded 121 g and 117 g of residue, respectively. The 1-butanol extract (117 g) was subjected to normal-phase silica-gel column chromatography {1.2 kg, $CHCl_3$–MeOH (10:1), $CHCl_3$–MeOH–H_2O [10:3:1, 7:3:1 (both lower layer), 6:4:1, 5:5:1]} to give six fractions [fr. 1 (6.8 g), fr. 2 (18.1 g), fr. 3 (15.7 g), fr. 4 (31.6 g), fr. 5 (20.5 g), Fr. 6 (14.7 g)]. Fraction 2 (17.5 g) was further separated by normal-phase silica-gel column chromatography [350 g, $CHCl_3$–MeOH–H_2O (15:3:1, 10:3:1, 7:3:1, all lower layer)] to provide four fractions [fr. 2-1 (13.0 g), fr. 2-2 (1.4 g), fr. 2-3 (1.5 g), fr. 2-4 (1.0 g)]. Fraction 2-2 (1.4 g) was purified by reversed-phase silica-gel column chromatography [27 g, H_2O–MeOH (7:3, 1:1, 1:4, 1:9), MeOH] and then HPLC [YMC-pack R & D-ODS-S-A, 35% aqueous MeOH) to give corchoionoside A (**1**, 39 mg), (6*S*,9*R*)-roseoside (**4**, 226 mg), betulalbuside A (**5**, 21 mg), and scopolin (**9**, 14 mg). Reversed-phase silica-gel column [30 g, H_2O–MeOH (9:1, 4:1, 3:2, 1:1, 1:4), MeOH] of fr. 2-3 (1.5 g) followed by HPLC (YMC-pack R & D-ODS-S-A, 30% aqueous MeOH) provided **1** (80 mg), **4** (57 mg), **5** (9 mg), corchoionosides B (**2**, 10 mg) and C (**3**, 7 mg), astragalin (**6**, 169 mg), and cichoriine (**8**, 39 mg). Fraction **6** (14.7 g) was subjected to reversed-phase silica-gel [300 g, H_2O–MeOH (1:1), MeOH] and ordinary-phase silica-gel column chromatography {16 g, $CHCl_3$–MeOH–H_2O [10:3:1 (lower layer), 6:4:1)]} to give isoquercitrin (**7**, 54 mg) and chlorogenic acid (**10**, 71 mg). Known compounds (**4**—**12**) were identified by comparison of their physical data ($[\alpha]_D$, mp, IR, ^{1}H-NMR, and ^{13}C-NMR) with reported values.[5,8-13]

Corchoionoside A (**1**): A white powder, $[\alpha]_D^{28}$ −24.8° (c=0.1, MeOH). High-resolution negative-mode FAB-MS: Calcd for $C_{19}H_{31}O_8$ $(M-H)^-$: 387.2019. Found: 387.2024. IR (KBr, cm^{-1}): 3425, 2965, 2926, 1655, 1638, 1076. ^{1}H-NMR (CD_3OD) δ: 0.96 (3H, s, 12-H_3), 1.14 (3H, s, 13-H_3), 1.19 (3H, s, 11-H_3), 1.22 (3H, d, J=6.3 Hz, 10-H_3), 1.35 (1H, dd, J=10.4, 13.0 Hz, 2β-H), 1.68 (1H, m, 2α-H), 1.74 (1H, dd, J=8.6, 14.5 Hz, 4α-H), 2.39 (1H, ddd, J=1.0, 5.9, 14.5 Hz, 4β-H), 3.12 (1H, dd, J=7.9, 7.9 Hz, 2′-H), 3.26—3.39 (3H, m, 3′, 4′, 5′-H), 3.67 (1H, dd, J=5.0, 11.9 Hz), 3.85 (1H, br d, J=11.9 Hz) (6′-H_2), 3.86 (1H, m, 3-H), 4.29 (1H, ddq, J=1.0, 5.6, 6.3 Hz, 9-H), 4.33 (1H, d, J=7.9 Hz, 1′-H), 5.66 (1H, dd, J=5.6, 15.5 Hz, 8-H), 5.91 (1H, dd, J=1.0, 15.5 Hz, 7-H). ^{13}C-NMR: given in Table 1. Negative-mode FAB-MS (m/z): 387 $(M-H)^-$. Positive-mode FAB-MS (m/z): 411 $(M+Na)^+$.

