Continuation of the chromatography with 2% EtOAc-C_6H_6 gave firstly 1 g of a 1:1 mixture (tlc) of compounds **7** and **9**, followed by 0.4 g of crude **9**. Crystallization from Et_2O-C_6H_{14} afforded 0.3 g (8.59%) of **9**: mp 173–175°; $[\alpha]D$ −154°. *Anal.* ($C_{25}H_{34}ClFO$) C, H, Cl, F.

16-Methylene-3β,5α,17α-trihydroxypregnane-6,20-dione 3,17-Diacetate (11). To a solution of 5 g (17.7 mmol) of 16-methylene-3β,17α-dihydroxypregnen-20-one diacetate (**10**) in 40 ml of EtOH-free $CHCl_3$ was added 26.7 ml of an Et_2O solution of monoperphthalic acid (115 mg/ml). After 12 hr at 0°, 100 ml of Et_2O was added and the solution was washed with 5% $NaHCO_3$ (until basic) and water, and then dried ($MgSO_4$). The solvent was removed under reduced pressure to give 5 g of crude 5,6-epoxide. To a suspension of the epoxide (4.5 g) in 50 ml of EtCOMe was added 5 ml of a solution composed of 15 g of chromium trioxide in 20 ml of H_2O. After stirring for 30 min the mixture was poured into 500 ml of ice H_2O and the precipitate was collected and air-dried for 1 hr. The product was then dissolved in a minimum of $CHCl_3$ and dried ($MgSO_4$). The solvent was removed under reduced pressure and the solid crystallized from EtOH to give 4 g (54%) of **11**: mp 256–258° dec; $[\alpha]D$ −159.2°. *Anal.* ($C_{26}H_{36}O_7$) C, H.

3β,17α-Dihydroxy-16-methylene-*B*-norpregn-5-en-20-one Diacetate (14). To a solution of 44 g (0.102 mol) of **11** dissolved in 300 ml of EtOH free $CHCl_3$ was added slowly 60 g of 87% *m*-chloroperbenzoic acid dissolved in 600 ml of $CHCl_3$. The temperature was maintained below 30° during the addition. The solution was then stirred for 1.5 hr at room temperature and cooled to 10°, and 250 ml of a 20% $NaHSO_3$ solution was added. The mixture was stirred for 10 min and then filtered to give 27.5 g of *m*-chlorobenzoic acid. The organic layer was separated and dried ($MgSO_4$), and the solvent was removed under reduced pressure. The residue was treated with 250 ml of C_6H_6 and the mixture again filtered to give a second crop (20 g) of *m*-chlorobenzoic acid. The remaining *m*-chlorobenzoic acid was extracted from the C_6H_6 filtrate by washing with 3 × 50 ml of 5% $NaHCO_3$ solution. The C_6H_6 solution was then washed with 2 × 100 ml of a 5% Na_2CO_3 solution and the extracts were immediately acidified in the cold. The mixture was extracted with Et_2O, the ether solution dried ($MgSO_4$), and the solvent removed under reduced pressure to give 21 g of 3β,17β-dihydroxy-5,20-dioxo-5,6-*seco*-16-methylenepregnan-6-oic acid diacetate (**12**) as a white foam. To a solution of the crude seco acid (21 g) in 50 ml of dry C_5H_5N was added 24 ml of PhCOCl. The dark mixture was then stirred overnight after which time 10 ml of dry MeOH was added. The mixture was stirred for an additional 30 min and then poured into 800 ml of ice H_2O. The mixture was extracted with Et_2O and the Et_2O solution washed with 1 *N* NaOH solution and H_2O and then dried ($MgSO_4$). Removal of the solvent under reduced pressure gave 42 g of a dark oil. The residue was then subjected to high vacuum (0.1 mm) and heated to 70° at which temperature the MeOCOPh distilled. The residue was then pyrolyzed at this pressure by heating to 200° for 10 min. Trituration of the residue with MeOH gave 7.4 g of a brown solid. Crystallization from MeOH afforded 5.4 g (13%) of **14**: mp 178.5–180.5°; $[\alpha]D$ −205°. *Anal.* ($C_{25}H_{34}O_5$) C, H.

6,7α-Dichloro-3β,17α-dihydroxy-16-methylenepregn-5-en-20-one Diacetate Etherate (15). A solution of 0.5 g (1.20 mmol) of **14** and 0.954 g (2.41 mmol) of phenyl(trichloromethyl)mercury in 5 ml of dry C_6H_6 was refluxed under nitrogen for 48 hr. The mixture was filtered and the solvent removed from the filtrate under reduced pressure. The residue was chromatographed through 15 g of silica gel. From a 2.5% EtOAc-C_6H_6 eluent was obtained 0.397 g of a yellow foam. Crystallization from Et_2O gave 0.186 g (31.5%) of **15**: mp 125° dec; $[\alpha]D$ −201.5°. *Anal.* ($C_{26}H_{34}Cl_2O_5 \cdot C_4H_{10}O$) C, H.

3β,17α-Diacetoxy-2′,2′,6,6-tetrafluoro-(16*R*)-spiro(5,7β-cyclopregnane-16,1′-cyclopropan)-20-one (16). To a refluxing solution of 1 g (2.41 mmol) of **14** dissolved in 50 ml of dry diglyme was added dropwise (over a 45-min period) a solution of 5.52 g (36.2 mmol) of sodium chlorodifluoroacetate dissolved in 50 ml of the same solvent. After the addition was completed the mixture was refluxed for an additional 15 min. The mixture was then cooled and filtered and the solvent removed under high vacuum. The residue was chromatographed on 25 g of silica gel. Elution with a 5% EtOAc-C_6H_6 solution afforded 0.6 g of crude product. Crystallization from CH_2Cl_2-Et_2O afforded 0.26 g (21.7%) of **16**: mp 190–192.5°; $[\alpha]D$ −31.1°. *Anal.* ($C_{27}H_{34}F_4O_5$) C, H, F.

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Synthesis and Biological Activities of Substituted Glycyrrhetic Acids

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18β-Glycyrrhetic acid (**1b**) was converted in good yield to the 3-oxo-4,4-bis(nor-18β-olean-4-ene) derivative **11a** in 25% overall yield. Derivatives of **1b** substituted in the A, B, C, and E ring were also prepared. Several 11-deoxoglycyrrhetic acid derivatives exhibited anti-DCA activity. In particular, when administered subcutaneously, 3-oxo-18β-olean-12-en-30-oic acid (**2d**) had about 75% the activity of spironolactone administered subcutaneously. Several compounds also exhibited weak antiviral and antiinflammatory properties.

The medicinal and biological properties associated with glycyrrhizin (**1a**) and its aglycone 18β-glycyrrhetic acid (**1b**) are well documented.[1] The biological properties in laboratory animals and *in vitro* assays which have been reported for 18β-glycyrrhetic acid and its derivatives are antiulcer,[2] antiinflammatory,[2a,3] sodium ion retention,[2a,4] antihormonal,[5] and antineoplastic.[6] In recent years the antigastric ulcer activity and the effects on mineral metabolism of glycyrrhetic acid derivatives have been reported in man.[2a,7] The ammonium salt of glycyrrhizin is also used as a commercial sweetening agent.[1]

Although the biological activities of glycyrrhetic acid

derivatives suggest that they act by their effect on enzyme synthesis or activation, their mechanism of action is not understood. A study of structure–activity relationships for **1b** was made with the purpose of separating the property of sodium ion retention from other potentially useful medicinal properties. The derivatives which were obtained were tested in biological assays modeled to measure anti-DCA, antiinflammatory, antiviral, and antiulcer properties.

Since there are data available on the metabolism of glycyrrhetic acid derivatives,[2a] the synthetic program was directed toward modifying positions in glycyrrhetic acid and 11-deoxoglycyrrhetic acid which are near those known to be accessible to mammalian cell enzyme systems.[2a] The positions which were selected to be modified in the oleane skeleton are those carbon atoms in ring A and C-30 in ring E.

Unsaturated Derivatives. Glycyrrhetic acid derivatives possessing an alkenyl linkage at carbon atoms 1, 4, and 5 were prepared. The C-1 unsaturated 3-ketone **3a** was prepared by treatment of the 3-enol acetate derived from **2a** followed by C-2 bromination and dehydrobromination with MgO–DMF (Scheme I).

