[Chem. Pharm. Bull. 22(7)1565—1570(1974)]

UDC 547.814.1.02:581.192

Studies on the Constituents of *Aloe arborescens* Mill. var. *natalensis* Berger. II.[1] The Structures of Two New Aloesin Esters

Kenji Makino, Akira Yagi, and Itsuo Nishioka

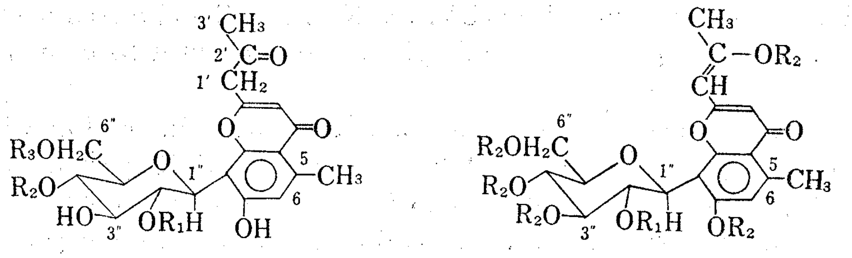
Faculty of Pharmaceutical Sciences, Kyushu University[2]

(Received December 14, 1973)

Two new aloesin esters were isolated from the fresh leaves of *Aloe arborescens* Mill. var. *natalensis* Berger and their structures were established to be 2″-O-*p*-coumaroylaloesin and 2″-O-feruloylaloesin on the basis of chemical and spectral evidences. These esters are the first naturally occurring 2″-O-acylated C-glucosyl compounds.

In the previous paper we reported the structure of aloearbonaside which was a novel chromenylglucoside in *Aloe arborescens* Mill. var. *natalensis* Berger.[3] Recently, Haynes, *et al.*, isolated a new C-glycosyl derivative, 2-acetonyl-8-glucopyranosyl-7-hydroxychromone, named aloesin, from *Aloe* spp.[4] Independently, Wagner, *et al.*, reported the isolation of two C-glucosylchromones, aloeresin A ("Aloe Harz"[5]) and aloeresin B, the latter of which had the same structure to aloesin,[6] and presumed aloeresin A to be 6″-O-*p*-coumaroylaloesin.[6] This paper deals with the structure determination of two new aloesin esters; 2″-O-*p*-coumaroylaloesin (I) and 2″-O-feruloylaloesin (II), isolated from *Aloe arborescens* Mill. var. *natalensis* Berger. These are the first 2″-O-acylated C-glycosyl compounds to be described.

The concentrated MeOH extract was treated with acetone and the acetone soluble portion was worked up by repeated silica gel column chromatography using acetone–EtOAc and $CHCl_3$–EtOH as solvents to give compound B (II). The fraction eluted with acetone–EtOAc



I: R_1=*p*-coumaroyl, $R_2=R_3$=H
II: R_1=feruloyl, $R_2=R_3$=H
III: $R_1=R_2=R_3$=H
IV: R_1=*p*-coumaroyl, R_2=H, R_3=trityl
V: R_1=*p*-coumaroyl, R_2-R_3=benzylidene
VI: R_1=feruloyl, R_2=H, R_3=trityl
VII: R_1=feruloyl, R_2-R_3=benzylidene

Ia: R_1=*p*-coumaroyl, R_2=Ac
IIa: R_1=feruloyl, R_2=Ac
IIIa: $R_1=R_2$=Ac

Chart 1

---

[1] This work was presented at the Annual Meeting of the Japanese Society of Pharmacognosy, Tokyo, October, Abstracts of Papers, 1973, p. 48.

[2] Location: *Katakasu, Higashi-ku, Fukuoka.*

[3] K. Makino, A. Yagi, and I. Nishioka, *Chem. Pharm. Bull.* (Tokyo), **21**, 149 (1973).

[4] L.J. Haynes, D.K. Holdsworth, and R. Russell, *J. Chem. Soc.* (*C*), **1970**, 2581.

