# Tannins and Related Compounds. CXVIII.[1] Structures, Preparation, High-Performance Liquid Chromatography and Some Reactions of Dehydroellagitannin–Acetone Condensates

Takashi TANAKA, Hiroshi FUJISAKI, Gen-ichiro NONAKA,* and Itsuo NISHIOKA

*Faculty of Pharmaceutical Sciences, Kyushu University 62, 3–1–1 Maidashi, Higashi-ku, Fukuoka 812, Japan.* *Received April 13, 1992*

**The dehydroellagitannins having a dehydrohexahydroxydiphenoyl ester group in the molecule were found to undergo highly regio- and stereospecific condensation with acetone in the presence of ammonium ion under almost neutral conditions. This reaction was successfully applied to high-performance liquid chromatographic analysis to detect the dehydroellagitannins in the plant extract. Furthermore, the reactions of the acetone condensates with L-cysteine methyl ester were examined, and appear to be applicable to structural studies of dehydroellagitannins.**

**Keywords** dehydroellagitannin; dehydrohexahydroxydiphenic acid; acetone condensate; ammonium ion; HPLC; L-cysteine methyl ester; hydrolyzable tannin; tannin

Recent progress in the chemistry of tannins has revealed that dehydroellagitannins [*e.g.*, geraniin (**1**),[2] *etc.*], a group of hydrolyzable tannins possessing a dehydrohexahydroxydiphenyl (DHHDP) group, are distributed widely in the plant kingdom, especially in members of the families Euphorbiaceae,[3] Cercidiphyllaceae,[4] Elaeocarpaceae,[5] Punicaceae,[6] Combretaceae,[7] *etc.* The dehydroellagitannin usually exists in solution as an equilibrium mixture of five- and six-membered hemiketal ring structures (**1a** and **1b**),[2] and hence gives a broad collapsed peak in high-performance liquid chromatography (HPLC) (Fig. 1a and b). In structural studies, since the presence of these two tautomers complicates the proton and carbon-13 nuclear magnetic resonance ($^1$H- and $^{13}$C-NMR, respectively) spectra, the phenazine derivatives (*e.g.*, **1d**),[2,8] which are prepared by condensation with *o*-phenylenediamine, are commonly used so that spectra which are simpler and more amenable to analysis can be obtained. However, some of the phenazine derivatives [*e.g.*, **4d**[3f]] are extremely unstable in solution and readily undergo solvolysis even during spectral measurement.

In the course of our chemical studies on tannins, we have sometimes isolated the acetone condensates of dehydroellagitannins, *e.g.*, acetonylgeraniin (phyllanthusiin D) (**1c**),[9] acetonylhelioscopinin A (**4c**),[9a] and acetonylcarpinusin (**5c**),[9a] from the aqueous acetone extracts.[10] Since these acetone condensates are invariably accompanied with the respective dehydroellagitannins, we consider that these compounds are artifacts. In the case of the roots of *Euphorbia adenochlora*, HPLC analysis of the aqueous acetone extract (Fig. 1b) showed peaks corresponding to the acetone condensates (**1c**, **4c** and **5c**), whereas these peaks were not observed in the aqueous methanol extract (Fig. 1a). These facts clearly indicated that these compounds are artifacts formed during the extraction with aqueous acetone (Chart 1).

These acetone condensates are fairly stable and are expected to become useful alternatives to the phenazine derivatives. Hence, we wish to describe herein details of their structural elucidation, efficient preparation and HPLC analysis. Furthermore, the reactions of these acetone

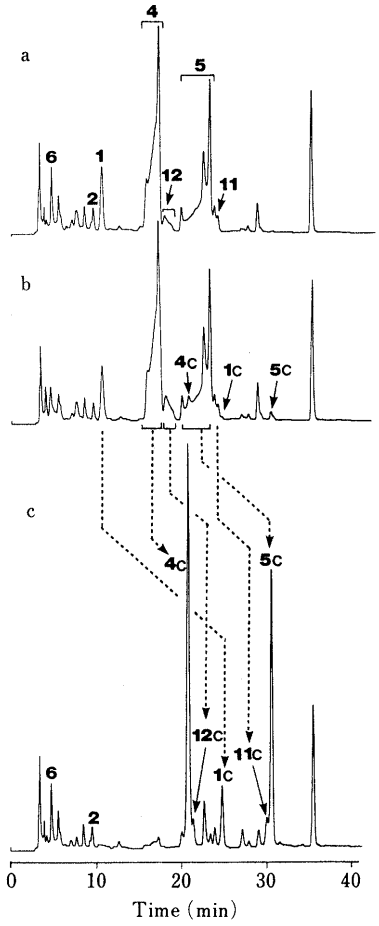



Fig. 1. HPLC Chromatograms of the Extracts of *Euphorbia adenochlora*

a) 80% aqueous MeOH extract; b) 80% aqueous acetone extract; c) 80% aqueous MeOH extract treated with 80% aqueous acetone containing ammonium formate (50 °C, 1 h).

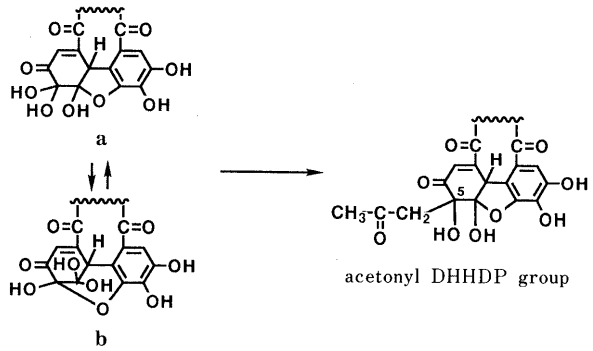



Chart 1

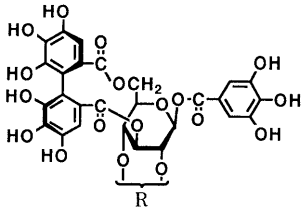
condensates with L-cysteine methyl ester,[11] which should be useful for structural studies of dehydroellagitannins, are also described.

**Structure Determination** The structures of the acetone condensates were established on the basis of spectroscopic evidence. For example, the $^1$H-NMR spectrum of acetonyl-helioscopinin A (**4c**) showed signals due to a galloyl ($\delta$ 7.12, 2H, s), a hexahydroxydiphenoyl ($\delta$ 6.63 and 6.93, each 1H, s) and a sugar moiety, the chemical shifts and the coupling patterns of which were closely related to those of the five-membered ring system (**4a**) of helioscopinin A.[3f] The observation of an aromatic singlet ($\delta$ 7.24) and mutually coupled methine ($\delta$ 4.95, d, $J=1$ Hz) and olefinic ($\delta$ 6.34, d, $J=1$ Hz) proton signals is also consistent with the features of **4a**. However, the appearance of isolated methyl ($\delta$ 2.16, 3H, s) and methylene ($\delta$ 2.93 and 3.48, each d, $J=15$ Hz) signals, together with the carbonyl carbon signal ($\delta$ 206.2) in the $^{13}$C-NMR spectrum, indicated the presence of an acetonyl group connected to a quaternary carbon through a carbon-to-carbon linkage. This was supported by the observation of the $(M-H)^-$ peak at $m/z$ 991 in the negative ion fast atom bombardment mass spectrum (FAB-MS). In the $^{13}$C-NMR spectrum, the chemical shifts of the signals due to the cyclohexenone ring are similar to those of **4a**, except for the appearance of the C-5 carbon signal at significantly upper field ($\delta$ 80.9) compared with that of **4a** ($\delta$ 92.4). This observation indicated that the acetonyl group is attached to this position. Furthermore, the configuration of the C-5 atom in **4c** was determined by measurement of the two-dimensional nuclear Overhauser effect (NOESY) spectrum, which showed a cross peak between the benzylmethine (H-1) ($\delta$ 4.95) and methylene ($\delta$ 2.93) signals. On the basis of the findings mentioned above, and the derivation of **4c** from **4** (*vide infra*), the structure of ace-

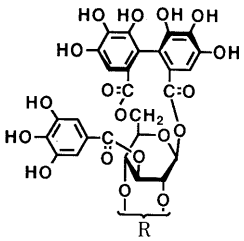
Table I. Condensation of Dehydroellagitannins (DET) with Acetone

| DET | Product | Yield (%)[a] |
|---|---|---|
| Geraniin (**1**) | **1c** | 87 |
| Granatin B (**3**)[6] | **3c** | 62 |
| Helioscopinin A (**4**) | **4c** | 85 |
| Carpinusin (**5**)[12] | **5c** | 67 |
| Furosin (**7**)[13] | **7c** | 82 |
| Terchebin (**8**)[7] | **8c** | 85 |
| Mallotusinic acid (**9**)[3a] | **9c** | 80 |
| Granatin A (**10**)[6] | **10c** | 74 |
| Euphorscopin (**11**)[3i] | **11c** | 86 |

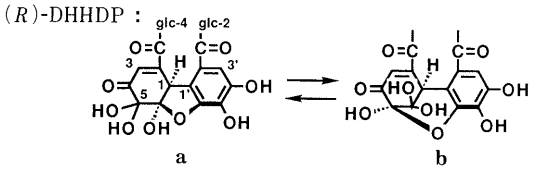
a) Isolated yield.