The synthesis of a glycyrrhetic acid derivative containing the 3-keto-Δ^4 moiety was an attractive goal in order to accomplish the desired transformations around C-4. It was achieved in good overall yield as follows. The known isopropylidene derivative **4** was selectively oxidized with osmium tetroxide in pyridine to the glycol **5** which was cleaved with lead tetraacetate in benzene in quantitative yield to the 5α-ketone **6a.** The ketone could be isomerized readily on brief treatment with alkoxide to the more stable 5β-ketone **6b.** The known steric hindrance at and diminished reactivity of the C-11 and C-30 carbonyl groups in glycyrrhetic acid allowed a selective addition of methylmagnesium bromide to the ketone **6a** in good yield. Dehydration of the resulting alcohol **7** with thionyl chloride and pyridine to **8** followed by cleavage of the double bond *via* **9a,** as described above for the glycol **5,** gave the triketone **10a** in excellent yield. Because of the steric hindrance about the C-12 double bond, the transformation of **8** to **10a** also could be accomplished by reductive ozonolysis. Cyclization of the triketone **10a** in the presence of KOH in methanol proceeded quantitatively to give the 3-keto-Δ^4-methyl ester **11a.** Alternatively, the addition of **10a** to refluxing collidine in the presence of lithium iodide yielded the carboxylic acid **11b.** In like manner, other methyl esters of glycyrrhetic acid derivatives were converted to their corresponding acids without isomerization at C-18 in fair to high yields. The overall yield of **11b** from **1b** was 25%. Epimerization at C-18 of **11b** to **11c** occurred in the presence of strong acid.

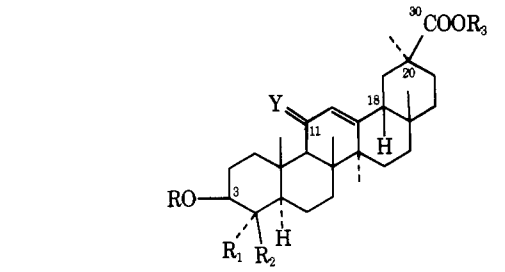
The introduction of a C-5 double bond in glycyrrhetic acid was accomplished by the alkylation of **11a** with methyl iodide in the presence of potassium *tert*-butoxide. A mixture of **11d** and **12a** was obtained, the proportion of which could be varied with reaction conditions. Reduction of the ketone **11d** with lithium tri-*tert*-butoxyaluminum hydride gave the alcohol **12b** (Scheme II).

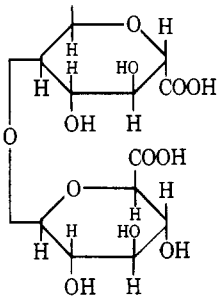
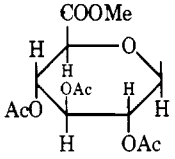
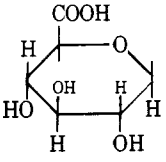
C-3 Substituted Derivatives. In addition to the known substances glycyrrhizin (**1a**) and 3-dehydroglycyrrhetic acid methyl ester (**2a**), two new ethers of glycyrrhetic acid derivatives were prepared for biological evaluation. Treatment of **1b** with TMS yielded the lipid-soluble ether **1c** and reaction of β-bromoglucose pentaacetate with the benzyl ester of **1b,** followed by hydrogenolysis and hydrolysis, gave the water-soluble glycoside **1i.**

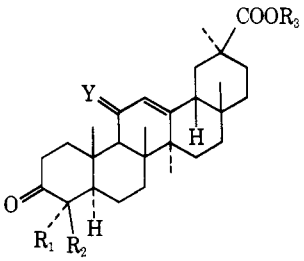
C-4 and C-5 Substituted Derivatives. The 4,4-bis(nor-glycyrrhetic acid) derivative **2b** was obtained by the reduction of **11b** with lithium and ammonia, followed by reduction with lithium tri-*tert*-butoxyaluminum hydride and oxidation of the resulting alcohol with chromic acid. The 3-hydroxy-4,4-bis(nor-5-ene) **12d** was obtained by the hydride reduction of an intermediate 3-keto-5-ene, which was derived by the formation of the enol of **11b** and acidification with acetic acid.

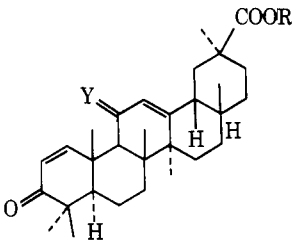
Epoxidation of **12b** gave the 5,6-oxide **13a.** The acids

Scheme I[a]



1	Y	R	R_1	R_2	R_3
a	O		CH_3	CH_3	H
b	O	H	CH_3	CH_3	H
c	O	$Si(CH_3)_3$	CH_3	CH_3	H
d	H_2	H	CH_3	CH_3	H
e	H_2	H	CH_3	CH_3	CH_3
f	H_2	$OC(CH_2)_2COOH$	CH_3	CH_3	H
g	H_2	$Si(CH_3)_3$	CH_3	CH_3	H
h	H_2		CH_3	CH_3	H
i	H_2		CH_3	CH_3	H
j	H_2	H	H	H	H



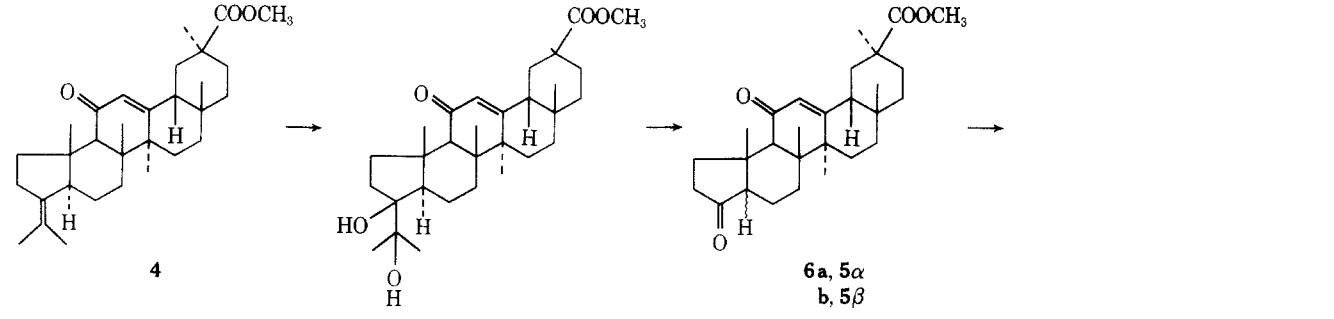


2	Y	R_1	R_2	R_3
a	O	CH_3	CH_3	CH_3
b	O	H	H	H
c	H_2	CH_3	CH_3	CH_3
d	H_2	CH_3	CH_3	H
e	H_2	CH_3	H	CH_3
f	H_2	CH_3	H	H

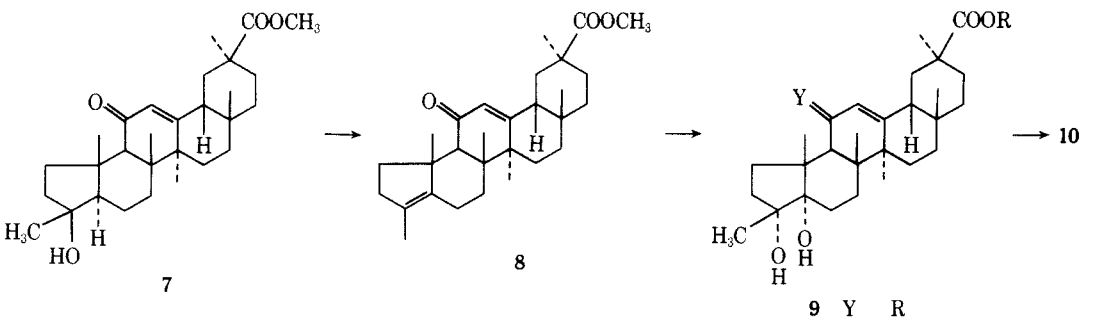
3	Y	R	
a	O	CH_3	(18ξ)
b	H_2	CH_3	
c	H_2	H	

[a] Bonds attached to positions 8, 10, 14, 17, and 20 denote methyl groups.

