Fungal Extractives. 12.[1a] Construction of the Vellerane Skeleton with Total Syntheses of Racemic Velleral, Vellerolactone, and Pyrovellerolactone. Revised Structures[1b]

Jan Froborg*[1c] and Göran Magnusson*

Contribution from Organic Chemistry 2, Chemical Center, The Lund Institute of Technology, Box 740, S-220 07 Lund 7, Sweden. Received April 24, 1978

Abstract: Velleral, vellerolactone, and pyrovellerolactone, three hydroazulenic sesquiterpenoids from basidiomycetes of the genus *Lactarius*, have been prepared by cycloaddition of enamines of bicyclo[3.3.0]octanones to 4-substituted tetrolic esters, followed by fission of the four-membered ring and subsequent hydrogenolytic deamination with diborane. Further functional group manipulation gave the natural compounds. X-ray crystallographic analyses of key synthetic intermediates, together with spectroscopic data on synthetic stereoisomers of the natural compounds, necessitate a revision of the earlier suggested structures for all three title compounds. Molecular mechanics calculations revealed that the configuration of a methyl group on carbon atom 3 in the seven-membered ring has a profound effect on the conformation of the hydroazulenic ring system. These findings are corroborated by the X-ray crystallographic results.

Sesquiterpenoid fungal metabolites with the hydroazulenic vellerane[2] skeleton, together with compounds with marasmane[3] and *seco*-vellerane[4] skeletons, have been reported in increasing numbers during recent years. Synthetic work toward the vellerane[5-7] and *seco*-vellerane[8] skeletons has been a main objective for several years in this laboratory while work on the marasmane skeleton[9,10] (including a total synthesis of marasmic acid[11]) has been done by others.

We now wish to report an efficient synthetic route to the title compounds (**1**, **2**, and **3**) and to stereoisomers of these. This has

velleral   vellerolactone   pyrovellerolactone
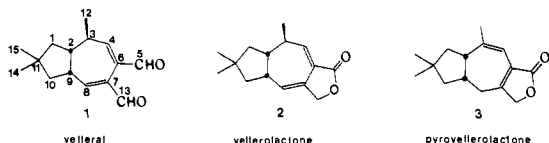
necessitated revision of the previously reported structures. The methyl group (Me-12 in velleral and vellerolactone; for numbering, see **1** and ref 6) is situated syn to the ring-junction hydrogen atoms, and not in the anti arrangement previously reported[2b-d] and the carbonyl group in vellerolactone and pyrovellerolactone is situated on C-5 instead of C-13.[2c,d] The latter accords with the recently revised structures for lactarorufin A and B[2h] and also with the structure of isolactarorufin.[12]

In the planning of the synthesis it was assumed that the Me-12 group was situated anti to the ring-junction hydrogen atoms and that C-13 carried the carbonyl group oxygen atom in the lactones (**2** and **3**). Accordingly, the general route shown in Scheme I was worked out and successfully carried through. However, after manipulation of the functional groups, the final products were found to be isomers of the natural products, showing very similar but significantly different spectral properties. For pyrovellerolactone (**3**) this could only mean that the carbonyl oxygen atom should be on C-5 instead of C-13 (the cis arrangement of the ring-junction hydrogen atoms had been confirmed by synthesis[5]). Compounds **1**, **2**, and **3** have been shown[2c,d,5] to have the same stereostructure (where applicable). As a consequence of this and of the synthetic results shown in Scheme I, two explanations were possible for the difference in stereostructure between the synthetic and natural compounds: either the previously determined C-3 stereostructure[2b] was incorrect or epimerization (at C-3 or C-9) had occurred in some synthetic step (presumably in the thermolysis of the four-membered ring or possibly in the BH₃ hydrogenolysis). To investigate this (and at the same time put the stereochemical outcome of the synthetic route on safe ground)

Scheme I
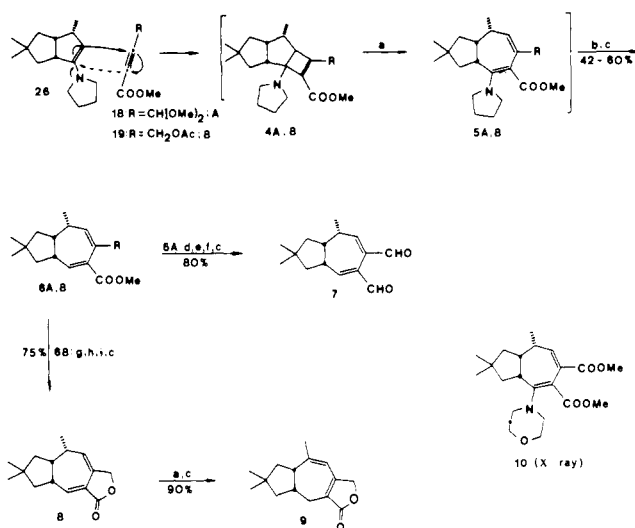
a) toluene reflux; b) BH₃; tetrahydrofuran; c) column chromatography; d) DIBAH; toluene; e) MnO₂; carbon tetrachloride; f) *p*-toluenesulfonic acid; acetone/water; g) NaOH; dioxane/water; h) HCl; water; i) *p*-toluenesulfonic acid; dioxane.

an X-ray crystallographic analysis was made[13] (Figure 1) of an intermediate[6] (**10**, Scheme I) obtained directly after the thermolysis step and thus still carrying the amino substituent (morpholine). (In spite of numerous attempts, we have been unable to obtain suitable crystals of the naturally occurring compounds or their derivatives for X-ray diffraction studies.) The analysis showed that **10** had retained its cis ring junction configuration through the thermolysis. A structural correlation had thus to be made between a BH₃-reduced compound and one with *known* ring-junction stereostructure in order to determine the stereochemical result of the BH₃ reaction[14] (Scheme II). Reduction of the ester function of **6A** with diisobutylaluminum hydride (DIBAH) followed by acid treatment in dry carbon tetrachloride gave the acetal **12**. Hydrolysis of **12** with acidic aqueous acetone gave the furan **13**, which had the same spectral properties as reported for the furan synthesized earlier[5] using a reaction analogous to one known[15] to give a hydroazulene with cis ring-junction hydrogen atoms (which has also been obtained from pyrovellerolactone (**3**) by DIBAH reduction[5]). This showed that also the BH₃-reduction step goes without epimerization at the neighboring asymmetric center. The results reported above thus showed that the original vel-
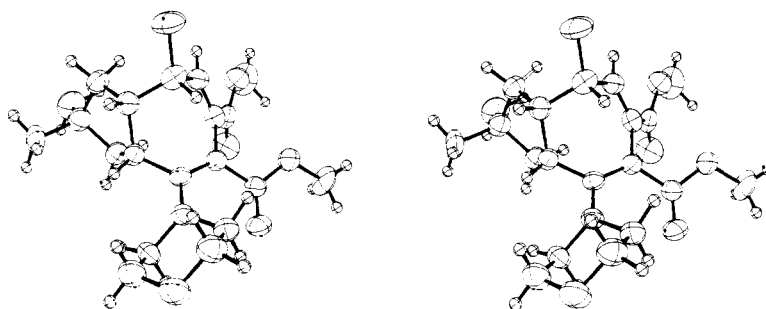
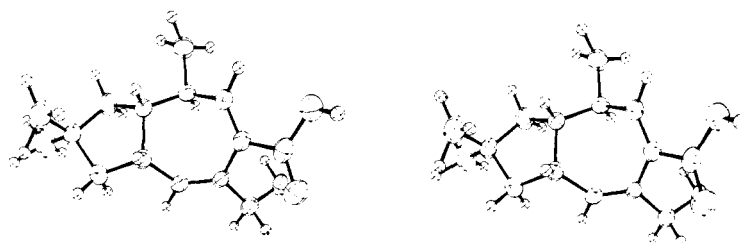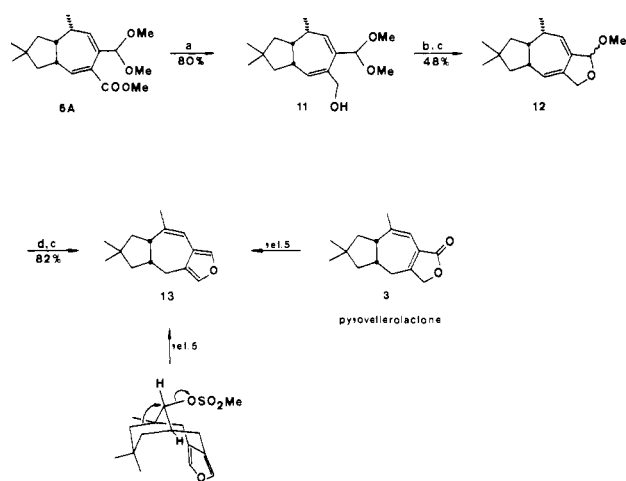**Figure 1.** Stereographic pair of drawings of **10**, obtained by X-ray crystallographic analysis.[13]