Corchoionoside B (**2**): A white powder, $[\alpha]_D^{23}$ +113.7° (c=0.4, MeOH). High-resolution negative-mode FAB-MS: Calcd for $C_{19}H_{27}O_9$ $(M-H)^-$: 399.1655. Found: 399.1654. IR (KBr, cm^{-1}): 3403, 2961, 2924, 2853, 1705, 1655, 1630, 1076. CD $[\theta]_{25}$ (c=0.0007, MeOH, nm): +57000 (246), −3600 (319). ^{1}H-NMR (CD_3OD) δ: 1.01 (3H, s, 13-H_3), 1.06 (3H, s, 12-H_3), 1.90 (3H, d, J=1.3 Hz, 11-H_3), 2.27 (1H, ABq, J=17.0 Hz, 2α-H), 2.60 (1H, ABq, J=17.0 Hz, 2β-H), 3.24—3.40 (4H, m, 2′, 3′, 4′, 5′-H), 3.63 (1H, dd, J=5.6, 11.9 Hz), 3.87 (1H, dd, J=1.3, 11.9 Hz) (6′-H_2), 4.33 (1H, d, J=7.6 Hz, 1′-H), 4.50, 4.74 (2H, ABq, J=17.2 Hz, 10-H_2), 5.93 (1H, d, J=1.3 Hz, 4-H), 6.66 (1H, d, J=15.8 Hz, 8-H), 7.11 (1H, d, J=15.8 Hz, 7-H). ^{13}C-NMR: given in Table 1. Negative-mode FAB-MS (m/z): 399 $(M-H)^-$. Positive-mode FAB-MS (m/z): 401 $(M+H)^+$, 423 $(M+Na)^+$.

Corchoionoside C (**3**): A white powder, $[\alpha]_D^{29}$ +25.3° (c=0.3, MeOH). High-resolution negative-mode FAB-MS: Calcd for $C_{19}H_{29}O_8$ $(M-H)^-$: 385.1863. Found: 385.1856. IR (KBr, cm^{-1}): 3432, 2940, 2926, 1655, 1654, 1645, 1076. ^{1}H-NMR (CD_3OD) δ: 1.01 (3H, s, 13-H_3), 1.03 (3H, s, 12-H_3), 1.29 (3H, d, J=6.3 Hz, 10-H_3), 1.94 (3H, d, J=1.3 Hz, 11-H_3), 2.17 (1H, br d, J=16.8 Hz, 2α-H), 2.61 (1H, br d, J=16.8 Hz, 2β-H), 3.14—3.32 (4H, m, 2′, 3′, 4′, 5′-H), 3.63 (1H, dd, J=6.1, 11.9 Hz), 3.85 (1H, dd, J=2.3, 11.9 Hz) (6′-H_2), 4.27 (1H, d, J=7.3 Hz, 1′-H), 4.53 (1H, dq, J=6.3, 7.3 Hz, 9-H), 5.73 (1H, dd, J=7.3, 15.5 Hz, 8-H), 5.86 (1H, br s, 4-H), 5.97 (1H, d, J=15.5 Hz, 7-H). ^{13}C-NMR: given in Table 1. Negative-mode FAB-MS (m/z): 385 $(M-H)^-$. Positive-mode FAB-MS (m/z): 409 $(M+Na)^+$.

