Bufadienolides. 10. 3β-Acetoxy-14β,21-epoxy-5β-bufanolide and Related Lactones[1,2a]

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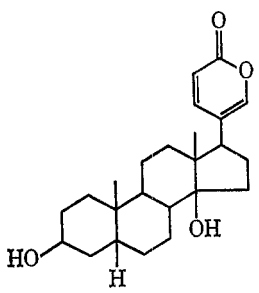
Methyl 3β-pyranyloxy-(21*S*)-methoxy-14β,21-epoxy-5β-(20*S*)-norcholanate (**6a**) and the axial acetal **6b** each gave isodigitoxigenin (**3a**) on treatment with hydrochloric acid in aqueous acetic acid. Extension of this reaction to cholanate ester **7b** gave 3β-acetoxy-14β,21-epoxy-5β-bufanolide (**4**). The epoxide **9** prepared from 3β-acetoxy-14β,21-epoxy-5β-chol-20(21)-enic acid (**8b**) spontaneously cyclized to give γ-lactones of type **11** rather than δ-lactone derivatives. In another potential approach to bufadienolides, 3β-pyranyloxy-23-mesyloxy-14β,21-epoxy-(21*S*)-methoxy-5β-(20*S*)-norcholane (**7d**) was converted into 3β-acetoxy-14β,21-epoxy-20-formylpregn-20(21)-ene (**15a**). Condensation of aldehyde **15a** with malonic acid took an unexpected course which culminated in formation of olefin **18**.

Two important considerations in a potentially useful synthesis of bufalin (**1**) are protection of the 14β-hydroxyl group and prevention of isomerization at position 17. In attempting to prepare bufalin from digitoxigenin (**2**) it seemed that both obstacles could be avoided by utilizing the cyclic acetal isodigitoxigenin (**3a**). Expansion of the lactone ring to give 14β,21-epoxybufanolide **4** followed by conversion into unsaturated lactone **5** and cleavage of the acetal linkage would then give bufalin (**1**).

The previous two papers in this series[2,3] described conversion of isodigitoxigenin into acetals **6a** and **6b** and homologation of compound **6a** to acetal **7a**. Model experiments determined that **6a** could be recyclized to isodigitoxigenin (**3a**) in excellent yield by treatment with hydrochloric acid in aqueous acetic acid.[4] Similar treatment of acetal **7b** gave a mixture of products, which after remethylation and reacetylation could be separated by column chromatography into three principal components. Vinyl ether **8a**[2] was present in largest amount (42%) together with a substance (28%) which showed two pmr acetate signals at δ 2.06 and



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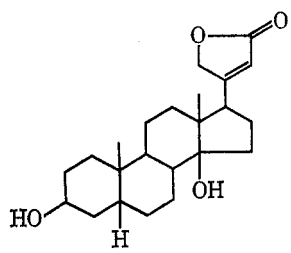

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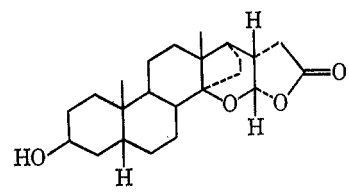
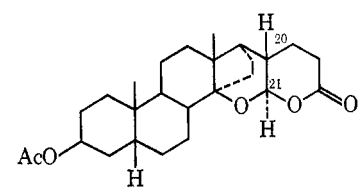


3a, R = H
b, R = $COCH_3$



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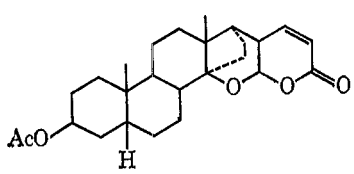

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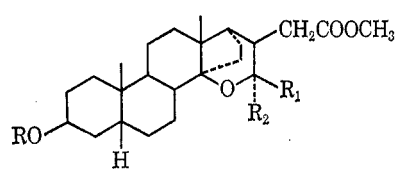
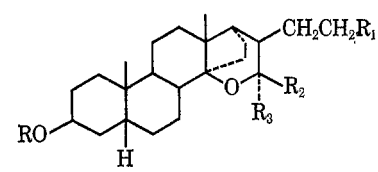
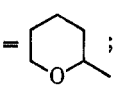
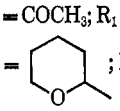


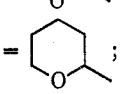
6a, R = tetrahydropyranyl; $R_1 = OCH_3$; $R_2 = H$
b, R = $COCH_3$; $R_1 = H$; $R_2 = OCH_3$