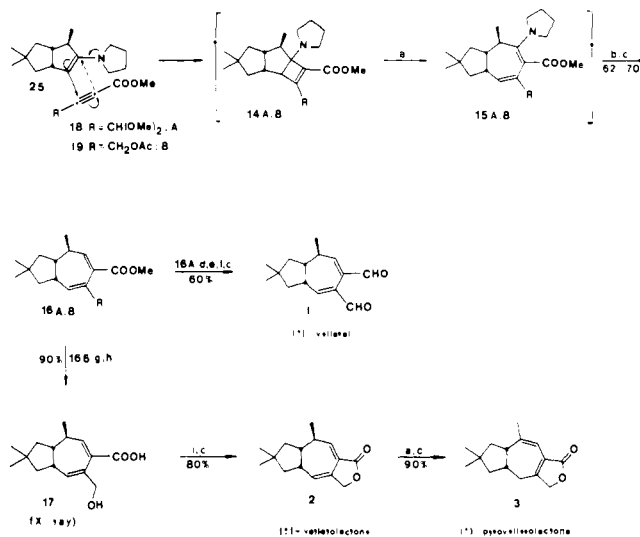**Figure 2.** Stereographic pair of drawings of **17**, obtained by X-ray crystallographic analysis.[16]

Scheme II

a) DIBAH; toluene; b) *p*-toluenesulfonic acid; carbon tetrachloride; c) column chromatography; d) HCl; acetone/water.

Scheme III

a) toluene reflux; b) BH$_3$; tetrahydrofuran; c) column chromatography; d) DIBAH; toluene; e) MnO$_2$; carbon tetrachloride; f) *p*-toluenesulfonic acid; acetone/water; g) NaOH; dioxane/water; h) HCl; water; i) *p*-toluenesulfonic acid; dioxane.

leral stereostructure[2b] was incorrect in the assignment of the C-3 configuration. Furthermore, the natural lactones (**2** and **3**) should have the carbonyl group in the C-5 position. Accordingly, the synthetic scheme was modified by changing from enamine **26** to **25** as starting material, thus permitting synthesis of the natural compounds with the Me-12 group syn to the ring-junction hydrogen atoms and with the lactone carbonyl group on C-5 (Scheme III).

To determine the stereostructure of the natural products (**1**, **2**, and **3**) unequivocally, an X-ray crystallographic structure[16] was determined for the hydroxy acid **17** (Figure 2 and Scheme III). During the synthetic work, molecular mechanics calculations[17] have been made on several derivatives having the hydroazulenic carbon skeleton. In essence, these compounds seem to have a strongly preferred conformation with the Me-12 group in a pseudoequatorial position. Thus, compounds with the Me-12 group syn to the ring-junction hydrogen atoms (e.g., **1**, **2**, **16**, and **17**) have an exo conformation for the hydroazulenic ring system (cf. Figures 2 and 3) and compounds with Me-12 anti to the ring-junction hydrogen atoms (e.g., **6**, **7**, **8**, and **10**) have an endo conformation (cf. Figures 1 and 3). The

calculated energy differences between the conformational pairs syn-exo: syn-endo or anti-exo:anti-endo lie in the range 3–5 kcal/mol. It is to be noted that an exo conformation could be derived for natural velleral[2b] from NMR spectral analysis.

Both the syn-exo vellerolactone (**2**) and the anti-endo isomer **8** have their hydrogen atom on C-3 in a favorable position for a 1,5-sigmatropic suprafacial hydride shift (cf. Figure 3). Both compounds gave the corresponding pyrolactones on heating[2c] (see Schemes I and III and Figure 3). The partly erroneous stereostructure determination of velleral,[2b] which indicated that the methyl group on C-3 should be situated anti to the ring-junction hydrogen atoms, has caused considerable confusion. For clarity: in the structures in our earlier papers, the methyl group on C-3 (in velleranes carrying a C-3 hydrogen atom) should be situated syn (not anti) to the ring-junction hydrogen atoms (see **1**) in the *natural* products and in deriv-
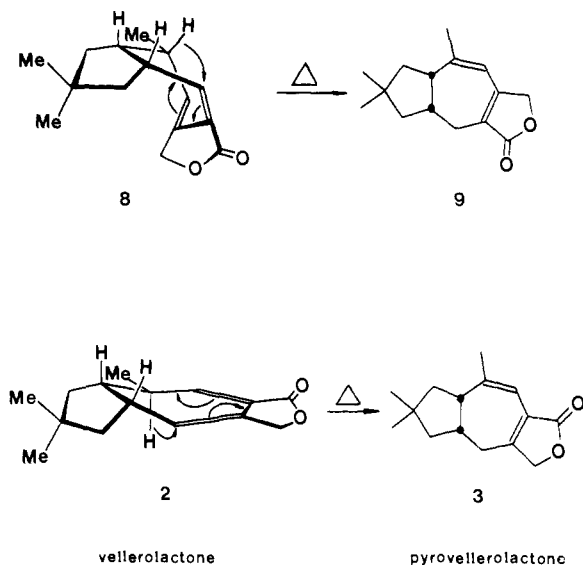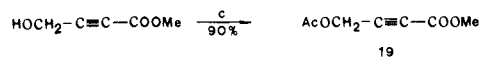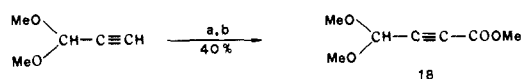
Figure 3. 1,5-Sigmatropic, suprafacial hydride shift in the most stable conformations of lactones **8** and **2**.