Acetylation of Corchoionoside A (1) A solution of **1** (2.0 mg) in pyridine (0.2 ml) was treated with Ac_2O (0.2 ml) in the presence of 4-DMAP (5.0 mg) and the reaction mixture was stirred at room temperature (20 °C) under an N_2 atmosphere for 3 h. The reaction mixture was poured into ice-water and the whole was extracted with AcOEt. The AcOEt extract was washed successively with 5% aqueous HCl, saturated aqueous $NaHCO_3$, and brine, then dried over $MgSO_4$, and filtered. After removal of the solvent from the filtrate under reduced pressure, the residue was purified on a reversed-phase silica-gel column (0.5 g, MeOH : H_2O = 50 : 50) to yield corchoionoside A pentaacetate (**1a**, 2.5 mg, 79.6%).

Corchoionoside A Pentaacetate (**1a**): A white powder, $[\alpha]_D^{21}$ −6.8° (c=0.3, MeOH). High-resolution positive-mode FAB-MS: Calcd for $C_{29}H_{43}O_{13}$ $(M+H)^+$: 599.2703. Found: 599.2717. IR (KBr, cm^{-1}): 3445, 2963, 2931, 2875, 1757. ^{1}H-NMR (CD_3OD) δ: 0.91 (3H, s, 12-H_3), 1.11 (3H, s, 13-H_3), 1.16 (3H, s, 11-H_3), 1.30 (3H, d, J=6.6 Hz, 10-H_3), 1.33 (1H, dd, J=9.6, 13.5 Hz, 2β-H), 1.65 (1H, ddd, J=1.3, 3.3, 13.5 Hz, 2α-H), 1.68 (1H, dd, J=7.9, 14.5 Hz, 4α-H), 2.27 (1H, ddd, J=1.3, 5.0, 14.5 Hz, 4β-H), 1.96, 2.00, 2.02, 2.02, 2.05 (15H, all s, Ac × 5), 3.85 (2H, m, 3, 5′-H), 4.11 (1H, dd, J=2.5, 12.2 Hz), 4.26 (1H, dd, J=5.0, 12.2 Hz) (6′-H_2), 4.74 (1H, d, J=7.9 Hz, 1′-H), 4.81 (1H, m, 2′-H), 4.97 (1H, dd, J=9.6, 9.6 Hz, 4′-H), 5.25 (1H, dd, J=9.6, 9.6 Hz, 3′-H), 5.33 (1H, ddq, J=1.0, 6.4, 6.6 Hz, 9-H), 5.61 (1H, dd, J=6.4, 15.5 Hz, 8-H), 5.96 (1H, dd, J=1.0, 15.5 Hz, 7-H). ^{13}C-NMR: given in Table 1. Positive-mode FAB-MS (m/z): 599 $(M+H)^+$.

Enzymatic Hydrolysis of Corchoionoside A (1) Giving Corchoionol A (11) A solution of **1** (30.0 mg) in acetate buffer (pH 4.4, 6.0 ml) was treated with β-glucosidase (Oriental Yeast Co., Ltd., 30.0 mg) and the solution was left standing at 38 °C for 16 h. The reaction solution was poured into water and the whole was extracted with AcOEt. The AcOEt extract was washed with brine, then dried over $MgSO_4$, and filtered. After removal of the solvent under reduced pressure, the residue was purified by HPLC (YMC Pack R & D, H_2O–MeOH) to yield corchoionol A (**11**, 16.1 mg, 92.1%).

11: A white powder, $[\alpha]_D^{27}$ −56.1° (c=0.3, MeOH). High-resolution positive-mode FAB-MS: Calcd for $C_{13}H_{22}O_3Na$ $(M+Na)^+$: 249.1467. Found: 249.1437. IR (KBr, cm^{-1}): 3414, 2967, 2928, 1655, 1051. ^{1}H-NMR ($CDCl_3$) δ: 0.97 (3H, s, 12-H_3), 1.13 (3H, s, 13-H_3), 1.19 (3H, s, 11-H_3), 1.22 (3H, d, J=10.7, 12.8 Hz, 2β-H), 1.28 (3H, d, J=6.4 Hz, 10-H_3), 1.61 (1H, ddd, J=1.8, 3.4, 12.8 Hz, 2α-H), 1.61 (1H, dd, J=8.9, 14.0 Hz, 4α-H), 2.36 (1H, ddd, J=1.8, 5.1, 14.0 Hz, 4β-H), 3.89 (1H, dd-like, 3-H), 4.38 (1H, dq, J=6.1, 6.4 Hz, 9-H), 5.75 (1H, dd, J=6.1, 15.3 Hz, 8-H), 5.89 (1H, d, J=15.3 Hz, 7-H). ^{13}C-NMR: given in Table 1. Positive-mode FAB-MS (m/z): 249 $(M+Na)^+$.