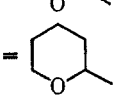


7a, R =  ; $R_1 = COOCH_3$; $R_2 = OCH_3$; $R_3 = H$

b, R = $COCH_3$; $R_1 = COOCH_3$; $R_2 = OCH_3$; $R_3 = H$
c, R = $COCH_3$; $R_1 = COOCH_3$; $R_2 = OCOCH_3$; $R_3 = H$

d, R =  ; $R_1 = OSO_2CH_3$; $R_2 = OCH_3$; $R_3 = H$

e, R =  ; $R_1 = I$; $R_2 = OCH_3$; $R_3 = H$

f, R =  ; $R_1 = OH$; $R_2 = OCH_3$; $R_3 = H$

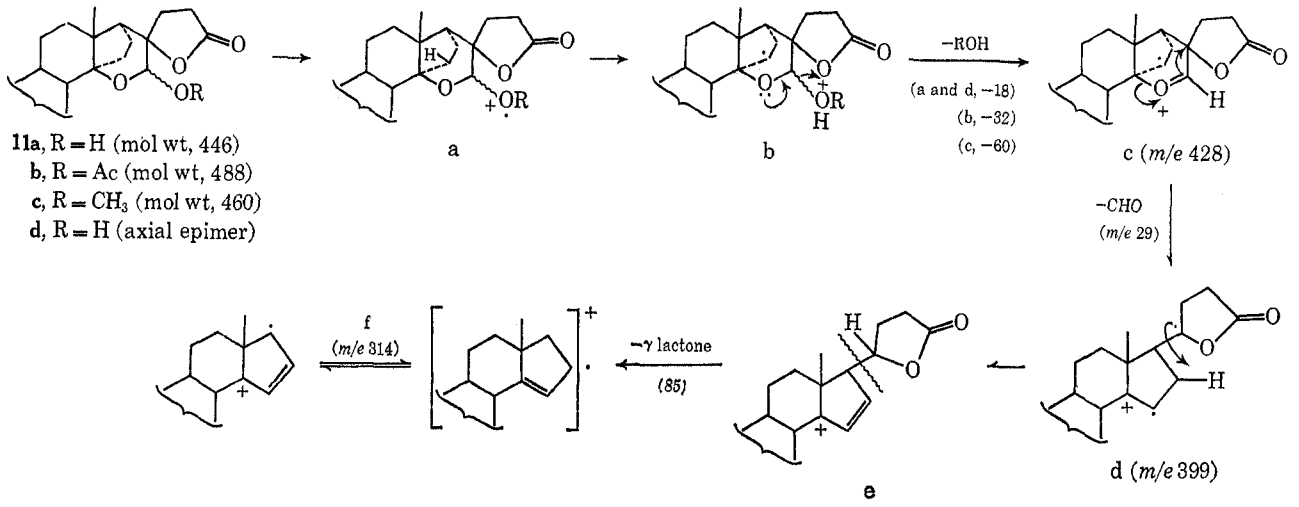
(1) This investigation was supported by Public Health Service Research Grants CA-10115-01 and CA-10115-02 from the National Cancer Institute. Purchase of the Atlas CH-4B and SM-1B mass spectrometers was made possible through Grants GB-4939 and GP-6979, respectively, from the National Science Foundation.

(2) (a) Preceding contribution: G. R. Pettit, T. R. Kasturi, J. C. Knight, and K. A. Jaeggi, *J. Org. Chem.*, **35**, 1410 (1970). (b) To whom inquiries should be addressed.

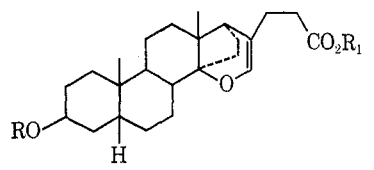
(3) G. R. Pettit, T. R. Kasturi, J. C. Knight, and J. Occolowitz, *J. Org. Chem.*, **35**, 1404 (1970).

(4) G. Buchi, D. M. Foulkes, M. Kurono, G. D. Mitchell, and R. S. Schneider, *J. Amer. Chem. Soc.*, **89**, 6745 (1967).

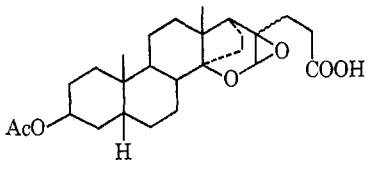
SCHEME I
FRAGMENTATION OF γ LACTONES 11a–11d



2.12. The mass spectrum also indicated the presence of two acetate groups, and the new compound was assigned structure **7c**. The most polar product, and that present in the smallest yield (1.6%), was the desired 3β-acetoxy-14β,21-epoxybufanolide **4**. Lactone **4** showed a pmr doublet at δ 5.08 for H-21 (acetal). The 8-Hz coupling constant indicated a *trans* fusion of the δ-lactone and tetrahydropyran rings, *i.e.*, a 20*S*,21*R* configuration.



8a, R = $COCH_3$; R_1 = CH_3
b, R = $COCH_3$; R_1 = H
c, R = R_1 = H



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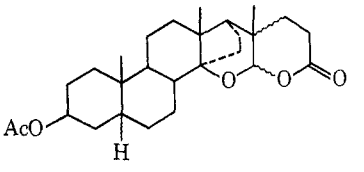

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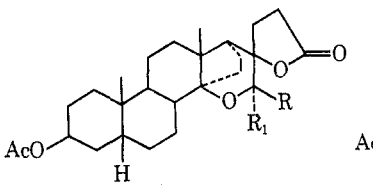
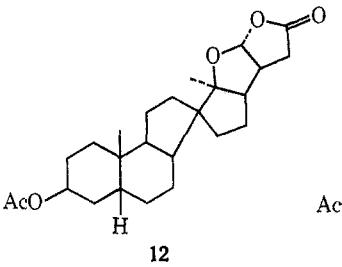