Scheme IV

a) EtMgCl; b) ClCOOMe; c) Ac$_2$O.

atives of these, and situated anti (see **7**) in all *synthetic* products.

Syntheses of Velleranes. Symmetrical acetylene derivatives (like dimethyl acetylenedicarboxylate) function well in addition reactions with enamines[6,18] and as dienophiles in Diels–Alder reactions.[19] Later selective manipulation of the functional groups originally present in the starting acetylenes can, however, be difficult (if not impossible; e.g., regiospecific formation of a γ-lactone ring as in **2** and **8**). A less obvious difficulty is the reduction of a symmetrical diester derivative to a symmetrical dialdehyde (such as velleral) either directly or by oxidation of a preformed diol. In both cases a plethora of products can be formed owing to the presence of a series of equilibria between cyclic and acyclic forms (e.g., hydroxy aldehydes, cyclic hemiacetals, lactones, alkoxy lactones). To avoid these difficulties, unsymmetrical acetylenes of the type shown in Scheme IV should be used. These will react in a regiospecifically predictive way, and furthermore, subsequent selective functional group manipulation should be possible under different reaction conditions. Acetylenes of the propiolic ester type have been used for addition to enamines;[20] tetrolic ester type acetylenes (like **18** and **19**) have been reported[21] (although not used for reaction with enamines).

Starting with the enamines **25** and **26**, the two synthetic routes leading to velleral (**1**), vellerolactone (**2**), and pyrovellerolactone (**3**) (Scheme III) and to the isomers **7**, **8**, and **9** (Scheme I) follow the same principal paths. Thus, to the freshly prepared pyrrolidine enamine (**25** or **26**) was added the appropriate acetylene ester (**18** or **19**). [In a previous synthesis[6] of inter alia the dienamine diester **10** (Scheme I), we added dimethyl acetylenedicarboxylate slowly to a morpholine enamine with toluene as solvent. However, with the less reactive

Scheme V
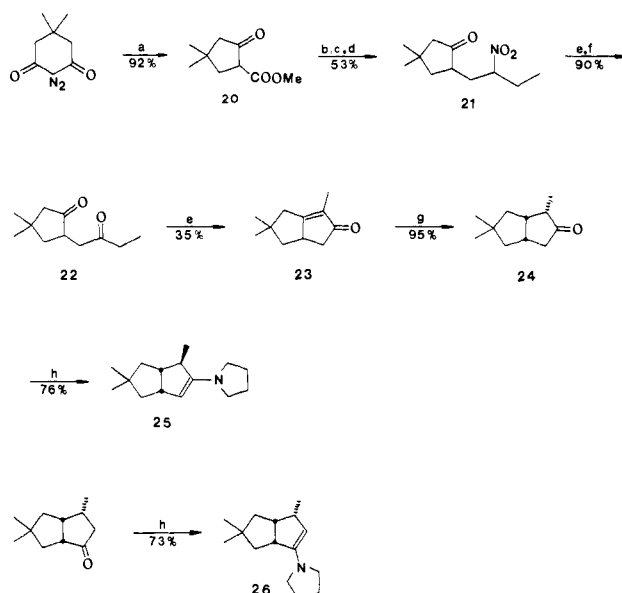
a) hν; methanol/tetrahydrofuran; b) NaH; tetrahydrofuran; c) 2-nitrobut-1-ene; d) conc. HCl/HOAc; e) NaOH; abs. ethanol; HCl; water; g) H$_2$, 760 mm; Rh/Al$_2$O$_3$; abs. ethanol; h) *p*-toluenesulfonic acid, pyrrolidine; benzene.

acetylenes used here (**18** and **19**), more reactive (pyrrolidine) enamines[22] had to be used. These enamines turned out to be very prone to decomposition and consequently, the shortest reaction times possible were desired. This was arranged by running the reaction without solvent.] After ca. 5 h at room temperature, the resulting viscous liquid was refluxed in toluene (opening the cyclobutene ring) and the dienamine thus obtained was reduced with diborane[14] to give the diene ester (**16** or **6**, Schemes I and III) in 40–70% overall yield (from **25** and **26**). The ester function of the diene acetal ester (**16A** or **6A**) was reduced with diisobutylaluminum hydride (DIBAH) and the resulting acetal alcohol was oxidized with active manganese dioxide, giving a diene acetal aldehyde. Hydrolysis of the latter gave velleral (**1**) or the isomeric diene dialdehyde **7**.

Hydrolysis of both ester functions in the methyl ester acetate (**16B** or **6B**) gave a hydroxy acid which could be transformed by *p*-toluenesulfonic acid in dioxane into vellerolactone (**2**) or the isomeric lactone (**8**). Both lactones were transformed into their pyro derivatives (pyrovellerolactone (**3**) and the isomeric lactone **9**; cf. Figure 3). A mixture of the pyrolactones has been prepared earlier[5] in this laboratory.

Syntheses of Acetylenes. The unsymmetrical acetylenes used here were prepared (Scheme IV) in analogy with a method previously reported.[21] Thus, 3,3-dimethoxypropyne (see Experimental Section) was converted into methyl 4,4-dimethoxytetrolate (**18**) by treatment with ethylmagnesium chloride followed by methyl chloroformate. Methyl 4-acetoxytetrolate (**19**) was prepared by acetylation of the corresponding alcohol.

An alternative route to compound **18** (and also to the corresponding amide and nitrile) was reported[23] after the completion of our acetylene syntheses.

Synthesis of Enamines. Ketone **24** (Scheme V) was used in a synthesis of the protoilludane skeleton.[24] In the present work, another route to **24** was used (Scheme V). Diazodimedon[25] was transformed into the β-keto ester **20** by photolysis in the presence of methanol (instead of water as in the preparation of the corresponding β-keto acid[26]). Treatment of **20** with sodium hydride, followed by a slow addition of 2-nitrobut-1-ene,[27] gave a crude product which was refluxed in concentrated

HCl/HOAc (2/5) giving the nitro ketone **21**. A Nef reaction with **21** gave the 1,4-diketone **22**,[24] which was transformed (without prior purification) into the bicyclic ketone **23**.[24] Hydrogenation of **23** over Rh/Al$_2$O$_3$ gave the saturated ketone **24**[24] (anti:syn, 94:6; GLC). Equilibration of **24** in either base or acid changed the isomer ratio (anti:syn, 3:7). The ketone **24** was transformed into the enamine **25**. Apparently only the more stable syn configuration of **25** was formed under the acidic reaction conditions used (the velleral isomer **7** could not be detected in the preparation of velleral (**1**); see Schemes I and III).

The morpholine analogue of enamine **26** was described in an earlier paper[6] where it was used for the preparation of **10** (Scheme I). The pyrrolidine enamine **26** (Scheme V) used here was prepared by the same method as **25**. In this case of course no epimerization of the methyl group is possible.