Scheme II

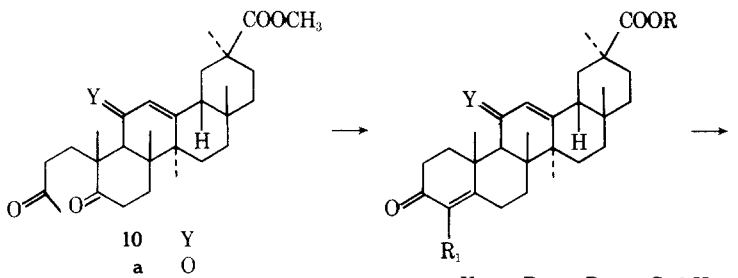


4 **5** **6a, 5α** **b, 5β**



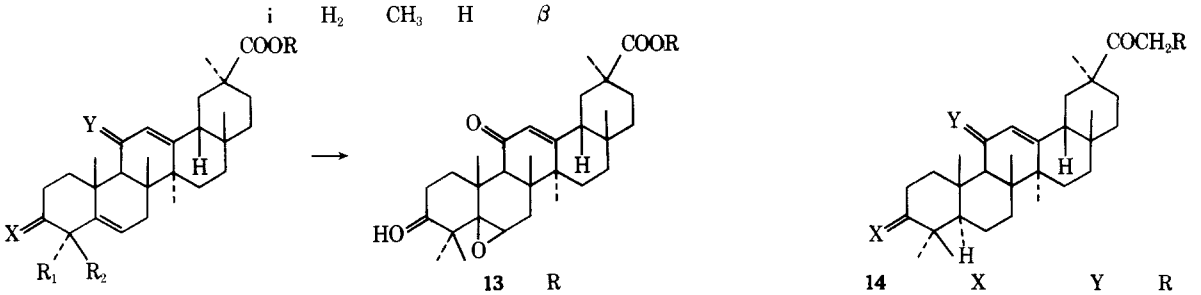
7 **8** **9** **10**

9	Y	R
a	O	CH_3
b	O	H
c	H_2	CH_3



10	Y
a	O
b	H_2

11	Y	R_1	R	C-18 H
a	O	H	CH_3	β
b	O	H	H	β
c	O	H	H	α
d	O	CH_3	CH_3	β
e	O	CH_3	H	β
f	H_2	H	CH_3	β
g	H_2	H	H	β
h	H_2	CH_3	CH_3	β
i	H_2	CH_3	H	β



12	X	Y	R_1	R_2	R
a	O	O	CH_3	CH_3	CH_3
b	OH, H	O	CH_3	CH_3	CH_3
c	OH, H	O	CH_3	CH_3	H
d	OH, H	O	H	H	H
e	O	H_2	CH_3	CH_3	CH_3
f	O	H_2	CH_3	CH_3	H
g	OH, H	H_2	H_2	H	H

13	R
a	CH_3
b	H

14	X	Y	R
a	O	O	OAc
b	-OAc, ...H	O	OH
c	-OH, ...H	O	OH
d	-OAc, ...H	O	H
e	-OH, ...H	O	H
f	O	O	H
g	-steroyl, ...H	O	H
h	-OAc, ...H	H_2	H
i	-OH, ...H	H_2	H
j	O	H_2	H
k	-*O*-steroyl, ...H	H_2	H

12c and 13b were obtained (*vide supra*) from the corresponding methyl esters.

C-30 Substituted Derivatives. The C-20 hydroxymethyl ketone **14a** derived from glycyrrhetic acid was reported to have antiinflammatory properties.[3g] For this reason the corresponding methyl ketone **14f** and the more lipid-like stearate ester **14g** were prepared.

11-Deoxoglycyrrhetic Acid Derivatives. The relative inertness of the double bond at C-12 in glycyrrhetic acid and its derivatives to catalytic hydrogenation and selective oxidation is well established (*vide supra*). Experiments in this laboratory have shown that even the reduction of the unsaturated ketone with metal and ammonia when carried out on the lithium salt proceeds slowly or not at all. Therefore, the preparation of 11-deoxoglycyrrhetic derivatives analogous to those described above for glycyrrhetic acid proceeded by reactions described above for the synthesis of substituted glycyrrhetic acids.

Biological Results. All compounds were tested for anti-DCA activity at 2.4 mg by the method of Kagawa[8] and measured against spironolactone which exhibited activity at a median effective dose (MED) of 0.33 mg when administered subcutaneously. Compounds **2d** and **1d**, when administered subcutaneously, exhibited anti-DCA activity with a MED of 0.4 and 1.33 mg, respectively. They were inactive when administered intragastrically (ig). Compounds **1e** and **1c** showed some anti-DCA activity at 2.4 mg and all other compounds were inactive.

Compounds were also tested for antiinflammatory activity in an adjuvant arthritis assay which was modified so that a compound was administered ig once daily starting on the day of inoculation and continuing through day 19.[9] The response was measured on day 20. Compounds **5**, **10a**, and **14h** exhibited some activity at 5 mg/kg. Compounds **1b,c**, **2c,d,g**, **9a**, **11b,c**, **12d**, **13b**, and **14h,j** were inactive when tested ig.

Compounds were tested for antiviral activity by the quantitative hemabsorption technique of Finter[10] modified to use primary cell cultures of rhesus monkey kidney for vaccinia and/or influenza A/575 viruses. Compounds **1b** and **3a** showed activity at 50 γ with slight cytotoxicity against vaccinia virus. Compounds **1c**, **2d**, and **12b** showed activity at 625 γ with slight cytotoxicity against influenza A virus. Most of all the other substances exhibited no antiviral activity in these assays.

When the compounds were tested in a Shay rat assay[11] for antiulcer activity ig four compounds, **1e**, **5**, **14g**, and **14h**, showed activity at 50, 50, 50, and 10 mg, respectively. Compounds **1,b,d,f,i**, **2,c,d,g**, **3a–c**, **9a,b**, **11b–i**, **12a–g**, **13a,b**, and **14f–j** were not active at 50 mg (ig).

Discussion

A study of structure and activity relationships of glycyrrhetic acid derivatives, particularly with respect to biological properties associated with sodium and potassium ion balance, showed that removal of the 11-oxo function leads to derivatives exhibiting anti-DCA properties in contrast to the DCA-like properties of the parent compound. However, structural specificity for this property, as well as antiulcer properties, appears high, for many minor modifications in the nucleus of the parent substances leads to loss of activity exhibited by the parent compound.

Experimental Section

Melting points were taken on a Fisher-Johns hot-stage apparatus and are uncorrected. Nmr spectra were taken on a Varian A-60A or a Varian T-60. All spectra are 60 MHz unless specified otherwise. Location of peaks are in hertz using TMS as an internal standard. Ir spectra were recorded in Chf unless specified otherwise. Uv spectra were in MeOH. Tlc runs were on 7.6-cm microscope slides covered with a 0.25-mm thickness of Woelm F silica, with a magnesium aluminum silicate binder. Visualization of spots was by phosphomolybdic acid, 5% in EtOH (wt/vol), followed by heat. Column chromatography used Mallinckrodt SilicAR CC-4 and CC-7 silicic acid and Baker SiO_2. The weight ratio of adsorbent to material was 100:1. Materials were applied as benzene solutions and, unless indicated otherwise, eluted with benzene containing increasing amounts of EtOAc. Where analyses are indicated only by symbols of the elements, analytical results obtained for those elements were within ±0.4% of the theoretical values.

3β-Trimethylsiloxy-11-oxo-18β-olean-12-en-30-oic Acid (1c). A solution of 1.3 g of **1b**, 50 ml of pyridine, 2.5 g of hexamethyldisilazane, and 1.5 g of trimethylchlorosilane was allowed to stand for 2 hr and then poured into 600 ml of water. After the mixture was cooled, the solid was collected by filtration and dried. The product was purified by extraction into benzene and evaporated to dryness. Product isolated in this manner gave **1c**: 1.07 g (71%); mp 244–246°. *Anal.* ($C_{33}H_{54}O_4Si$) C, H.

3β-Hydroxy-18β-olean-12-en-30-oic Acid 3-Hemisuccinate (1f). A mixture of 0.5 g of **1d**, 500 mg of succinic anhydride, and 5 ml of pyridine was heated at reflux for 6 hr, cooled, and diluted with H_2O. The precipitate was collected by filtration and dried to yield 500 mg of crude **1f**. Recrystallization from CH_2Cl_2–MeOH gave mp 289–292°. *Anal.* ($C_{34}H_{52}O_6$) C, H.