11a, R = OH; R_1 = H
b, R = $OCOCH_3$; R_1 = H
c, R = OCH_3; R_1 = H
d, R = H; R_1 = OH



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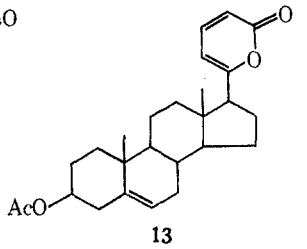

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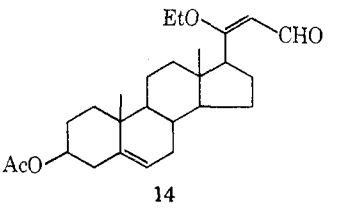

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The low yield in the cyclization step leading to lactone **4** was not encouraging, and other more promising approaches were considered. For example, by analogy with the ready formation of tetrahydropyranyl esters from dihydropyran and carboxylic acids, acid **8b** should undergo intramolecular cyclization to lactone **4**. However, no observable lactone formation could be demonstrated under conditions normally used for this reaction.[5]

In view of the work of Stevens and coworkers on formation and reactions of epoxy ethers,[6] we expected that an epoxypyran such as **9**, formed by *m*-chloroperbenzoic acid oxidation of dihydropyran **8b**, would undergo rapid intramolecular cyclization yielding lactone **10**. The rate of intramolecular reaction would presumably be much faster than the competing intermolecular reaction with *m*-chlorobenzoic acid present in the reaction mixture. Experimentally, reaction rapidly occurred, and after a few minutes acid **8b** was consumed. The product was separated into neutral and sodium hydroxide soluble fractions. The neutral material was a high-melting solid which showed the expected molecular ion at *m/e* 446 ($C_{26}H_{38}O_6$) and a sharp pmr signal at δ 5.32 for an acetal-type proton, but the infrared spectrum showed absorption at 1760 cm^{-1} more typical of a γ-lactone than a δ-lactone. Structure **11a** therefore seemed probable and is supported by mass spectral evidence (Figure 1). A possible scheme which accounts for prominent fragmentation is shown in Scheme I. Empirical formulae of the major fragments were checked by high-resolution mass spectrometry (Table I).

(5) See, *e.g.*, W. S. Johnson, R. C. Christiansen, and R. E. Ireland, *J. Amer. Chem. Soc.*, **79**, 1995 (1957).

(6) C. L. Stevens and J. Tazuma, *ibid.*, **76**, 715 (1954). We wish to thank Professor M. E. Munk for a valuable discussion concerning such epoxy ether reactions.

TABLE I
HIGH-RESOLUTION MASS SPECTRAL DATA

Empirical formula	Calcd mass	Measured mass	
		Lactone **11a**	Lactone **11b**
$C_{23}H_{31}O_2$	339.2323	339.2339	339.2352
$C_{22}H_{31}O_3$	343.2273	343.2269	...
$C_{25}H_{35}O_4$	399.2535	399.2523	...
$C_{26}H_{36}O_5$	428.2562	428.2577	428.2559
$C_{26}H_{38}O_6$	446.2668	446.2711	...

Additional evidence was obtained by examining the acidic fraction. Although it had been extractable by strong base, isolation by acidification and reextraction gave a neutral substance, presumably a lactone, which readily opened and recyclized. As an infrared spectrum of the crude product indicated loss of the 3β-acetate, the alcohol was reacetylated and purified by chromatography. The pmr spectrum showed signals for two acetate groups at δ 2.02 and 2.12, and infrared peaks at 1790 (γ lactone), 1762 (acetal acetate), and 1734 cm^{-1} (acetate) were consistent with a structure such as **11b**. The mass spectrum showed a molecular ion at *m/e* 488, and exhibited the same fragmentation as found in that of hemiacetal lactone **11a**. The major peak at *m/e* 339 in each spectrum is presumably due to further loss of acetic acid from the 3-acetate group of the ions at *m/e* 399 (d).

The γ-lactone **11a** tended to open readily, and even on heating with aqueous methanol was converted into a complex mixture. Two compounds could be isolated by preparative layer chromatography, mp 209–222° and 266–271°, respectively. The former (**11c**) showed typical infrared γ-lactone absorption at 1782 cm^{-1}, a pmr methyl ether peak at δ 3.40, and a molecular ion at *m/e* 460, which indicated methylation of the hemiacetal hydroxyl. In addition, ions at *m/e* 428 (c), 399 (d), 339 (d − 60), and 314 (f) indicated a close structural similarity to starting material **11a** (see Scheme I).

The higher melting compound, of which only a few milligrams was obtained, showed typical γ-lactone infrared absorption at 1772 cm^{-1} and hydroxyl absorption at 3500 cm^{-1}. Therefore the substance with a melting point of 266–271° may be hemiacetal lactone **11d**, the C_{21} epimer of **11a**. The mass spectrum did not show a molecular ion, but prominent ions at *m/e* 428, 399, 339, and 314 were present. Lack of a molecular ion is noteworthy in view of the strong molecular ion shown by **11a**. If a mechanism such as that in Scheme I is operative, then hydrogen transfer (a → b) with subsequent loss of water might be expected to take place much more readily in the isomer with the axial OH-21.