[5] H. Auterhoff and B. Ball, *Arzneim. Forsch.*, **4**, 725 (1954); L. Hörhammer, H. Wagner, and G. Bittner, *ibid.*, **13**, 537 (1963); T.J. McCarthy and C.H. Price, *Planta Med.*, **14**, 146 (1966).

[6] H. Wagner, M. Rattenberger, A. Prox, and H. Inouye, *Mitt. Dtsch. Pharmz. Ges.*, **40**, 94 (1970).

Table I. NMR Data[a)]

Compd.	S.[b)]	C_3–H	C_6–H		Acetyl					Methyl		*p*-Coumaroyl (Feruloyl)					
					$C_{2''}$	$C_{3'',4''}$	$C_{6''}$	C_7	$C_{2'}$	$C_{3'}$	C_5	vinyl ($J=16$)			C_3C_5	C_2C_6	
I	a	6.15	6.60	3.86						2.28	2.60	6.10	7.35		6.75	7.45	
II	b	6.18	6.84	3.92						2.34	2.75	6.06	7.30	–	6.64	7.18	6.94
III	a	6.10	6.65	3.74						2.18	2.60			–	–	–	–
Ia	c	6.40	6.82	5.81		2.05	2.00	2.42	2.28	2.22	2.76	6.00	6.90		7.36	7.06	
IIa	c	6.33	6.97	5.84		2.08	2.00	2.41	2.24	2.16	2.80	6.04	7.36	–	7.04	6.86	6.67
IIIa	c	6.44	6.84	5.80	1.72	2.08	2.04	2.42	2.28	2.24	2.80			–	–	–	–
IV	b	6.02	6.69	3.80						2.30	2.67	6.15	7.40		6.82	7.52	
V	a	6.20	6.60	3.90						2.31	2.55	6.15	7.36		6.75	7.45	
VI	c	6.10	6.88	3.66						2.27	2.80	6.00[c)]			6.65[c)]		
VII	c	6.20	6.80	3.70						2.32	2.78	6.12[c)]			6.60[c)]		

a) δ in ppm

b) solvent: a, $(CD_3)_2SO$ b, acetone-d_6 c, $CDCl_3$

c) The other coupled proton signal patterns are obscured due to the overlapping by the signal of the solvent.

was rechromatographed over a silica gel column, followed by preparative thin-layer chromatography (TLC) and recrystallization to give compound A (I).

I, mp 156—162°, $C_{28}H_{28}O_{11}$, $[\alpha]_D^{14}$ $-82.7°$, showing blue fluorescence under ultraviolet (UV) lamp, indicated the positive coloration reaction to Echtblausalz B-KOH and sodium nitroprusside reagent. The UV spectrum (228, 244, 253 and 300 nm) and the infrared (IR) spectrum (3400, 1715 and 1650 cm^{-1}) of I revealed a remarkable similarity to those of aloesin.[7)] The nuclear magnetic resonance (NMR) spectrum of I exhibited two vinyl proton signals ($J=16$ Hz) at δ 6.10 and 7.35, two pairs of AB splitting aromatic proton signals ($J=8$ Hz) at δ 6.75 and 7.45, and phenolic proton signals at δ 9.95 and 10.68, besides the proton signals due to aloesin. The presence of ester bond in I was suggested by IR spectral analysis (1715 cm^{-1}) and by the positive coloration to hydroxamate–ferric chloride. On hydrolysis with 10% HCl in dioxane–H_2O (1: 1) I gave aloesin (III), mp 142—144° and *p*-coumaric acid which were identified by the direct comparison (UV, IR, NMR, TLC and mixed melting point) with an authentic sample, respectively. Accordingly, I was confirmed to be mono *p*-coumaroylaloesin.

The location of *p*-coumaroyl moiety in I was determined in the following way. Since the phenolic proton signals appeared at δ 9.95 and 10.68 in NMR spectrum of I, it was suggested that the acyl residue was linkaged at the hydroxyl group in sugar moiety.

When tritylated with trityl chloride in pyridine I gave trityl ether (IV), mp 140—144°, $C_{47}H_{42}O_{11}\cdot1^1/_2H_2O$ indicative of the characteristic trityl group at δ 7.2—7.6 in NMR spectrum and at 700 cm^{-1} in IR spectrum, and it was verified that the acyl group did not connect with a primary alcohol.