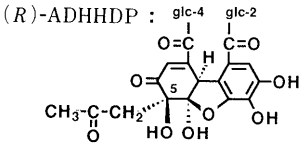
**1** : R=(*R*)-DHHDP
**1c**: R=(*R*)-ADHHDP
**1d**: R=(*R*)-phenazine
**1e**: R=AP-DHHDP
**1f** : R=P-DHHDP
**2** : R=H, H
**3** : R=(*S*)-DHHDP
**3c**: R=(*S*)-ADHHDP



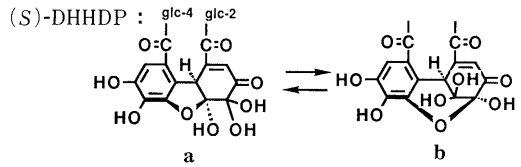
**4** : R=(*S*)-DHHDP
**4c**: R=(*S*)-ADHHDP
**4d**: R=(*S*)-phenazine
**5** : R=(*R*)-DHHDP
**5c**: R=(*R*)-ADHHDP
**6** : R=H, H



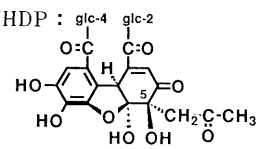
(*R*)-DHHDP : a ⇄ b



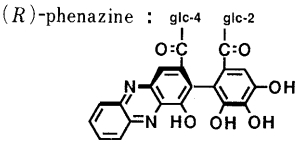
(*R*)-ADHHDP



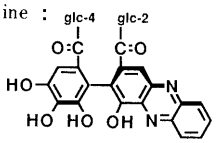
(*S*)-DHHDP : a ⇄ b



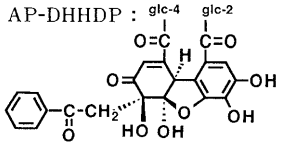
(*S*)-ADHHDP



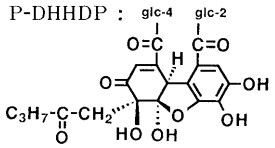
(*R*)-phenazine



(*S*)-phenazine



AP-DHHDP



P-DHHDP

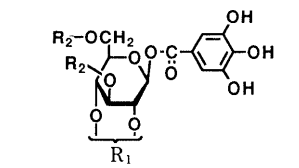
tonylhelioscopinin A was concluded to be as represented by the formula **4c**.

**Preparation** Although as mentioned above, condensation of acetone with the DHHDP group was proved to occur during the extraction, purified dehydroellagitannins alone did not react with acetone even at elevated temperature. This observation suggested that a substance catalyzing this aldol-type condensation under mild conditions exists in the plant material. We examined, therefore, whether various salts catalyze the reaction in aqueous acetone, and found that some ammonium salts (ammonium formate, ammonium acetate and ammonium bicarbonate) efficiently catalyze the condensation. Other related salts, such as sodium formate, sodium acetate, ammonium sulfate and ammonium chloride, and amino acids, such as glycine, did not catalyze the reaction. These results indicated that the presence of ammonium ion under neutral or weakly alkaline condition is required to promote this condensation. Furthermore, the structural similarity between the hydrated cyclohexenetrione ring of the DHHDP group and ninhydrin suggested that the ammonium ion acts as a Schiff base to form an imine at the C-5 position. Under similar conditions, acetophenone and 2-pentanone reacted with the DHHDP group in **1** to give the condensates **1e** and **1f**, respectively. However, reaction of **1** with acetoaldehyde, cyclohexanone and acetylacetone afforded complex mixtures.

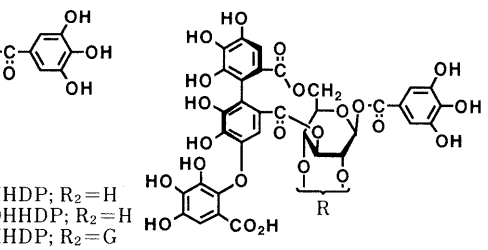
Nine dehydroellagitannins possessing the *R*- or *S*-DHHDP group afforded the respective acetone condensates in high yields (Table I). In each case, the reaction yielded a single acetone condensate, suggesting that this reaction occurs regio- and stereospecifically at the C-5 position of the DHHDP group.

Trapain (**13**),[14] however, did not afford the corresponding acetone condensate, and a large portion of the starting material was recovered. This is probably because the DHHDP in this tannin exists as a rigid six-membered ring system,[14] the C-5 position being protected.

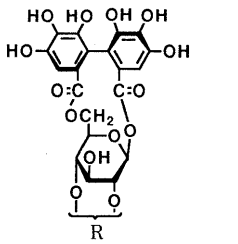
**Application to HPLC Analysis of Dehydroellagitannins** As mentioned above, the equilibrium between the five- and six-membered hemiketal ring structures of the DHHDP group causes broadening or splitting of the peaks on HPLC analysis (Fig. 1a and b), and this phenomenon always makes it troublesome to identify the peaks. This problem was solved by application of the above-mentioned reaction. Figure 1c shows a chromatogram of the aqueous methanol extract of *Euphorbia adenochlora*, treated with aqueous acetone containing ammonium formate at 50 °C for one hour. In the chromatogram, the peaks due to the dehydroellagitannins [**1**, **4**, carpinusin (**5**),[12] euphorscopin (**11**)[3i] and jolkianin (**12**)[3i]] disappeared, and instead, sharp peaks arising from the respective acetone condensates (**1c**, **4c**, **5c**, **11c** and **12c**) were observed. This result indicated that the dehydroellagitannins in the extract are almost



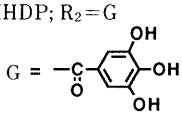
**7** : $R_1$=(*R*)-DHHDP; $R_2$=H
**7c**: $R_1$=(*R*)-ADHHDP; $R_2$=H
**8** : $R_1$=(*R*)-DHHDP; $R_2$=G
**8c**: $R_1$=(*R*)-ADHHDP; $R_2$=G



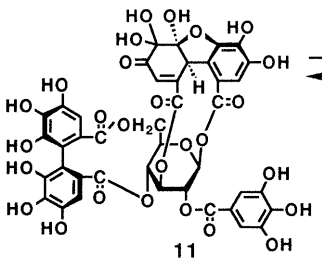
**9** : R=(*R*)-DHHDP
**9c**: R=(*R*)-ADHHDP



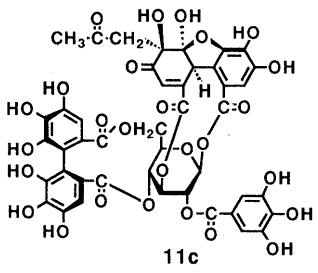
**10** : R=(*S*)-DHHDP
**10c**: R=(*S*)-ADHHDP



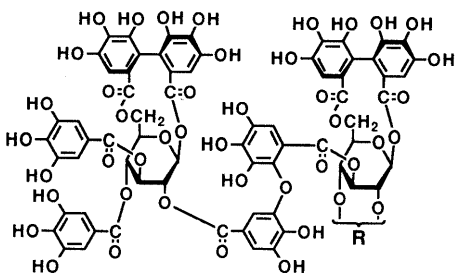
G =



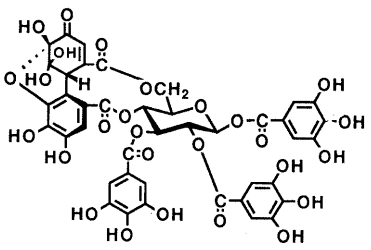
**11**



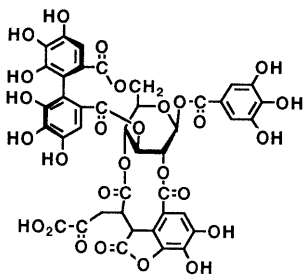
**11c**



**12** : R=(*S*)-DHHDP
**12c**: R=(*S*)-ADHHDP



**13**



**14**

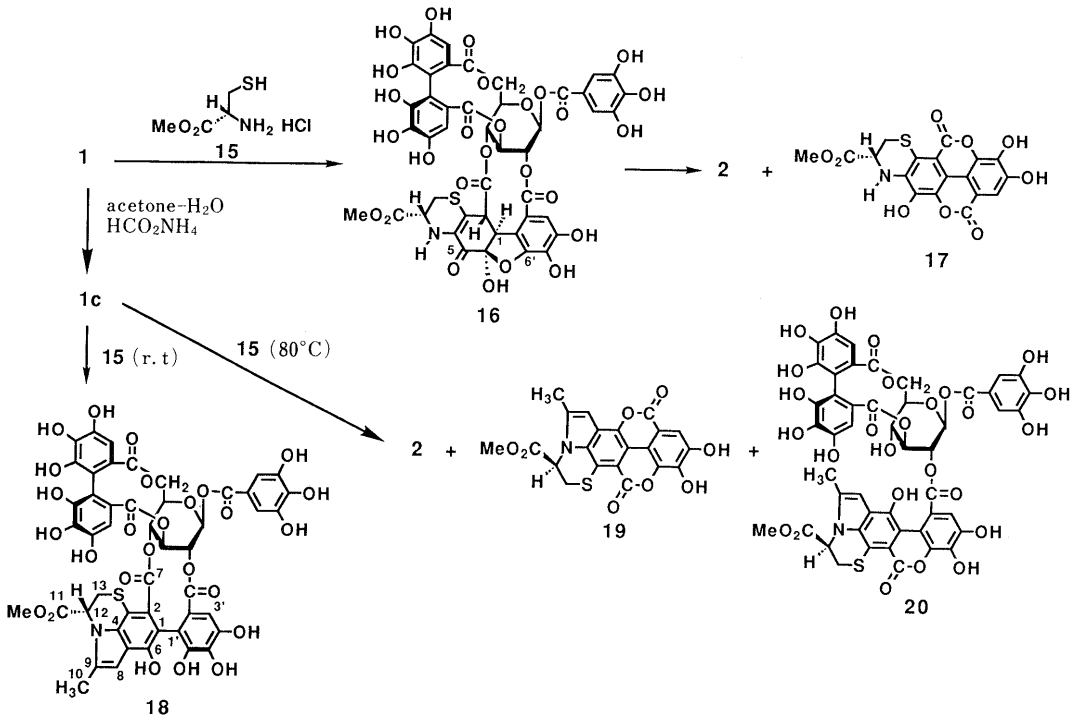


Chart 2

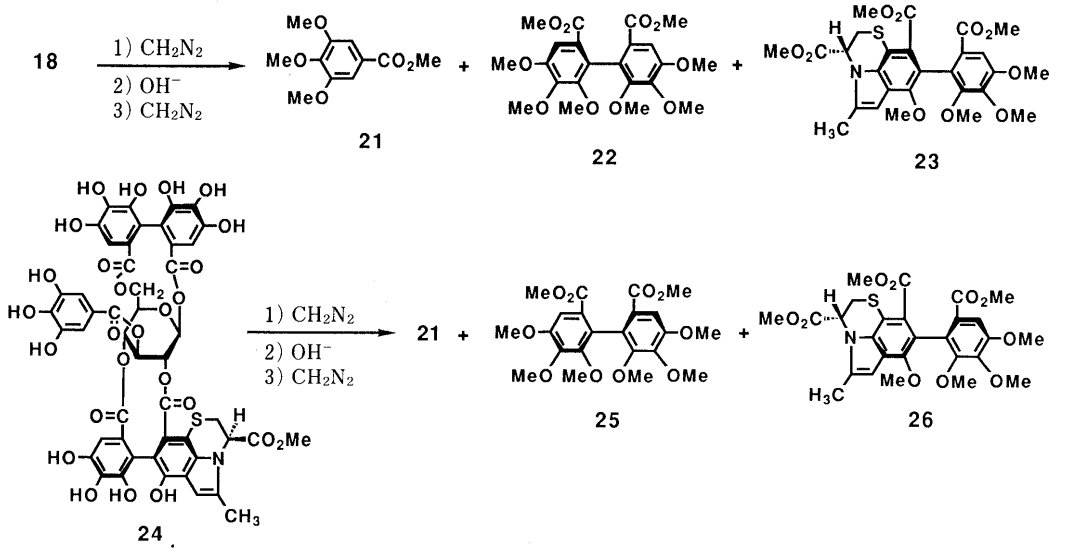


Chart 3

quantitatively converted into the respective acetone condensates by simple derivatization. Moreover, by comparison of the chromatograms before and after reaction, the peaks due to the dehydroellagitannins are apparently distinguishable from those of other tannins, such as corilagin (**2**)[15] and helioscopinin B (**6**).[3f]