3β-Trimethylsiloxy-18β-olean-12-en-30-oic Acid (1g). When 1.3 g of **1d** was converted to **1g** as in the procedure for the preparation of **1c**, 1 g (70%), mp 216–219°, was obtained. *Anal.* ($C_{33}H_{56}O_3Si$) C, H.

3β-[(Methyl-β-D-glucopyranuronosyl-2,3,4-*O*-triacetyl)oxy]olean-12-en-30-oic Acid Benzyl Ester (1h). The technique of Becker was used.[12] A solution of 5 g of the benzyl ester of **1b**, prepared from benzyl chloride and the sodium salt of **1b**, in 250 ml of anhydrous benzene was heated to reflux and a solution of 15 g of methylbromotriacetyl glucuronate in 225 ml of benzene was added over a period of 30 min while benzene was collected by distillation to maintain a constant volume. During the addition of the bromide 20.7 g of freshly prepared dry Ag_2CO_3 suspended in benzene was added in portions. The mixture was refluxed for 20 min, cooled, and filtered. The filtrate was evaporated to dryness and the residue purified by column chromatography on silica gel in benzene. The desired material, 3.27 g (34%), was eluted from the column with benzene–EtOAc (9:1). A small amount of **1h** was purified for analysis by slow evaporation of a solution of **1h** in MeOH in a stream of N_2 which yielded crystals: mp 118–121°; λ_{max} 5.68 and 5.79 μ; nmr max at 120 (9 H, $COCH_3$), 224 (3 H, OCH_3), 308 (H, C-12 H), 440 Hz (5 H). *Anal.* ($C_{50}H_{70}O_{12}$) C, H.

3β-[(β-D-Glucopyranuronosyl)oxy]olean-12-en-30-oic Acid (1i). A solution of 1.5 g of crude **1h** and 75 ml of 0.1 *N* KOH was refluxed for 1 hr, cooled, and poured into a mixture of 400 ml of 0.5 *N* HCl and 150 ml of saturated brine. The suspension was extracted three times with EtOAc. The extract was dried over $MgSO_4$ and evaporated to dryness. The solid was hydrogenolyzed in 100 ml of HOAc in the presence of palladium black catalyst at 37 psi at 25° for 2.5 hr. The catalyst was removed by filtration and the filtrate distilled to a small volume and partitioned between 0.5 *N* HCl–brine and EtOAc and isolated in a manner described above. The product, **1i**, weighed 712 mg (73%): mp 220° dec; λ_{max} (KBr) 2.92, 3.57–4.16, and 5.81 μ. *Anal.* ($C_{36}H_{60}O_{11}$) C, H.

3β-Hydroxy-4,4-desmethyl-18β-olean-12-en-30-oic Acid (1j). The ketone **11g** (4.25 g) was treated with lithium in liquid NH_3 as described for **12d**. After that the reaction mixture was warmed to room temperature, diluted with H_2O (1.2 l.), and acidified with concentrated HCl. The resulting solid was washed with water and dried. The dry solid (4.1 g) was dissolved in 150 ml of THF and treated with 9.1 g of lithium tri-*tert*-butoxyaluminum hydride overnight at room temperature. The reaction mixture was poured on ice, acidified with concentrated HCl, and extracted twice with Et_2O–EtOAc. The combined organic extract was washed twice with H_2O and once with saturated brine, decolorized, evaporated to a solid, and recrystallized (EtOAc) to give two pure crops of **1j** with an aggregate weight of 2.48 g (58%): mp 278–280°; λ_{max} 2.71, 2.76, 2.84, 5.74, and 5.88 μ; nmr (C_5D_5N) max at 230 (C-3 H) and 330 Hz (C-12 H). *Anal.* ($C_{28}H_{44}O_3$) C, H.

4,4-Desmethyl-3,11-dioxo-18β-olean-12-en-30-oic Acid (2b). Liquid NH_3 (200 ml) was distilled into a 500-ml round-bottom flask which was insulated. Then 11 ml of 2-propanol and Li wire (3.2 mm in diameter, 5 mm in length) were added. After complete

solution, 4.00 g of **11b** was suspended in 100 ml of THF and added to the NH_3 solution. Li wire (4-cm lengths) was added in increments, the addition always being made when the blue color disappeared. After seven additions, the NH_3 was allowed to boil off. The residual material was diluted to 1.1 l. with H_2O and extracted with three 150-ml portions of 1:1 Et_2O–EtOAc. The organic fractions were combined, washed twice with H_2O and once with saturated brine, filtered, and evaporated to a solid. The solid was suspended in 500 ml of acetone. CrO_3–H_2SO_4 (8 *N*) was added slowly with vigorous stirring until the acetone solution was dark brown. The solution was decanted from the green gum, diluted to 1.7 l. with H_2O, and extracted with three 200-ml portions of 1:1 Et_2O–EtOAc. The combined organic fractions were washed as above, decolorized, filtered, and evaporated to yield 4 g of solid. The solid was dissolved in $CHCl_3$ and applied to a CC-4 silicic acid column. Elution with $CHCl_3$ was followed by elution with 99:1 $CHCl_3$–EtOH, which eluted 862 mg of the desired material **2b** and, slightly before it, the undesired 12,13-dihydro compound. Recrystallization from benzene containing a small amount of MeOH gave 750 mg (18%): mp 299–303°; λ_{max} 249.5 nm (ϵ 11,800); λ_{max} 5.85, 6.02, shoulders at 5.73 and 6.15 μ. *Anal.* ($C_{28}H_{40}O_4$) C, H.

Methyl 3-Oxo-11β-olean-12-en-30-oate (2c). When 2 g of **1e** was oxidized with CrO_3 according to the procedure described above, 2 g of **2c** was obtained. Recrystallization from MeOH gave mp 179–181°. *Anal.* ($C_{31}H_{48}O_3$) C, H.

3-Oxo-18β-olean-12-en-30-oic Acid (2d). To a stirred mixture of 0.5 g of **1d** in 40-ml parts of acetone was added slowly a solution of 0.5 ml of 4 *N* aqueous chronic acid in 8 ml of acetone. The reaction mixture was stirred until tlc indicated that the starting material was consumed and then 3 ml of 2-propanol and 100 ml of water were added successively. The mixture was concentrated by evaporation under reduced pressure, diluted with water, and filtered. The product was collected by filtration, washed with water, and air-dried to yield 480 mg (98%) of **2d.** Recrystallization from acetone gave mp 270–273°: λ_{max} 2.8, 3.0, 5.8, 5.89, and 6.0 μ. *Anal.* ($C_{30}H_{46}O_3$) C, H.

3-Oxo-4-desmethyl-18β-olean-12-en-30-oic Acid (2e). This substance was prepared in the same manner as **2f,** starting with **11i. 11i** (2.7 g) yielded, after recrystallization from CH_2Cl_2–MeOH, 333 mg of product: mp 230–236°; λ_{max} 2.84, 3.57–4.17, 5.74, and 5.86 μ; nmr max 59 (C-4 Me), >71 Hz (no Me groups). Comparison of the ORD–CD (Jasco J-20, 93 mg % in dioxane, 1-cm cell path length) of **2e** with that of **11i** shows that the 4-methyl group is in an equatorial (α) position, as expected. *Anal.* ($C_{29}H_{44}O_3 \cdot 0.25H_2O$) C, H.

A precise mass determination was made on a CEC 21-110B, using Nier peak matching. Perfluorokerosene (*m/e* 442.972837) was used as a reference: calcd ($C_{29}H_{44}O_3$), 440.3290; found, 440.3283.

3-Oxo-4,4-desmethyl-18β-olean-12-en-30-oic Acid (2f). 11g (2 g) was treated with Li–NH_3 and CrO_3 as described for the preparation of **2b.** The product **2f** was obtained. Recrystallization from CH_2Cl_2–MeOH gave 500 mg (25%): mp 254–255°; λ_{max} 5.75, 5.88, and 10.98 μ; nmr (C_5D_5N) max at 321 Hz. *Anal.* ($C_{28}H_{42}O_3$) C, H.