In summary, the enamines **25** and **26** can be used for stereospecific preparation of hydroazulenes with cis ring-junction hydrogen atoms and with a C-3 methyl group syn or anti to these.

Experimental Section

NMR spectra (60 or 100 MHz) were run in CDCl$_3$ (except for **17**) with Me$_4$Si as internal standard. IR spectra were run as liquid films unless otherwise stated. Melting points are uncorrected. GLC was performed on a 50-m SE-30 glass capillary column. High-resolution mass spectra were run on a Varian MAT 311 instrument (Department of Clinical Chemistry, University Hospital, Lund).

3,3-Dimethoxypropyne was prepared as described[28] for the diethoxy analogue: bp 57–58 °C (125 mm); NMR δ 5.19 (d, 1 H, *J* = 1.5 Hz), 3.41 (s, 6 H), 2.59 ppm (d, 1 H, *J* = 1.5 Hz).

Methyl 4,4-dimethoxytetrolate (18) was prepared from 3,3-dimethoxypropyne by the method described for methyl 4-tetrahydropyranyloxymethyltetrolate:[21] yield 40%; bp 52 °C (1 mm); IR ν 2250, 1725 cm^{-1}; NMR δ 5.27 (s, 1 H), 3.83 (s, 3 H), 3.42 ppm (s, 6 H).

Methyl 4-Acetoxytetrolate (19). The hydroxytetrolate[21] and an excess of acetic anhydride were refluxed for 12 h. Evaporation and distillation gave **19**: yield 90%; bp 67–69 °C (0.3 mm); IR ν 2250, 1755, 1725, 1270, 1225 cm^{-1}; NMR δ 4.80 (s, 2 H), 3.77 (s, 3 H), 2.10 ppm (s, 3 H).

2-Methoxycarbonyl-4,4-dimethylcyclopentanone (20). An ice-cooled, cylindrical, three-necked flask (sampling, water condenser and insertion lamp; quartz-jacketed, air-cooled mercury lamp; 400 W/150 V) was charged with 2-diazodimedone[25] (50 g, 0.307 mol), dry methanol (30 mL), and dry tetrahydrofuran (1500 mL). The progress of the reaction was followed by the evolution of nitrogen (ca. 2 L/h). Removal of the solvent gave practically pure **20** (92%): bp 61 °C (0.5 mm); IR ν 1763, 1735 cm^{-1}; NMR δ 3.75 (s, 3 H), 3.40 (t, 1 H, *J* = 10 Hz), 2.22 (s, broad, 2 H), 1.25 (s, 3 H), 1.08 ppm (s, 3 H).

4,4-Dimethyl-2-(β-nitrobutyl)cyclopentanone (21). The keto ester **20** (6.8 g, 40 mmol) was added dropwise to a cooled (−10 °C) suspension of sodium hydride (45 mmol) in dry tetrahydrofuran (50 mL). After stirring for 20 min, 2-nitrobut-1-ene[27] (4.04 g, 40 mmol) was slowly added dropwise (temperature increase to ca. 20 °C). After 1 h at 20 °C, acetic acid (10 mL) was added and the tetrahydrofuran was evaporated. The residue was refluxed in a mixture of concentrated hydrochloric acid and acetic acid (2:5, 70 mL) for 10 h. Extraction with ether, drying (Na$_2$SO$_4$), and distillation gave the nitro ketone **21** (4.5 g, 53%): bp 90–95 °C (0.2 mm); IR ν 1745, 1555 cm^{-1}; NMR δ 4.67 (m, 1 H), 1.20 (s, 3 H), 1.07 ppm (s, 3 H).

4,4-Dimethyl-2-(β-oxybutyl)cyclopentanone (22).[24] The nitro ketone **21** (4 g) was stirred for 30 min in a solution of sodium hydroxide (1 g) in absolute ethanol (30 mL). Evaporation of the solvent gave a salt that was dissolved in water (10 mL) and added to a hydrogen chloride solution (2 M, 20 mL). After 15 h at room temperature, the aqueous solution was extracted with ether. The ether extract was dried (Na$_2$SO$_4$), filtered, and evaporated, giving a yellow residue (3 g, 90%) containing no starting nitro ketone (GLC). The crude diketone **22** decomposed on heating and was consequently used in the next step without prior distillation.

2,7,7-Trimethylbicyclo[3.3.0]oct-1-en-3-one (23).[24] The crude diketone **22** (3 g) was dissolved in ethanolic sodium hydroxide (0.5 M, 40 mL) and the solution was stirred for 20 h at 40 °C. The reaction

mixture was evaporated, diluted with water, acidified, and extracted with ether. Drying of the ether phase (Na$_2$SO$_4$), filtration, and distillation gave the ketone (**23**), yield 1.12 g (35%), bp 69–71 °C (0.7 mm).

2,7,7-Trimethylbicyclo[3.3.0]octan-3-one (24).[24] The unsaturated ketone **23** (1.1 g) was hydrogenated (Rh/Al$_2$O$_3$, 10%, 100 mg) in absolute ethanol (30 mL) at atmospheric pressure and room temperature. Filtration and distillation gave the saturated ketone **24** (95%), bp 60–61 °C (1 mm); GLC analysis showed a 94:6 ratio between the anti and the syn epimers. Equilibration of **24** (catalytic amount of MeONa in MeOH or 2 M HCl in dioxane) changed the epimer ratio to 3:7 between the anti and syn epimers, respectively.

4,7,7-Trimethyl-3-(N-pyrrolidino)bicyclo[3.3.0]oct-2-ene (25). The ketone **24** (0.8 g), pyrrolidine (1.5 g), and *p*-toluenesulfonic acid (ca. 1 mg) were dissolved in benzene (25 mL) and refluxed in a Soxhlet apparatus (Linde 3A molecular sieve) for 10 h. Evaporation of the benzene and distillation gave the unstable pyrrolidine enamine **25** (0.8 g, 76%): bp 95–97 °C (0.5 mm); IR ν 3055, 1660, 1630, 1390, 1370 cm^{-1}; NMR δ 4.00 (s, broad, 1 H), 1.15 (d, 3 H, *J* = 7 Hz), 1.02 (s, 3 H), 0.92 ppm (s, 3 H).

4,7,7-Trimethyl-2-(N-pyrrolidino)bicyclo[3.3.0]oct-2-ene (26) was prepared from 4,7,7-trimethylbicyclo[3.3.0]octan-2-one[6] as described above for enamine **25**: yield 73%, bp 82 °C (0.2 mm); IR ν 3070, 1630, 1390, 1370 cm^{-1}; NMR δ 3.80 (s, broad, 1 H), 1.08 (s, 3 H), 0.99 (d, 3 H, *J* = 7 Hz), 0.97 ppm (s, 3 H). The enamines **25** and **26** are unstable and had to be used immediately.

