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Studies of Oligosaccharides. XII.[1] Hydrophilization of Glycyrrhetinic Acid by Coupling to Gentio Oligosaccharides

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Three kinds of analogues of glycyrrhezin (ester–, ether- and ester-ether-type glycosides) were synthesized by coupling gentio oligosachacrides to the carboxyl group, the hydroxyl group and both of its aglycone, and their hydrophilic properties were compared. The solubilities in water as well as water–oil partition coefficients increased with the increasing number of the D-glucose unit, and the introduction of sugars to the hydroxyl group was more effective to the solubilization of the aglycone than to the carboxyl group.

There have been a number of works on the structure-activity relationship in glycosides. However, most discussions have been confined to aglycone structures, and sugar parts have been too much ignored. In an attempt to elucidate the role of sugar moieties in glycosides we have synthesized some analogues of natural glycosides, in which gentio oligosaccharides with various chain length are coupled to hydroxyl and/or carboxyl groups of their aglycones, and have compared the physical and pharmacological properties of such synthetic analogues. In this paper we describe, as a preliminary work of this series, syntheses of ester-, ether- and ester-ether-type glycosides of glycyrrhetinic acid (I), the aglycone of glycyrrhezin (II), a triterpenoid glycoside of liquorice root, and present fundamental physical data concerning the hydrophilicities of these glycosides.

Syntheses of Ester-Type Glycosides, IV, V and VI

Both of the C-3 bound hydroxyl and the C-30 carboxyl groups in I (3β-hydroxy-11-oxo-olean-12-en-30-oic acid) are reactive to acetobromo sugars under the ordinary Königs-Knorr conditions. The reactivity to the hydroxyl group was, however, greatly depressed, as reported,[3] when pyridine was used as reaction medium. Thus, the mono-, di- and tri-saccharides were introduced in considerable yields to the carboxyl group in I by condensation of I with acetobromo-glucose, -gentiobiose and -gentiotriose in the presence of silver oxide. Deacetylation of the condensates with dilute methanolic sodium methoxide afforded ester-type glycosides, IV, V and VI. Glycosides of this type could be obtained alternatively by the reaction of the sodium salt of I with acetobromo sugars in N,N-dimethylformamide. Infrared (IR) and proton magnetic resonance (PMR) spectroscopic evidences indicated that the α,β-unsaturated ketone in the aglycone was unattacked during these reactions. The anomeric configurations of newly formed sugar-triterpene glycosidic linkages were analyzed based on optical data as well as PMR spectroscopy. The ester-type glycoside IV has been reported to be an anomeric mixture of α- and β-linked glycosides.[3] Indeed, the specific rotation of the acetate of IV in chloroform (+99.6°) was between the predicted values[4] for α- (+154°) and β- (+50°) linked

1) Part XI: K. Koizumi and T. Utamura, *Carbohyd. Res.*, 33, 127 (1974).
2) *a*) Location: *6–1–1 Toneyama, Toyonaka, Osaka*; *b*) *3–777 Nakahozumi, Ibaraki, Osaka.*
3) A.M. Juodvirsis and A.T. Troshchenko, *Khim. Prir. Soedin*, 4, 13 (1968).
4) T. Reichstein, *Angew. Chem.*, 63, 412 (1951). These values were calculated from the following specific rotations: I, +163°; methyl α-D-glucopyranoside tetraacetate, +130.5°; methyl β-D-glucopyranoside tetraacetate, −18.2°.

glycosides, calculated on the basis of the isorotation rule, and hence glycoside IV appeared to be a mixture composed of approximately equal amounts of both anomers. However, in the PMR spectrum of the acetate of IV, there was observed a doublet at τ 4.3—4.4 with a spacing of 7—8 Hz, which should be ascribed to an axially oriented H-1′ signal on the analogy of the H-1 chemical shifts and $J_{1,2}$ coupling constants of α- and β-D-glycopyranose pentaacetates (τ 3.66, 3.2 Hz and τ 4.24, 7.8 Hz), whereas there was detected no equatorially oriented H-1′ signal around τ 3.6. This observation indicated that glycoside IV is linked β to the aglycone rather than α. Glycosides V and VI were considered to be also β-anomers from PMR data.