Methyl 3,11-Dioxo-18ξ-oleana-1,12-dien-30-oate (3a). A solution of 27 g of **2a,** 3 g of *p*-TsOH, and 800 ml of isopropenyl acetate was concentrated by slow distillation over a period of 16 hr to a volume of 400 ml. The solution was diluted with 8 ml of pyridine and 400 ml of benzene. The solution was washed with water, dried over $MgSO_4$, and distilled to dryness. The crystalline residue was purified by column chromatography on 1.8 kg of silica gel in benzene. Elution with EtOAc–benzene (3:97) gave 19.3 g of 3-enol acetate of **2a:** λ_{max} 249 nm (ϵ 11,500). To a solution of the crude enol acetate in 1 l. of HOAc was added a solution of 8 g of Br_2 in 74 ml of HOAc over a period of 1.5 hr. The solution was then added dropwise with stirring to 1 l. of H_2O. The bromide was collected by filtration, washed with H_2O, and dried at 20° to give 20.1 g of crude product. The crude bromide was added in portions to a refluxing mixture of 18 g of MgO and 306 ml of DMF over a period of 15 min with vigorous stirring in an atmosphere of N_2. The mixture was stirred at reflux for 15 min, cooled, and added with stirring to a mixture of 305 ml of concentrated HCl and 0.5 kg of ice. The crude product was collected by filtration and dried to yield 16.7 g of crude **3a.** Purification of the crude product was purified by column chromatography on 1.2 kg of silica gel. Elution of the column with EtOAc–benzene (1:19) gave 4.4 g of crude **3a.** Recrystallization from CH_2Cl_2–MeOH gave mp 213–215°. *Anal.* ($C_{31}H_{44}O_4$) C, H.

Methyl 3-Oxo-18β-oleana-1,12-dien-30-oate (3b). The 1-dehydro ester **3b** was prepared according to the procedure described for **3a** starting with **2c.** Accordingly, 5.75 g of **2c** yielded 4.75 g of crude 3-enol acetate of **2c,** which was converted by bromination and dehydrobromination to 4.15 g of crude **3b.** Purification of **3b** by column chromatography on 450 g of silica gel yielded 1.2 g of **3b** when the column was eluted with benzene–ethyl acetate (99:1). Recrystallization from MeOH gave mp 147–150°: λ_{max} 228 nm (ϵ 10,000). *Anal.* ($C_{31}H_{40}O_3$) C, H.

3-Oxo-18β-oleana-1,12-dien-30-oic Acid (3c). When a mixture of 250 mg of **3b** and KOH (10%) in MeOH was refluxed for 24 hr and acidified with HOAc, 200 mg (79%) of **3c** was obtained. Recrystallization from CH_2Cl_2–Me_2CO gave mp 273–283°: λ_{max} 228.5 nm (ϵ 10,040). *Anal.* ($C_{30}H_{44}O_3$) C, H.

Methyl 3-Nor-4α-hydroxy-4β-(hydroxylisopropyl)-4,4-desmethyl-11-oxo-18β-olean-12-en-30-oate (5). To a stirred mixture of 5 g of methyl 3-nor-4-isopropylidene-4,4-desmethyl-11-oxo-18β-olean-12-en-30-oate† (4) and 28.1 ml of pyridine was added over a period of 30 min, 57.1 ml of a solution of 50 g of OsO_4 dissolved in 982 ml of pyridine. Stirring was continued for an additional 30 min and then 3 ml of the above OsO_4–pyridine solution was added. After stirring for an additional 30 min, 5 ml of the OsO_4–pyridine solution was added and the mixture stirred for 10 min. The mixture was quenched with a solution of 5.7 g of $NaHSO_3$ in 86 ml of H_2O and 42.2 ml of pyridine. The mixture was heated on a steam bath for about 2 hr and then added rapidly to a mixture of about 1500 ml of water and ice. Stirring was continued for about 64 hr after which time 1 l. of water was added. The mixture was filtered and the collected precipitate was washed with water and air-dried. The crude product was dissolved in a 1:1 mixture of $CHCl_3$ and MeOH and filtered. Benzene was added to the filtrate and that solution was evaporated to dryness to yield 5.3 g (99%) of crude **5.** Recrystallization from CH_2Cl_2–MeOH–benzene gave mp 199–201°: nmr max 51, 60, 68, 69, 76, 85, 223, and 342 Hz. *Anal.* ($C_{31}H_{44}O_5$) C, H.

Methyl 3-Nor-4,4-desmethyl-4,11-dioxo-18β-olean-12-en-30-oate (6a). **5** (17 g) was dissolved in 879 ml of benzene and filtered. The filtrate was treated with a slurry of 18 g of $Pb(OAc)_4$ in 220 ml of benzene in portions until tlc indicated that the starting material was depleted. After the addition of 5.5 ml of ethylene glycol, the mixture was stirred for 15 min and filtered. The filtrate was washed with water, dried over anhydrous $MgSO_4$, and evaporated to dryness to yield crude product **6a.** Recrystallization from CH_2Cl_2–MeOH gave 13.3 g (89%): mp 210–213°; λ_{max} 249 nm (ϵ 11,000); nmr max 50, 60, 69, 84, 223, and 347 Hz. *Anal.* ($C_{28}H_{40}O_4$) C, H. The 5α-ketone **6a** could be isomerized with KOH–MeOH to the more stable 5β-ketone **6b** as previously described.[14]

Methyl 3-Nor-4-hydroxy-4-desmethyl-11-oxo-18β-olean-12-en-30-oate (7) and **Methyl 3-Nor-4-desmethyl-11-oxo-18β-oleana-4,12-dien-30-oate (8).** To a solution of 10 g of **6a** and 300 ml of a 1:1 THF–ethyl ether solution was added dropwise 8 ml of 3 *N* MeMgBr in ether solution. That mixture was allowed to react until tlc indicated that the starting material was consumed. AcOH and water were added to the mixture which was then concentrated to a small volume and diluted with benzene. The benzene solution was washed with water and then with $NaHCO_3$ and dried over $MgSO_4$. After evaporation to dryness, there was obtained 10 g (97%) of crude **7,** mp 201–203°, which exhibited nmr max at 50, 67, 78, 82, 222, and 345 Hz.

A stirred solution of 36.1 g of crude **7** in 283 ml of pyridine was cooled under a nitrogen atmosphere to −15° in a NaCl–ice water bath. To that solution was added 11.8 ml of $SOCl_2$ and the solution was allowed to stir for about 20 min. Then 633 ml of benzene containing 25.3 ml of EtOH was added in one portion. The resulting solution was washed with 4 *N* HCl until the reaction mixture was acidic and then it was washed successively with water and aqueous $NaHCO_3$, dried over anhydrous $MgSO_4$, filtered, and evaporated to dryness to yield **8.** Recrystallization from CH_2Cl_2–MeOH gave 16.9 g (49%): mp 199–202°; λ_{max} 248.5 nm (ϵ 11,100); λ_{max} 5.78, 6.02, and 6.20 μ. *Anal.* ($C_{29}H_{42}O_3$) C, H.

Methyl 3-Nor-4α,5α-dihydroxy-4-desmethyl-11-oxo-18β-olean-12-en-30-oate (9a). A mixture of 1.0 g of **8,** 9.82 ml of pyridine, and 0.675 g of OsO_4 was stirred for 30 min. Then 9.82 ml of pyridine and 1.2 g of sodium bisulfite in 20 ml of water were added and that solution was stirred at 50° for 30 min. The reaction mixture was diluted with 110 ml of water and the precipitate

† This compound was prepared in 70% yield by the method described in ref 3.

which formed was collected by filtration, washed with water, and dried under reduced pressure to yield crude **9a**, 1.1 g (99%).‡ Recrystallization from CH_2Cl_2–benzene gave mp 204–215°: λ_{max} (KBr) 2.90, 5.78, 6.00, and 6.19 μ. *Anal.* ($C_{29}H_{44}O_5$) C, H.

3-Nor-4,5-dihydroxy-4-desmethyl-11-oxo-11β-olean-12-en-30-oic Acid (9b). **9a** (2 g), 55 ml of 2,4,6-trimethylpyridine, and 2.0 g of lithium iodide were combined with stirring under a nitrogen atmosphere at room temperature. That mixture was heated slowly to reflux and the refluxing was continued for 3.5 hr. After cooling slightly, the reaction mixture was added to a stirred slurry of ice–water and hydrochloric acid. The precipitate which formed was recovered by filtration and dried to yield 1.3 g of **9b**. Recrystallization from MeOH gave mp 275–277°: λ_{max} 3.3 (broad), 5.85, 6.0, and 6.1 μ. *Anal.* ($C_{28}H_{42}O_5$) C, H.