**Some Reactions of the Acetone Condensates** For the purpose of developing a new method for the structural elucidation of dehydroellagitannins, the reactivities of the acetone condensates were investigated. On treatment of the acetone condensate (**1c**) with 1 N sulfuric acid (80 °C, 1 h), no reaction occurred and the starting material was recovered, whereas heating of **1c** in phosphate buffer (pH 7.0, 80 °C, 2 h) afforded the partial hydrolysates, corilagin (**2**) (30%) and another product (**14**) (32%). The structure of **14** was determined as follows. The negative ion FAB-MS showed the $(M-H)^-$ peak at *m/z* 951, in accord with that of geraniin (**1**), suggesting the loss of the acetonyl moiety. The $^1$H-NMR spectrum exhibited aliphatic ABMX-type signals [$\delta$ 5.02 (d, $J=3$ Hz); 3.76 (ddd, $J=2$, 3, 11 Hz); 2.37 (dd, $J=2$, 19 Hz) and 2.90 (dd, $J=11$, 19 Hz)], while the $^{13}$C-NMR spectrum showed signals due to a carbonyl ($\delta$ 195.5) and seven carboxyl carbons ($\delta$ 163.4, 164.5, 165.5, 166.6, 169.0, 171.3, 176.2), among which the chemical shift of the carbonyl signal at $\delta$ 176.2 suggested the presence of a $\beta,\gamma$-unsaturated $\gamma$-lactone ring in the molecule. This was supported by the infrared absorption at 1800 cm$^{-1}$. Furthermore, the relatively upfield shifts of the carboxyl and carbonyl signals at $\delta$ 163.4 and 195.5, which are similar to those of oxalic acid ($\delta$ 160.1) and diacetyl ($\delta$ 197.7),

respectively, suggested the presence of an α-ketocarboxylic acid moiety. On the basis of these spectral findings, the structure of the product was concluded to be as represented by the formula **14**. This compound was considered to have been formed by a retro-aldol-type cleavage and fission of the cylohexanone ring.

Recently, we have reported on the condensation reaction of the dehydroellagitannins with L-cysteine methyl ester (**15**),[11] which was found to be applicable to the specific hydrolysis of the DHHDP group, *e.g.* geraniin (**1**) afforded in excellent yields corilagin (**2**) and the bislactone (**17**) *via* the condensation product (**16**) (Chart 2). Application of this reaction to the acetone condensate (**1c**) at room temperature gave a single product (**18**) (63%). The $^1$H-NMR spectrum of **18** showed signals corresponding to the cysteine methyl ester moiety [$\delta$ 5.65 (1H, dd, $J=3$, 4 Hz, methine), 3.88 (3H, s, methyl), 3.69 (1H, dd, $J=3$, 14 Hz, methylene) and 3.35 (1H, dd, $J=4$, 14 Hz, methylene)]. The signals due to benzyl methine, olefinic and methylene protons originally found in the acetonyl DHHDP group disappeared, and instead, mutually coupled olefinic ($\delta$ 6.38, d, $J=1$ Hz) and methyl signals ($\delta$ 2.46, d, $J=1$ Hz) were observed. The small coupling constant indicated long-range coupling between these protons. In the $^{13}$C-NMR spectrum, the absence of a carbonyl signal and the appearance of olefinic carbon signals at $\delta$ 98.9 and 131.8 suggested the presence of an enamine structure in the molecule of **18**. Taking into account the structure of the condensation product (**16**) of **1**, these spectral observations led us to formulate the structure (**18**) for this product, and the result of negative ion FAB-MS, exhibiting the $(M-H)^-$ peak at *m/z* 1072, was in agreement with this proposed structure. Similarly, application of this reaction to acetonylhelioscopinin A (**4c**) afforded **24** in a high yield (75%).

The reaction of **1c** with **15** at 80 °C afforded the products **19** (46%) and **20** (38%), togehter with corilagin (**2**) (55%). The negative ion FAB-MS of **19** showed the $(M-H)^-$ peak at *m/z* 438, suggesting that this compound originated from the 2,4-acyl group. The $^1$H- and $^{13}$C-NMR spectra exhibited signals similar to those of the 2,4-acyl group, and the observation of two $\delta$-lactone signals at $\delta$ 159.0 and 159.1 led us to conclude the structure to be as shown by the formula **19**.

The product **20** was shown to have the same molecular mass as **18** by negative ion FAB-MS [*m/z*: 1072 $(M-H)^-$]. The $^1$H-NMR spectrum was closely related to that of **18**, except for the significant upfield shift of the glucopyranose H-4 signal. The $^{13}$C-NMR spectrum of **18** also showed a close resemblance to that of **20**, but differed in the chemical shifts of the sugar signals and the aromatic C-6′ signal ($\delta$ 148.7), the latter being shifted downfield as compared with that of **20** ($\delta$ 136.2—146.1). Furthermore, heating of **20** in aqueous acetonitrile yielded **2** and **19**. On the basis of these findings, the structure of this product was confirmed as **20**. The reaction of acetonylhelioscopinin A (**4c**) with **15** under the same conditions similarly afforded **19** (42%), helioscopinin B (**6**) (35%) and **24** (43%).

Methylation of **1c** with dimethyl sulfate and potassium carbonate in dry acetone yielded the dodecamethylate (30%) [field-desorption mass spectrum (FD-MS) *m/z*: 1160 $(M^+)$]. Subsequent alkaline methanolysis of the methyl ether formed a complex mixture, and gave, among others, methyl trimethoxybenzoate (**21**) and dimethyl (*R*)-hexamethoxydiphenate (**22**). Alternatively, methylation of **18** with diazomethane, followed by alkaline hydrolysis and methylation, afforded compound **23** [electron impact mass spectrum (EI-MS) *m/z*: 559 $(M^+)$], together with **21** and **22** (Chart 3). Similar treatment of **24** furnished **26** [EI-MS *m/z*: 559 $(M^+)$], **21** and dimethyl (*S*)-hexamethoxydiphenate (**25**) (Chart 3). Compounds **23** and **26** differed in specific optical rotation [**23**: $[\alpha]_D$ −117.6° ($CHCl_3$), **26**: $[\alpha]_D$ −180.4° ($CHCl_3$)] and also slightly in the chemical shifts of the methoxyl signals in their $^1$H-NMR spectra, thus confirming that these compounds are diastereoisomeric.

These results indicated that the formation of the acetone condensates, followed by reaction with L-cysteine methyl ester, is useful for the selective hydrolysis and determination of the chirality of the DHHDP group. Taking their stability into account, the acetone condensates are expected to become useful alternatives to the phenazine derivagives for the structure elucidation of the dehydroellagitannins.

## Experimental

Details of the instruments and chromatographic conditions used throughout this work was essentially the same as described in the previous paper,[1] except in the following respects. HPLC was performed on a Tosoh apparatus equipped with a CCPM solvent delivery system, a UV-8000 spectrometer and a Cosmosil 5$C_{18}$-AR (Nacarai Tesque) column (4.6 mm i.d. × 250 mm) [mobile phase, acetonitrile–50 mM $H_3PO_4$ aqueous solution (gradient elution of 15%→40% acetonitrile for 90 min); flow rate, 0.8 ml/min; column temperature, 20 °C; detection, 280 nm].

**HPLC Analysis of the Extract of *E. adenochlora*** The underground parts collected in Saga prefecture, Japan, in February, were dried at room temperature for one week. Two portions (2.0 g) of the milled roots were separately extracted with 80% aqueous acetone (20 ml) and 80% aqueous methanol (20 ml) at room temperature for 5 d. Each extact was concentrated, passed through Toyopak ODS (Tosoh) with 60% aqueous methanol, and concentrated to dryness. The residues were dissolved in 50% aqueous acetonitrile (20 ml) and subjected to HPLC analysis (Fig. 1a and b). A portion (2.0 ml) of the solution of the aqueous methanol extract was concentrated to dryness and treated with 80% aqueous acetone (5.0 ml) containing ammonium formate (30 mg) at 50 °C for 1 h. The mixture was concentrated to dryness, then the residue was dissolved in 2.0 ml of 50% aqueous acetonitrile and subjected to HPLC (Fig. 1c).

**General Procedures for Preparing the Acetone Condensates (1c, 3c, 4c, 5c and 7c—11c)** A solution of each dehydroellagitannin (50—200 mg) and ammonium formate (25—100 mg) in 80% aqueous acetone (5—20 ml) was stirred at 50 °C for 2 h. The reaction mixture was concentrated until all the acetone was removed. In the cases of **1**, **4**, **5** and **8**, the precipitates of the products were formed, and these were collected by filtration. In other cases, the mixture was subjected to Sephadex LH-20 chromatography (2.0 cm i.d. × 25 cm) with 80% aqueous methanol to give the products. The yields are shown in Table I. Similar treatment of trapain (**13**) (100 mg) resulted in recovery of the starting material (70 mg).