Syntheses of Ether-Type Glycosides, VII and VIII

Direct syntheses of ether-type glycosides was unsuccessful due to the preferential introduction of sugars to the carboxyl group. For this reason the carboxyl group in I was transiently protected as its methyl ester III. Glycosylation of III in chloroform in the presence of silver oxide afforded the methyl ester acetates of VII and VIII in fairly good yields. Treatment of these condensates with methanolic sodium methoxide resulted in deacetylation without hydrolysis of the methyl ester group, whereas heating in 5% aqueous ethanolic sodium hydroxide caused simultaneous elimination of both protecting groups, acetyl and methyl, giving ether-type glycosides, VII and VIII. The anomeric configurations of VII and VIII were determined as β from the $J_{1',2'}$, coupling constants (7.2 and 7.4 Hz, respectively) of their methyl ester acetates. Comparison of the specific rotations of the methyl ester acetate of VII in chloroform (+61.8°) with the theoretical values[5] for α- (+133°) and β- (+67°) linked glycosides also suggested that the anomeric configuration in VII is β rather than α. No optical rotation data were not available for the derivatives of VIII. These assignments of anomeric configuration conform to the general course of the Königs-Knorr condensation in which β-glycosylation is predominant due to neighboring-group participation.

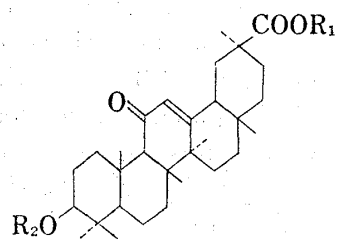
Syntheses of Ester-Ether-Type Glycosides, IX and X

Condensation of I with more than two moles of acetobromo sugars in chloroform in the presence of silver oxide, followed by chromatographic fractionation of the condensates, afforded the acetates of ester–ether-type glycosides, IX and X. The anomeric configurations of both types of sugar-triterpene linkages were considered to be β from PMR spectra of their acetates; they gave two doublets of anomeric protons at *ca.* τ 4.2 (ester) and 5.5 (ether) with spacings of 7—8 and *ca.* 7 Hz, respectively. The sugar moieties linked to the carboxyl group were extremely labile, and the deacetylation with 0.1—0.01 M methanolic sodium methoxide caused simultaneous cleavage of the sugar-triterpene linkages, giving ether-type glycosides. However, the deacetylation using dilute methanolic barium methoxide for short reaction times yielded quantitatively ester–ether-type glycosides without scission of ester linkages.

Solubilities in Water of Synthetic Glycosides

As shown in Table I the solubilities in water of these synthetic glycosides increased with the increasing number of the D-glucose unit. It is noticeable that, in the cases of ester-type glycosides, the increase in solubilities is not linear but rather exponential with respect to the D-glucose number. It is also observed that the introduction of sugars to the hydroxyl group is more effective to the solubilization of the aglycone than to the carboxyl group (comparison of IV with VII and V with VIII). Since the striking difference of solubilities between the ether-type glycoside VII and the natural glycoside II arises from the difference of the sugar moieties, glucobiose and glucobiuronate residues, the solubilizing effect of the latter appears to be much greater than that of the former. However, the introduction of the former to both functional groups (X) brought about a strong solubilizing effect comparable to that in II.

5) These values were calculated from the following specific rotations: III, +133°; methyl α-D-glucopyranoside tetraacetate, +130.5°; methyl β-D-glucopyranoside tetraacetate, −18.2°.