Methyl 3-Nor-4α,5α-dihydroxy-4-desmethyl-18β-olean-12-en-30-oate (9c). A solution of 1.04 g of **9a** in 200 ml of HOAc was shaken with 0.1 g of prereduced Pt in an atmosphere of H_2 (56 psi) for 3 days. The catalyst was removed by filtration. The filtrate was distilled to dryness under reduced pressure to yield **9c**. Recrystallization from EtOH gave 370 mg: mp 225–230°; λ_{max} (KBr) 2.89, 3.00, and 5.77 μ; nmr max at 222 (singlet, $-OCH_3$) and 325 (multiplet, C-12 H). *Anal.* ($C_{29}H_{46}O_4$) C, H.

Methyl 4,5-*seco*-4,4-Desmethyl-3,5,11-trioxo-18β-olean-12-en-30-oate (10a). **9a** (17.4 g) was treated with $Pb(OAc)_4$ according to the procedure for the preparation of **6a**. Crude **10a** (14.4 g, 77%) was obtained. Recrystallization from CH_2Cl_2–MeOH gave mp 173.5–177°: λ_{max} 249.5 nm (ε 12,700); nmr max at 51, 70, 81.5, 84, 127.5, 223, 350 Hz. *Anal.* ($C_{29}H_{42}O_5$) C, H.

Alternate Method. To a stirred solution of 52.8 g of **8** in 158.4 ml of MeOH, cooled in a 2-propanol–Dry Ice bath, was added ozone for about 1.5 hr until the solution acquired a blue color. The system was flushed with oxygen for about 10 min, and 40 g of Zn and 420 ml of HOAc were added. Stirring was continued while the reaction mixture was heated on a steam bath to a temperature of about 27°. Then 745 ml of $CHCl_3$ was added and the mixture was filtered. The filtrate was washed with water and then with aqueous $NaHCO_3$, dried over anhydrous $MgSO_4$ and Na_2SO_4, and evaporated to dryness under reduced pressure to yield 54 g (95%) of crude **10a**.

Methyl 4,4-Desmethyl-3,11-dioxo-18β-oleane-4,12-dien-30-oate (11a). A mixture of 8.8 g of **10a**, a solution of 2 g of KOH dissolved in 79.2 ml of MeOH, and 158.4 ml of MeOH was stirred at reflux under N_2 for 30 min, then cooled, and neutralized with 7.35 ml of HOAc. To that solution was added 50 ml of water and the mixture was evaporated almost to dryness under reduced pressure. The crystalline material was triturated with water, filtered, and dried under reduced pressure to yield 7.70 g (100%) of **11a**, mp 224–228°. Recrystallization from Me_2CO–hexane gave mp 237.5–238°; λ_{max} 243 nm (ε 12,500); nmr max at 51 Hz. *Anal.* ($C_{29}H_{40}O_4$) C, H.

4,4-Desmethyl-3,11-dioxo-18β-oleana-4,12-diene-30-oic Acid (11b). When **11a** was refluxed with 2,4,6-trimethylpyridine and LiI according to the procedure outlined for the preparation of **9b**, **11b** was obtained in 90% yield. Recrystallization from Me_2CO–CH_2Cl_2 gave mp 290–300°: λ_{max} 243 nm (ε 25,000); nmr max at 53, 72, 82.5, 84, and 91 Hz. *Anal.* ($C_{28}H_{38}O_4$) C, H.

4,4-Desmethyl-3,11-dioxo-18α-oleana-4,12-dien-30-oic Acid (11c). A mixture of 7.4 g of **11a**, 155 ml of HOAc, and 17.6 ml of concentrated HCl was heated on a steam bath for about 45 hr. After that time the reaction mixture was rapidly poured into a vigorously stirred mixture of ice and water. That mixture was filtered and the precipitate which collects was washed with water and air-dried. The resulting product was triturated with MeOH and filtered to yield crude **11c**. Recrystallization from MeOH gave 3.4 g (40%): mp >300°; λ_{max} 241 nm (ε 13,000); nmr max at 47, 78, 82, 95, and 348 Hz. *Anal.* ($C_{28}H_{38}O_4$) C, H.

Methyl 4-Desmethyl-3,11-dioxo-18β-oleana-4,12-dien-30-oate (11d). Continuation of elution of the column mentioned in the preparation of **12a** gave **11d** when the eluent was PhH–EtOAc (95:5). The product weighed 5.89 g (46%). Recrystallization from CH_2Cl_2–MeOH gave mp 219–222°: λ_{max} 249 nm (ε 26,800); λ_{max} 5.78 and 6.02 μ; nmr max at 107 Hz (3 H, C-4 Me). *Anal.* ($C_{30}H_{42}O_4$) C, H.

4-Desmethyl-3,11-dioxo-18β-oleana-4,12-dien-30-oic Acid (11e). The substance was prepared in 47% yield from 3.91 g of **11d**. Recrystallization from aqueous MeOH gave 1.80 g: mp 278–284° dec; λ_{max} 249 nm (ε 25,000); λ_{max} 3.57–4.16, 5.73, 5.78, and 6.02 μ. *Anal.* ($C_{29}H_{40}O_4$) C, H.

Methyl 3-Oxo-4,4-desmethyl-18β-oleana-4,12-dien-30-oate (11f). When 920 mg of **9c** was cleaved with $Pb(OAc)_4$ as in the procedure for the preparation of **10a**, about 920 mg of crude diketone **10b** was obtained. The crude product then was treated with KOH–MeOH according to the procedure for the preparation of **11a** to yield **11f**. Recrystallization from MeOH gave 106 mg: mp 207–209°; λ_{max} 241 nm (ε 16,200); λ_{max} 5.78, 5.99, and 6.17 μ; nmr max at 221 ($-OCH_3$), 324 (broad, C-12 H) and 345 Hz (C-4 H). *Anal.* ($C_{29}H_{42}O_3$) C, H.

3-Oxo-4,4-desmethyl-18β-oleana-4,12-dien-30-oic Acid (11g). When 1.75 g of **11f** was treated with 2.0 g of LiI according to the procedure for the preparation of **9b** and the product was recrystallized from CH_2Cl_2–MeOH, 1.0 g of **11g**, mp 259–264°, was obtained: λ_{max} 241 nm (ε 15,700); λ_{max} 3.57–4.16, 5.74, 5.88, and 5.98 μ. *Anal.* ($C_{28}H_{40}O_3$) C, H.

Methyl 4-Desmethyl-3-oxo-18β-oleana-4,12-dien-30-oate (11h). Elution of the column mentioned in **12e** yielded **11h** immediately after **12e**. **11h** was recovered from the column in 44% yield and was recrystallized from CH_2Cl_2–MeOH: mp 170–172°; λ_{max} 249.5 nm (ε 16,200); λ_{max} 5.79 and 6.03 μ; nmr max at 109 (C-4 Me) and 328 Hz (C-12 H). *Anal.* ($C_{30}H_{44}O_3$) C, H.

3-Oxo-4-Desmethyl-18β-oleana-4,12-dien-30-oic Acid (11i). This substance was prepared from 829 mg of **11h** in the same manner as **11e** was prepared from **11d**. **11i** was recrystallized from aqueous MeOH and then from PhH–cyclohexane. Two crops gave an aggregate yield of 53% of pure material: mp 216–218°; λ_{max} 251 nm (ε 13,590); λ_{max} 3.57–4.17, 5.76, 5.90, and 6.04 μ; nmr max at 109 Hz (C-4 Me). *Anal.* ($C_{29}H_{42}O_3$) C, H.

Methyl 3,11-Dioxo-18β-oleana-5,12-dien-30-oate (12a). The technique of Atwater, *et al.*,[15] was used. A three-neck 1-l. round-bottom flask was charged with 150 ml of *t*-BuOH and 4.8 g of *t*-BuOK. The mixture was heated to reflux, and a solution of 12.4 g of **11a** in boiling (165 ml) *t*-BuOH was added. MeI (5.1 g) in 125 ml of *t*-BuOH was added dropwise over 2.5 hr. The reaction mixture was cooled, 19 ml of H_2O, 12 g of K_2CO_3, and 20 ml of MeI were added, and refluxing was resumed for 3 hr to reesterify any saponified material. The reaction mixture was then cooled and evaporated to a small volume and partitioned between 400 ml of H_2O and 200 ml of EtOAc; the organic phases were combined, washed with H_2O and saturated brine, filtered, and evaporated to 12.6 g of solid. The solid was chromatographed on silica gel. Elution with benzene–EtOAc (96:4) gave 2.65 g (20%) of **12a**, mp 272–273°. Recrystallization occurred from CH_2Cl_2–MeOH: λ_{max} 250.5 nm (ε 11,500); λ_{max} 5.79, 5.85, and 6.06 μ; nmr max at 344 Hz (C-6 H). *Anal.* ($C_{31}H_{44}O_4$) C, H.