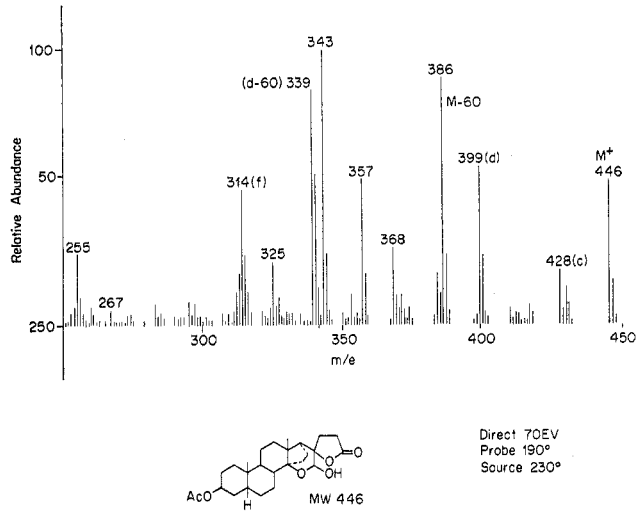
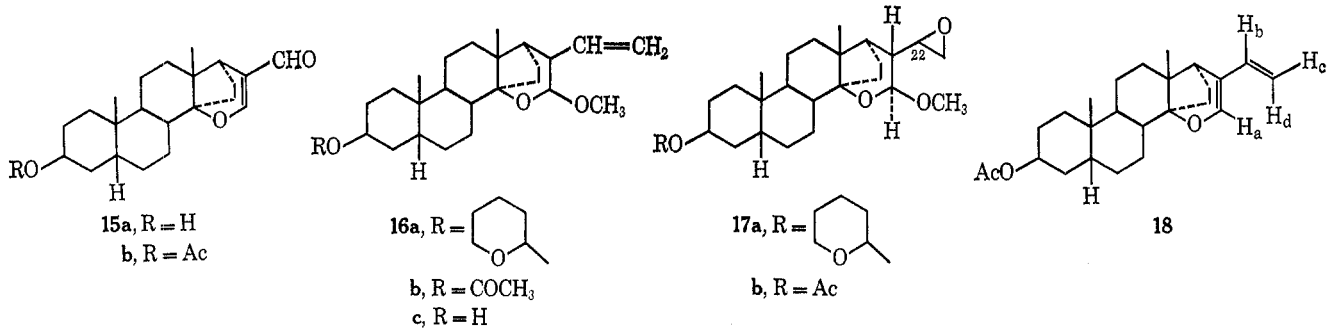


Figure 1.

At this stage, model experiments designed to cleave the 14β,21-epoxy linkage in isodigitoxigenin (**3**) showed that acidic conditions were prone to generate a carbonium ion at C-14, resulting in rearrangement to C-norcardenolides[3] such as **12**. Thus considerable difficulty was expected in effecting conversion of unsaturated lactone **5** into bufalin (**1**), and an alternative approach was investigated. An earlier contribution of this series[7] summarized synthesis of isobufadienolides such as 6′-substituted 2-pyrone **13** from unsaturated aldehyde **14** by condensation with malonic acid in pyridine–piperidine solution. With these experiments in mind we decided to explore an analogous transformation with unsaturated aldehyde **15**. Similar reaction leading to formation of a 5′-substituted 2-pyrone would give bufalin or the 14-dehydro derivative. Aldehyde **15** was prepared as follows. The previously described mesylate **7d**[2] was converted into **7e** with sodium iodide in refluxing acetone, and the product was dehydrohalogenated to olefin **16** with potassium *t*-butoxide in dimethyl sulfoxide.[8] The structure of olefin **16a** was confirmed by the pmr spectrum which showed a complex pattern in the olefinic proton region at δ 4.8–6.2, integrating for three protons and strongly resembling in overall appearance the spectra of compounds with an allylic grouping –CHRCH=CH$_2$.[9a] Ozonolysis of



(7) G. R. Pettit, J. C. Knight, and C. L. Herald, *J. Org. Chem.*, **35**, 1393 (1970).

(8) N. F. Wood and F. C. Chang, *ibid.*, **30**, 2054 (1965).