On treatment with benzaldehyde and anhydrous $ZnCl_2$ I provided benzylidene derivative (V), mp 218—222°, $C_{35}H_{32}O_{11}\cdot2H_2O$ exhibiting the aromatic proton signals at δ 7.2—7.6 and a benzylidene methine proton signal[10)] at δ 5.64 in the NMR spectrum. These observations excluded the possibility for the location of the acyl group at $C_{6''}$ and $C_{4''}$ in sugar moiety.

[7)] The β-linkage of sugar in aloesin has been demonstrated by doublets ($J=9$ Hz) due to the anomeric proton at δ 4.68[8)] and the spin decoupling experiment of hexaacetate (IIIa).[9)]

[8)] B. Casu, M. Reggiani, G.G. Gallo, and A. Vigevani, *Tetrahedron Letters*, **1964**, 2839.

[9)] By the spin decoupling experiments the methine and methylene proton signals in IIIa were assigned as follows: δ 4.00 ($C_{6''}$-H, m.), 4.42 ($C_{4''}$-H, m.), 5.20 ($C_{5''}$-H, m.), 5.28, 5.38 ($C_{2''}$- or $C_{3''}$-H, t. $J=8$ Hz) and 5.62 ($C_{1''}$-H, d. $J=8$ Hz).

[10)] F.A. Bovey, "NMR Data Tables for Organic Compounds," Vol. II, Interscience Pub., a division of John Wiley & Sons, New York, London, Sydney, 1967, Compound No. 3282, 3455.

Oxidation of I with periodate at 5° resulted in the consumption of 1.05 mole equivalent of periodate during 24 hr. Therefore, the above findings led to the conclusion that *p*-coumaric acid must be attached at $C_{2''}$ of sugar moiety in I. This was in good accord with the following NMR spectral examination on the acetate (Ia).

The NMR spectrum of hexaacetylaloesin (IIIa), mp 116—118°, indicated acetyl proton signals at δ 1.72 ($C_{2''}$), 2.04 ($C_{6''}$), 2.08 ($C_{3'',4''}$), 2.28 (C-2') and 2.42 (C-7),9) while hexaacetate of I, (Ia), mp 124—128°, $C_{40}H_{40}O_{17}$ exhibited acetyl proton signals at δ 2.00 ($C_{6''}$), 2.05 ($C_{3'',4''}$), 2.42 (C-7), 2.26 (C-4 in *p*-coumaroyl group) and 2.28 ($C_{2'}$).

Since the 2″-O-acetyl methyl group in acetylated C-glycosyl-xanthone,11) -flavone12) and -chromone4) can be expected to be affected by the magnetic anisotropy of the aromatic ring, the higher field signal (δ 1.7—1.8) than signals due to $C_{3''}$,$C_{4''}$,$C_{6''}$-OAc was assigned to the $C_{2''}$-OAc.13) Indeed, the acetyl proton signal due to $C_{2''}$-OAc appeared at δ 1.72 in the NMR spectrum of IIIa. However, in the NMR spectrum of Ia no diagnostic signal due to $C_{2''}$-OAc could be detected. This information was consistent with the results of the above chemical evidences.

Consequently, the structure of compound A should be presented as 2″-O-*p*-coumaroylaloesin (I).

II, mp 153—156°, $C_{29}H_{30}O_{12}\cdot H_2O$, $[\alpha]^{22}_{D}$ −77.5° showing blue fluorescence under UV lamp, expressed the positive coloration reaction to Echtblausalz B-KOH, sodium nitroprusside and hydroxamate-ferric chloride, and presented almost identical absorption bands to those of I in UV spectrum (230, 244, 253, 300 and 320 nm) and IR spectrum (3400, 1715 and 1650 cm^{-1}). The NMR spectrum of II exhibited two phenolic proton signals at δ 8.16 and 8.90, two vinyl proton signals (J=16 Hz) at δ 6.06 and 7.30, a methoxyl proton signal at δ 3.88 and ABX splitting three aromatic proton signals (J_{ortho}=10 Hz, J_{meta}=2 Hz) at δ 6.64, 6.94 and 7.18, besides the signals attributed to the protons of aloesin. Comparative studies on UV, IR and NMR spectra of I and II clearly revealed a close relationship between their structures.

