2, J = 6.0, 7.5 Hz, α-H); IR (CHCl$_3$) 1040, 1080, 1110, 1310, 1560, 1635 cm^{-1}.

Anal. (C$_9$H$_{15}$N$_2$O$_3$P) C, H, N, P.

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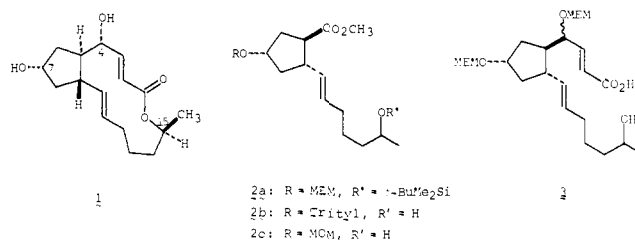
Total Synthesis of Brefeldin A

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Abstract: A total synthesis of (±)-brefeldin A has been achieved in 15 steps from *trans*-4-oxo-1,2-cyclopentanedicarboxylic acid via the key intermediate **2c**. Two distinct syntheses of **2** have been developed, leading to the trityl ether **2b** and the methoxymethyl ether **2c**. The former involves stereoselective conjugate addition of vinylalane **16** to hydroxy enone **4** and a surprisingly regioselective trityl ether isomerization. The latter synthesis employs the bicyclic lactone **21** to establish the stereochemistry around the five-membered ring and incorporates a new carboxyl to acetylene conversion. The ester **2c** is further elaborated via a β-keto sulfoxide alkylation–elimination sequence to the γ-ketoacrylic acid **34**, which is lactonized using Mukaiyama's procedure. Selective reduction and deprotection provide racemic brefeldin A.
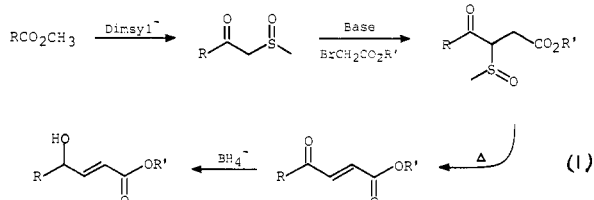
Brefeldin A (**1**) possesses a diverse spectrum of antifungal,[2] antiviral,[3] antimitotic,[4] and antitumor[2a] activity in addition to the synthetic attractions of its macrocyclic framework. This fungal metabolite has been isolated from a variety of organisms,[2a,5] and was known variously as ascotoxin,[5f] cyanein,[5b] and decumbin,[5a] before the identity of these materials with brefeldin A was established.[5f,6] The complete structure was revealed by X-ray diffraction,[7] after chemical and spectroscopic studies had elucidated all features except the configuration at C-4.[5f,8] Biosynthetic studies[5j,9] have established that the molecule is derived entirely from acetate and have ruled

1

2a: R = MEM, R' = t-BuMe$_2$Si
2b: R = Trityl, R' = H
2c: R = MOM, R' = H

3

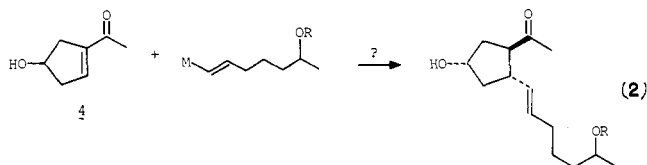out a number of more detailed postulates. In 1976, Corey and Wollenberg reported the first total synthesis of racemic bre-

feldin A, via the intermediates **2a** and **3**.[10b] Further refinements have been reported recently,[10c] as has an alternative synthesis of intermediate **3** by Crabbé and co-workers.[11]

We have completed a synthesis of (±)-brefeldin A in 15 steps from *trans*-4-oxo-1,2-cyclopentanedicarboxylic acid. Our synthetic planning reflected two desires: to use the β-keto sulfoxide alkylation–elimination sequence of eq 1[12] to intro-