(9) (a) N. S. Bhacca, L. F. Johnson, and J. N. Shoolery, "Varian N.M.R. Spectra Catalog," Varian Associates, 1962, Spectra No. 26, 32, 38, and 136; (b) Spectrum No. 32.

olefin **16** followed by treatment with zinc–acetic acid did not give very satisfactory yields of aldehyde **15a**. Another approach was more effective. Epoxidation of olefin **16a** using *m*-chloroperbenzoic acid gave oily epoxide **17a**, but analogous oxidation of **16b** gave crystalline acetate **17b**. Both epoxides showed a complex three-proton pattern in the pmr spectrum at δ 2.5–3.0 resembling that of propylene oxide.[9b] The 60-MHz spectra were difficult to resolve, and at 100 MHz it was apparent that **17b** was a 1:1 mixture of two epimeric (at C-22) epoxides. For instance, what appeared to be a doublet with J = 8.5 Hz at δ 4.56 in the 60-MHz spectrum (owing to H-21) appeared in the 100-Mz spectrum as a pair of closely spaced doublets with coupling constants of 8.8 and 9.5 Hz. Cleavage of the epoxide with periodic acid[10] and simultaneous loss of methanol from the acetal grouping gave aldehyde **15**. Unfortunately, the aldehyde could not be induced to react under conditions developed for conversion of aldehyde **14** into pyrone **13**. More severe conditions, *i.e.*, refluxing pyridine–piperidine for extended periods, gave complex mixtures, and the only crystalline product isolated was diene **18**. Structure **18** was deduced mainly from the pmr spectrum, which was reminiscent of a compound containing an isolated vinyl group. The splitting pattern was complicated by presence of signals owing to the olefinic H-21 (H_a) and H-3α, but coupling constants of the expected magnitude could still be assigned. Fine splitting owing to allylic coupling was not observed in the 60-MHz spectrum. Structure **18** was further supported by the mass spectrum, which showed a molecular ion at m/e 384, correct for $C_{25}H_{36}O_3$.

Meanwhile other approaches to bufalin began showing greater promise than those summarized above. Further efforts to utilize lactone **4** and aldehyde **15** were discontinued when a useful synthesis of bufalin was achieved employing another route[11].

Experimental Section

Low-resolution mass spectra were secured by E. S. Bebee using an Atlas CH-4B mass spectrometer equipped with "molecular beam" direct probe inlet system and operating under the following conditions: electron energy 70 eV, trap current 19 μA, source temperature 230°, probe temperature 120–190°, accelerating voltage 3 kV. High-resolution measurements were made with an Atlas SM-1B instrument, again using a direct probe inlet system. Other operational parameters follow: electron energy 70 eV, trap current 290 μA, source temperature 230°, probe temperature 120–190°, accelerating voltage 8 kV, apparent resolution 12,500. The mass reference compound was perfluorokerosene.

Unless otherwise stated, introduction to the Experimental Section of part 9[2] provides other necessary general information for the following experimental summaries.

Recyclization of Methyl 3β-Pyranyloxy-(21*S*)-methoxy-14β,21-epoxy-5β-(20*S*)-norcholanate (6a) and Methyl 3β-Acetoxy-(21*R*)-methoxy-14β,21-epoxy-5β-(20*S*)-norcholanate (6b) to Isodigitoxigenin Acetate (3b).—To a solution of acetal **6a** (0.16 g) in acetic acid (10 ml) was added water (5 ml) and concentrated hydrochloric acid (0.5 ml). The clear solution was stirred overnight at room temperature, and the resulting cloudy suspension was diluted with water (20 ml) and filtered. The precipitate (0.11 g) was dried and purified by preparative layer chromatography on one 20 × 20 cm plate with 4:1 chloroform–ethyl acetate. Two distinct zones were eluted. The least polar material (64 mg) crystallized from methanol–chloroform, giving shining needles (60 mg) of isodigitoxigenin acetate (**3b**), mp 258–260°. The more polar material (24 mg) crystallized from the same solvent, providing isodigitoxigenin (**3a**) as small flakes (17 mg), mp 265–267°. The specimens thus prepared were identical[12] with authentic samples of isodigitoxigenin and acetate derivative.

Similar treatment of acetal **6b** (0.21 g) gave a solid product (0.148 g), which was separated into isodigitoxigenin acetate (0.10 g) and isodigitoxigenin (12.5 mg) as above.