Preparation of the (*R*)-MTPA Ester (12) and the (*S*)-MTPA Ester (13) from Corchoionol A (11) A solution of **11** (3.7 mg, 0.016 mmol) in CH_2Cl_2 (1.0 ml) was treated with (*R*)-MTPA (51.1 mg, 0.22 mmol) in the presence of EDC·HCl (41.9 mg, 0.22 mmol) and 4-DMAP (18.0 mg, 0.15 mmol), and the mixture was stirred at room temperature (20 °C) under an N_2 atmosphere for 8 h. It was poured into ice-water and the whole was extracted with AcOEt. The AcOEt extract was successively washed with 5% aqueous HCl, aqueous saturated $NaHCO_3$, and brine, then dried over $MgSO_4$ and filtered. Evaporation of the solvent from the filtrate under reduced pressure furnished a residue, which was purified on a silica-gel column (0.6 g, *n*-hexane : AcOEt = 20 : 1) to give **12** (6.6 mg, 13%). Through a similar procedure, **13** (5.0 mg) was prepared from **11** (3.9 mg) by the use of (*S*)-MTPA (40.4 mg), EDC·HCl (33.1 mg), and 4-DMAP (12.6 mg).

Corchoionol A (*R*)-MTPA Ester (**12**): A white powder. ^{1}H-NMR (CD_3OD) δ: 0.95 (3H, s, 12-H_3), 1.05 (3H, s, 13-H_3), 1.10 (3H, s, 11-H_3), 1.34 (3H, d, J=6.3 Hz, 10-H_3), 1.45 (1H, dd, J=8.9, 13.5 Hz, 2β-H), 1.70 (1H, ddd, J=1.3, 3.3, 13.5 Hz, 2α-H), 1.80 (1H, dd, J=6.9, 14.9 Hz, 4α-H), 2.33 (1H, ddd, J=1.0, 5.3, 14.9 Hz, 4β-H), 5.14 (1H, m, 3-H), 5.59 (1H, dd, J=6.3, 6.9 Hz, 9-H), 5.66 (1H, dd, J=6.9, 14.9 Hz, 8-H), 6.06 (1H, d, J=14.9 Hz, 7-H).

Corchoionol A (*S*)-MTPA Ester (**13**): A white powder. ^{1}H-NMR (CD_3OD) δ: 0.90 (3H, s, 12-H_3), 0.92 (3H, s, 13-H_3), 1.15 (3H, s, 11-H_3), 1.31 (1H, m, 2β-H), 1.40 (3H, d, J=6.3 Hz, 10-H_3), 1.62 (1H, ddd, J=1.3, 3.6, 13.2 Hz, 2α-H), 1.91 (1H, dd, J=7.6, 14.8 Hz, 4α-H), 2.43 (1H, ddd, J=1.3, 5.6, 14.8 Hz, 4β-H), 5.60 (1H, d, J=14.5 Hz, 8-H), 5.60 (1H, m, 9-H), 5.93 (1H, dd, J=3.6, 14.5 Hz, 7-H).

Enzymatic Hydrolysis of Corchoionoside C (3) Giving Corchoionol C (14) A solution of **3** (4.2 mg) in acetate buffer (pH 4.4, 1.0 ml) was treated with β-glucosidase (Oriental Yeast Co., Ltd., 5.0 mg) and the solution was left standing at 38 °C for 7 h. It was poured into water and the whole was extracted with AcOEt. The AcOEt extract was washed with brine, then dried over $MgSO_4$, and filtered. Removal of the solvent from the filtrate under reduced pressure gave **14** (2.4 mg, 99%).