I: (glycyrrhetinic acid) $R_1=R_2=H$
II: (glycyrrhezin) $R_1=H, R_2=$ O-β-D-glucopyranurosyl-O-β-glucopyranuranurosido
III: $R_1=Me, R_2=H$
IV: $R_1=\beta$-D-glucopyranosyl, $R_2=H$
V: $R_1=\beta$-gentiobiosyl, $R_2=H$
VI: $R_1=\beta$-gentiotriosyl, $R_2=H$
VII: $R_1=H$, $R_2=\beta$-D-glucopyranosyl
VIII: $R_1=H$, $R_2=\beta$-gentiobiosyl
IX: $R_1=R_2=\beta$-D-glucopyranosyl
X: $R_1=R_2=\beta$-gentiobiosyl

Chart 1

Table I. Hydrophilic Properties of Synthetic Glycosides

Glycoside	Solubility in water mg/100 ml (37°)	Water/*n*-BuOH partition coefficient (37°)
I	1.06	1.02×10^{-2}
II	$>20\times10^{3}$	97.0×10^{-2}
IV	1.17	0.00×10^{-2}
V	1.44	0.63×10^{-2}
VI	10.6	12.1×10^{-2}
VII	4.02	1.48×10^{-2}
VIII	8.85	3.72×10^{-2}
IX	6.55	1.93×10^{-2}
X	$>20\times10^{3}$	17.9×10^{-2}

Water-Oil Distribution of Synthetic Glycosides

The partitition of these synthetic glycosides between aqueous and organic phases is of pharmacological interest, since it is closely related to the incorporation of such drugs in animal bodies. Therefore the partition coefficients of these glycosides were measured using water/*n*-butanol as a model system, and the results are tabulated in Table I. They reflected their solubilities in water and there was observed a parallelism between these two parameters; the partition coefficients increased again with the increasing number of the D-glucose unit and the relative hydrophilicities of glycosides with the D-glucose number 2 were in the order of ester type<ester-ether type<ether type. The ester type glycosides, IV and V had weaker hydrophilicities than the aglycone I for unknown reasons, and even the highest hydrophilic glycoside X had a lower value of partition coefficient than the natural glycoside II.

Experimental[6]

3β-Hydroxy-11-oxo-olean-12-en-30-oic Acid β-D-Gulcopyranosyl Ester (IV)——The acetate of IV was prepared by condensation of I with acetobromoglucose according to the literature.[3] The crude product was purified by silica gel column chromatography, followed by recrystallization from EtOH, mp 233—235°; $[\alpha]_D^{21}$ +99.6° ($c=0.95$, $CHCl_3$). Lit.[3] mp 227—229°; $[\alpha]_D^{20}$ +111° ($c=2.01$, $CHCl_3$). *Anal.* Calcd. for $C_{44}H_{64}O_{13}$: C, 66.03; H, 8.16. Found: C, 65.98; H, 8.05. IR ν_{max}^{KCl} cm^{-1}: 1650 (CO of C=C–C=O), 1750 (–COO–), 3500 (OH). PMR ($CDCl_3$) τ: 4.3—4.4 (1H, d, H-1′, $J_{1',2'}=7$—8 Hz), 4.37 (1H, s, =C<u>H</u>–C=O), 4.6—5.1 (3H, m, H-2′,3′,4′), 5.6—6.3 (3H, m, H-5′, 2×H-6′), 6.7—9.3 (23H, m, triterpene-ring protons), 7.96, 8.01 (3H, each, s, $COC\underline{H}_3$), 8.03 (6H, 2×$COC\underline{H}_3$), 9.22 (6H, s, 2×C-$C\underline{H}_3$).

In an alternative synthesis the sodium salt of I (0.60 g, 1.2 mmoles) was dissolved in a minimum volume of dimethyl formamide (DMF), and to this solution was added acetobromoglucose (0.52 g, 1.3 mmoles). After 3 hr $CHCl_3$ (20 ml) and water (20 ml) were added to the reaction solution, and the mixture was shaken vigorously. The $CHCl_3$ layer was washed with satd. $NaHCO_3$, followed by water. The solid mass obtained

6) All evaporations were effected below 40° under diminished pressure. Melting points were measured on a hot stage with a Yanagimoto micro melting-point apparatus and are uncorrected. Specific rotations were measured in a 1-dm tube. Ultraviolet (UV) and IR spectra were obtained with a Shimazu UV-200 and Hitachi EPI G-2 spectrophotometer, respectively. PMR spectra were observed at 90 MHz with a Hitachi R-22 spectrometer using tetramethylsilane as the internal standard.

after evaporation of solvent was purified on a silica gel column (Wakogel C-200, 100 g) with 7: 3 benzene–AcOEt. Recrystallization of the product from methanol afforded the pure acetate of IV (0.70 g, 73% based on the sodium salt of I), mp and mixed mp 233—235°.