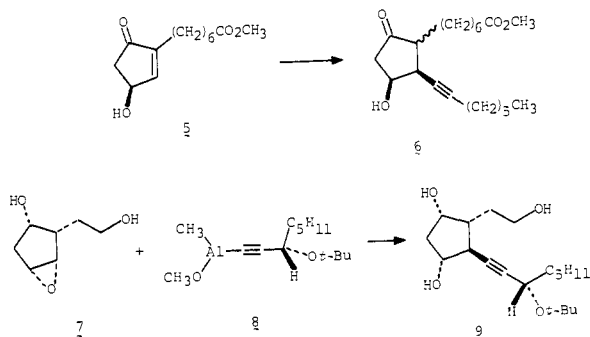duce the γ-oxygenated α,β-unsaturated ester moiety and to streamline the synthesis by limiting the number of steps concerned solely with functional-group protection/deprotection. In the latter regard, protecting groups were avoided or removed in conjunction with another transformation in the synthetic sequence, as far as possible. The former intention led to our choice of ester **2** as the key intermediate, and we developed two distinct syntheses of this structure, protected as the trityl ether **2b** and as the methoxymethyl ether **2c**. During the course of this work, we have also uncovered additional examples of stereoselective transformations on the bicyclic framework.[10a]

Synthesis of Trityl Ether 2b Via Conjugate Addition. Our first synthesis of a derivative of **2** sought to establish the substitution pattern and stereochemistry of the five-membered ring by a hydroxyl-directed conjugate addition reaction (eq 2). Precedent from the prostaglandin field for this proposed
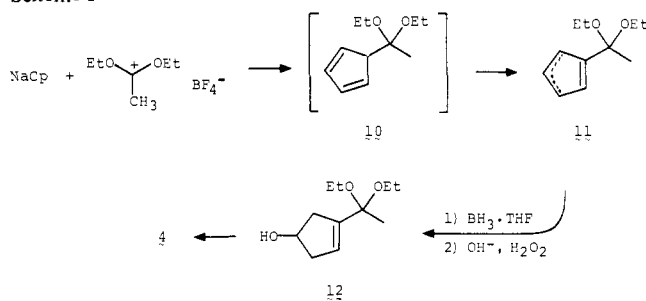

step was found in the work of Pappo and Collins[13] and Fried and Sih.[14] Trioctynylalane delivers an alkynyl group to the hydroxy enone **5** specifically from the side cis to the hydroxyl,[13] and alane **8** reacts with epoxydiol **7** to give triol **9** exclusively.[14] In each case, protection of the directing hydroxyl group reverses or destroys the stereo- or regioselectivity.
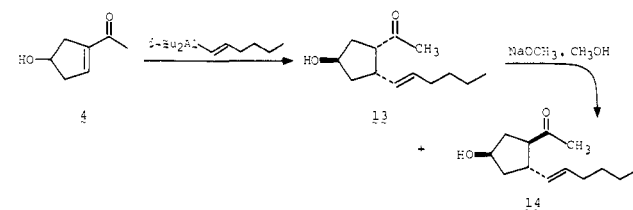

The synthesis of hydroxycyclopentenyl ketone **4** is outlined in Scheme I. Alkylation of sodium cyclopentadienide with diethoxymethylcarbenium tetrafluoroborate[15] provides, initially, the 5-substituted compound **10**, which rearranges on standing at room temperature to a mixture of the 1- and 2-substituted cyclopentadienes (**11**) (78% yield). Hydroboration of this mixture proceeds with attack at the least hindered end of the conjugated system of both isomers, to give a single hydroxy ketal **12** after oxidation. This acid-labile material is simultaneously purified and hydrolyzed by chromatography through silica gel, affording the hydroxy enone **4** in 74% overall yield from the mixture of dienes (**11**).

Scheme I

Two stereoisomeric conjugate addition products, **13** and **14** (5:1 ratio), are isolated from the reaction of (*E*)-1-hexenyl-diisobutylalane with hydroxy enone **4**. Conversion of the major product **13** to its epimer **14** with sodium methoxide in methanol