Corchoionol C (**14**): A white powder, $[\alpha]_D^{28}$ +184.3° (c=0.1, $CHCl_3$), High-resolution positive-mode FAB-MS: Calcd for $C_{13}H_{21}O_3$ $(M+H)^+$: 225.1490. Found: 225.1480. IR (KBr, cm^{-1}): 3422, 2965, 2924, 2853, 1655. ^{1}H-NMR (CD_3OD) δ: 1.01 (3H, s, 13-H_3), 1.03 (3H, s, 12-H_3), 1.24 (3H, d, J=6.6 Hz, 10-H_3), 1.92 (3H, d, J=1.3 Hz, 11-H_3), 2.15 (1H, ABq, J=17.0 Hz, 2α-H), 2.51 (2H, ABq, J=17.0 Hz, 2β-H), 4.32 (1H,

q, J=6.6 Hz, 9-H), 5.83 (3H, m, 4, 7, 8-H). ^{13}C-NMR: given in Table 1. Negative-mode FAB-MS (m/z): 223 $(M-H)^-$. Positive-mode FAB-MS (m/z): 225 $(M+H)^+$, 207 $(M-H_2O+H)^+$], 247 $(M+Na)^+$.

Enzymatic Hydrolysis of (6*S*,9*R*)-Roseoside (4) Followed by PCC Oxidation A solution of **4** (40.2 mg) in acetate buffer (pH 4.4, 6.0 ml) was treated with β-glucosidase (Oriental Yeast Co., Ltd., 55.0 mg) and the solution was left standing at 38 °C for 55 h. Work-up of the reaction solution as described above yielded **15** (18.6 mg, 80%), which was identified by comparison of the physical data (^{1}H-NMR, IR, $[\alpha]_D$, CD) with reported values.[27] A solution of **15** (16.2 mg, 0.0072 mmol) in CH_2Cl_2 was treated with PCC (31.2 mg, 0.14 mmol) and the mixture was stirred at room temperature (20 °C) for 4 h. After removal of the insoluble part from the reaction mixture by filtration, the filtrate was evaporated under reduced pressure to give a residue (52.9 mg, quant.). This was purified on a normal-phase silica-gel column (2.0 g, $CHCl_3$) to furnish **16** (14.0 mg, 86.4%), which was identified by comparison of the physical data (^{1}H-NMR, IR, $[\alpha]_D$, CD) with reported values.[27]

$NaBH_4$ Reduction of the 9-Keto Derivative (16) A solution of **16** (13.2 mg, 0.059 mmol) in EtOH (1.0 ml) was treated with $NaBH_4$ (2.0 mg) and the mixture was left standing at 0 °C for 30 min. It was treated with acetone (0.5 ml) to quench excess $NaBH_4$ and the whole was neutralized with Dowex HCR-W2 (H^+ form). After removal of the resin by filtration, the filtrate was evaporated under reduced pressure to yield a mixture of reduction products (13.1 mg, 98.4%), which were separated by HPLC [Capcell pack C_{18}, H_2O–MeOH (65:35)] to give **14** (7.0 mg, 51.7%) and **15** (3.1 mg, 21.9%).

Histamine Release from Rat Peritoneal Exudate Cells This experiment was performed according to the method previously reported.[20c] Briefly, peritoneal exudate cells from male Wistar rats weighing 350 to 500 g were sensitized with diluted anti-DNP IgE (×100) at 37 °C for 1 h. The cell suspension (10^4 cells/1.62 ml) and 180 μl of test compound were preincubated for 15 min; 200 μl of phosphatidylserine (1 mg/ml) and 222 μl of DNP-BSA (1 mg/ml) were added at the same time. The incubation was continued for 20 min. The test tube was placed in an ice-cold bath to stop the reaction. The supernatant was obtained by centrifugation for 10 min at 100×***g***, 4 °C, and the histamine concentration of the supernatant was measured by the method of Imada *et al.*[28]

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