The acetate of IV (11 g) was dissolved in a mixture of MeOH (200 ml) and CH_2Cl_2 (50 ml). To this solution was added 0.1 M methanolic sodium methoxide (60 ml). After 10 min the reaction solution was decationized with Amberlite IR-120 (H^+) and evaporated to dryness. The residual syrup was purified on a silica gel column with 1: 1 benzene–AcOEt to give amorphous powder of IV, mp 185—189.5°; $[\alpha]_D^{17}$ +143° (c=1.01, EtOH). *Anal.* Calcd. for $C_{36}H_{56}O_9 \cdot 2H_2O$: C, 64.65; H, 9.04. Found: C, 64.38; H, 9.11. UV λ_{max}^{EtOH} nm (ε): 250 (11000). IR ν_{max}^{Nujol} cm^{-1}: 1080 (–C–O–C–), 1650 (CO of C=C–C=O), 1740 (–COO–), 3400 (OH).

3β-Hydroxy-11-oxo-olean-12-en-30-oic Acid β-Gentiobiosyl Ester (V)——The acetate of V was obtained from the reaction of I (1.2 g, 2.5 mmoles), Ag_2O (2.0 g), acetobromogentiobiose (3.5 g, 4.9 mmoles), and pyridine (10 ml) in a similar manner as in the synthesis of the acetate of IV. Chromatographic purification, followed by recrystallization from MeOH afforded the pure acetate of V (1.5 g, 57%), mp 157—159°; $[\alpha]_D^{21}$ +58.2° (c=1.0, $CHCl_3$). *Anal.* Calcd. for $C_{56}H_{80}O_{21}$: C, 61.75; H, 7.40. Found: C, 61.53; H, 7.30. IR ν_{max}^{KCl} cm^{-1}: 1650 (CO of C=C–C=O), 1750 (–COO–), 3450 (OH). PMR ($CDCl_3$) τ: 4.3—4.4 (1H, d, H-1′, $J_{1',2'}$= 7—8 Hz), 4.34 (1H, s, =C<u>H</u>–C=O), 4.6—6.4 (13H, m, sugar-ring protons), 6.7—9.3 (23H, m, triterpene-ring protons), 7.95—8.05 (21H, m, 7×COC<u>H</u>$_3$), 8.65, 9.01 (3H, each, s, C–C<u>H</u>$_3$), 8.86 (9H, s, 3×C–C<u>H</u>$_3$), 9.20 (6H, s, 2×C–C<u>H</u>$_3$).

Deacetylation of the acetate of V in a similar manner as in the deacetylation of the acetate of IV afforded amorphous V in nearly quantitative yield, mp 219—222°; $[\alpha]_D^{18}$ +82.3° (c=1.64, EtOH). *Anal.* Calcd. for $C_{42}H_{66}H_{14} \cdot 2H_2O$: C, 60.71; H, 8.49. Found: C, 60.94; H, 8.62. UV λ_{max}^{EtOH} nm (ε): 250 (12000). IR ν_{max}^{Nujol} cm^{-1}: 1080 (–C–O–C–), 1650 (CO of C=C–C=O), 1740 (–COO–), 3400 (OH).