established that these materials are isomeric at the acetyl position and that the major product **13** possesses the cis relationship between the acetyl and hexenyl substituents. Kinetically controlled protonation of the enolate intermediate to give predominantly the cis-1,2 relationship is well precedented in analogous cyclohexyl systems.[16]
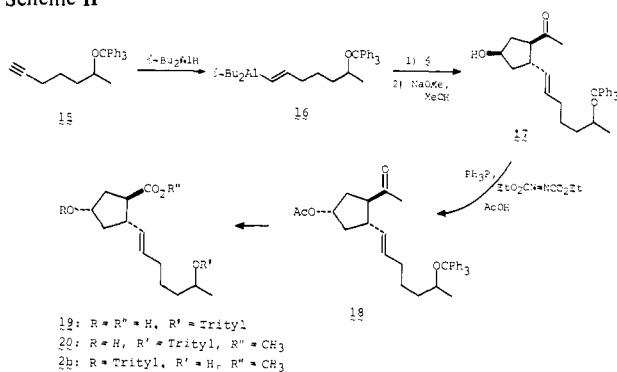
Addition of a europium shift reagent (Eu[fod]$_3$) to the isomeric mixture causes a much greater downfield shift in the ^{1}H NMR spectrum for the acetyl resonance of **14** than it does for its epimer **13**, suggesting that the acetyl and hydroxyl groups are cis in isomer **14**. This assignment was corroborated by IR spectroscopy, using samples of **13** and **14** which had been purified by VPC. Isomer **13** shows only a single sharp O–H band at 3630 cm^{-1} in dilute solution (0.005 M in CCl$_4$); under similar conditions, epimer **14** shows in addition a broad band at 3460 cm^{-1}, indicative of an intramolecular hydrogen bond.

These results indicate that conjugate attack is directed to the face of the enone system opposite to the hydroxyl. The alane must bind to the hydroxyl of **4** in a nonproductive manner (either with preferential loss of the vinyl group[17] or so that intramolecular addition is prevented) and subsequently serve merely as a bulky substituent, deflecting intermolecular reaction by another molecule to the opposite face for steric reasons. Consistent with this interpretation is the requirement of 2 equiv of alane for optimization of the yield in this reaction.

A number of modifications in this initial approach failed to reverse the undesired stereochemical result. The transoid enone 2-cyclohexenone fails to react with alane reagents[18] but does undergo conjugate addition with the "ate" complex formed by combination of the alane with lithium isopropoxide.[19] Heterocuprates are also effective in 1,4 additions.[20] Moreover, the lithium salt of **4** can be generated without decomposition with lithium diisopropylamide (LDA) in tetrahydrofuran (THF) at 0 °C. However, treatment of this lithium alkoxide with (*E*)-1-hexenyldiisobutylalane or with (*E*)-1-hexenyl-copper did not lead to intramolecular addition via the "ate" complexes. Even the alkynylalane reagents (tri-1-hexynylalane and diethyl-1-hexynylalane), which are so well directed in the examples above,[13,14] failed to give any of the conjugate addition product.

In spite of the undesired sense of the addition, it proceeds with high stereoselectivity: less than 5% of the product arises
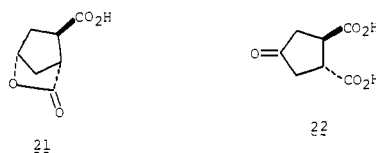
Scheme II

19: R = R" = H, R' = Trityl
20: R = H, R' = Trityl, R" = CH₃
2b: R = Trityl, R' = H, R" = CH₃

from addition cis to the hydroxyl group. The desired stereo-isomer may then be obtained cleanly by inversion of the configuration at the hydroxyl position (Scheme II). Accordingly, the functionalized alkenylalane 16 was prepared (by hydroalumination[21] of the trityl-protected alkyne 15) and allowed to react with 0.5 equiv of hydroxy enone 4, to give the adduct 17 in 40% yield after base-catalyzed epimerization and chromatographic purification. The configuration of the hydroxyl group was inverted by the procedure of Mitsunobu and Bose[22] (Ph₃P, EtO₂CN=NCO₂Et, AcOH; 80% yield of 18 after chromatography), and the methyl ketone was oxidized to the carboxylic acid 19 using alkaline hypobromite[23] (91% yield). The trityl ether is a necessary protecting group for the oxygen function during the hydroalumination reaction, because less sterically congested derivatives, such as ethylenedioxy- or 2-tetrahydropyranyl are strongly coordinated by diisobutyl-alane.[21] This coordination retards the rate of hydroalumination so severely that condensation of these derivatives with only 1 equiv of diisobutylalane requires an impractically long reaction period.