**Acetonylgeraniin (1c)** A white powder ($H_2O$), mp 220—222 °C (dec.), $[\alpha]_D^{22}$ −132.8° ($c=1.0$, MeOH). *Anal.* Calcd for $C_{44}H_{32}O_{27}\cdot 6H_2O$: C, 48.01; H, 4.03. Found: C, 48.05; H, 3.95. Negative ion FAB-MS *m/z*: 991 $(M-H)^-$. $^1$H-NMR (acetone-$d_6$, 100 MHz) $\delta$: 2.19 (3H, s, $CH_3$), 2.99, 3.48 (each 1H, d, $J=16$ Hz, $CH_2$), 4.39 (1H, dd, $J=12$, 14 Hz, glc-6), 4.70—4.94 (2H in total, m, glc-5, 6), 4.92 [1H, d, $J=1$ Hz, acetonyl DHHDP (ADHHDP)-1], 5.43 (1H, br s, glc-4), 5.57 (2H, br s, glc-2, 3), 6.32 (1H, d, $J=1$ Hz, ADHHDP-3), 6.58 (1H, s, glc-1), 6.66, 7.08 [each 1H, s, hexahydroxydiphenoyl (HDDP)-H], 7.19 (2H, s, galloyl-H), 7.23 (1H, s, ADHHDP-3′). $^{13}$C-NMR (acetone-$d_6$, 25.05 MHz) $\delta$: 31.8 ($CH_3$), 49.8 ($CH_2$), 51.8 (ADHHDP-1), 62.2 (glc-3), 63.7 (glc-6), 66.5 (glc-4), 70.1 (glc-2), 73.0 (glc-5), 80.8 (ADHHDP-5), 91.7 (glc-1), 107.9, 110.3 (HHDP-3, 3′), 109.7 (ADHHDP-6), 110.7 (galloyl-2, 6), 113.3 (ADHHDP-3′), 115.1, 116.9, 117.8 (HHDP-1, 1′, ADHHDP-2′), 124.4, 125.4 (HHDP-2, 2′), 126.9 (ADHHDP-3), 136.5, 137.2, 137.8 (HHDP-5, 5′, ADHHDP-5′), 139.9 (galloyl-4), 144.4, 144.6, 144.9, 145.4 (HHDP-4, 4′, 6, 6′, ADHHDP-2), 145.9 (galloyl-3, 5), 146.9 (ADHHDP-6′), 147.5 (ADHHDP-4′), 164.6, 165.0, 165.3, 166.1, 168.4 (COO), 197.6 (ADHHDP-4), 206.2 (CO).

**Acetonylgranatin B (3c)** A white amorphous powder, $[\alpha]_D^{20}$ −137.2° ($c$=1.1, MeOH). *Anal.* Calcd for $C_{44}H_{32}O_{27}\cdot 2H_2O$: C, 51.37; H, 3.53. Found: C, 51.57; H, 3.30. Negative ion FAB-MS $m/z$: 991 $(M-H)^-$. $^1$H-NMR (acetone-$d_6$, 100 MHz) $\delta$: 2.19 (3H, s, $CH_3$), 2.99, 3.50 (each 1H, d, $J$=15 Hz, $CH_2$), 4.23 (1H, dd, $J$=8, 11 Hz, glc-6), 4.85 (1H, dd, $J$=8, 11 Hz, glc-5), 4.92 (1H, d, $J$=1 Hz, ADHHDP-1), 5.10 (1H, br s, glc-4), 5.31 (1H, t, $J$=11 Hz, glc-6), 5.55 (1H, br s, glc-2), 6.22 (1H, d, $J$=3 Hz, glc-3), 6.39 (1H, d, $J$=1 Hz, ADHHDP-3), 6.59 (1H, s, glc-1), 6.74, 6.95 (each 1H, s, HHDP-H), 7.15 (2H, s, galloyl-H), 7.23 (1H, s, ADHHDP-3′). $^{13}$C-NMR (acetone-$d_6$, 67.8 MHz) $\delta$: 32.1 ($CH_3$), 49.9 ($CH_2$), 51.7 (ADHHDP-1), 61.1, 63.7, 63.8, 69.6, 72.8 (glc-2, 3, 4, 5, 6), 81.2 (ADHHDP-5), 91.5 (glc-1), 107.7, 110.4 (HHDP-3, 3′), 109.4 (ADHHDP-6), 111.0 (galloyl-2, 6), 113.9 (ADHHDP-3′), 115.9, 117.1, 117.3 (ADHHDP-1′, HHDP-1, 1′), 119.5, 119.7 (ADHHDP-2′, galloyl-1), 124.4, 125.4 (HHDP-2, 2′), 128.1 (ADHHDP-3), 136.8, 137.0, 137.7 (ADHHDP-5′, HHDP-5, 5′), 139.9 (galloyl-4), 144.9, 145.1, 145.5, 145.9 (galloyl-3, 5, ADHHDP-2, HHDP-4, 4′, 6, 6′), 147.0 (ADHHDP-6′), 147.6 (ADHHDP-4′), 164.6, 165.3, 165.4, 169.2 (COO), 198.0 (ADHHDP-4), 206.8 (CO).

**Acetonylhelioscopinin A (4c)** A white powder ($H_2O$), mp 258 −260 °C (dec.), $[\alpha]_D^{18}$ +138.2° ($c$=0.7, MeOH). *Anal.* Calcd for $C_{44}H_{32}O_{27}\cdot 5/2H_2O$: C, 50.92; H, 3.59. Found: C, 50.68; H, 3.28. Negative ion FAB-MS $m/z$: 991 $(M-H)^-$. $^1$H-NMR (acetone-$d_6$, 100 MHz) $\delta$: 2.16 (3H, s, $CH_3$), 2.93, 3.48 (each 1H, d, $J$=15 Hz, $CH_2$), 4.24 (1H, dd, $J$=5, 12 Hz, glc-6), 4.79 (1H, dd, $J$=5, 12 Hz, glc-5), 4.95 (1H, d, $J$=1 Hz, ADHHDP-1), 5.08 (1H, br s, glc-4), 5.32 (1H, br s, glc-2), 5.40 (1H, t, $J$=12 Hz, glc-6), 6.00 (1H, br s, glc-3), 6.21 (1H, s, glc-1), 6.34 (1H, d, $J$=1 Hz, ADHHDP-3), 6.63, 6.93 (each 1H, s, HHDP-H), 7.12 (2H, s, galloyl-H), 7.24 (1H, s, ADHHDP-3′). $^{13}$C-NMR (acetone-$d_6$, 25.05 MHz) $\delta$: 31.9 ($CH_3$), 49.9 ($CH_2$), 51.7 (ADHHDP-1), 60.2, 63.9, 66.9, 68.1, 70.9 (glc-2, 3, 4, 5, 6), 80.9 (ADHHDP-5), 89.9 (glc-1), 109.4, 109.6 (2C) (HHDP-3, 3′, ADHHDP-6), 111.0 (galloyl-2, 6), 113.6 (ADHHDP-3′), 116.4 (2C), 117.5 (ADHHDP-1′, HHDP-1, 1′), 119.3, 120.0 (ADHHDP-2′, galloyl-1), 124.8, 125.4 (HHDP-2, 2′), 127.3 (ADHHDP-3), 137.1 (3C) (ADHHDP-5′, HHDP-5, 5′), 139.4 (galloyl-4), 144.5, 144.7, 144.9, 145.1, 145.7, 145.9 (ADHHDP-2, galloyl-3, 5, HHDP-4, 4′, 6, 6′), 147.0 (ADHHDP-6′), 147.5 (ADHHDP-4′), 164.6, 164.9, 165.0, 166.3, 169.5 (COO), 197.5 (ADHHDP-4), 206.2 (CO).

**Acetonylcarpinusin (5c)** A white powder ($H_2O$), mp 230—235° (dec.), $[\alpha]_D^{18}$ +38.6° ($c$=0.4, MeOH). *Anal.* Calcd for $C_{44}H_{32}O_{27}\cdot H_2O$: C, 52.29; H, 3.39. Found: C, 52.25; H, 3.07. Negative ion FAB-MS $m/z$: 991 $(M-H)^-$. $^1$H-NMR (acetone-$d_6$, 100 MHz) $\delta$: 2.20, (3H, s, $CH_3$), 3.06, 3.51 (each 1H, d, $J$=16 Hz, $CH_2$), 4.12 (1H, dd, $J$=5, 12 Hz, glc-6), 4.71 (1H, dd, $J$=5, 12 Hz, glc-5), 4.95 (1H, d, $J$=1 Hz, ADHHDP-1), 5.05 (1H, br d, $J$=3 Hz, glc-4), 5.46 (1H, br s, glc-2), 5.55 (1H, t, $J$=12 Hz, glc-6), 6.04 (1H, br s, glc-3), 6.27 (1H, s, glc-1), 6.34 (1H, d, $J$=1 Hz, ADHHDP-3), 6.84, 6.87 (each 1H, s, HHDP-H), 7.17 (2H, s, galloyl-H), 7.26 (1H, s, ADHHDP-3′). $^{13}$C-NMR (acetone-$d_6$, 25.05 MHz) $\delta$: 31.9 ($CH_3$), 50.1 ($CH_2$), 51.8 (ADHHDP-1), 60.2, 63.8, 68.0 (2C), 72.0 (glc-2, 3, 4, 5, 6), 80.9 (ADHHDP-5), 90.4 (glc-1), 109.0, 109.6, 109.8 (ADHHDP-6, HHDP-3, 3′), 111.0 (galloyl-2, 6), 113.7 (ADHHDP-3′), 116.2, 116.7, 117.2 (ADHHDP-1′, HHDP-1, 1′), 119.5, 120.0 (ADHHDP-2′, galloyl-1), 125.1, 125.5 (HHDP-2, 2′), 127.3 (ADHHDP-3), 137.0, 137.2, 137.3 (ADHHDP-5′, HHDP-5, 5′), 139.6 (galloyl-4), 144.7, 144.9, 145.2, 145.3, 145.7, 146.0 (ADHHDP-2, galloyl-3, 5, HHDP-4, 4′, 6, 6′), 147.0 (ADHHDP-6′), 147.5 (ADHHDP-4′), 164.6 (2C), 165.3, 165.9, 169.7 (COO), 197.7 (ADHHDP-4), 206.5 (CO).