3β-Hydroxy-11-oxo-olean-12-en-30-oic Acid β-Gentiotriosyl Ester (VI)——The acetate of VI was obtained from the reaction of I (0.6 g, 1.3 mmoles), Ag_2O (1.0 g), acetobromogentiotriose[7] (2.5 g, 2.5 mmoles), and pyridine (5 ml) in a similar manner as in the synthesis of the acetate of IV. Chromatographic purification, followed by recrystallization from EtOH afforded the pure acetate of VI, mp 177—179°; $[\alpha]_D^{21}$ +44.5° (c=1.37, EtOH). *Anal.* Calcd. for $C_{68}H_{96}O_{29}$: C, 59.29; H, 7.02. Found: C, 59.25; H, 6.86. IR ν_{max}^{KCl} cm^{-1}: 1650 (CO of C=C–C=O), 1750 (–COO–), 3450 (OH). PMR ($CDCl_3$) τ: 4.3—4.4 (1H, d, H-1′, $J_{1',2'}$=*ca.* 8 Hz), 4.34 (1H, s, =C<u>H</u>–C=O), 4.6—6.4 (20H, m, sugar-ring protons), 6.7—9.3 (23H, m, triterpene-ring protons), 7.95—8.05 (30H, m, 10×COC<u>H</u>$_3$), 8.65, 9.01 (3H, each, s, C–C<u>H</u>$_3$), 8.86 (9H, s, 3×C–C<u>H</u>$_3$), 9.20 (6H, s, 2×C-C<u>H</u>$_3$).

Deacetylation of the acetate of VI in a similar manner as in the deacetylation of the acetate of IV afforded amorphous VI in nearly quantitative yield, mp 205.5—206.5°; $[\alpha]_D^{19}$ +57.0° (c=1.58, EtOH). *Anal.* Calcd. for $C_{48}H_{66}O_{14} \cdot 2H_2O$: C, 60.71; H, 8.49. Found: C, 60.56; H, 8.62. UV λ_{max}^{EtOH} nm (ε): 250 (12300). IR ν_{max}^{Nujol} cm^{-1}: 1080 (–C–O–C–), 1650 (CO of C=C–C=O), 1740 (–COO–), 3400 (OH).

3β-O-β-D-Glucopyranosyl-11-oxo-olean-12-30-oic Acid (VII)——A mixture of methyl glycyrrhetinate (III, 2.9 g, 6.0 mmoles), Ag_2O (2.8 g, 12 mmoles), Drierite (6.5 g), I_2 (0.3 g) and $CHCl_3$ (15 ml) was stirred for 1 hr. A $CHCl_3$ solution (5 ml) containing acetobromoglucose (4.9 g, 12 mmoles) was added gradually with vigorous stirring. After stirring for additional 3 hr the reaction mixture was filtered, and the precipitate washed with $CHCl_3$ (30 ml). The combined filtrate and the washing were evaporated to dryness, and the residual syrup was crystallized from EtOH–acetone–$CHCl_3$ to give needles of the methyl ester acetate of VII (2.8 g, 58%), mp 232.5—234°; $[\alpha]_D^{22}$ +61.8° (c=1.04, $CHCl_3$). *Anal.* Calcd. for $C_{45}H_{66}O_{13}$: C, 66.30; H, 8.18. Found: C, 66.12; H, 8.12. IR ν_{max}^{KCl} cm^{-1} of carbonyls: 1660 (C=C–C=O), 1730 (–COO–), 1760 (–OCOCH$_3$). PMR ($CDCl_3$) τ: 4.36 (1H, s, =C<u>H</u>–C=O), 4.7—5.1 (3H, m, H-2′,3′,4′), 5.46 (1H, d, H-1′, $J_{1',2'}$=7.2 Hz), 5.6—6.4 (3H, m, H-5′, 2×H-6′), 6.8—9.3 (23H, m, triterpene-ring protons), 6.33 (3H, s, OC<u>H</u>$_3$), 7.85, 7.94, 7.98, 8.01 (3H, each s, COC<u>H</u>$_3$), 8.66, 9.07, 9.20, 9.23 (3H, each s, C-C<u>H</u>$_3$), 8.87 (9H, s, 3×C-C<u>H</u>$_3$).