Cyclization of Methyl 3β-Acetoxy-(21*S*)-methoxy-14β,21-epoxy-5β-(20*S*)-cholanate (7b) to 3β-Acetoxy-14β,21-epoxybufanolide (4).—To acetal **7b** (1.0 g) dissolved in acetic acid (45 ml) was added water (5 ml) containing concentrated hydrochloric acid (2.5 ml). The mixture was stirred for 2 days at room temperature, diluted with water, and extracted with chloroform. An infrared spectrum of the crude product suggested the presence of free carboxylic acid groups and deacetylation at C-3. Therefore, a solution of the product in diethyl ether was treated for 2 hr with diazomethane at 0°. Excess reagent was destroyed by adding a few drops of acetic acid. The solution was evaporated to dryness and the residue was reacetylated with 10 ml of 1:1 acetic anhydride–pyridine overnight at room temperature. Chromatography of the crude product in ligroin on silica containing increasing amounts of ethyl acetate gave three fractions, which follow in order of increasing polarity. (a) **Methyl 3β-acetoxy-14β,21-epoxy-5β-chol-20(21)-enate (8a)** was eluted by 9:1 ligroin–ethyl acetate and crystallized from methanol as large prisms (0.50 g), mp 105–107°, identical[12] with an authentic specimen. (b) **Methyl 3β-(21*R*)-diacetoxy-14β,21-epoxy-5β-(20*S*)-cholanate (7c)** was eluted by 4:1 ligroin–ethyl acetate and crystallized from ligroin–ethyl acetate as long needles (0.30 g), mp 154–162°, mol wt, 504 (mass spectrum). On crystallization, ester **7c** tended to decompose to unsaturated ester **8a** and elemental analysis was not performed, but the mass and other spectral data support the assigned structure: pmr δ 1.0 (C-18 methyl), 1.10 (C-19 methyl), 2.06 (C-3 O acetate), 2.12 (C-21 O acetate), 3.70 (-COOCH$_3$), 5.10 (H-3α), 5.66 (doublet, J = 8.5 cps, acetal -OCHO-). (c) **3β-Acetoxy-14β,21-epoxybufanolide (4)** was eluted by 1:1 ligroin–ethyl acetate and crystallized from ligroin–ethyl acetate as small prisms (13 mg): mp 207–218°; ν_{max}^{KBr} 1745 and 1735 cm^{-1}; RD (c 0.583, dioxane) $[\alpha]_{600}$ −60°, $[\alpha]_{589}$ −67°, $[\alpha]_{500}$ −94°, $[\alpha]_{400}$ −163°, $[\alpha]_{350}$ −240°, $[\alpha]_{300}$ −420°, $[\alpha]_{250}$ −960°, and $[\alpha]_{234}$ −1561°; pmr δ 0.94 (C-18 methyl), 1.08 (C-19 methyl), 2.0 (O acetate), 2.64 (triplet, J = 7 Hz, -CH$_2$CO-), 5.06 (H-3α), and 5.08 (doublet, J = 8 Hz, -OCHO-).

Anal. Calcd for $C_{26}H_{38}O_5$: C, 72.52; H, 8.90; mol wt, 430. Found: C, 72.79; H, 8.94; mol wt, 430 (mass spectrum).

Attempted Cyclization of 3β-Hydroxy-14β,21-epoxy-5β-chol-20(21)-enic Acid (8c).—Acid **8c** (0.11 g) was dissolved in benzene (12 ml), and p-toluenesulfonic acid (11 mg) was added. The solution was stirred at room temperature for 73 hr and examined periodically by tlc without any observable reaction. Also, 24 hr at reflux followed by 48 hr at room temperature had no effect. The benzene solution was washed with water, dried, and evaporated to yield an oil with the same infrared and pmr spectrum as those of the starting material.

Peracid Oxidation of 3β-Acetoxy-14β,21-epoxy-5β-chol-20(21)-enic Acid (8b).—To cholenic acid **8b** (2.78 g) dissolved in chloroform (50 ml) was added *m*-chloroperbenzoic acid (1.4 g). After 15 min at room temperature the solution was washed with saturated sodium bicarbonate solution to remove *m*-chlorobenzoic acid, then with 2 *N* sodium hydroxide solution. Neutral material, obtained by evaporation of the washed and dried chloroform solution, was an amorphous solid (1.96 g). The alkaline extract was acidified and precipitated material was extracted with diethyl ether, washed with water, dried, and evaporated to give a crystalline solid (0.67 g).

The neutral material (0.75 g) was chromatographed on silica, and elution with 4:1 benzene–ethyl acetate gave γ lactone **11a** as a solid which crystallized from chloroform–ligroin as minute needles (0.11 g): mp 251–253°; ν_{max} 1758 (γ lactone), 1725, and 1260 cm^{-1} (acetate); $[\alpha]_D$ +63° (c 0.44); pmr δ 1.02 (C-18 methyl), 1.18 (C-19 methyl), 2.04 (O acetate), 5.10 (H-3α), and 5.32 (-OCHO-).

Anal. Calcd for $C_{26}H_{38}O_6$: mol wt, 446.266822. Found: mol wt, 446.271129 (mass spectrum).

(10) (a) G. Maerker and E. T. Haeberer, *J. Amer. Oil Chem. Soc.*, **48**, 97 (1966); (b) S. G. Wyllie and C. Djerassi, *J. Org. Chem.*, **33**, 305 (1968).

(11) G. R. Pettit, L. E. Houghton, J. C. Knight, and F. Brushweiler, *Chem. Commun.*, 93 (1970); *J. Org. Chem.*, in press.

(12) Mutual identity was confirmed by comparison of infrared spectra and thin layer chromatography R_f values.

An ir and pmr spectrum of the base-soluble material indicated deacetylation and the presence of small amounts of *m*-chlorobenzoic acid. The crude product was redissolved in diethyl ether and the ethereal solution was washed with sodium bicarbonate solution and evaporated. The resultant crystalline solid was acetylated with 1:1 acetic anhydride–pyridine overnight at room temperature and chromatographed on silica gel. Elution with 4:1 ligroin–ethyl acetate gave γ lactone **11b**, which crystallized from chloroform–ligroin as short needles (0.15 g): mp 224–232°; ν_{max}^{KBr} 1790, 1762, 1734, and 1240 cm^{-1}; $[\alpha]D$ +29.5° (*c* 3.15); $\nu_{max}^{CHCl_3}$ 1780 and 1740 cm^{-1}; pmr δ 1.00 (C-18 methyl), 1.10 (C-19 methyl), 2.04 (C-3β O acetate), 2.12 (C-21 O acetate), 5.12 (H-3α), and 5.89 (–OCHO–).