Addition Reaction between the Enamines and the Acetylenes. Opening of the Cyclobutene Ring. The acetylene ester (**18** or **19**, 4 mmol) was added dropwise to the freshly distilled enamine (**25** or **26**, 4 mmol) containing a trace of hydroquinone (room temperature, N$_2$ blanket). Stirring for ca. 5 h gave a viscous cycloaddition product which was almost pure [e.g., **4A**: NMR δ 5.35 (s, 1 H), 3.78 (s, 3 H), 3.45 (s, 6 H), 3.10 (s, 1 H), 1.07 (s, 3 H), 1.02 (d, 3 H, *J* = 6 Hz), 1.00 ppm (s, 3 H)]. The crude product was refluxed (1–2 h) in dry toluene in order to open up the cyclobutene ring. The reaction was followed by TLC (SiO$_2$, ethyl acetate). After removal of the solvent, the crude dienamine ester [3.g., **5A**: NMR δ 5.87 (d, 1 H, *J* = 5 Hz), 5.00 (s, 1 H), 3.75 (s, 3 H), 3.50 (s, 3 H), 3.42 (s, 3 H), 1.08 (s, 3 H), 1.05 (d, 3 H, *J* = 6 Hz), 0.92 ppm (s, 3 H)] was used directly in the next step.

General Deamination Procedure by Diborane Reduction. The crude dienamine ester (**5** or **15**) obtained above was reduced with diborane as previously[14] described. Column chromatography (SiO$_2$, 75 g, hexane/ethyl acetate, 7/3) gave the pure hydroazulenic diene ester (**6** or **16**) in 42–70% overall yield.

7-Methoxycarbonyl-6-dimethoxymethyl-2,2,4-trimethyl-1,2,3,-3a,4,8a-hexahydroazulene (6A).[29] Yield 42%; IR ν 1720, 1620, 1465, 1440, 1250, 1076 cm^{-1}; NMR δ 7.48 (d, 1 H, *J* = 8 Hz), 6.17 (dd, 1 H, *J* = 6 and 1.5 Hz), 5.33 (m, 1 H), 3.78 (s, 3 H), 3.40 (s, 3 H), 3.30 (s, 3 H), 1.00 (s, 3 H), 0.98 (d, 3 H, *J* = 7.5 Hz), 0.90 ppm (s, 3 H); mass spectrum *m/e* (rel intensity) 308 (M$^+$, 8, C$_{18}$H$_{28}$O$_4$), 261 (100, base peak).

Anal. Calcd for C$_{18}$H$_{28}$O$_4$: mol wt, 308.1988. Found: mol wt, 308.1989.

6-Acetoxymethyl-7-methoxycarbonyl-2,2,4-trimethyl-1,2,3,3a,-4,8a-hexahydroazulene (6B). Yield 60%; IR ν 1750, 1725, 1645, 1620, 1465, 1440 cm^{-1}; NMR δ 7.58 (d, 1 H, *J* = 8 Hz), 6.00 (d, 1 H, *J* = 6 Hz), 4.98, 4.68 (AB q, 2 H, *J*$_{AB}$ = 12 Hz), 3.80 (s, 3 H), 2.00 (s, 3 H), 1.02 (s, 3 H), 0.98 (d, 3 H, *J* = 7.5 Hz), 0.91 ppm (s, 3 H); mass spectrum *m/e* (rel intensity) 306 (M$^+$, 13, C$_{18}$H$_{26}$O$_4$), 246 (100, base peak).

Anal. Calcd for C$_{18}$H$_{26}$O$_4$: mol wt, 306.1831. Found: mol wt, 306.1834.

6-Methoxycarbonyl-7-dimethoxymethyl-2,2,4-trimethyl-1,2,3,3a,4,8a-hexahydroazulene (16A). Yield 62%; IR ν 1721, 1640, 1617, 1461, 1438 cm^{-1}; NMR δ 7.07 (d, 1 H, *J* = 6 Hz), 6.32 (s, broad, 1 H), 5.38 (m, 1 H), 3.77 (s, 3 H), 3.40 (s, 3 H), 3.15 (s, 3 H), 1.10 (s, 3 H), 1.08 (d, 3 H, *J* = 6 Hz), 0.95 ppm (s, 3 H); mass spectrum *m/e* (rel intensity) 308 (M$^+$, 20, C$_{18}$H$_{28}$O$_4$), 261 (100, base peak).

Anal. Calcd for C$_{18}$H$_{28}$O$_4$: mol wt, 308.1988. Found: mol wt, 308.2035.

7-Acetoxymethyl-6-methoxycarbonyl-2,2,4-trimethyl-1,2,3,3a,-4,8a-hexahydroazulene (16B). Yield 70%; IR ν 1747, 1723, 1642, 1618, 1460, 1439 cm^{-1}; NMR δ 7.17 (d, 1 H, *J* = 6 Hz), 6.10 (s, broad, 1 H), 5.01, 4.61 (AB q, 2 H, *J*$_{AB}$ = 12.5 Hz), 3.78 (s, 3 H), 2.02 (s, 3

H), 1.10 (s, 3 H), 1.07 (d, 3 H, J = 6 Hz), 0.93 ppm (s, 3 H); mass spectrum *m/e* (rel intensity) 306 (M+, 7, $C_{18}H_{26}O_4$), 246 (100, base peak).

Anal. Calcd for $C_{18}H_{26}O_4$: mol wt, 306.1831. Found: mol wt, 306.1830.

Preparation of Velleral (1) and Its Anti Isomer (7). Diisobutylaluminum hydride (DIBAH, 2 mmol in 5 mL of dry toluene) was added dropwise to the diene acetal ester (**16A** or **6A**, 1 mmol) dissolved in dry toluene (15 mL) and cooled to −50 °C. After ca. 1 h, the reaction was quenched by addition of H_2O (0.5 mL), Na_2SO_4 (ca. 100 mg), and 10% NaOH solution (0.5 mL). Ether (30 mL) was added and the reaction mixture was stirred for 1 h to granulate the precipitated salts. Filtration and evaporation gave almost pure diene acetal alcohol [from **16A**, NMR δ 6.12 (d, 1 H, J = 3 Hz), 5.96 (d, 1 H, J = 5 Hz), 4.68 (s, 1 H), 4.36, 3.98 (AB q, 2 H, J_{AB} = 12 Hz), 3.39 (s, 6 H), 1.10 (s, 3 H), 1.01 (d, 3 H, J = 7 Hz), 0.96 ppm (s, 3 H); from **6A**; NMR δ 6.40 (d, 1 H, J = 7.5 Hz), 6.00 (d, 1 H, J = 5.5 Hz), 4.66 (s, 1 H), 4.36, 3.94 (AB q, 2 H, J_{AB} = 12 Hz), 3.43 (s, 3 H), 3.40 (s, 3 H), 1.03 (s, 3 H), 1.02 (d, 3 H, J = 7.5 Hz), 0.90 ppm (s, 3 H)]. The alcohol was oxidized with active manganese dioxide (0.5 g, Merck, *gefällt aktiv*) in carbon tetrachloride (7 mL, room temperature, ca. 1 h). The crude diene acetal aldehyde [e.g., from **16A**: NMR δ 9.44 (s, 1 H), 6.88 (d, 1 H, J = 5.5 Hz), 6.28 (s, broad, 1 H), 5.47 ppm (m, 1 H)] was treated with a trace of *p*-toluenesulfonic acid in acetone/water (20/1, 10 mL) for 2 h. After neutralization (K_2CO_3), the solvent was evaporated and the residue extracted with ether. After drying and evaporation of the ether phase, the residue was chromatographed (SiO_2 column, 12 g; hexane/ethyl acetate, 7/3). This gave pure, racemic velleral (**1**) and the isomer **7** in 80% overall yield (from **16A** and **6A**).