The methyl ester acetate of VII (2.2 g) was dissolved in MeOH–CH_2Cl_2 (3: 1, 40 ml), and to this solution was added 0.1 M methanolic MeONa (10 ml). The reaction solution was allowed to stand overnight, and then decationized by stirring with Amberlite IR-120 (H^+) resin. Upon gradual evaporation of solvent the crystalline methyl ester of VII (1.5 g, 85%) was separated. Recrystallization from aq. MeOH afforded prisms, mp 260—262°; $[\alpha]_D^{22}$ +84.5° (c=1.29, EtOH). *Anal.* Calcd. for $C_{37}H_{58}O_9 \cdot H_2O$: C, 66.84; H, 9.10. Found: C, 66.54; H, 8.94. IR ν_{max}^{Nujol} cm^{-1}: 1660 (CO of C=C–C=O), 1730 (–COO–), 3300 (OH).

The methyl ester acetate of VII (0.5 g) was dissolved in 5% aqueous ethanolic (1: 1) NaOH (20 ml) and the solution was refluxed for 3 hr. The reaction solution was decationized with Amberlite IR-120 (H^+) resin, and evaporated to dryness. The residual syrup was triturated in aq. EtOH to give amorphous VII (0.39 g, 93%), $[\alpha]_D^{22}$ +91.8° (c=1.04, EtOH). Glycoside VII decomposed gradually over 200° without melting. *Anal.* Calcd. for $C_{36}H_{56}O_9 \cdot 2H_2O$: C, 64.65; H, 9.04. Found: C, 64.99; H, 8.89. UV λ_{max}^{EtOH} nm (ε): 248 (13000). IR ν_{max}^{Nujol} cm^{-1}: 1650 (CO of C=C–C=O), 1710 (–COOH), 3400 (OH).

3β-O-β-Gentiobiosyl-11-oxo-olean-12-en-30-oic Acid (VIII)——The reaction mixture obtained from the reaction of III (3.0 g, 6.2 mmoles), Ag_2O (2.9 g, 12.4 mmoles), Drierite (6 g), I_2 (0.3 g), $CHCl_3$ (15 ml) and

7) K. Takiura, S. Honda, T. Endo, and K. Kakehi, *Chem. Pharm. Bull.* (Tokyo), **20**, 438 (1972).

acetobromogentiobiose (8.7 g, 12.4 mmoles) for 4.5 hr was worked up in a similar manner as in the synthesis of the methyl ester acetate of VII. The crude product (3.5 g, 51%) was purified on a silica gel column (Wakogel C-200, 200 g) with 1:1 benzene–AcOEt to give a refined product of the methyl ester acetate of VIII as amorphous powder, mp 167—168°; $[\alpha]_D^{22}$ +49.8° (c=1.09, $CHCl_3$). *Anal.* Calcd. for $C_{57}H_{82}O_{21}$: C, 62.04; H, 7.51. Found: C, 61.79; H, 7.58. IR ν_{max}^{KCl} cm^{-1} of carbonyls: 1660 (C=C–C=O), 1730 (–COO–), 1760 (–$COCH_3$). PMR ($CDCl_3$) τ: 4.47 (1H, s, =C<u>H</u>–C=O), 4.8—5.4 (7H, m, H-2′,3′,4′,1″,2″,3″,4″), 5.49 (1H, d, H-1′, $J_{1',2'}$=7.4 Hz), 5.6—6.5 (6H, m, H-5′, 2×H-6′, H-5″, 2×H-6″), 6.8—9.3 (23H, m, triterpene-ring protons), 6.33 (3H, s, OC<u>H</u>$_3$), 7.85—8.07 (21H, m, 7×COC<u>H</u>$_3$), 8.65, 9.08, 9.17, 9.25 (3H, each, s, C-C<u>H</u>$_3$), 8.89 (9H, s, 3×C-C<u>H</u>$_3$).