On hydrolysis II yielded aloesin and ferulic acid which were identified with an authentic sample by the direct comparison (UV, IR, NMR, TLC and mixed melting point). Thus, the structure of II was proved to be monoferuloylaloesin. In the NMR spectrum of II the presence of two phenolic protons at δ 8.16 and 8.90 revealed the hydroxyl group in sugar moiety was esterified.

The location of feruloyl moiety was determined by the similar manner to that of I.

The formation of trityl derivative (VI), mp 132—135°, $C_{48}H_{44}O_{12}\cdot 2H_2O$ indicative of the characteristic trityl group at δ 7.1—7.5 (NMR) and at 700 cm^{-1} (IR), and of benzylidene derivative (VII), mp 188—191°, $C_{36}H_{34}O_{12}$ revealing the aromatic proton signals at δ7.2—7.6 and a benzylidene methine proton signal at δ 5.72 (NMR) led to the conclusion that ferulic acid was linkaged at $C_{2''}$ or $C_{3''}$ position.

Oxidation of II with periodate at 5° resulted in the consumption of 1.15 mole equivalent of periodate during 24 hr.

Thus, it was established that ferulic acid should be attached at $C_{2''}$ position in sugar moiety of II. The above chemical evidences agreed with the following NMR spectral examinations on the acetate (IIa).

The NMR spectrum of hexaacetate of II, (IIa), mp 106—108°, $C_{41}H_{42}O_{18}$ indicated the acetyl proton signals at δ 2.00 ($C_{6''}$), 2.08 ($C_{3''}$, $C_{4''}$), 2.24 ($C_{2'}$), 2.29 and 2.41 (two phenol ace-

---

11) D. Billet, J. Massicot, C. Mercier, D. Anker, A. Matschenko, C. Mentzer, M. Chaigneau, G. Valdener, and H. Pacheco, *Bull. Soc. Chim. Fr.*, **1965**, 3006; L.J. Haynes and D.R. Taylor, *J. Chem. Soc.*, (*C*), **1966**, 1685; M. Aritomi and T. Kawasaki, *Chem. Pharm. Bull.* (Tokyo), **18**, 2224 (1970).

12) R.M. Horowitz and B. Gentili, *Chem. Ind.* (London), **1964**, 498; W.E. Hillis and D.H.S. Horn, *Australian J. Chem.*, **18**, 531 (1965).

13) D.K. Holdsworth, *Phytochemistry*, 12, 2011 (1973).

tates). The absence of the diagnostic $C_{2''}$-OAc proton signal led to the conclusion that feruloyl moiety must be attached at $C_{2''}$-O position in sugar moiety.

Accordingly, the structure of compound B should be 2″-O-feruloylaloesin (II).

Although Haynes, *et al.*, reported the presence of aloesin in Aloe spp.[4] no aloesin was detected from the fresh leaves of this plant.

As the O-glycosides having *p*-coumaroyl or feruloyl functions at C-2 position in sugar moiety, grandidentatin[14] and petunoside[15] were reported. The C-glycosides with *p*-coumaroyl or feruloyl substituents at C-2 position in glucose moiety apparently are the first naturally occurring compounds.

Biogenetically, 2″-O-*p*-coumaroyl and 2″-O-feruloylaloesin, together with barbaloin and aloearbonaside were speculated to be derived from polyketides[3,16] in this plant. The biosynthetic studies are in progress.