(±)-**Velleral (1)** had mp 71–72 °C (from hexane); spectral data (IR, NMR, MS, UV) were the same as for the natural compound.[2b]

6,7-Bisformyl-2,2,4-trimethyl-1,2,3,3a,4,8a-hexahydroazulene (7). IR ν 1708, 1690, 1630, 1615, 1388, 1371 cm⁻¹; NMR δ 9.51 (s, 1 H), 9.48 (s, 1 H), 7.50 (d, 1 H, J = 8 Hz), 6.93 (d, 1 H, J = 6 Hz), 1.15 (d, 3 H, J = 6 Hz), 1.03 (s, 3 H), 0.97 ppm (s, 3 H); mass spectrum *m/e* (rel intensity) 232 (M+, 100, base peak, $C_{15}H_{20}O_2$), 204 (57), 119 (73), 105 (72), 95 (60), 91 (83).

Anal. Calcd for $C_{15}H_{20}O_2$: mol wt, 232.1463. Found: mol wt, 232.1469.

Preparation of Vellerolactone (2) and the Isomer 8. The diene acetate ester (**16B** or **6B**, 1 mmol) was hydrolyzed with sodium hydroxide (5 mmol) in dioxane/water (1/1, 10 mL) at room temperature. After ca. 15 h, the mixture was acidified with hydrochloric acid (2 M), saturated with sodium chloride, and extracted with ether. Drying of the ether phase and evaporation gave crude diene alcohol acid (e.g., **17**; see below). The hydroxy acid was stirred in dioxane with a catalytic amount of *p*-toluenesulfonic acid (room temperature, 36 h). Column chromatography (SiO_2, 10 g; hexane/ethyl acetate, 7/3) gave the pure lactone (**2** or **8**) in ca. 70% overall yield.

6-Carboxy-7-hydroxymethyl-2,2,4-trimethyl-1,2,3,3a,4,8a-hexahydroazulene (17). The crude acid was recrystallized from ethyl acetate for X-ray crystallography.[16] Larger crystals were obtained by letting hexane vapor diffuse into a semisaturated solution of **17** in ethyl acetate (test tube with **17** in a closed vessel containing hexane): mp 142 °C; IR (KBr) ν 3342, 1697, 1638, 1610, 1387, 1377, 1370, 1020, 788, 700 cm⁻¹; NMR (acetone-d_6) δ 7.12 (d, 1 H, J = 6 Hz), 6.03 (s, broad, 1 H), 4.33, 4.13 (AB q, 2 H, J_{AB} = 13 Hz), 1.10 (s, 3 H), 1.06 (d, 3 H, J = 6 Hz), 0.96 ppm (s, 3 H).

(±)-**Vellerolactone (2)** had mp 78–80 °C; spectroscopic data (IR, NMR, MS, UV) were the same as for the natural compound.[2c]

2,2,4-Trimethyl-1,2,3,3a,4,8a-hexahydroazulene-6,7-carbolactone (8). UV λ_{max} (EtOH) 284 nm (ε 7300); IR ν 1765, 1675, 1648, 1370, 1355 cm⁻¹; NMR δ 6.85 (d, broad, 1 H, J = 3 Hz), 5.53 (s, broad, 1 H), 4.76 (m, 2 H), 1.14 (d, 3 H, J = 8 Hz), 1.08 (s, 3 H), 0.99 ppm (s, 3 H); mass spectrum *m/e* (rel intensity) 232 (M+, 58, $C_{15}H_{20}O_2$), 217 (100, base peak).

Anal. Calcd for $C_{15}H_{20}O_2$: mol wt, 232.1463. Found: mol wt, 232.1484.

Preparation of the Pyrolactones 3 and 9. The diene lactone (**2** or **8**, 60 mg) was refluxed in dry toluene (5 mL) for 4 h. Evaporation and column chromatography (SiO_2, 3 g; hexane/ethyl acetate, 7/3) of the residue gave the pure lactone (**3** or **9**) in 90% yield.

(±)-**Pyrovellerolactone (3)** had mp 72–74 °C; spectroscopic data (IR, NMR, MS, UV) were the same as for the natural compound.[2c]

2,2,4-Trimethyl-1,2,3,3a,8,8a-hexahydroazulene-6,7-carbolactone (9) had mp 90–92 °C; UV λ_{max} (EtOH) 281 nm (ε 13 900); IR (CCl₄) ν 1763, 1667, 1632 cm⁻¹; NMR δ 5.80 (s, broad, 1 H), 4.66 (s, broad, 2 H), 2.00 (s, broad, 3 H), 1.10 (s, 3 H), 1.05 ppm (s, 3 H); mass spectrum *m/e* (rel intensity) 232 (M+, 68, $C_{15}H_{20}O_2$), 217 (100, base peak), 187 (18) 148 (30), 131 (20), 95 (73), 91 (25).

Anal. Calcd for $C_{15}H_{20}O_2$: mol wt, 232.1463. Found: mol wt, 232.1474.

2,2,4-Trimethylfuro[6,7-*c*]-1,2,3,3a,8,8a-hexahydroazulene (13). The diene acetal ester **6A** (550 mg) was reduced with DIBAH as described above. The resulting diene acetal alcohol (**11**, 400 mg) was dissolved in carbon tetrachloride (10 mL) and a catalytic amount of *p*-toluenesulfonic acid was added. After stirring for ca. 2 h (TLC) the CCl₄ solution was washed with Na_2CO_3 solution and water, dried (Na_2SO_4), evaporated, and chromatographed (SiO_2 column, 15 g; hexane/ethyl acetate, 8/1). This gave the cyclic acetal **12** (170 mg) as a mixture of diastereomers [NMR δ 3.43, 3.39 ppm (s)]. The cyclic acetal **12** (70 mg) was dissolved in acetone (10 mL) and water (0.5 mL), and hydrochloric acid (2 m, 0.5 mL) was added. After ca. 2 h (TLC) the reaction mixture was neutralized with saturated NaHCO₃ solution, evaporated, and partitioned between ether and water. The ether phase was dried (Na_2SO_4), evaporated, and chromatographed (SiO_2 column, 15 g, hexane) giving pure furan **13** (82%). Spectral data (IR, NMR) were the same as previously[5] reported for this furan.