Deacetylation of the methyl ester acetate of VIII with MeONa in a similar manner as in the deacetylation of the methyl ester acetate of VII gave the methyl ester of VIII in nearly quantitative yield. Recrystallization from aq. MeOH afforded prisms, mp 195—196°; $[\alpha]_D^{19}$ +57.7° (c=1.30, EtOH). *Anal.* Calcd. for $C_{43}H_{68}O_{14}\cdot H_2O$: C, 62.45; H, 8.53. Found: C, 61.96; H, 8.67. IR ν_{max}^{Nujol} cm^{-1}: 1660 (CO of C=C–C=O), 1730 (–COO–), 3300 (OH).

Heating the methyl ester acetate of VIII in a similar manner as in the deprotection of the methyl ester acetate of VII gave glycoside VIII as amorphous powder in nearly quantitative yield, $[\alpha]_D^{22}$ +36.6° (c=1.04, EtOH). Glycoside VIII decomposed gradually over 200° without melting. *Anal.* Calcd. for $C_{42}H_{66}O_{14}\cdot 2H_2O$: C, 60.71; H, 8.49. Found: C, 60.35; H, 8.61. UV λ_{max}^{EtOH} nm (ε): 251 (9700). IR ν_{max}^{Nujol} cm^{-1}: 1650 CO of C–C–C–O), 1710 (–COOH), 3400 (OH).

3β-O-β-D-Glucopyranosyl-11-oxo-olean-12-en-30-oic Acid β-D-Glucopyranosyl Ester (IX)——To a mixture of I (2.4 g, 5.0 mmoles), Ag_2O (2.6 g), Drierite (2.6 g), I_2 (0.5 g) and $CHCl_3$ (30 ml) was added with stirring a $CHCl_3$ solution (10 ml) containing acetobromoglucose (4.0 g, 10 mmoles). The reaction mixture was stirred for 1 hr, and acetobromoglucose (4.0 g, 10 mmoles) and Ag_2O (2.6 g) were supplemented. After stirring overnight the reaction mixture was filtered, and the precipitate washed with $CHCl_3$ (20 ml). The combined filtrate and the washing were shaken with satd. NaCl (100 ml) five times, and dried over $CaCl_2$. The solid mass obtained after evaporation of solvent was purified on a silica gel column (Wakogel C-200, 300 g) with 1:1 benzene–AcOEt. Recrystallization from CH_2Cl_2–ether–hexane afforded the pure acetate of IX (0.94 g, 17%), mp 153—155°; $[\alpha]_D^{20}$ +71.4° (c=0.99, $CHCl_3$). *Anal.* Calcd. for $C_{58}H_{82}O_{22}$: C, 61.58; H, 7.31. Found: C, 61.28; H, 7.36. IR ν_{max}^{KCl} cm^{-1} of carbonyls: 1660 (C=C–C=O), 1750 (–COO– and –$COCH_3$). PMR ($CDCl_3$) τ: 4.24 (1H, d, H-1′ of the sugar ring bound to C-30 COOH, $J_{1',2'}$=7—8 Hz), 4.34 (1H, s, =C<u>H</u>–C=O), 5.46 (1H, d, H-1′ of the sugar ring bound to OH at C-3, $J_{1',2'}$=*ca.* 7 Hz), 4.7—6.4 (12H, m, sugar-ring protons), 7.95—8.05 (24H, m, 8×COC<u>H</u>$_3$), 8.68, 9.07 (3H, each, s, C-C<u>H</u>$_3$), 8.89 (9H, s, 3×C-C<u>H</u>$_3$), 9.23 (6H, s, 2×C-C<u>H</u>$_3$).

The acetate of IX (0.3 g) was dissolved in 0.0025 M methanolic barium methoxide (25 ml), which was prepared by dissolving BaO in methanol, followed by filtering impurities, and the solution was allowed to stand for 45 min. The reaction solution was decationized with Amberlite IR-120 (H^+) resin and evaporated to dryness. The residual syrup was dissolved in a small volume of EtOH, and this solution was dropped into acetone. Glycoside IX was obtained as amorphous powder in nearly quantitative yield, mp 205—212°; $[\alpha]_D^{20}$ +145° (c=1.10, EtOH). *Anal.* Calcd. for $C_{42}H_{66}O_{14}\cdot 4H_2O$: C, 58.18; H, 8.60. Found: C, 58.19; H, 8.41. UV λ_{max}^{EtOH} nm (ε): 249 (11800). IR ν_{max}^{Nujol} cm^{-1}: 1650 (CO of C=C–C=O), 1740 (–COO–), 3350 (OH).