Experimental

Melting points were determined on a Yanagimoto melting point apparatus and uncorrected. IR spectra were obtained with a KOKEN DS-301 and UV spectra were recorded with a Shimadzu SV-50A. NMR spectra were taken with a JEOL C-60H or PS-100. Chemical shifts were expressed in ppm from Me_4Si as internal reference and coupling constants (J) in Hz. The spin decoupling experiments were demonstrated in $CDCl_3$ by JEOL PS-100. TLC were performed on silica gel (Merck) employing the following solvent systems: 1) EtOAc: MeOH: H_2O (20: 3: 2), 2) $CHCl_3$: EtOH: H_2O (30: 7: 3), 3) $CHCl_3$: EtOAc (2: 1). As spraying reagent 1% Echtblausalz B (Merck)–KOH solution and the blue fluorescence under UV light when alcoholic alkaline solution was sprayed, were used for detection.

Isolation of 2″-O-*p*-Coumaroylaloesin (I) and 2″-O-Feruloylaloesin (II)——The homogenate of the fresh leaves (5 kg), cultivated in the herbal garden of this university, was extracted with MeOH (3 liter) at room temperature and the filtrate was concentrated to syrup. To the syrup acetone (5 liter) was added and the precipitate was removed by filtration. The filtrate was evaporated to dryness and the residue (14.5 g) was chromatographed over a silica gel column (150 g) using EtOAc and EtOAc–acetone (4: 1) subsequently as the solvent. The EtOAc–acetone (4: 1) eluate (Fraction A) (4.8 g) was rechromatographed over a silica gel column (100 g) using $CHCl_3$ and $CHCl_3$–EtOH (9: 1) as the solvent. The $CHCl_3$–EtOH (9: 1) (2.1 g) eluate was rechromatographed over a silica gel column (70 g) using EtOAc and EtOAc–acetone (9: 1) as the solvent and the EtOAc–acetone (9: 1) eluate was recrystallized from acetone–hexane to give colorless amorphous II (385 mg), mp 153—156°, *Anal.* Calcd. for $C_{29}H_{30}O_{12}\cdot H_2O$: C, 59.18: H, 5.48. Found: C, 59.12; H, 5.34. $[\alpha]_D^{22}$ −77.5° (MeOH, $c=1.7$), UV λ_{max}^{MeOH} nm (log ε): 230 (4.3), 244 (4.2), 253 (4.2), 300 (4.2), 320 (sh.). IR ν_{max}^{KBr} cm^{-1}: 3400 (OH), 1715 (C=O), 1650 (C=O). NMR (d_6-acetone) δ ppm: 3.0—5.0 ($C\underline{H}_2$, $C\underline{H}OH$, m.), 3.88 (s. 3H, OCH_3), 8.16, 8.90 (br. s. 2 OH, exchanged with D_2O) and the others as given in Table I.

The fraction A was purified by preparative TLC using the lower layer of $CHCl_3$–EtOH–H_2O (30: 7: 3) as the solvent to afford II (108 mg) and I (5.6 mg). I, colorless amorphous (from acetone–hexane), mp 156—162°, *Anal.* Calcd. for $C_{28}H_{28}O_{11}$: C, 62.22; H, 5.22. Found: C, 62.25; H, 5.59. $[\alpha]_D^{14}$ −82.7° (MeOH, $c=1.4$). UV λ_{max}^{MeOH} nm (log ε): 228 (4.5), 244 (4.3), 253 (4.5), 300 (4.4). IR ν_{max}^{KBr} cm^{-1}: 3400 (OH), 1715, (C=O), 1650 (C=O). NMR (d_6-DMSO) δ ppm: 3.0—5.0 ($C\underline{H}_2$, $C\underline{H}OH$, m.), 9.95, 10.68 (br. s. 2 OH, exchanged with D_2O), and the others as given in Table I.