Anal. Calcd for $C_{28}H_{40}O_7$: mol wt, 488. Found: mol wt, 488 (mass spectrum).

When crystallization of lactone **11a** from aqueous methanol was attempted, a viscous oil was obtained which showed many spots on tlc examination. The oil from 0.32 g of lactone **11a** was chromatographed in 9:1 ligroin–ethyl acetate on silica gel. Elution with 2:1 ligroin–ethyl acetate gave a crystalline solid (50 mg), mp 208–220°, which was separated into two components by preparative layer chromatography with 1:4 ethyl acetate–chloroform. The major component was lactone **11c**, crystallized from methanol as long needles (30 mg): mp 209–222°; ν_{max}^{KBr} 1782 (γ lactone), 1738, and 1240 cm^{-1} (acetate); $\nu_{max}^{CHCl_3}$ 1785 (γ lactone), 1735, and 1270 cm^{-1} (acetate); pmr δ 1.02 (C-18 methyl), 1.08 (C-19 methyl) 2.06 (O acetate), 3.40 (acetal OCH_3), 4.65 (acetal –OCHO–), and 5.10 (H-3α).

Anal. Calcd for $C_{27}H_{40}O_6$: mol wt, 460. Found: mol wt, 460 (mass spectrum).

The minor component was lactone **11d**, which crystallized also from methanol as short, sparkling needles (7 mg): mp 266–271° dec; ν_{max}^{KBr} 1750, 1740, and 1235 cm^{-1}; $\nu_{max}^{CHCl_3}$ 1772 (γ lactone), 1735, and 1270 cm^{-1} (acetate).

Anal. Calcd for $C_{26}H_{38}O_6$: mol wt, 446. Found: mol wt, 446 (mass spectrum).

3β-Pyranyloxy- and 3β-Acetoxy-14β,21-epoxy-(21*S*)-methoxy-5β-(20*S*)-norchol-22(23)-ene (16a and 16b).—To a solution of 3β-pyranyloxy-23-mesyloxy-14β,21-epoxy-(21*S*)-methoxy-5β-(20*S*)-norcholane (**7d**, 1.3 g) in redistilled acetone (50 ml) was added sodium iodide (0.7 g). The solution was heated at reflux for 5 hr, by which time tlc showed no starting material. After cooling, dilution with water, and extraction with diethyl ether, the ethereal solution was washed with water and evaporated, yielding iodide **7e** as a colorless glass (1.33 g). Without further purification, the iodide was dissolved in benzene (10 ml) and added to a stirred 1 *N* solution of potassium *t*-butoxide (2.24 g) in dimethyl sulfoxide (20 ml). The yellow solution was stirred at room temperature for 15 min, poured into ice–water, and extracted with ether. The ether solution was evaporated to a yellow gum (1.03 g) which was applied in ligroin to a column of silica gel. Elution with 19:1 ligroin–ethyl acetate gave olefin **16a** as a colorless oil: pmr δ 0.98 (C-18 methyl), 1.09 (C-19 methyl), 3.50 (–OCH_3), 4.0 (THP –OCHO–), 4.44 (doublet, J = 8 Hz, –OCHO), 4.66 (H-3α), 5.0 (multiplet), 5.22 (sharp singlet), and 5.5–6.1 (multiplet).

The oily tetrahydropyranyl ether was converted into the crystalline acetate **16b** in the following manner. To a solution of ether **16a** (0.40 g) in methanol (20 ml)–water (0.4 ml) was added *p*-toluenesulfonic acid (40 mg). The solution was stirred at room temperature for 1.25 hr. Alcohol **16c** was isolated by dilution with water followed by extraction with ether. Following acetylation with 1:1 acetic anhydride–pyridine (5 ml) overnight at room temperature, acetate **16b** was obtained as an oil that crystallized on standing, and recrystallized from methanol as well-formed prisms (0.25 g): mp 146–148°; ν_{max}^{KBr} 1738, 1240 (acetate), and 1640 cm^{-1} (weak, C=C); $[\alpha]D$ +35.8° (*c* 1.54); pmr δ 1.02 (C-18 methyl), 1.10 (C-19 methyl), 2.06 (O-acetate), 3.50 (–OCH_3), 4.44 (doublet, J = 8 cps, –OCHO–), 5.10 (H-3α), and 4.90–6.10 (complex 3 H region).

Anal. Calcd for $C_{26}H_{40}O_4$: C, 74.96; H, 9.68; mol wt, 416. Found: C, 74.94; H, 9.59; mol wt, 416 (mass spectrum).

In both tetrahydropyranyl ether **16a** and acetate **16b**, the region in the pmr spectrum between δ 4.90 and 6.10 integrated for three protons and bore a strong resemblance to that shown by compounds containing an allylic grouping.