Our plans for further elaboration of 19 to brefeldin A called for conversion of the ester 20 to the β-keto sulfoxide (as in eq 1) and then, at appropriate points, acetylation of the cyclopentyl hydroxyl and selective cleavage of the trityl protecting group. Esterification with dimethyl sulfate and potassium carbonate in refluxing acetone[24] proceeded smoothly to give the methyl ester 20. However, traces of acid, arising from dimethyl sulfate contamination of the crude product, catalyzed its transformation to a new substance on standing at room temperature. This material was the major component (>90%) of the mixture and was isolated in 58% yield from the carboxylic acid 19 after chromatographic purification. As evidence for the assigned structure, the doublet resonance for the terminal methyl group in 2b is found at 1.17 ppm in the ¹H NMR spectrum, similar to its position at 1.20 ppm in the spectrum of 6-heptyn-2-ol and in contrast to its position between 0.83 and 0.92 ppm in the spectra of compounds 15–18.
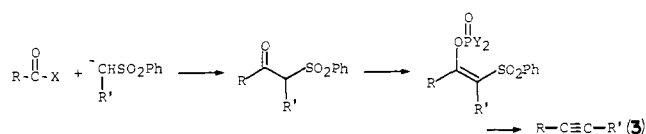
This unanticipated selectivity of a trityl group for a cyclopentyl vs. a secondary-alkyl ether position could be demonstrated with cyclopentanol and 6-heptyn-2-ol as well. Treatment of an equimolar solution of cyclopentanol and trityl ether 15 in chloroform with a trace of sulfuric acid resulted in rapid equilibration to a mixture of 95% cyclopentyl trityl ether and 5% 15. The isomerization 20 to 2b would be valuable in the synthesis of brefeldin A, because by simultaneously releasing the side-chain hydroxyl and blocking the cyclopentyl hydroxyl groups it obviates two subsequent protection/deprotection steps. The generality of this selective protection procedure remains to be established.

Synthesis of Methoxymethyl Ether 2c via Bicyclic Lactone 21. Although the key intermediate was accessible by the route outlined above, the disappointing yield and stereochemical

outcome of the conjugate addition reaction and the need for several chromatographic purification steps made an alternative synthesis of 2 desirable. In this connection, the bicyclic lactone 21 intrigued us: although no stereoselective transformations would be required for its synthesis from the known diacid 22,[25]
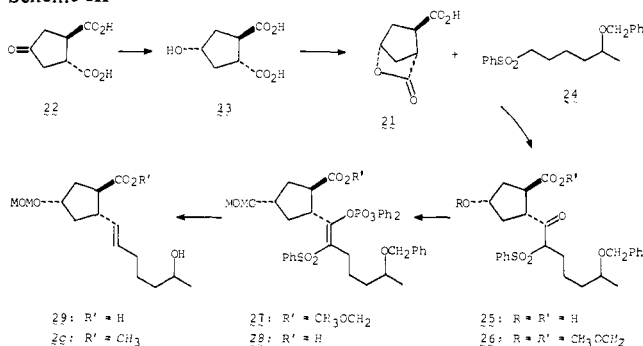

it would be ideally suited for the stereospecific introduction of substituents around the five-membered ring of brefeldin A. For instance, elaboration of the lactone carboxyl group to an acetylene followed by reduction to the trans alkene would complete a direct and stereospecific synthesis of a derivative of 2. However, no general method for the conversion of a carboxyl group to an acetylene was available. This impetus led to our development of the sequence illustrated by eq 3 as a general

alkyne synthesis which could be applied specifically to the problem at hand. Condensation of a carboxylic acid derivative with a sulfonyl-stabilized carbanion and phosphorylation of the enol of the resulting β-keto sulfone provide an intermediate which undergoes reductive elimination to an alkyne with sodium in ammonia or sodium amalgam in THF. This sequence has been applied to the synthesis of a variety of alkyl- and aryl-, mono- and disubstituted acetylenes.[26]
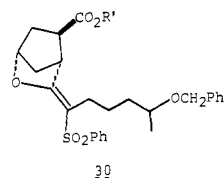
The synthesis of 2c via the bicyclic lactone 21, incorporating this alkyne synthesis, is outlined in Scheme III. Reduction of keto diacid 22[25] with sodium borohydride in methanol affords the hydroxy diacid 23 in 95% yield. Lactonization of this material is complicated by the fact that polymerization (via the trans carboxyl group) occurs in preference to lactonization under conditions which allow equilibration; i.e., refluxing dioxane or benzene with acid catalysis or high-vacuum distillation from naphthalenesulfonic acid. However, lactonization proceeds smoothly with tosyl chloride in pyridine or, more conveniently, with ethyl chloroformate and 1 equiv of triethylamine in THF/acetone (0 °C for 30 min), to give the bicyclic compound 21 in 76% (isolated) yield. Because equilibration of the mixed anhydride intermediates[27] is not likely to occur in this kinetically controlled process, formation of lactone in preference to polymer indicates that the cis carboxyl group is activated more readily than the trans. The acidifying influence of the hydroxyl group via hydrogen bonding is probably responsible for this increased reactivity under the mildly basic reaction conditions.