Acid Hydrolysis of I Yielding Aloesin (III) and *p*-Coumaric Acid——I (170 mg) was hydrolyzed with 10% HCl in dioxane–H_2O (1: 1) (50 ml) for 3 hr under reflux and after the evaporation of the solvent the residue (137 mg) was extracted with hot acetone (50 ml) three times. The extract was chromatographed over a silica gel column (10 g) using EtOAc and EtOAc–acetone (4: 1) as the solvent. The EtOAc eluate was recrystallized from EtOAc to give *p*-coumaric acid (12.3 mg), mp 202—204°, UV λ_{max}^{MeOH} nm: 225, 290, 310. IR ν_{max}^{KBr} cm^{-1}: 3360 (OH), 1670 (C=O), 1630 (C=C), which was identified by the direct comparison (UV, IR, TLC and mixed melting point) with an authentic sample. The EtOAc–acetone (4: 1) eluate (93.5 mg) was rechromatographed over a silica gel column (5 g) using EtOAc and EtOAc–acetone (4: 1) as the solvent and the EtOAc–acetone (4: 1) eluate (42 mg) was purified by preparative TLC (solvent 1 and 2) to give III, colorless amorphous (9 mg) (from acetone), mp 142—144°, UV λ_{max}^{MeOH} nm (log ε): 245 (4.4), 253 (4.4), 297 (4.3), IR ν_{max}^{KBr} cm^{-1}: 3400 (OH), 1710 (C=O), 1650 (C=O). NMR (d_6-DMSO) δ ppm: 3.2—4.8 ($C\underline{H}_2$, $C\underline{H}OH$, m.), 10.20 (s. OH, exchanged with D_2O), and the others as given in Table I, which was identified by the direct comparison (UV, IR, TLC and mixed melting point) with the authentic sample isolated from commercial aloin (Merck).

---

14) I.A. Pearl and S.F. Darling, *J. Org. Chem.*, **27**, 1806 (1962).

15) L. Birkofer, C. Kaiser, and H. Kosmol, *Z. Naturforschg.*, 20, b. 605 (1965).

16) A.J. Birch, *Science*, **156**, 202 (1967).

Hydrolyses of II Yielding III and Ferulic Acid——a) Acid Hydrolysis: II (24 mg) was hydrolyzed with 10% HCl–EtOH for 1 hr under reflux and after evaporation of the solvent the residue was chromatographed over a silica gel column (5 g) using $CHCl_3$ and EtOH by the gradient elution. The $CHCl_3$–EtOH (19: 1) eluate afforded a spot of ferulic acid on TLC (solvent 1). The $CHCl_3$–EtOH (4: 1) eluate was developed four times by preparative TLC (solvent 1) to give III, colorless amorphous (3 mg) (from acetone), mp 143—146°, which was identified by the direct comparison (UV, IR, TLC and mixed melting point) with an authentic sample.

b) Alkaline Hydrolysis: II (98 mg) was hydrolyzed with 2% Na_2CO_3 in EtOH–H_2O (1: 1) for 1 hr under reflux and after neutralization of the solution the solvent was evaporated *in vacuo*. The residue was extracted with EtOAc and after evaporation of the solvent the residue was chromatographed over a silica gel column (5 g) using $CHCl_3$–EtOH as the solvent. The $CHCl_3$–EtOH (19: 1) eluate gave ferulic acid, colorless needles (11 mg), mp 170—172°, which was identified by the direct comparison (UV, IR, TLC and mixed melting point) with an authentic sample.

Tritylation of I Yielding IV——To a solution of I (168 mg) dissolved in pyridine (7 ml) trityl chloride (227 mg) was added and after heating for 4 hr at 110°, the excess reagent was decomposed by adding H_2O, and then the reaction mixtures were extracted with ether. The extract (315 mg) was chromatographed over a silica gel column (15 g) using hexane–EtOAc (1: 2) as the solvent. The hexane–EtOAc (1: 2) eluate was recrystallized from EtOAc to give IV pale yellow amorphous (20 mg), mp 140—144°, *Anal.* Calcd. for $C_{47}H_{42}O_{11}\cdot 1^1/_2H_2O$: C, 69.74; H, 5.60. Found: C, 69.66; H, 5.88. UV λ_{max}^{MeOH} nm (log ε): 243 (4.4), 254 (4.3), 302 (4.4). IR ν_{max}^{KBr} cm^{-1}: 3300 (OH), 1710 (C=O), 1650 (C=O), 700 (aromatic). NMR (d_6-acetone) δ ppm: 3.5—5.0 ($C\underline{H}_2$, $C\underline{H}OH$, m.), 7.2—7.6 (aromatic), 9.00 (s. 2 OH, exchanged with D_2O), and the others as given in Table I.