References and Notes

(1) (a) Part 11: see ref 7. (b) Presented in part at the Euchem Conference on Structure, Synthesis and Biosynthesis of Mono- and Sesquiterpenoids, Varenna, Italy, Aug 25–31, 1977. (c) Astra Pharmaceuticals AB, 151 85 Södertälje, Sweden.
(2) (a) S. Nozoe, H. Matsumoto, and S. Urano, *Tetrahedron Lett.*, 3125 (1971); (b) G. Magnusson, S. Thorén, and T. Drakenberg, *Tetrahedron*, **29**, 1621 (1973); (c) G. Magnusson and S. Thorén, *Acta Chem. Scand.*, **27**, 1573 (1973); (d) *ibid.*, **27**, 2396 (1973); (e) G. Magnusson, S. Thorén, J. Dahmén, and K. Leander, *Acta Chem. Scand. Ser. B*, **28**, 841 (1974); (f) W. M. Daniewski, A. Ejchart, J. Jurczak, L. Kozerski, and J. S. Pyrek, *Bull. Acad. Pol. Sci., Ser. Chim.*, **20**, 131 (1972); (g) W. M. Daniewski, M. Kocór, and B. Zoltowska, *ibid.*, **21**, 785 (1973); (h) M. Bogucka- Ledóchowska, A. Hempel, Z. Dauter, A. Konitz, E. Borowski, W. Daniewski, and M. Kocór, *Tetrahedron Lett.*, 2267 (1976); (i) G. Vidari, L. Garlaschelli, M. de Bernardi, G. Fronza, and P. Vita-Finzi, *ibid.*, 1773 (1975); (j) M. de Bernardi, G. Fronza, G. Vidari, and P. Vita-Finzi, *Chim. Ind. (Milan)*, **58**, 177 (1976).
(3) (a) J. J. Dugan, P. de Mayo, M. Nisbeth, J. R. Robinson, and M. Anchel, *J. Am. Chem. Soc.*, **88**, 2838 (1966); (b) G. Magnusson, S. Thorén, and B. Wickberg, *Tetrahedron Lett.*, 1105 (1972); (c) S. Nozoe, H. Kobayashi, S. Urano, and J. Furukawa, *ibid.*, 1381 (1977).
(4) (a) G. Magnusson and S. Thorén, *Tetrahedron*, **30**, 1431 (1974); (b) ref 2j.
(5) J. Froborg, G. Magnusson, and S. Thorén, *J. Org. Chem.*, **40**, 1595 (1975).
(6) T. Fex, J. Froborg, G. Magnusson, and S. Thorén, *J. Org. Chem.*, **41**, 3518 (1976).
(7) J. Froborg and G. Magnusson, *Tetrahedron*, in press.
(8) J. Froborg, G. Magnusson, and S. Thorén, *Acta Chem. Scand., Ser. B*, **28**, 265 (1974).
(9) D. Helmlinger, P. de Mayo, M. Nye, L. Westfelt, and R. B. Yeats, *Tetrahedron Lett.*, 349 (1970).
(10) S. R. Wilson and R. B. Turner, *J. Org. Chem.*, **38**, 2870 (1973).
(11) W. J. Greenlee and R. B. Woodward, *J. Am. Chem. Soc.*, **98**, 6075 (1976).
(12) A. Konitz, M. Bogucka-Ledóchowska, Z. Dauter, A. Hempel, and E. Borowski, *Tetrahedron Lett.*, 3401 (1977).
(13) J. Albertson, B. Briggman, and Å Oskarsson, *Cryst. Struct. Commun.*, submitted.
(14) J. Froborg, G. Magnusson, and S. Thorén, *Tetrahedron Lett.*, 1621 (1975).
(15) J. A. Marshall and J. J. Partridge, *Tetrahedron*, **25**, 2159 (1969).
(16) K. Ståhl, *Cryst. Struct. Commun.*, submitted.
(17) N. L. Allinger, *Adv. Phys. Org. Chem.*, **13**, 1 (1976).
(18) (a) G. A. Berchtold and G. F. Uhlig, *J. Org. Chem.*, **28**, 1459 (1963); (b) K. C. Brannock, R. D. Burpitt, V. W. Goodlett, and J. G. Thweatt, *ibid.*, **28**, 1464 (1963).
(19) E. g., L. F. Fieser and M. Fieser, "Reagents for Organic Synthesis", Vol. I–V, Wiley, New York, N.Y., 1967.
(20) (a) C. F. Huebner, L. Dorfman, M. M. Robison, E. Donoghue, W. G. Pierson, and P. Strachan, *J. Org. Chem.*, **28**, 3134 (1963); (b) K. C. Brannock, R. D. Burpitt, V. W. Goodlett, and J. G. Thweatt, *ibid.*, **29**, 818 (1964); (c) W.

E. Parham and R. J. Sperley, *ibid.*, **32**, 926 (1967).
(21) R. N. Warrener and E. N. Cain, *Aust. J. Chem.*, **24**, 785 (1971).
(22) E. J. Stamhuis in "Enamines", A. G. Cook, Ed., Marcel Dekker, New York, N.Y., 1969, p 112.
(23) F. Farina, M. V. Martin, M. R. Martin, and F. Sanchez, *Synthesis*, 642 (1977).
(24) S. Kagawa, S. Matsumoto, S. Nishida, S. Yu, J. Morita, A. Ichihara, H. Shirahama, and T. Matsumoto, *Tetrahedron Lett.*, 3913 (1969).
(25) M. Regitz, *Justus Liebigs Ann. Chem.*, **676**, 101 (1964).

(26) I. K. Korobitsyna and V. A. Nikolaev, *J. Org. Chem. USSR (Engl. Transl.)*, **7**, 411 (1974).
(27) A. Blomquist and T. H. Shelley, Jr., *J. Am. Chem. Soc.*, **70**, 147 (1948).
(28) L. Brandsma, "Preparative Acetylenic Chemistry", American Elsevier, New York, N.Y., 1971, p 118.
(29) Compounds **6A, B, 16A, B, 7, 17, 8, 9**, and **13** are named as hydroazulene derivatives in the Experimental Section. This differs from the numbering used in the text and in the schemes where the compounds are treated as vellerane derivatives following the suggestion of Professor Sukh Dev.

Stereochemistry of Macrolides. 3. X-ray Crystal Structure Analysis of 11,4″-Bis[O-(p-bromobenzoyl)]oleandomycin

Haruo Ogura,*[1a] Kimio Furuhata,[1a] Yoshinori Harada,[1b] and Yoichi Iitaka[1b]

Contribution from the School of Pharmaceutical Sciences, Kitasato University, Shirokane, Minato-ku, Tokyo 108, and Faculty of Pharmaceutical Sciences, University of Tokyo, Hongo, Bunkyo-ku, Tokyo 113, Japan. Received January 16, 1978

Abstract: Molecular structures of oleandomycin and its derivatives both in solid and solution have been determined by means of X-ray analysis of 11,4″-bis[O-(p-bromobenzoyl)]oleandomycin and its CD and NMR spectra. It was shown that the compound, $C_{49}H_{67}O_{14}NBr_2 \cdot H_2O \cdot CH_3OH \cdot C_2H_5OH$, is orthorhombic with space group $P2_12_12_1$ and unit cell dimensions of $a = 14.283$ (8), $b = 32.057$ (12), and $c = 12.767$ (7) Å, $V = 5843$ Å^3, and $Z = 4$. The crystal structure was solved by the heavy-atom method and refined by the block-diagonal least-squares method including anisotropic thermal parameters to a final R value of 0.112. The absolute configuration of the macrolactone moiety of oleandomycin was determined to be $2R,3S,4R,5S,6S,10R,11S,12R,13R$, based on the already determined configuration of the D-desosamine moiety. A new "diamond lattice" conformation model F for 11,4″-bis[O-(p-bromobenzoyl)]oleandomycin was proposed based on the results of X-ray analysis. Conformations of oleandomycin and its derivatives in solution were discussed on the basis of CD and NMR spectra.