3β-Pyranyloxy- and 3β-Acetoxy-14β,21;22,23-diepoxy-(21*S*)-methoxy-5β-(20*S*)-norcholane (17a and 17b).—A solution of 3β-pyranyloxy-14β,21-epoxy-(21*S*)-methoxy-5β-(20*S*)-norchol-22(23)-ene (**16a**, 0.10 g) in chloroform (5 ml) was stirred with *m*-chloroperbenzoic acid (35 mg) at room temperature for 18.5 hr. As reaction was still incomplete by tlc with 39:1 chloroform–ethyl acetate, a further 35 mg of peracid was added and stirring was continued for a total of 45 hr. No starting material remained and the mixture was washed with sodium bicarbonate solution and water and evaporated to a colorless oil which was purified by preparative layer chromatography on one plate (40 × 20 × 0.2 cm) in 19:1 ligroin–ethyl acetate. Epoxide **17a** was obtained as an oil (91 mg) which did not crystallize: pmr δ 0.96 (C-18 methyl), 1.00 (C-19 methyl), 2.5–3.00 (complex 3 H region, protons α to oxide), 3.48 (OCH_3), 4.00 (THP acetal H), 4.56 (doublet, J = 8 Hz, –OCHO–), and 4.65 (H-3α).

Since the tetrahydropyranyl ether **17a** did not crystallize, acetate **17b** was prepared by epoxidation of 3β-acetoxy olefin **16b**. The olefin (0.27 g) was stirred at room temperature in chloroform (10 ml) with *m*-chloroperbenzoic acid (0.15 g) for 97 hr, and the product was isolated as described above. Epoxide **17b** crystallized from methanol as well-formed prisms (0.16 g): mp 182–184°; $[\alpha]D$ +19° (*c* 1.84); pmr δ 1.02 (C-18 methyl), 1.06 (C-19 methyl), 2.06 (O acetate), 2.55–3.00 (complex 3 H region, protons α to oxide), 3.52 (–OCH_3), 4.56 (doublet, J = 8 Hz, –OCHO–), and 5.12 (H-3α).

Anal. Calcd for $C_{26}H_{40}O_5$ (mol wt, 432): C, 72.19; H, 9.32. Found: C, 72.30; H, 9.15. The mass spectrum showed a peak at *m/e* 400 for loss of 32 (CH_3OH) from the molecular ion at *m/e* 432.

The pmr spectra at δ 2.5–3.00 of tetrahydropyranyl ether **17a** and the acetate **17b** resembled that of propylene oxide.

Condensation of 3β-Acetoxy-14,21-epoxy-20-formylpregn-20(21)-ene (15a) with Malonic Acid.—To a solution of epoxide **17b** (0.16 g) in acetone (15 ml) was added a solution of periodic acid (0.45 g) in acetone (8 ml)–water (1.5 ml). The mixture was heated at reflux for 1 hr, cooled, diluted with water, and extracted with diethyl ether. The extract was washed well with water and evaporated to a yellow, acrid-smelling, lacrymatory oil. The oil was held at 60° (0.1 mm) for 1 hr and chromatographed on silica gel. Elution with 9:1 ligroin–ethyl acetate gave some unchanged epoxide, followed by a more polar oily fraction (99 mg) identified by spectral characteristics as aldehyde **15a**: ν_{max}^{film} 1720, 1650, and 1600 cm^{-1}; pmr δ 0.94 (C-18 methyl), 1.02 (C-19 methyl), 2.06 (O acetate), 2.66 (multiplet, H-17), 5.12 (H-3), 7.08 (H-21), and 9.32 (–CHO).

Aldehyde **15a** (0.10 g) was dissolved in pyridine (3 ml) and morpholine (0.10 g), and malonic acid (0.10 g) was added. The mixture was warmed on a steam bath for 1 hr, cooled, acidified with 2 *N* hydrochloric acid, and extracted with diethyl ether. Removal of solvent gave an oil with an infrared spectrum superimposable on that of starting material. Extending the reaction time had no effect. Recovered aldehyde was redissolved in pyridine (3 ml) with morpholine (0.20 g) and malonic acid (0.20 g), and the solution was heated at reflux for 1 hr. The product was isolated as noted directly above. Tlc with 7:3 ligroin–ethyl acetate indicated the presence of relatively nonpolar diene **18**, which was isolated by preparative thin layer chromatography with 4:1 ligroin–ethyl acetate mobile phase and crystallized from methanol as needles (15 mg): pmr δ 1.02 (combined C-18 and -19 methyls), 2.06 (O acetate), 4.7 (J_{bc} = 10 Hz, J_{cd} = 2 Hz, H_c), 4.94 (J_{bd} = 17 Hz, J_{cd} = 2 Hz, H_d), 5.1 (H-3α), 6.22 (H_a), and 6.24 (J_{bd} = 17 Hz, J_{bc} = 10 Hz, H_b).

Registry No.—**3a**, 464-82-4; **3b**, 23337-56-6; **4**, 23337-57-7; **7c**, 23359-75-3; **11a**, 23359-76-4; **11b**, 23337-58-8; **11c**, 23337-59-9; **11d**, 23359-77-5; **15a**, 23359-78-6; **16a**, 23337-60-2; **16b**, 23337-61-3; **17a**, 23337-62-4; **17b**, 23337-63-5; **18**, 23359-79-7.