**Acetonylfurosin (7c)** A white amorphous powder, $[\alpha]_D^{18}$ −151.7° ($c$=0.6, MeOH). *Anal.* Calcd for $C_{30}H_{26}O_{19}\cdot H_2O$: C, 50.85; H, 3.98. Found: C, 50.85; H, 4.05. Negative ion FAB-MS $m/z$: 689 $(M-H)^-$. $^1$H-NMR (acetone-$d_6$, 100 MHz) $\delta$: 2.19 (3H, s, $CH_3$), 2.97, 3.49 (each 1H, d, $J$=15 Hz, $CH_2$), 3.94—4.39 (3H in total, m, glc-5, 6, 6), 4.55 (1H, br s, glc-3), 4.94 (1H, d, $J$=1 Hz, ADHHDP-1), 5.11 (1H, br d, $J$=3 Hz, glc-4), 5.30 (1H, dd, $J$=2, 3 Hz, glc-2), 6.31 (1H, d, $J$=1 Hz, ADHHDP-3), 6.42 (1H, d, $J$=2 Hz, glc-1), 7.22 (3H, s, ADHHDP-3′, galloyl-H). $^{13}$C-NMR (acetone-$d_6$, 25.05 MHz) $\delta$: 31.9 ($CH_3$), 49.9 ($CH_2$), 51.8 (ADHHDP-1), 62.6, 62.9, 72.0, 73.1, 78.2 (glc-2, 3, 4, 5, 6), 80.9 (ADHHDP-5), 92.4 (glc-1), 109.8 (ADHHDP-6), 110.3 (galloyl-2, 6), 113.3 (ADHHDP-3′), 117.6 (ADHHDP-1′), 120.2, 120.7 (ADHHDP-2′, galloyl-1), 127.2 (ADHHDP-3), 137.0 (ADHHDP-5′), 139.5 (galloyl-4), 145.7 (ADHHDP-2), 146.1 (galloyl-3, 5), 147.0 (ADHHDP-6′), 147.4 (ADHHDP-4′), 165.0 (2C), 166.0 (COO), 197.6 (ADHHDP-4), 206.2 (CO).

**Acetonylterchebin (8c)** A white powder ($H_2O$), mp 234—236° (dec.), $[\alpha]_D^{18}$ −24.6° ($c$=0.4, MeOH). *Anal.* Calcd for $C_{44}H_{34}O_{27}\cdot 2H_2O$: C, 51.27; H, 3.72. Found: C, 51.00; H, 3.23. Negative ion FAB-MS $m/z$: 993 $(M-H)^-$. $^1$H-NMR (acetone-$d_6$, 100 MHz) $\delta$: 2.17 (3H, s, $CH_3$), 3.01, 3.50 (each 1H, d, $J$=16 Hz, $CH_2$), 4.68 −4.92 (3H in total, m, glc-5, 6, 6), 4.97 (1H, d, $J$=1 Hz, ADHHDP-1), 5.32 (1H, br d, $J$=3 Hz, glc-4), 5.55 (1H, br s, glc-2), 6.02 (1H, br s, glc-3), 6.34 (1H, d, $J$=1 Hz, ADHHDP-3), 6.59 (1H, s, glc-1), 7.05, 7.26, 7.31 (each 2H, s, galloyl-H), 7.28 (1H, s, ADHHDP-3′). $^{13}$C-NMR (acetone-$d_6$, 25.05 MHz) $\delta$: 32.0 ($CH_3$), 49.8 ($CH_2$), 52.0 (ADHHDP-1), 62.4, 64.6, 69.1, 70.4, 74.7 (glc-2, 3, 4, 5, 6), 80.9 (ADHHDP-5), 92.2 (glc-1), 109.9 (ADHHDP-6), 110.1, 110.6 (galloyl-2, 6), 113.5 (ADHHDP-3′), 117.1 (ADHHDP-1′), 119.8, 120.2, 120.9 (ADHHDP-2′, galloyl-1), 127.4 (ADHHDP-3), 137.3 (ADHHDP-5′), 139.1, 139.9, 140.0 (galloyl-4), 145.3 (ADHHDP-2), 145.9, 146.0, 146.1 (galloyl-3, 5), 147.0 (ADHHDP-6′), 147.5 (ADHHDP-4′), 164.3, 164.9, 165.1, 165.7, 166.5 (COO), 197.5 (ADHHDP-4), 206.2 (CO).

**Acetonylmallotusinic Acid (9c)** A white amorphous powder, $[\alpha]_D^{18}$ −49.5° ($c$=0.7, MeOH). *Anal.* Calcd for $C_{51}H_{36}O_{32}\cdot H_2O$: C, 51.18; H, 3.37. Found: C, 50.87; H, 3.38. Negative ion FAB-MS $m/z$: 1159 $(M-H)^-$. $^1$H-NMR (acetone-$d_6$+$D_2O$, 100 MHz) $\delta$: 2.21 (3H, s, $CH_3$), 2.99, 3.48 (each 1H, d, $J$=15 Hz, $CH_2$), 4.34 (1H, dd, $J$=12, 14 Hz, glc-6), 4.64—4.82 (2H in total, m, glc-5, 6), 4.90 (1H, d, $J$=1 Hz, ADHHDP-1), 5.46 (3H, br s, glc-2, 3, 4), 6.29 (1H, d, $J$=1 Hz, ADHHDP-3), 6.54 (1H, s, glc-1), 6.65, 7.01, 7.15, 7.20 (each 1H, s, valoneoyl-H, ADHHDP-3′), 7.23 (2H, s, galloyl-H). $^{13}$C-NMR (acetone-$d_6$+$D_2O$, 25.05 MHz) $\delta$: 32.2 ($CH_3$), 49.6 ($CH_2$), 51.9 (ADHHDP-1), 63.1 (glc-3), 63.9 (glc-6), 66.9 (glc-4), 71.0 (glc-2), 73.5 (glc-5), 80.8 (ADHHDP-5), 91.6 (glc-1), 107.9, 109.5, 111.0 (valoneoyl-3, 3′, 6″), 109.7 (ADHHDP-6), 111.1 (galloyl-2, 6), 113.4 (ADHHDP-3′), 115.5, 116.8, 117.6, 119.1 (ADHHDP-1′, valoneoyl-1, 1′, 1″), 119.9 (galloyl-1, ADHHDP-2′), 123.8, 124.7 (valoneoyl-2, 2′), 126.9 ADHHDP-3), 136.4, 137.4, 138.3, 138.8 (ADHHDP-5′, galloyl-5, valoneoyl-5, 5′, 3″), 140.2, 142.8 (valoneoyl-2″, 5″), 145.0, 145.4, 146.0, 146.1 (galloyl-3, 5, valoneoyl-4, 6, 6′, ADHHDP-2), 146.8, 147.6 (ADHHDP-4′, 6′, valoneoyl-4′), 164.7, 165.1, 165.4, 166.4, 168.6, 170.1 (COO), 197.7 (ADHHDP-4), 206.5 (CO).

**Acetonylgranatin A (10c)** A white amorphous powder, $[\alpha]_D^{18}$ +23.9° ($c$=0.7, MeOH). *Anal.* Calcd for $C_{37}H_{28}O_{23}\cdot 2H_2O$: C, 50.69; H, 3.68. Found: C, 50.54; H, 3.47. Negative ion FAB-MS $m/z$: 839 $(M-H)^-$. $^1$H-NMR (acetone-$d_6$, 100 MHz) $\delta$: 2.15 (3H, s, $CH_3$), 2.90, 3.46 (each 1H, d, $J$=15 Hz, $CH_2$), 4.12 (1H, dd, $J$=5, 11 Hz, glc-6), 4.59 (1H, br s, glc-3), 4.62 (1H, dd, $J$=5, 12 Hz, glc-5), 4.92 (1H, d, $J$=1 Hz, ADHHDP-1), 4.99 (1H, br s, glc-4), 5.09 (1H, dd, $J$=11, 12 Hz, glc-6), 5.18 (1H, br s, glc-2), 6.11 (1H, s, glc-1), 6.30 (1H, d, $J$=1 Hz, ADHHDP-3), 6.84 (2H, s, HHDP-H), 7.20 (1H, s, ADHHDP-3′). $^{13}$C-NMR (acetone-$d_6$, 67.8 MHz) $\delta$: 32.0 ($CH_3$), 50.0 ($CH_2$), 51.6 (ADHHDP-1), 61.0, 64.2, 69.3, 70.8, 71.0 (glc-2, 3, 4, 5, 6), 81.1 (ADHHDP-5), 90.0 (glc-1), 108.7, 109.5, 109.6 (HHDP-3, 3′, ADHHDP-6), 113.5 (ADHHDP-3′), 116.0, 117.2 (HHDP-1, 1′), 117.8 (ADHHDP-1′), 119.7 (ADHHDP-2′), 125.4, 125.7 (HHDP-2, 2′), 127.4 (ADHHDP-3), 136.4, 136.9, 137.0 (ADHHDP-5′, HHDP-5, 5′), 144.6, 144.8, 145.1, 145. 5 (ADHHDP-2, HHDP-4, 4′, 6, 6′), 147.0 (ADHHDP-6′), 147.6 (ADHHDP-4′), 165.3, 165.9, 166.6, 168.8 (COO), 198.0 (ADHHDP-4), 206.7 (CO).