Methyl 3β-Hydroxy-11-oxo-18β-oleana-5,12-dien-30-oate (12b). **12a** (1.35 g) was treated with 3.5 g of lithium tri-*tert*-butoxyaluminum hydride in 30 ml of THF for 2 hr at 22°. The reaction mixture was poured on ice, carefully acidified with concentrated HCl, and extracted with Et_2O and then with $CHCl_3$. The organic extracts were combined, dried ($CaSO_4$), filtered, and evaporated to a solid, **12b**, which was crystallized from CH_2Cl_2–MeOH: 1.08 g (79%); mp 250–254°; λ_{max} 250.5 nm (ε 11,100); λ_{max} 2.74, 5.76, and 6.04 μ. *Anal.* ($C_{31}H_{46}O_4$) C, H.

3β-Hydroxy-11-oxo-18β-oleana-5,12-dien-30-oic Acid (12c). When 4.5 g of **12b** was treated with LiI and 2,4,6-collidine (*vide supra* **9b**), crude **12c** was obtained. Recrystallization from CH_2Cl_2–MeOH gave 2.08 g (46%): mp 273–276°; λ_{max} 251 nm (ε 10,300); λ_{max} 2.70, 2.76, 2.84, 3.57–4.16, 5.73, and 5.86 μ; nmr max (C_5D_5N) 346 Hz (C-6 H). *Anal.* ($C_{30}H_{44}O_4$) C, H.

3β-Hydroxy-4,4-desmethyl-11-oxo-18β-oleana-5,12-dien-30-oic Acid (12d). **11b** (5 g) was suspended in 200 ml of warm *t*-BuOH and this suspension added to a suspension of 20 g of *t*-BuOK in 200 ml of *t*-BuOH. The mixture was stirred 13 min at 22° and then poured into 500 ml of 4.2 *N* AcOH. The *t*-BuOH was evaporated and the resulting suspension was diluted with H_2O and extracted twice with EtOAc–Et_2O. The organic phases were combined, washed twice with H_2O and once with saturated brine, decolorized, filtered, and evaporated to a solid.

The solid was dissolved in 100 ml of THF and treated with 10 g of lithium tri-*tert*-butoxyaluminum hydride for 18 hr at 22°. The reaction mixture was poured on ice, diluted with H_2O, acidified with concentrated HCl, and extracted twice with 250-ml portions of 1:1 Et_2O–EtOAc. The organic extracts were combined and washed as above and then evaporated to an oil. This oil was extracted several times with hot 2:3 cyclohexane–benzene (50-ml portions). The extracts were poured into pentane. Pure product **12d**, 1.89 g (38%), precipitated: mp 168–169°; λ_{max} 250.5 nm (ε

‡ Hydroxylation is assumed to proceed cis and α, *i.e.*, from the least hindered site. An attempt to reduce **9** with sodium borohydride gave a borate diester of the diol.

11,000); λ_{max} 2.77, 2.84, 5.86, and 6.05 μ; nmr max at 214 (C-3 α-H) and 322 Hz (C-6, H). *Anal.* ($C_{28}H_{40}O_4$) C, H.

Methyl 3-Oxo-18β-oleana-5,12-dien-30-oate (12e). This substance was prepared from **11e** in the same manner as **12a** and **12b** were prepared from 4 g of the analogous 11-oxo material. The isolate from the reaction mixture was applied to a Baker SiO_2 column, and elution of the column with PhH–EtOAc (98:2) gave first **12e** and then **11h** (*vide infra*). **12e** was recovered from the column in 9% yield. Recrystallization from CH_2Cl_2–MeOH gave mp 161–163°: λ_{max} 5.79 and 5.85 μ; nmr max at 321 (C-12 H) and 336 Hz (C-6 H). *Anal.* ($C_{31}H_{46}O_3$) C, H.

3-Oxo-18β-oleana-5,12-dien-30-oic Acid (12f). This substance was synthesized in the same manner as **12c**, starting from 450 mg of **12e**. Crystallization from MeOH–H_2O (7:2) gave a 78% yield of **12f**: mp 208–210°; λ_{max} 3.57–4.17, 5.74, 5.86, and 5.99 μ; nmr max at 322 Hz (C-6 H). *Anal.* ($C_{30}H_{44}O_3$) C, H.

3β-Hydroxy-4,4-desmethyl-18β-oleana-5,12-dien-30-oic Acid (12g). When 1 g of **11g** was treated with *t*-BuOK and reduced according to the procedure for the preparation of **12d**, crude **12g** was obtained. Recrystallization from CH_2Cl_2–MeOH gave 323 mg (32%): mp 267–270°; λ_{max} 2.92, 3.84, and 5.86 μ; nmr (C_5D_5N) max at 230 (C-3 H) and 330 Hz (C-6 and C-12 H). *Anal.* ($C_{28}H_{42}O_3$) C, H.

Methyl 3β-Hydroxy-5α,6α-oxa-11-oxo-18β-olean-12-en-30-oate (13a). **12b** (4 g) was dissolved in 100 ml of $CHCl_3$ and cooled to −15°. *m*-Chloroperbenzoic acid (2.50 g) was added with stirring. After 30 min, the reaction mixture was removed to a cold room (5°) and allowed to stand 30 hr. Aqueous $NaHSO_3$ was added, and the mixture was evaporated to a wet mass, which was partitioned between H_2O and EtOAc. The organic phase was washed twice with 5% KOH (H_2O), twice with H_2O, and once with saturated brine; it was then filtered and evaporated to a solid. Recrystallization from CH_2Cl_2–MeOH gave 3.8 g of **13a** (92%): mp 240–242°; λ_{max} 246.5 nm (ϵ 12,000); λ_{max} 2.75, 6.13, and 13.58 μ. *Anal.* ($C_{31}H_{46}O_5$) C, H.

3β-Hydroxy-5α,6α-oxa-11-oxo-18β-olean-12-en-30-oic Acid (13b). When 3.4 g of **13a** and 3.4 g of LiI were treated with 2,4,6-collidine as in the procedure for the preparation of **9b**, the product **13b** was obtained. Recrystallization from CH_2Cl_2–MeOH yielded 2.6 g (79%): mp 242–243°; λ_{max} 247 nm (ϵ 11,200); λ_{max} 2.7, 2.74, 2.82, 3.57–4.16, 5.84, and 5.98 μ; nmr max at 225–240 Hz (broad, C-6 H). *Anal.* ($C_{30}H_{44}O_5$) C, H.

A small amount of **13b** was converted to the Me ester with CH_2N_2 in Et_2O. The ester showed λ_{max} at 5.77 and 5.99 μ, with no band at 5.85 μ or anywhere else close to 5.88 μ. Therefore, the epoxide did not rearrange to the 6-ketone under the saponification conditions.

30-Methyl-18β-olean-12-ene-3,30-dione (14f). When 500 mg of **14e**[16] was oxidized with 0.6 ml of 8 *N* CrO_3–H_2SO_4 in acetone at 0°, 450 mg (90%) of crude **14f** was obtained. Recrystallization from MeOH gave mp 296–298°. *Anal.* ($C_{31}H_{46}O_3$) C, H.

3β-Stearoyloxy-30-methyl-18β-olean-12-ene-11,20-dione (14g). By substituting an equivalent quantity of **14d**[16] in the procedure for the preparation of **14k**, 2.3 g of **14g** was obtained. Recrystallization from EtOH gave mp 108–109°. *Anal.* ($C_{49}H_{82}O_4$) C, H.

3β-Acetoxy-30-methyl-18β-olean-12-en-30-one (14h). To a suspension containing 3 g of the acetate of **1d**, 214 ml of anhydrous ethyl ether, and 14.7 ml of pyridine was added, dropwise at room temperature, 21.9 g of thionyl chloride. After stirring the reaction mixture for 4 hr, 100 ml by volume of an ethereal anhydrous HCl solution was added. The resulting mixture was filtered and the filtrate was evaporated to dryness. Then 46.2 ml of *n*-hexane was added to the solid and the solution was brought to reflux temperature. After cooling, the solid was recovered by filtration and dried to yield crude 3β-acetoxy-18β-olean-12-en-30-oyl chloride, which was used without purification.