Tritylation of II Yielding VI——To a solution of II (52 mg) dissolved in pyridine (2 ml) trityl chloride (108 mg) was added and the solution was heated for 4 hr at 100°. After working up in the same way as for IV, the crude product (129 mg) was chromatographed over a silica gel column (10 g) using hexane–EtOAc (1: 2) by the gradient elution. The hexane–EtOAc eluate (24 mg) was purified by preparative TLC (solvent 3) to afford VI, pale yellow amorphous (13 mg), (from $CHCl_3$), mp 132—135°. *Anal.* Calcd. for $C_{48}H_{44}O_{12}\cdot 2H_2O$: C, 67.91; H, 5.72. Found: C, 67.90; H, 5.70. UV λ_{max}^{MeOH} nm (log ε): 228 (4.4), 244 (4.3), 253 (4.3), 298 (4.3), IR ν_{max}^{KBr} cm^{-1}: 3400 (OH), 1720 (C=O), 1650 (C=O), 700 (aromatic). NMR ($CDCl_3$) δ ppm: 3.84 (s. 3H, OCH_3), 3.5—5.0 ($C\underline{H}_2$, $C\underline{H}OH$, m.), 7.1—7.5 (aromatic), 9.00 (s. 2 OH, exchanged with D_2O), and the others as given in Table I.

Benzylidene Derivative (V) of I——To a solution of I (207 mg) dissolved in benzaldehyde (4 ml) anhydrous $ZnCl_2$ (1.4 g) was added and the reaction mixture was left stand for 24 hr at room temperature. The excess reagent was decomposed by adding ice H_2O and the precipitate was collected by filtration. The product was chromatographed over a silica gel column (10 g) using MeOH–$CHCl_3$ as the solvent. The $CHCl_3$–MeOH (49: 1) eluate (54 mg) was further rechromatographed over a silica gel column (6 g) using hexane–EtOAc (2: 3) as the solvent. Finally, the fraction (25 mg) was rechromatographed over a silica gel column (5 g) using $CHCl_3$–MeOH (49: 1) as the solvent to yield V, colorless amorphous (16 mg), (from $CHCl_3$), mp 218—222°. *Anal.* Calcd. for $C_{35}H_{32}O_{11}\cdot 2H_2O$: C, 63.24; H, 5.46. Found: C, 63.70; H, 5.29. UV λ_{max}^{MeOH} nm (log ε): 228 (4.4), 244 (4.3), 253 (4.3), 300 (4.3). IR ν_{max}^{KBr} cm^{-1}: 3200 (OH), 1720 (C=O), 1650 (C=O), 700 (aromatic), NMR (d_6-DMSO) δ ppm: 3.5—5.2 ($C\underline{H}_2$, $C\underline{H}OH$, m.), 5.64 (s. 1H, benzylidene methine), 7.2—7.6 (aromatic), 10.00, 10.08 (s. 2OH, exchanged with D_2O), and the others as given in Table I.

Benzylidene Derivative (VII) of II——To a solution of II (53 mg) dissolved in benzaldehyde (3 ml) anhydrous $ZnCl_2$ (1.1 g) was added and the reaction mixture was left stand for 5 hr at 80°. After working up in the same way as for V, the crude product was chromatographed over a silica gel column (5 g) using EtOH and $CHCl_3$ by the gradient elution. The $CHCl_3$–EtOH (20: 1) eluate (21 mg) was purified by preparative TLC (solvent 3) to give VII, colorless amorphous (12 mg), (from $CHCl_3$), mp 188—191°. *Anal.* Calcd. for $C_{36}H_{34}O_{12}\cdot {}^1/_2H_2O$: C, 64.76; H, 5.30. Found: C, 64.34; H, 5.32. UV λ_{max}^{MeOH} nm (log ε): 229 (4.4), 243 (4.3), 252 (4.2), 302 (4.3). IR ν_{max}^{KBr} cm^{-1}: 3400 (OH), 1720 (C=O), 1650 (C=O), 700 (aromatic). NMR ($CDCl_3$) δ ppm: 3.88 (s. 3H, OCH_3), 5.72 (s. 1H, benzylidene methine), 3.4—5.4 ($C\underline{H}_2$, $C\underline{H}OH$, m.), 7.2—7.6 (aromatic) and the others as given in Table I.