**Acetonyleuphorscopin (11c)** A white amorphous powder, $[\alpha]_D^{18}$ +106.3° ($c$=0.4, MeOH). *Anal.* Calcd for $C_{44}H_{32}O_{27}\cdot 3/2H_2O$: C, 51.82; H, 3.46. Found: C, 51.72; H, 3.64. Negative ion FAB-MS $m/z$: 991 $(M-H)^-$. $^1$H-NMR (acetone-$d_6$, 100 MHz) $\delta$: 2.15 (3H, s, $CH_3$), 2.95, 3.44 (each 1H, d, $J$=15 Hz, $CH_2$), 3.91 (1H, dd, $J$=7, 11 Hz, glc-6), 4.84 (1H, ddd, $J$=7, 7, 7 Hz, glc-5), 4.88 (1H, d, $J$=1 Hz, ADHHDP-1), 5.03 (1H, dd, $J$=7, 11 Hz, glc-6), 5.24 (1H, br d, $J$=4 Hz, glc-3), 5.44 (1H, d, $J$=7 Hz, glc-4), 5.98 (1H, dd, $J$=1, 4 Hz, glc-2), 6.34 (1H, d, $J$=1 Hz, ADHHDP-3), 6.37, 6.79 (each 1H, s, HHDP-H), 6.57 (1H, d, $J$=1 Hz, glc-1), 7.23 (2H, s, galloyl-H), 7.24 (1H, s, ADHHDP-3′). $^{13}$C-NMR (acetone-$d_6$, 25.05 MHz) $\delta$: 32.2 ($CH_3$), 49.7 ($CH_2$), 51.9 (ADHHDP-1), 64.5 (2C), 69.4 (2C), 72.0 (glc-2, 3, 4, 5, 6), 80.9 (ADHHDP-5), 96.4 (glc-1), 108.5, 110.2 (2C) (ADHHDP-6, HHDP-3, 3′), 110.7 (galloyl-2, 6), 112.7 (ADHHDP-3′), 114.5, 115.8 (HHDP-1, 1′), 117.1 (ADHHDP-1′), 119.6 (ADHHDP-2′), 120.7 (galloyl-1), 125.1, 125.7 (HHDP-2, 2′), 127.2 (ADHHDP-3), 136.5, 137.1, 137.4 (HHDP-5, 5′, ADHHDP-5′), 139.9 (galloyl-4), 144.0, 144.4, 144.9, 145.1, 145.4 (ADHHDP-2, galloyl-3, 5, HHDP-4, 4′, 6, 6′), 147.0 (ADHHDP-6′), 147.5 (ADHHDP-4′), 163.6, 165.0, 166.1, 167.4, 168.3 (COO), 197.4 (ADHHDP-4), 206.0 (CO).

**Reaction of 1 with Acetophenone** A mixture of **1** (200 mg), acetophenone (1.0 ml) and ammonium formate (100 mg) in methanol (3 ml) was heated at 50 °C with stirring for 4 h. The reaction mixture was directly subjected to Sephadex LH-20 chromatography (2.0 cm i.d. × 25 cm) with 80% aqueous methanol to yield **1e** (59.3 mg, 27%). **1e**: A white amorphous powder, $[\alpha]_D^{18}$ −106.9° ($c$=1.0, MeOH). *Anal.* Calcd for $C_{49}H_{34}O_{27}\cdot 2H_2O$: C, 53.95; H, 3.51. Found: C, 53.75; H, 3.42. Negative ion FAB-MS

*m/z*: 1053 $(M-H)^-$. $^1$H-NMR (acetone-$d_6$, 100 MHz) $\delta$: 3.32, 4.30 (each 1H, d, $J$=15 Hz, $CH_2$), 4.37 (1H, dd, $J$=12, 14 Hz, glc-6), 4.67—4.89 (2H, m, glc-5, 6), 5.05 [1H, d, $J$=1 Hz, 2, 4-acetyl group (acyl)-1], 5.42 (1H, br s, glc-4), 5.58 (2H, br s, glc-2, 3), 6.42 (1H, d, $J$=1 Hz, acyl-3), 6.59 (1H, s, glc-1), 6.63, 7.10 (each 1H, s, HHDP-H), 7.20 (2H, s, galloyl-H), 7.25 (1H, s, acyl-3′), 7.45—7.75 (3H, m, phenyl-3, 4, 5), 8.04 (2H, dd, $J$=2, 8 Hz, phenyl-2, 6). $^{13}$C-NMR (acetone-$d_6$, 25.05 MHz) $\delta$: 45.2 ($CH_2$), 52.0 (acyl-1), 62.3 (glc-3), 63.8 (glc-6), 66.5 (glc-4), 70.2 (glc-2), 73.1 (glc-5), 81.0 (actyl-5), 91.9 (glc-1), 108.1, 109.7, 110.2 (acyl-6, HHDP-3, 3′), 110.7 (galloyl-2, 6), 113.3 (acyl-3′), 115.1, 117.1 (2C) (HHDP-1, 1′, acyl-1′), 120.3 (galloyl-2, acyl-2′), 125.4, 125.6 (HHDP-2, 2′), 127.2 (acyl-3), 129.4 (phenyl-2, 3, 5, 6), 134.3 (phenyl-4), 136.3, 137.1, 137.6 (acyl-5, HHDP-5, 5′), 138.1 (phenyl-1), 139.6 (galloyl-4), 144.6, 144.9, 145.1, 145.3 (HHDP-4, 4′, 6, 6′, acyl-2), 145.8 (galloyl-3, 5), 147.0 (acyl-6′), 147.4 (acyl-4′), 164.6, 164.9, 165.4, 166.1, 168.3 (COO), 197.6 (2C) (acyl-4, CO).

**Reaction of 1 with 2-Pentanone** A mixture of **1** (500 mg), ammonium formate (200 mg) and 2-phentanone (5 ml) in 15% aqueous EtOH (6 ml) was heated at 50 °C with stirring for 4 h. The reaction mixture was concentrated *in vacuo* and subjected to Sephadex LH-20 chromatography (2.0 cm i.d. × 25 cm) with 80% aqueous methanol to yield **1f** (130 mg, 24%). **1f**: A white powder ($H_2O$–MeOH), mp 218—220 °C (dec.), $[\alpha]_D^{18}$ −107.5° ($c$=0.4, MeOH). *Anal.* Calcd for $C_{46}H_{36}O_{27}\cdot 3H_2O$: C, 51.40; H, 3.94. Found: C, 51.53; H, 3.66. Negative ion FAB-MS *m/z*: 1019 $(M-H)^-$. $^1$H-NMR (acetone-$d_6$, 100 MHz) $\delta$: 0.88 (3H, t, $J$=7 Hz, $CH_3$), 1.52 (2H, sextet, $J$=7 Hz, $CH_2$), 2.53 (2H, t, $J$=7 Hz, $CH_2$), 2.94, 3.46 (each 1H, d, $J$=15 Hz, $CH_2$), 4.38 (1H, dd, $J$=12, 14 Hz, glc-6), 4.68—4.83 (2H, m, glc-5, 6), 4.91 (1H, d, $J$=1 Hz, acyl-1), 5.44 (1H, br s, glc-4), 5.56 (2H, br s, glc-2, 3), 6.32 (1H, d, $J$=1 Hz, acyl-3), 6.57 (1H, s, glc-1), 6.65, 7.09 (each 1H, s, HHDP-H), 7.19 (2H, s, galloyl-H), 7.23 (1H, s, H-3′).

**Treatment of 1c with Phosphate Buffer** A suspension of **1c** (200 mg) in 0.2 M phosphate buffer (pH 7.0, 50 ml) was heated at 80 °C with stirring for 2 h. After cooling, the mixture was acidified with 2 N HCl, and subjected to Cosmosil 75$C_{18}$-OPN chromatography (3.0 cm i.d. × 20 cm) with water containing increasing proportions of methanol to afford **2** (40.5 mg, 30%) and **14** (63.7 mg, 32%). **14**: A tan amorphous powder. $[\alpha]_D^{18}$ −80.8° ($c$=1.1, MeOH). *Anal.* Calcd for $C_{41}H_{28}O_{27}\cdot H_2O$: C, 50.73; H, 3.11. Found: C, 50.82; H, 3.33. Negative ion FAB-MS *m/z*: 951 $(M-H)^-$. IR $\nu_{max}^{KBr}$ cm$^{-1}$: 1800, 1710, 1705, 1600, 1420. $^1$H-NMR (acetone-$d_6$+$D_2O$, 270 MHz) $\delta$: 2.37 [1H, dd, $J$=2, 19 Hz, 2, 4-acyl group (acyl)-4], 2.90 (1H, dd, $J$=11, 19 Hz, acyl-4), 3.76 (1H, ddd, $J$=2, 3, 11 Hz, acyl-3), 4.46 (1H, dd, $J$=8, 11 Hz, glc-6), 4.78 (1H, $J$=11 Hz, glc-6), 4.95 (1H, dd, $J$=8, 11 Hz, glc-5), 5.02 (1H, d, $J$=3 Hz, acyl-2), 5.32 (1H, br d, $J$=4 Hz, glc-4), 5.43 (1H, br s, glc-2), 5.97 (1H, br s, glc-3), 6.44 (1H, s, glc-1), 6.69, 7.03 (each 1H, s, HHDP-H), 7.15 (2H, s, galloyl-H), 7.33 (1H, s, acyl-3′). $^{13}$C-NMR (acetone-$d_6$+$D_2O$, 25.05 MHz), $\delta$: 35.2 (acyl-4), 43.9 (acyl-3), 47.7 (acyl-2), 61.2 (glc-3), 64.0 (glc-6), 67.1 (glc-4), 70.1 (glc-2), 73.4 (glc-5), 92.0 (glc-1), 107.7, 110.4 (HHDP-3, 3′), 110.6 (2C) (galloyl-2, 6), 114.5 (acyl-3′), 115.3, 117.2 (2C), 117.4 (HHDP-1, 1′, acyl-1′, 2′), 119.7 (galloyl-1), 124.1, 125.3 (HHDP-2, 2′), 136.3 (2C), 137.8, 140.1 (HHDP-5, 5′, galloyl-4, acyl-5′), 143.1, 144.7, 144.9, 145.2, 145.5 (HHDP-4, 4′, 6, 6′, acyl-4′), 145.9 (2C) (galloyl-3, 5), 147.9 (acyl-6′), 163.4 (acyl-6), 164.5, 165.5, 166.6, 169.0, 171.3 (COO), 176.2 (acyl-1), 195.5 (acyl-5).