Oleandomycin is a 14-membered macrolide antibiotic isolated by Celmer et al.[2] and its structure and configuration were reported by the same author.[3] In this paper, we wish to report the conformation of oleandomycin by means of an X-ray analysis of 11,4″-bis[O-(p-bromobenzoyl)]oleandomycin and propose a new "diamond lattice" conformation model F (diamond model F). In relation to the conformation in solid state, CD curves and NMR spectra of oleandomycin (1), 11,4″-bis[O-(p-bromobenzoyl)]oleandomycin (2), triacetyloleandomycin (3), and the diacetylanhydroaglycon (4) of oleandomycin elucidated the stereochemistry of the 14-membered macrolactone of oleandomycin derivatives in solution (see Figure 1).

Bis[O-(p-bromobenzoyl)]oleandomycins were obtained by treating a benzene solution of oleandomycin (1) with p-bromobenzoyl chloride and pyridine, and bis-benzoate 2 was isolated from the above reaction mixture by silica-gel column chromatography.

Crystals of the compound suitable to X-ray work were obtained from an ethanol–methanol (1:1) solution in the form of colorless prisms elongated along the c axis. The density was measured by the floatation method in an aqueous solution of potassium iodide. Preliminary cell dimensions were obtained from Weissenberg photographs.

Experimental Section

11,4″-Bis[O-(p-bromobenzoyl)]oleandomycin[4] (2). To a solution of oleandomycin (1 g) in benzene (100 mL), a mixture of p-bromobenzoyl chloride (0.5 g) in benzene (20 mL) and pyridine (0.5 mL) was added dropwise under cooling. The reaction mixture was kept at room temperature for 20 h and treated with methanol to decompose the excess reagent. After removal of the solvent, the residue was dissolved in minimum amount of water, neutralized with 2% $NaHCO_3$, and then extracted with chloroform. The chloroform extract was washed, dried, and evaporated to leave a white powder. The bis-benzoate mixture thus obtained was purified by silica-gel chromatography. Elution with benzene–acetone (9:1) and evaporation of the eluate left white powder, and a second recrystallization from ethanol–methanol gave 150 mg of **2** as colorless prisms: mp 109–110 °C; IR ν_{max}^{KBr} 3420 (OH), 1745 (ester), 1720 (ketone), 1610 and 1590 cm^{-1} (phenyl); UV λ_{max}^{MeOH} 243.0 nm (log ε 4.27). Anal. Calcd for $C_{49}H_{67}O_{14}NBr_2 \cdot H_2O \cdot CH_3OH \cdot C_2H_5OH$: C, 54.22; H, 7.09; N, 1.22. Found: C, 54.15; H, 7.11; N, 1.23.

Diacetylanhydroaglycon (4) of Oleandomycin. The anhydroaglycon of **1** was prepared from **1** (0.85 g) according to modified procedures of Hochstein et al.[5] The yield was 0.072 g, mp 219–220 °C (lit.[5] mp 228–230 °C). The diacetate **4** was prepared from the anhydroaglycon (0.05 g) by treating with acetic anhydride (1 mL) and pyridine (1 mL) at 60 °C. Recrystallization of the product from ether yielded 0.02 g (32%) of **4** as colorless needles: mp 196–197 °C; IR ν_{max}^{KBr} 1725 (ester), 1692 and 1640 cm^{-1} (α,β-unsaturated ketone); UV λ_{max}^{MeOH} 234.0 nm (log ε 4.12); CD (MeOH) $[\theta]_{333}^{20}$ −1194, $[\theta]_{294}$ 1683, $[\theta]_{233}$ 27 151; NMR[6] (CDCl$_3$, 100 MHz) δ 0.88 (3 H, d, $J_{4\text{-Me},4}$ = 7.0 Hz, 4-Me), 0.93 (3 H, d, $J_{6\text{-Me},6}$ = 7.0 Hz, 6-Me), 1.05 (3 H, d, $J_{12\text{-Me},12}$ = 7.0 Hz, 12-Me), 1.13 (3 H, d, $J_{2\text{-Me},2}$ = 7.0 Hz, 2-Me), 1.37 (3 H, d, $J_{13\text{-Me},13}$ = 6.5 Hz, 13-Me), 1.90 (1 H, m, 6-H), 1.92 (1 H, ddq, $J_{4,3}$ = 2.0 Hz, $J_{4,5}$ = 10.5 Hz, $J_{4,4\text{-Me}}$ = 7.0 Hz, 4-H), 1.98 (3 H, d, $J_{10\text{-Me},11}$ = 1.5 Hz, 10-Me), 2.69 (1 H, dd, $J_{2,3}$ = 8.5 Hz, $J_{2,2\text{-Me}}$ = 7.0 Hz, 2-H), 2.84 (1 H, d, $J_{14ax,14eq}$ = 5.5 Hz, 14-H$_{ax}$), 2.90 (1 H, d, $J_{14eq,14ax}$ = 5.5 Hz, 14-H$_{eq}$), 3.18 (1 H, ddq, $J_{12,11}$ = 10.7 Hz, $J_{12,13}$ = 4.5 Hz, $J_{12,12\text{-Me}}$ = 7.0 Hz, 12-H), 4.65 (1 H, dd, $J_{5,4}$ = 10.5 Hz, $J_{5,6}$ = 1.5 Hz, 5-H), 4.96 (1 H, dd, $J_{13,12}$ = 4.5 Hz, $J_{13,13\text{-Me}}$ = 6.5 Hz, 13-H), 5.09 (1 H, dd, $J_{3,2}$ = 8.5 Hz, $J_{3,4}$ = 2.0 Hz, 3-H), 6.54 (1 H, dq, $J_{11,12}$ = 10.7 Hz, $J_{11,10\text{-Me}}$ = 1.5 Hz, 11-H). Anal. Calcd for $C_{24}H_{36}O_8$: C, 63.56; H, 8.18. Found: C, 63.70; H, 8.02.

Crystal data for 11,4″-bis[O-(p-bromobenzoyl)]oleandomycin: mp 109–111 °C; $C_{49}H_{67}O_{14}NBr_2 \cdot H_2O \cdot CH_3OH \cdot C_2H_5OH$, MW 1150.1; orthorhombic; $a = 14.283$ (8), $b = 32.057$ (12), $c = 12.767$ (7) Å; $V = 5843$ Å^3; $d_{obsd} = 1.295$ g/cm^3, $d_{calcd} = 1.310$ g/cm^3; $Z = 4$; $F(000) = 2416$; space group $P2_12_12_1$ (absent reflections; $h00$ when h is odd, $0k0$ when k is odd, and $00l$ when l is odd).

The diffraction intensities of a 0.2 × 0.3 × 0.4 mm^3 parallelpiped crystal were measured with a Rigaku four-circle diffractometer using graphite monochromated Cu Kα radiation. The accurate cell dimensions were determined by the least-squares method using the