Scheme III

29: R' = H
2c: R' = CH₃

27: R' = CH₃OCH₂
28: R' = H

25: R = R' = H
26: R = R' = CH₃OCH₂

The synthesis of the requisite sulfone **24** from 6-bromo-2-hexanone[28] is unexceptional. Formation of the α-lithio derivative with *n*-butyllithium and subsequent condensation with the bicyclic lactone **21** in THF at -78 °C affords the β-keto sulfone **25** in over 90% yield. This condensation is carried out in the presence of 2 equiv of lithium diisopropylamide, to deprotonate the carboxylic acid and the initially formed β-keto sulfone.[26]

The hydroxyl and carboxyl groups of the adduct **25** must be protected before generation of the enol phosphate moiety; however, derivatization under alkaline conditions (acetic anhydride/pyridine or MEM-Cl/ethyldiisopropylamine) leads to significant reaction via the sulfonyl enol tautomer, as well as formation of the bicyclic enol ether **30**. Esterification with

30

diazomethane and hydroxyl protection under acidic conditions (dimethoxymethane/P_4O_{10}[29]) avoid this difficulty, and enol phosphorylation (diphenyl phosphorochloridate/4-dimethylaminopyridine/acetonitrile) proceeds smoothly to give **27** (R$'$ = CH$_3$ instead of CH$_3$OCH$_2$). Reduction of this material with sodium amalgam in THF,[26] protection of the carboxyl group by hydrolysis, and simultaneous cleavage of the benzyl ether and reduction to the trans alkene with sodium in ammonia lead straightforwardly to the hydroxy acid **29**.
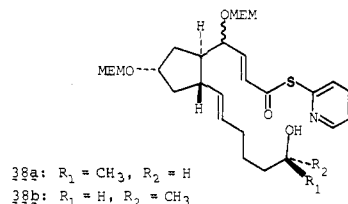
This overall transformation is considerably streamlined, however, by protecting both the hydroxyl and carboxyl groups as their methoxymethyl derivatives in one step, because after formation of the enol phosphate **27** the carboxylic ester can be selectively hydrolyzed with trifluoroacetic acid in acetic acid. Reductive elimination, alkyne reduction, and benzyl ether cleavage are all performed at once on the carboxylic acid **28** using sodium in liquid ammonia, to afford the hydroxy acid **29** in 55% overall yield for the four steps from β-keto sulfone **25**.

Esterification of **29** completes the alternative synthesis of the key intermediate. Although the syntheses of both **2b** and **2c** require a similar number of steps, the overall yield via lactone **21** is more than twice as high as that by the conjugate addition sequence, and no chromatographic purification is required.

Conversion of 2c to Brefeldin A. The further elaboration of hydroxy ester **2c** to brefeldin A is outlined in Scheme IV. Reaction of **2c** with 4 equiv of dimsylsodium gives the β-keto sulfoxide **31** in 87% yield. Because the β-ketosulfoxide moiety is more acidic than a secondary alcohol,[30] alkylation of **31** with potassium *tert*-butoxide and methyl bromoacetate can be

performed in good yield without protecting the side-chain hydroxyl group. The alkylated intermediate **32** is not purified but is pyrolyzed directly to provide the keto acrylate **33** in 70% yield from **31**.[12]