Then 0.25 g of Mg turnings was covered by 36 ml of anhydrous ether and 1.4 g of MeI in 7 ml of ether was added to that mixture over 1 hr. Then the mixture was cooled in an ice bath and 1.0 g of $CdCl_2$, which was dried at 110° for 1 hr, was added over a 5-min period. The mixture was removed from the ice bath, stirred at room temperature for 5 min, and refluxed for 45 min. Then the ether was removed by distillation and 8.8 ml of anhydrous benzene was added. After cooling the mixture to room temperature, the acid chloride was added and the mixture was refluxed for 2 hr. Cooling of the reaction mixture followed by the addition of ice water and 20% sulfuric acid afforded two phases. The organic phase was separated, successively washed with water, aqueous $NaHCO_3$ solution, and water until neutral, and dried over $MgSO_4$. The benzene was evaporated and the material remaining was recrystallized from methanol to yield **14h.** Recrystallization from EtOH gave 0.53 g (50%), mp 232–233°. *Anal.* ($C_{33}H_{52}O_3$) C, H.

3β-Hydroxy-30-methyl-18β-olean-12-ene-30-dione (14i). A solution of 350 mg of **14h**, 16.3 ml of EtOH, and 15 ml of a 0.2 *N* KOH in EtOH solution was refluxed for 1 hr. Then the EtOH was evaporated and the organic material extracted with $CHCl_3$, washed with water until neutral, and dried. Recrystallization from EtOH gave 2.2 g (73%), mp 235–237°. *Anal.* ($C_{31}H_{50}O_2$).

30-Methyl-18β-olean-12-ene-3,30-dione (14j). When 500 mg of **14i** was oxidized with a small excess of 8 *N* $CrO_3H_2SO_4$ in acetone, 400 mg (80%) of crude **14j** was obtained. Recrystallization from EtOH gave mp 169–170°. *Anal.* ($C_{31}H_{48}O_2$) C, H.

3β-Steroyloxy-30-methyl-18β-olean-12-en-30-one (14k). To a solution of 250 mg of **14i** in 3 ml of pyridine was added 150 mg of stearoyl chloride and the mixture was stirred at room temperature for 2 hr. Then an additional 200 mg of stearoyl chloride was added and, after stirring the solution for an additional 30 min, ether and ice water were added. The organic layer was washed with water until neutral and then dried over Na_2SO_4. The solvent was evaporated and the oily material which remained was extracted with MeOH. The volume of the MeOH extract was reduced to half its original volume by heating and the solution was cooled to room temperature. The remaining solvent was evaporated under reduced pressure to afford crude product which, when chromatographed on silica gel with benzene as eluent and recrystallized from methanol, yielded pure **14k:** 1.3 g (50%); mp 119–120°. *Anal.* ($C_{49}H_{82}O_4$) C, H.

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Synthesis of 2-Oxygenated Glycyrrhetic Acid Derivatives

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2β,3α-Dihydroxy and 2β,3β-dihydroxy derivatives of glycyrrhetic acid and 11-deoxoglycyrrhetic acid were synthesized. Unsaturation was introduced at C-18 in both the 2β,3α and in the 2β,3β series of glycyrrhetic acid. Some of the compounds exhibited low levels of antibacterial and antifungal activity.

Studies in this laboratory to achieve a separation of activities for derivatives of glycyrrhetic acid have focused on rings A and E.[1] Since the mammalian system metabolizes many triterpenes by oxygenation,[1] it was felt that a synthesis of some oxygenated derivatives of glycyrrhetic acid might yield products with a different spectrum of activities from the parent compound. This paper reports oxygenations at positions 2 and 3 of the oleane skeleton, as well as the introduction of unsaturation at C-18.

2β,3α-Dihydroxy Derivatives. Mousseron-Canet and Crouzet[2] reported the synthesis of **1a** and **1b**. Hydrogenolysis of this mixture with PtO_2 in acetic acid yielded a mixture of the corresponding 11-deoxo compounds **2** and **3**, which were separated by column chromatography. The dihydroxy compound **4** could be derived from a mixture of **2** and **3** by refluxing in methanolic KOH. The C-30 ester is sufficiently hindered so that no saponification occurs at this site. The 11-oxo analog of **4**, compound **5**, was made by direct treatment of a mixture of **1a** and **1b** with refluxing methanolic KOH. Treatment of the C-30 ester to yield the free acid **6** was effected with LiI in refluxing 2,4,6-collidine.

Unsaturation at C-18 was introduced by the method of Ruzicka and Jeger.[3] Thus, acetylation of a mixture of **1a** and **1b** yielded the 2β,3α-diacetoxy compound, which was treated with Br_2 in hot glacial acetic acid in the presence of HBr. Separation by column chromatography yielded **7**, which was completely hydrolyzed to the free acid **8** with refluxing methanolic KOH (Scheme I).

2β,3β-Dihydroxy Substitutions. Treatment of **9b**[1] with *t*-BuOK and O_2 in *t*-BuOH and hexamethylphosphoric triamide by the method of Hanna and Ourisson[4] yielded the Δ^1-2-hydroxy-3-oxo compound **10b**. Reduction of **10b** with $NaBH_4$ yielded the 2β,3β-dihydroxy compound **12b**. Reaction of **10b** with Br_2 in THF with an HBr catalyst yielded **11**. In the 11-oxo series, **10a** was synthesized from **9a**[5] in the same manner as **10b** was derived from **9b**. The preparation of **12a** from **10a** was performed with $NaBH_4$ at 0–12° for 75 min. Acetylation of **12a** yielded **13**, which was converted to the Δ^{18} compound **14** (*vide supra*) (Scheme II).

An alternative method of *cis* hydroxylation involved reaction of the Δ^2 compound **15**[2] with OsO_4. Hydroxylation followed by crystallization resulted in the isolation of only the 2α,3α isomer **16**. When the products of hydroxylation were hydrogenolyzed and then separated by chromatography, isomers **17** and **18** were isolated (Scheme III).

Biology. All compounds were tested for anti-DCA activity by the method of Kagawa.[6] The test drug was injected subcutaneously at a dose of 2.4 mg/rat in a series of eight rats per dose. The standard drug in this assay is spironolactone, which displays a median effective dose of 0.33 mg/rat in this test. None of the compounds showed activity in this test.

Many of the compounds were tested for antiulcer activity in the pylorus ligated Shay rat,[7] tested in groups of six. Only **13** showed activity (at 50 mg/rat), as it did in the Shay rat antisecretory test,[8] in which it caused a 29% reduction in acid secreted in a 5-hr period. A group of six rats received 25 mg/rat in this test.

A number of compounds were tested for antiviral activity against influenza A (strain 575). The test compound was added to cell cultures of primary rhesus monkey kidney 1 hr before challenge with virus. The quantitative hemagglutination technique of Finter[9] was then used to assay activity. Compounds **6** and **13** were active at 625 μg/ml, but their cytotoxicity precluded their pursuit as leads.

Some of these compounds showed weak *in vitro* antibacterial and antifungal activity. **8**, **12a**, **13**, and **14** were active at 100 ppm against *Erwinia* species in a beef extract growth medium, and **4**, **6**, and **11** were active at 1000 ppm against the same bacteria. **6** and **14** were active against *Bacillus subtilis* in beef extract at 1000 ppm. **11** was active against *Trichophyton mentagrophytes* in a dextrose agar gel medium at 1000 ppm, and **8** and **14** were active against *Verticillium albo atrium* in the same agar medium at 1000 ppm.

In summary, introduction of an oxygen function into the 2 position of glycyrrhetic acid does not confer anti-DCA activity or antiviral activity to the compound. In addition, very little antiulcer activity is achieved, and only very low levels of antiinfective potency are attained.

Experimental Section

Melting points were taken on a Fisher-Johns hot-stage apparatus and are uncorrected. Melting points over 300° were taken on a Mel-Temp hot block apparatus and are also uncorrected. Nmr spectra were taken on a Varian A-60A, a Varian T-60, and Varian XL-100. All spectra are 60 MHz unless specified otherwise. Location of peaks (δ) is by parts per million away from TMS as an internal standard. Ir spectra were recorded in Chf unless specified otherwise, on a Beckman IR 12. Uv spectra were in MeOH on a Beckman DK-2A. Tlc runs were on 7.6-cm microscope slides covered with a 0.25-mm thickness of Woelm F silica, with a magne-