**Reaction of 1c with L-Cysteine Methyl Ester (15)** 1) At Room Temperature: A mixture of **1c** (300 mg), L-cysteine methyl ester hydrochloride (**15**) (300 mg) and ammonium formate (300 mg) in water–acetonitrile (5 : 2, 7 ml) was stirred at room temperature for 1 h. The reaction mixture was directly subjected to Sephadex LH-20 chromatography (2.0 cm i.d. × 25 cm) with 80% aqueous methanol to afford **18** (213 mg, 63%) as a yellow powder ($H_2O$), mp 214—217 °C (dec), $[\alpha]_D^{18}$ −65.9° ($c$=0.7, MeOH). *Anal.* Calcd for $C_{48}H_{35}NO_{26}S\cdot 2H_2O$: C, 51.94; H, 3.54; N, 1.26. Found: C, 51.51; H, 3.09; N, 1.43. Negative ion FAB-MS *m/z*: 1072 $(M-H)^-$. $^1$H-NMR (acetone-$d_6$, 270 MHz) $\delta$: 2.46 (3H, d, $J$=1 Hz, acyl-10), 3.35 (1H, dd, $J$=4, 14 Hz, acyl-13), 3.69 (1H, dd, $J$=3, 14 Hz, acyl-13), 3.88 (3H, s, $CH_3$), 4.01 (1H, dd, $J$=4, 12 Hz, glc-6), 4.75 (1H, dd, $J$=8, 12 Hz, glc-6), 4.85 (1H, dd, $J$=4, 8 Hz, glc-5), 5.35, (2H, s, glc-3, 4), 5.56 (1H, d, $J$=6 Hz, glc-2), 5.65 (1H, dd, $J$=3, 4 Hz, acyl-12), 6.07 (1H, d, $J$=6 Hz, glc-1), 6.38 (1H, d, $J$=1 Hz, H-8), 6.66, 6.94 (each 1H, s, HHDP-H), 7.09 (2H, s, galloyl-H), 7.35 (1H, s, acyl-3′). $^{13}$C-NMR (acetone-$d_6$, 25.05 MHz) $\delta$: 12.1 (acyl-10), 30.4 (acyl-13), 53.0 ($CH_3$), 55.0 (acyl-12), 65.3 (glc-6), 68.0 (2C), 76.4 (2C), (glc-2, 3, 4, 5), 92.1 (glc-1), 98.9 (acyl-8), 108.0, 108.3, 109.6, 110.2 (2C), 110.7, 113.1, 113.5, 114.8, 116.4, 117.9, 120.0, 120.3, 121.2, 124.6, 125.5 (acyl-1, 2, 3, 5, 1′, 2′, 3′, HHDP-1, 1′, 2, 2′, 3, 3′, galloyl-1, 2, 6), 131.8 (acyl-9), 136.2, 137.2, 138.8, 138.9, 139.5 (acyl-4, 5′, HHDP-5, 5′, galloyl-4), 144.6, 144.9, 145.0, 145.2, 146.0, 146.1 (acyl-6, 4′, 6′, HHDP-4, 4′, 6, 6′, galloyl-3, 5), 164.7, 166.1, 166.3, 168.3, 168.6, 170.2 (COO).

2) At 80 °C: A mixture of **1c** (300 mg), **15** (200 mg) and sodium acetate (200 mg) in acetonitrile–water (3 : 2, 25 ml) was stirred at 80 °C for 3 h, then allowed to cool. The resulting yellow needles of **19** (63.8 mg, 46%) were collected by filtration. The filtrate was concentrated and subjected to MCI-gel CHP-20P chromatography (3.0 cm i.d. × 25 cm) with water containing increasing proportions of methanol to yield **2** (109.6 mg, 55%) and **20** (127 mg, 38%). **19**: Yellow needles, mp 283 °C (dec). $[\alpha]_D^{23}$ −190.0° ($c$=0.4, pyridine). *Anal.* Calcd for $C_{21}H_{13}NO_8S$: C, 57.40; H, 2.98; N, 3.19. Found: C, 56.91; H, 3.03; N, 3.15. Negative ion FAB-MS *m/z*: 438 $(M-H)^-$. IR $\nu_{max}^{KBr}$ cm$^{-1}$: 3450, 1738, 1720, 1617, 1538. $^1$H-NMR (acetone-$d_6$+$D_2O$, 100 MHz) $\delta$: 2.57 (3H, d, $J$=1 Hz, H-10), 3.40—3.70 (H-13, overlapped with HOD signal), 3.76 (3H, s, $CH_3$), 5.92 (1H, t, $J$=3 Hz, H-12), 6.72 (1H, d, $J$=1 Hz, H-8), 7.61 (1H, s, H-3′). $^{13}$C-NMR (DMSO-$d_6$, 25.05 MHz) $\delta$: 11.9 (C-10), 28.2 (C-13), 53.0 (2C), (C-12, $CH_3$), 97.4 (C-8), 104.5, 107.8, 109.1, 109.8, 112.9, 119.3, 120.2 (C-1, 2, 3, 5, 1′, 2′, 3′), 130.3 (C-9), 135.6 (C-5′), 137.0 (C-4), 139.1 (C-6′), 143.0 (C-6), 147.7 (C-4′), 159.0, 159.1 (COO), 168.6 (C-11). **20**: A yellow amorphous powder. $[\alpha]_D^{27}$ −95.4° ($c$=0.7, MeOH). *Anal.* Calcd for $C_{48}H_{35}NO_{26}S\cdot 2H_2O$: C, 51.94; H, 3.54; N, 1.26. Found: C, 51.98; H, 3.27; N, 1.08. Negative ion FAB-MS *m/z*: 1072 $(M-H)^-$. $^1$H-NMR (acetone-$d_6$, 100 MHz) $\delta$: 2.31 (3H, d, $J$=1 Hz, acyl-10), 3.08 (1H, dd, $J$=4, 14 Hz, acyl-13), 3.17 (1H, dd, $J$=3, 14 Hz, acyl-13), 3.75 (3H, s, $CH_3$), 4,16 (1H, br s, glc-4), 4.38 (1H, dd, $J$=8, 11 Hz, glc-6), 4.52 (1H, dd, $J$=8, 11 Hz, glc-6), 4.72 (1H, t, $J$=8 Hz, glc-5), 5.13 (1H, dd, $J$=3, 4 Hz, acyl-12), 5.17 (1H, br s, glc-2), 5.74 (1H, d, $J$=4 Hz, glc-3), 6.34 (2H, d, $J$=1 Hz, glc-1, acyl-8), 6.58, 6.75, 6.84 (each 1H, s, acyl-3′, HHDP-H), 7.14 (2H, s, galloyl-H). $^{13}$C-NMR (acetone-$d_6$, 25.05 MHz) $\delta$: 12.1 (acyl-10), 26.7 (acyl-13), 53.4 ($CH_3$), 54.6 (acyl-12), 64.5 (glc-6), 65.5, 68.7, 69.8, 75.3 (glc-2, 3, 4, 5), 91.7 (glc-1), 99.1 (acyl-8), 104.4, 105.5, 107.7, 109.8, 110.5 (2C), 113.7, 115.2, 116.4, 119.0, 120.1, 124.2 (2C), 125.7, 125.8 (acyl-1, 2, 3, 5, 1′, 2′, 3′, HHDP-1, 2, 3, 1′, 2′, 3′, galloyl-1, 2, 6). 132.9 (acyl-9), 135.5, 136.4, 137.4, 138.4 (acyl-4, 5′, HHDP-5, 5′), 139.8 (galloyl-4), 144.7, 145.1, 145.3, 145.7 (2C), 145.9, 146.1, 146.5 (acyl-5′, 6, HHDP-4, 6, 4′, 6′, galloyl-3, 5), 148.7 (acyl-6′), 165.1, 166.4, 167.2, 168.4, 168.7, 170.3 (COO).

**Partial Hydrolysis of 20** A solution of **20** (50 mg) in water–acetonitrile (2 : 1, 5 ml) was heated at 80 °C for 3 h. After cooling, the resulting yellow needles of **19** (12 mg, 60%) were collected by filtration. The filtrate was subjected to MCI-gel CHP-20P chromatography ($H_2O$–MeOH) to afford **2** (18 mg, 61%).

**Reaction of 4c with 15** 1) At Room Temperature: A mixture of **4c** (1.0 g), **15** (1.0 g) and ammonium formate (1.0 g) in water–acetonitrile (5 : 2, 30 ml) was stirred at room temperature for 1 h. The reaction mixture was directly applied to a column of Sephadex LH-20 (3.0 cm i.d. × 30 cm) with 80% aqueous methanol to give **24** (810 mg, 75%) as a yellow powder ($H_2O$), mp 231—233 °C (dec), $[\alpha]_D^{18}$ +63.4° ($c$=0.7, MeOH). *Anal.* Calcd for $C_{48}H_{35}NO_{26}S$: C, 53.69; H, 3.28; N, 1.30. Found: C, 53.90; H, 3.08; N, 1.39. Negative ion FAB-MS *m/z*: 1072 $(M-H)^-$. $^1$H-NMR (acetone-$d_6$, 100 MHz) $\delta$: 2.41 (3H, d, $J$=1 Hz, acyl-10), 3.31 (1H, dd, $J$=4, 14 Hz, acyl-13), 3.65 (1H, dd, $J$=3, 14 Hz, acyl-13), 3.76 (3H, s, $CH_3$), 4.09 (1H, dd, $J$=5, 12 Hz, glc-6), 4.68 (1H, dd, $J$=5, 12 Hz, glc-5), 4.84 (1H, br s, glc-4), 5.16 (1H, br s, glc-2), 5.35 (1H, br s, glc-3), 5.43 (1H, t, $J$=12 Hz, glc-6), 5.65 (1H, dd, $J$=3, 4 Hz, acyl-12), 5.99 (1H, s, glc-1), 6.41 (1H, d, $J$=1 Hz, acyl-8), 6.73, 6.82, 6.83 (each 1H, s, acyl-3′, HHDP-H), 7.07 (2H, s, galloyl-H).

2) At 80 °C: A mixture of **4c** (300 mg), **15** (200 mg) and sodium acetate (200 mg) in acetonitrile–water (3 : 2, 25 ml) was heated at 80 °C for 1 h. The reaction mixture was worked up in the same way as described for **1c** to give **19** (55.3 mg, 42%), **6** (66.9 mg, 35%) and **24** (139 mg, 43%).