Acetylation of I Yielding Hexaacetate (Ia)——I (100 mg) was acetylated with Ac_2O–pyridine (1: 3) (8 ml) at room temperature for 14 hr, followed by the usual work up. The product was chromatographed overa silica gel column (10 g) using hexane and EtOAc by the gradient elution. The hexane–EtOAc (3: 2) eluate was recrystallized from hexane–$CHCl_3$ to afford Ia, colorless amorphous (23 mg), mp 124—128°. *Anal.* Calcd. for $C_{40}H_{40}O_{17}$: C, 60.62; H, 5.08. Found: C, 60.48; H, 5.08. UV λ_{max}^{MeOH} nm (log ε): 226 (4.3), 252 (4.3), 262 (4.3), 292 (4.4). IR ν_{max}^{KBr} cm^{-1}: 1760 (C=O), 1640 (C=O). NMR ($CDCl_3$) δ ppm: 3.8—5.5 ($C\underline{H}_2$, $C\underline{H}OAc$, m.), and the others as given in Table I.

Acetylation of II Yielding Hexaacetate (IIa)——II (51 mg) was acetylated with Ac_2O–pyridine (3: 1) (4 ml) at room temperature for 14 hr, followed by the usual work up. The product was purified by preparative TLC using hexane–EtOAc (1: 2) as the solvent to give IIa, colorless amorphous (13 mg) (from hexane–$CHCl_3$), mp 118—120°, *Anal.* Calcd. for $C_{41}H_{42}O_{18}$: C, 59.85; H, 5.14. Found: C, 60.24; H, 5.28. UV λ_{max}^{MeOH} nm (log ε): 260 (4.2), 296 (4.2). IR ν_{max}^{KBr} cm^{-1}: 1750 (C=O), 1640 (C=O). NMR ($CDCl_3$), δ ppm: 3.81 (s. 3H, OCH_3), 3.8—5.5 ($C\underline{H}_2$, $C\underline{H}OAc$, m.), and the others as given in Table I.

Acetylation of III Yielding Hexaacetate (IIIa)——III (110 mg) was acetylated with Ac_2O–pyridine (3:1) (8 ml) at room temperature for 14 hr, followed by the usual work up. After working up in the same way as for Ia, IIIa was recrystallized from $CHCl_3$–hexane to give IIIa, colorless needles (38 mg), mp 116—118°. UV $\lambda_{\max}^{\text{MeOH}}$ nm (log ε): 220 (4.1), 253 (4.2), 260 (4.2), 295 (4.1), 310 (sh.). IR $\nu_{\max}^{\text{KBr}}$ cm^{-1}: 1750 (C=O), 1640 (C=O).

Periodate Oxidation of I and II——To each solution of I or II (1/30—1/40 m mole) dissolved in MeOH (10 ml) and H_2O (5 ml), 0.02 M $NaIO_4$ aq. solution (5 ml) was added and the reaction mixture was left stand at 5° in darkness. As a blank test, 0.02 M $NaIO_4$ aq. solution (5 ml) without addition of sample was left stand in the same condition. At the definite time intervals 0.01 N solution of sodium arsenite (25 ml) and 2% KI solution (1 ml) were added to each solution. After keeping for 10 min, excess arsenite was titrated with 0.01 N I_2 solution. The results were given in Table II.

TABLE II. Molar Consumption of $NaIO_4$

Period of oxidation Sample	10 min	1 hr	24 hr
I	0.83	0.92	1.05
II	1.01	1.03	1.15

Acknowledgement This work was supported in part by a Grant-in-Aid for Scientific Research from the Ministry of Education, which is gratefully acknowledged. The authors express their thanks to Mr. Matsui and Miss Soeda for UV and IR spectral measurements, to Mr. Tanaka for NMR spectral measurement, to Mr. Ishimura for the microanalyses and to Miss Iwasa for her technical assistance. We are also grateful to Prof. H. Inouye of Kyoto University for his kind advice.