Alkaline hydrolysis of **33** and lactonization of the hydroxy acid **34** using Mukaiyama's procedure[31] give a 37% yield of macrocyclic material as a 1:1 mixture of the C-15 epimers **35** and **36**. This result contrasts with that of Corey and Wollenberg, who found that cyclization of the 2-pyridylthiol esters of the γ-*alkoxy* derivatives **38a**, with the natural configuration at C-15, occurred much more rapidly than cyclization of the C-15 epimers **38b**.[10a] The diastereomeric ketones **35** and **36**
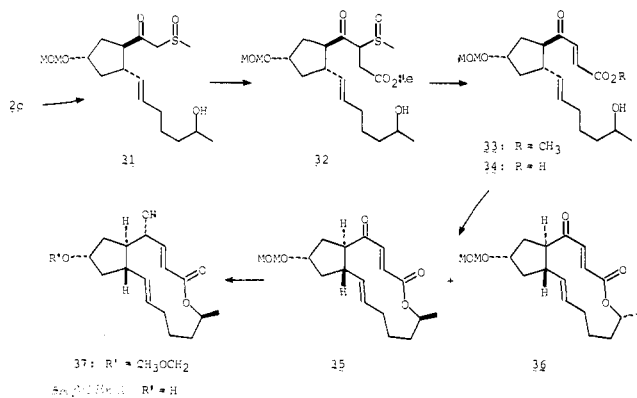

38a: R$_1$ = CH$_3$, R$_2$ = H
38b: R$_1$ = H, R$_2$ = CH$_3$

may be separated by preparative thin-layer chromatography or by high-pressure liquid chromatography and are readily distinguished by their ^{1}H NMR spectra. The more polar isomer (**35**) has the natural relative configuration at C-15, as shown by its conversion to racemic brefeldin A.

Alternative lactonization procedures involving carboxyl activation via the *tert*-butylthiol[33] and 2-pyridylthiol[34] esters could not be applied to the ketoacrylic acid **34**, because of competing Michael addition of the thiols to the activated double bond.[35] Another approach to the *tert*-butylthiol ester by alkylation of the β-keto sulfoxide **31** with *tert*-butyl bromoacetothiolate only led to destruction of the thiol ester and recovery of **31**. A more circuitous route via an intermediate such as **41** (R^1, R^2 = OR, H) would allow intersection with the previously published syntheses of brefeldin A[10,11] but would require a minimum of nine steps for the conversion of **33** to **37**, instead of the three steps illustrated in Scheme IV.

Reduction of **35** with borohydride in methanol at -75 °C[10a] affords a greater than 10:1 mixture of the methoxymethyl ether of brefeldin A and its C-4 epimer in quantitative yield. Finally, deprotection of the C-7 hydroxyl with trifluoroacetic acid in aqueous ethanol and recrystallization from ethyl acetate provide racemic brefeldin A, mp 175–175.5 °C, which is identical with a sample of the natural material by TLC, ^{1}H NMR, and infrared spectroscopy.[32]

The relative configuration at C-15 has a pronounced effect on the rate of lactonization of the γ-alkoxyacrylic acid derivatives **38**[10a] but not on the lactonization of the γ-ketoacrylic acids **34**, as noted above. In addition, this chiral center affects the stereochemistry of borohydride reduction at C-4: the 15-*epi*-7-methoxymethyl ether **36** gives the epimeric alcohols **39** and **40** in a ratio of more than 10:1 with sodium borohydride

Scheme IV

31

32

33: R = CH$_3$
34: R = H

37: R$'$ = CH$_3$OCH$_2$
          R$'$ = H

35

36

39: R^1 = OH, R^2 = H
40: R^1 = H, R^2 = OH

41: R^1 = OH, R^2 = H
42: R^1 = H, R^2 = OH

in methanol at -75 °C, a stereoselectivity opposite to that observed with isomer **35**, which has the natural relative configuration at C-15. Although the origin of this remarkable selectivity is not obvious, it indicates that substituents in a macrocyclic structure can have an important effect on the conformation and reactivity of a seemingly remote center.[36]

The configurations at C-4 were assigned by cleaving the

macrocycles with sodium carbonate in methanol and comparing the resulting mixture of esters **41** and **42** with that derived from the C-15 epimer **37**. In this way, the two series can be related, because the monocyclic compounds which are stereoisomeric only at C-15 are indistinguishable by TLC or 180-MHz ^{1}H NMR. This comparison allowed us to show that reduction of the monocyclic keto acrylate **33** also proceeds stereoselectivity, favoring the unnatural C-4 epimer **41** over **42** by a ratio of 5:1.