**Methylation of 1c** A mixture of **1c** (480 mg), dimethyl sulfate (2 ml) and anhydrous potassium carbonate (2 g) in dry acetone (30 ml) was stirred under reflux for 2 h. The inorganic salts were removed by filtration, and the filtrate, after concentration, was applied to a silica gel column. Elution with benzene containing increasing proportions of acetone yielded the dodecamethylate (177 mg, 30%) as a white amorphous powder, $[\alpha]_D^{18}$ −103.7° ($c$=0.8, $CHCl_3$). *Anal.* Calcd for $C_{56}H_{56}O_{27}$: C, 57.93; H, 4.86. Found: C, 57.46; H, 5.10. FD-MS *m/z*: 1160 ($M^+$). $^1$H-NMR ($CDCl_3$, 270 MHz) $\delta$: 2.27 (3H, s, $CH_3$), 2.82, 3.52 (each 1H, d, $J$=16 Hz, $CH_2$), 3.34, 3.62, 3.69 (6H), 3.70, 3.71, 3.89, 3.90, 3.95, 3.98, 3.99, 4.03 (each 3H, s, $CH_3$), 4.43 (1H, dd, $J$=8, 11 Hz, glc-6), 4.85 (1H, d, $J$=1 Hz, acyl-1), 4.86 (1H, dd, $J$=8, 11 Hz, glc-5), 5.14 (1H, t, $J$=11 Hz, glc-6), 5.56 (1H, dd, $J$=2, 3 Hz, glc-4), 5.61 (1H, br s, glc-2), 5.63 (1H, dd, $J$=1, 2 Hz, glc-3), 6.44 (1H, d, $J$=1 Hz, acyl-3), 6.69 (2H, s, glc-1, aromatic-H), 6.73 (1H, s, aromatic-H), 7.25 (2H, s, trimethoxybenzoyl-H), 7.28 (1H, s, acyl-3′).

**Alkaline Methanolysis** A solution of the dodecamethyl ether of **1c** (100 mg) in 1% methanolic sodium methoxide (5 ml) was left standing for 20 h at room temperature. The reaction mixture was treated with Amberlite IR-120B ($H^+$ form), and separated by silica gel chromatography with benzene–acetone (19 : 1) to give **21** (10 mg) and **22** (28 mg), $[[\alpha]_D^{27}$ +23.3° ($c$=0.5, $CHCl_3$)].

**Methylation of 18, Followed by Alkaline Hydrolysis** A solution of **18** (60 mg) in methanol (3 ml) was treated with ethereal diazomethane at 5 °C for 14 h. The mixture was concentrated to dryness, and the residue was subjected to silica gel chromatography with benzene–acetone (9 : 1) to afford the crude methyl ether (35.9 mg). Without further purification, the product (24 mg) was hydrolyzed in 5% aqueous sodium hydroxide (1 ml) and methanol (3 ml) under reflux for 30 min. The reaction mixture was concentrated *in vacuo*, acidified with 1 N HCl (5 ml), and extracted with ether. The organic layer was washed with water, dried ($Na_2SO_4$), and concentrated. The residue was treated with ethereal diazomethane for 1 h. After concentration, the products were separated by silica gel chromatography with benzene containing increasing proportions of acetone to give **21** (2 mg), **22** (7 mg) and **23** (6.4 mg) as colorless needles (MeOH), mp 190—191 °C, $[\alpha]_D^{25}$ −117.6° ($c$=0.2, $CHCl_3$). *Anal.* Calcd for $C_{27}H_{29}NO_{10}S$: C, 57.95; H, 5.22; N, 2.50. Found: C, 57.96; H, 5.26; N, 2.51. EI-MS $m/z$: 559 ($M^+$). $^1$H-NMR ($CDCl_3$, 100 MHz) $\delta$: 2.41 (3H, d, $J$=1 Hz, H-10), 3.29, 3.60 (each 1H, dd, $J$=4, 13 Hz, H-13), 3.44, 3.52, 3.68, 3.76, 3.81, 3.94, 3.97 (each 3H, s, $CH_3$), 5.28 (1H, t, $J$=4 Hz, H-12), 6.44 (1H, d, $J$=1 Hz, H-8), 7.25 (1H, s, H-3′).

**Methylation of 24, Followed by Alkaline Hydrolysis** A solution of **24** (500 mg) in methanol (20 ml) was treated with ethereal diazomethane for 20 h. The mixture was concentrated to dryness and passed through a silica gel column to afford the crude methyl ether (226 mg). Without further purification, the product (172 mg) was treated with 5% aqueous sodium hydroxide (2 ml) and methanol (8 ml) under reflux for 1 h. The reaction mixture was worked up in the same way as described above to afford the carboxylic acids, which were treated with ethereal diazomethane. Purification of the products by silica gel chromatography afforded **21** (27.9 mg), **25** (48.8 mg), $[[\alpha]_D^{25}$ −25.4° ($c$=0.5, $CHCl_3$)], and **26** (54.7 mg) as colorless granules (MeOH), mp 132—134 °C, $[\alpha]_D^{23}$ −180.4° ($c$=0.3, $CHCl_3$). *Anal.* Calcd for $C_{27}H_{29}NO_{10}S$: C, 57.95; H, 5.22; N, 2.50. Found: C, 57.73; H, 5.30; N, 2.49. EI-MS $m/z$: 559 ($M^+$). $^1$H-NMR ($CDCl_3$, 100 MHz) $\delta$: 2.39 (3H, d, $J$=1 Hz, H-10), 3.32, 3.65 (each 1H, dd, $J$=4, 13 Hz, H-13), 3.49, 3.54, 3.60, 3.74, 3.82, 3.90, 3.95 (each 3H, s, $CH_3$), 5.27 (1H, t, $J$=4 Hz, H-12), 6.42 (1H, d, $J$=1 Hz, H-8), 7.26 (H-3′, overlapped with solvent signal).

**Acknowledgement** The authors are grateful to Mr. Y. Tanaka, Miss Y. Soeda and Dr. R. Isobe for $^1$H- and $^{13}$C-NMR and MS measurements, and to the staff of the Central Analysis Room of this university for elemental analyses.

**References and Notes**

1) Part CXVII: T. Tanaka, H.-H. Tong, Y.-M. Xu, K. Ishimaru, G. Nonaka and I. Nishioka, *Chem. Pharm. Bull.*, **40**, 2975 (1992).
2) T. Okuda, T. Yoshida and T. Hatano, *J. Chem. Soc., Perkin Trans. 1*, **1982**, 9.
3) *a*) R. Saijo, G. Nonaka and I. Nishioka, *Chem. Pharm. Bull.*, **37**, 2063 (1989); *b*) *Idem*, *ibid.*, **37**, 2624 (1989); *c*) R. Saijo, G. Nonaka, I. Nishioka, I.-S. Chen and T.-H. Hwang, *ibid.*, **37**, 2940 (1989); *d*) G. Nonaka, M. Hayashi, T. Tanaka, R. Saijo and I. Nishioka, *ibid.*, **38**, 861 (1990); *e*) J.-H. Lin, G. Nonaka and I. Nishioka, *ibid.*, **38**, 1218 (1990); *f*) S.-H. Lee, T. Tanaka, G. Nonaka and I. Nishioka, *ibid.*, **38**, 1518 (1990); *g*) J.-H. Lin, M. Ishimatsu, T. Tanaka, G. Nonaka and I. Nishioka, *ibid.*, **38**, 1844 (1990); *h*) J.-H. Lin, T. Tanaka, G. Nonaka, I. Nishioka and I.-S. Chen, *ibid.*, **38**, 2162 (1990); *i*) S.-H. Lee, T. Tanaka, G. Nonaka and I. Nishioka, *ibid.*, **39**, 630 (1991).
4) G. Nonaka, M. Ishimatsu, M. Ageta and I. Nishioka, *Chem. Pharm. Bull.*, **37**, 50 (1989).
5) T. Tanaka, G. Nonaka, I. Nishioka, K. Miyahara and T. Kawasaki, *J. Chem. Soc., Perkin Trans. 1*, **1986**, 369.
6) T. Okuda, T. Yoshida, K. Mori and T. Hatano, *Heterocycles*, **15**, 1323 (1981); T. Tanaka, G. Nonaka and I. Nishioka, *Phytochemistry*, **24**, 2075 (1985); T. Tanaka, G. Nonaka and I. Nishioka, *Chem. Pharm. Bull.*, **38**, 2424 (1990).
7) J. C. Jochims, G. Taigel and O. T. Schmidt, *Justus Liebigs Ann. Chem.*, **717**, 169 (1968); T. Okuda, T. Hatano, H. Nitta and R. Fujii, *Tetrahedron Lett.*, **21**, 4361 (1980).
8) O. T. Schmidt, R. Schanz, R. Eckert and R. Wurmb, *Justus Liebigs Ann. Chem.*, **706**, 131 (1967).
9) *a*) T. Tanaka, G. Nonaka and I. Nishioka, Abstracts of Papers, Meeting of the Kyushu Branch, Pharmaceutical Society of Japan, Fukuoka, December 1990, p. 47; *b*) K. Yazaki, T. Yoshida and T. Okuda, *Phytochemistry*, **30**, 501 (1991); *c*) L. Y. Foo and H. Wong, *ibid.*, **31**, 711 (1992).
10) Compounds **1c**, **4c** and **5c** were isolated from *Euphorbia helioscopia* and *E. adenochlora*, S.-H. Lee, T. Tanaka, G. Nonaka and I. Nishioka, unpublished data.
11) T. Tanaka, H. Fujisaki, G. Nonaka and I. Nishioka, *Heterocycles*, **33**, 375 (1992).
12) G. Nonaka, M. Akazawa and I. Nishioka, *Heterocycles*, **33**, 597 (1992).
13) K. Yazaki, T. Hatano and T. Okuda, *J. Chem. Soc., Perkin Trans. 1*, **1989**, 2289.
14) G. Nonaka, Y. Matsumoto and I. Nishioka, *Chem. Pharm. Bull.*, **29**, 1184 (1981).
15) O. T. Schmidt, D. M. Schmidt and J. Herok, *Justus Liebigs Ann. Chem.*, **587**, 67 (1954).