3β-O-β-Gentiobiosyl-11-oxo-olean-12-en-30-oic Acid β-Gentiobiosyl Ester (X)——The reaction mixture obtained from the reaction of I (1.4 g, 3.0 mmoles), Ag_2O (1.6 g), Drierite (1.5 g), I_2 (0.3 g), $CHCl_3$ (25 ml), and acetobromogentiobiose (4.3 g, 6.0 mmoles) was worked up in a similar manner as in the synthesis of the acetate of IX. Recrystallization of the product from MeOH afforded the pure acetate of X (1.1 g, 21%), mp 185—186°; $[\alpha]_D^{20}$ +12.5° (c=0.92, $CHCl_3$). *Anal.* Calcd. for $C_{82}H_{134}O_{38}$: C, 57.67; H, 6.73. Found: C, 57.63; H, 6.87. IR ν_{max}^{KBr} cm^{-1} of carbonyls: 1660 (C=C–C=O), 1750 (–COO– and –$OCOCH_3$). PMR ($CDCl_3$) τ: 4.23 (1H, d, H-1′ of the sugar ring bound to C-30 COOH, $J_{1',2'}$=7—8 Hz), 4.33 (1H, s, =C<u>H</u>–C=O), 5.46 (1H, d, H-1′ of the sugar ring bound to OH at C-3, $J_{1',2'}$=*ca.* 7 Hz), 4.7—6.4 (26H, m, sugar-ring protons), 6.7—9.3 (23H, m, triterpene-ring protons), 7.95—8.05 (42H, m, 14×COC<u>H</u>$_3$), 8.68, 9.07 (3H, each, s, C-C<u>H</u>$_3$), 8.90 (9H, s, C-C<u>H</u>$_3$), 9.23 (6H, s, 2×C-C<u>H</u>$_3$).

The acetate of X was deacetylated with 0.0025 M methanolic barium methoxide in a similar manner as in the deacetylation of the acetate of IX gave X in nearly quantitative yield as amorphous powder, mp 212—215°; $[\alpha]_D^{20}$ +45.8° (c=0.96, EtOH). *Anal.* Calcd. for $C_{54}H_{86}O_{24}\cdot 3H_2O$: C, 55.28; H, 7.90. Found: C, 55.59; H, 8.01. UV λ_{max}^{EtOH} nm (ε): 250 (12400). IR ν_{max}^{Nujol} cm^{-1}: 1650 (CO of C=C–C=O), 1740 (–COO–), 3350 (OH).

Solubilities in Water of Synthetic Glycosides——Suspensions of finely pulverized glycoside (1—2 mg) in water (10 ml) were shaken constantly on a temperature-controlled water bath for several days. The insoluble parts were centrifuged and the supernatants (5.00 ml) were evaporated to dryness. The residues were dissolved in appropriate volumes of ethanol (spectroscopic grade) so that their concentrations were below 1.2×10^{-4} M, at which concentrations all glycosidess obeyed the Beer's law. Solubilities in water were calculated from observed absorbancies. The glycosides XII as well as II had too great solubilities to be measured by this method.

Water/*n*-BuOH Partition Coefficients of Synthetic Glycosides——Glycosides (1—2 mg) were dissolved in water-satd. *n*-BuOH (5.00 ml) and to these solutions were added *n*-BuOH–satd. water (5.00 ml). The

mixtures were shaken constantly on a temperature-controlled water bath for 10 hr, and then allowed to stand for 24 hr. Aliquots of both phases (2.00 ml) were evaporated to dryness and the residues were dissolved in appropriate volumes of ethanol and the UV absorption was read at λ_{max}'s. Partition coefficients were obtained as the ratios of absorbancies of both phases.

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