Experimental Section

Melting points were determined with a Büchi melting-point apparatus and are uncorrected. Infrared spectra were recorded on a Perkin-Elmer Model 710 spectrophotometer. Routine (60-MHz) ^{1}H NMR spectra were recorded on a Varian Associates Model T-60 or a Hitachi-Perkin-Elmer Model R-24B spectrometer; high-field (180-MHz) FT ^{1}H NMR spectra were acquired on a system equipped with a Bruker magnet and Nicolet computer. Unless otherwise noted, the NMR solvent was CDCl$_3$. Chemical shifts are reported in parts per million on the δ scale, relative to tetramethylsilane as internal standard. Data are presented as follows: chemical shift (multiplicity, integrated intensity, coupling constants, assignment). For the macrocyclic lactones, NMR assignments employ the numbering scheme of brefeldin A (see **1**).

Tetrahydrofuran (THF) was dried by distillation from sodium benzophenone ketyl, and ammonia by distillation from sodium. Unless otherwise specified: reaction workups culminated in drying the solvent over anhydrous MgSO$_4$ and removing the solvent by evaporation at reduced pressure; distillations involved bulb to bulb distillation using a Kugelrohr oven, at the oven temperature and pressure indicated; and the chromatographic adsorbant was Davison Grade 923 silica gel, 100–200 mesh, eluted with the indicated solvent.

5-(1,1-Diethoxyethyl)cyclopentadiene (10) and the 1- and 2-Substituted Isomers (11). To a stirred suspension of 0.25 mol of diethoxymethylcarbenium tetrafluoroborate[15] in 150 mL of THF at −78 °C was added 100 mL of a 2.1 M solution of sodium cyclopentadienide in THF over a 30-min period. After warming to −10 °C, the mixture was partitioned between ether and cold saturated NaHCO$_3$, and the organic layer was washed with ice water and brine, dried (K$_2$CO$_3$), and concentrated at 0 °C to give 30 g (78%) of the 5-substituted isomer as a dark oil: ^{1}H NMR (CCl$_4$) δ 0.87 (s, 3), 1.18 (t, 6), 3.57 (m, 5, OCH$_2$ and >CH–), 6.36 (br s, 4).

On standing at room temperature in either solution (10 mL/g) for 24 h, this material rearranged to a mixture of the 1- and 2-substituted isomers **11**: ^{1}H NMR δ 0.93 (t, 6), 1.22 and 1.25 (s, 3), 2.70 (m, 2), 3.13 (m, 4), 6.12 (m, 3).

1-(4-Hydroxy-1-cyclopentenyl)ethanone (4). To the mixture of rearranged cyclopentadienes **11** (30 g, 165 mmol) in 150 mL of THF at 0 °C was added 83 mL of a 1.0 M solution of BH$_3$·THF over a 3.5-h period. After an additional 2 h at room temperature, 18.5 mL of 3 N NaOH and 18.5 mL of 30% aqueous hydrogen peroxide (0.18 mmol) were added cautiously, at such a rate that the solvent was kept at gentle reflux. The two-phase mixture was stirred at room temperature overnight and diluted with ether, and the ether layer was washed with water and brine, dried (K$_2$CO$_3$), and evaporated to give 27.0 g (80%) of the allylic ketal **12**: ^{1}H NMR (CCl$_4$) δ 1.18 (t, 6), 1.35 (s, 3), 2.32 (m, 4), 3.37 (q, 4), 4.37 (m, 1), 5.63 (br t, 1, J = 2 Hz).

An 8.5-g sample of this material was chromatographed on 260 g of silica gel, eluting with chloroform/methanol (9:1), to give 4.9 g (92%) of hydroxy enone **4** as an oil: bp 58–62 °C (0.04 Torr); ^{1}H NMR δ 2.32 (s, 3), 2.65 (m, 4), 3.38 (br s, 1, OH), 4.52 (m, 1), 6.65 (m, 1); IR (film) 1280 (CO), 1620 (w, C=C), 1660 (C=O), 3400 (OH) cm^{-1}.

Anal. (C$_7$H$_{10}$O$_2$) C, H.

6-Triphenylmethoxy-1-heptyne (15). A solution of 12.9 g (46.5 mmol) of trityl chloride and 5.2 g (46.5 mmol) of 6-heptyn-2-ol in 50 mL of dry pyridine was heated at 75 °C for 2 days. After cooling and removing the pyridinum hydrochloride by filtration, the mixture was diluted with benzene; washed with aqueous NaHCO$_3$, with saturated CuSO$_4$ until the pyridine had been removed, and with water and brine; dried; and concentrated to give a yellow oil which solidified on trituration with hexane. Further purification by removal of CCl$_4$-insoluble material and trituration with hexane afforded 11.8 g (72%) of the trityl ether: mp 80–84 °C; ^{1}H NMR δ 0.92 (d, 3), 1.90 (t, 1, J = 1 Hz, ≡CH), 1.97 (m, 2, –CH$_2$C≡), 3.27 (m, 1, >CHO), 7.2–7.7 (m, 15);

IR (nujol) 2100 (w, C≡C), 3240 (w, ≡CH) cm^{-1}. A sample was further purified for analysis by recrystallization from methanol: mp 83.5–85 °C.

Anal. (C$_{26}$H$_{26}$O) C, H.

1-([1α,2β(E),4α]-4-Hydroxy-2-[6-triphenylmethoxy-1-hepten-yl]cyclopentyl)ethanone (17). A mixture of 3.89 g (11.0 mmol) of alkyne **15** and 11.0 mmol of diisobutylalane in 12 mL of hexane was kept at 50 °C for 5 h, diluted with 24 mL of ether, and cooled to −10 °C. To this solution of the alkenylalane **16** was added 0.63 g (5.0 mmol) of hydroxy enone **4** in 3 mL of THF over 10 min. After 45 min at 0 °C, the mixture was treated sequentially with 0.44 mL of water, 0.44 mL of 15% aqueous NaOH, and 1.32 mL of water, and the resulting slurry was stirred for 1 h before filtering through MgSO$_4$ and concentrating at reduced pressure. The residue was dissolved in 75 mL of methanol with 0.2 mmol of NaOH and kept overnight to epimerize the acetyl group. The solution was neutralized with 0.2 mmol of acetic acid and concentrated, and the residue was chromatographed on 200 g of Florisil, eluting with ether/hexane (3:2), to give 0.94 g (40%) of hydroxy ketone **17** as an oil: bp 140 °C (0.03 Torr); ^{1}H NMR δ 0.87 (d, 3), 2.17 (s, 3), 3.53 [m, 1, >CHOC(C$_6$H$_5$)$_3$], 4.27 (m, 1, >CHOH), 5.30 (m, 2); IR (film) 970 (trans CH=CH), 1704 (C=O), 3460 (OH) cm^{-1}.

Anal. (C$_{33}$H$_{38}$O$_3$) C, H.

1-([1α,2β(E),4β]-4-Acetoxy-2-[6-triphenylmethoxy-1-heptenyl]-cyclopentyl)ethanone (18). Diethyl azodicarboxylate (509 mg, 2.92 mmol) in 3 mL of dry THF was added to a solution of 939 mg (1.95 mmol) of hydroxy ketone **17**, 509 mg (2.92 mmol) of triphenylphosphine, and 145 μL (2.54 mmol) of acetic acid in 7 mL of THF, and the mixture was stirred at room temperature overnight. After concentration at reduced pressure, the residue was purified by chromatography on Florisil, eluting with 20–30% ether/hexane, to give 817 mg (80%) of the inverted acetate **18** as an oil: ^{1}H NMR δ 0.90 (d, 3), 2.03 and 2.13 (s, 3), 3.48 [m, 1, >CHOC(C$_6$H$_5$)$_3$], 5.12 (m, 1, >CHOAc), 5.37 (m, 2); IR (film) 975 (trans CH=CH), 1715, 1738 (C=O) cm^{-1}.

Anal. (C$_{35}$H$_{40}$O$_4$) C, H.

Methyl [1α,2β(E),4β]-2-(6-Hydroxy-1-heptenyl)-4-triphenyl-methoxycyclopentanecarboxylate (2b). A solution of 2.14 g (4.08 mmol) of the methyl ketone **18** in 50 mL of dioxane and 15 mL of water was cooled to 8 °C, and 25.4 mL of a 0.53 M solution of NaOBr in aqueous dioxane was introduced. After 3 h at 10 °C, the mixture was brought to room temperature, treated with 5 mL of 10% aqueous Na$_2$SO$_3$, diluted with water, and washed with ether. The product was recovered from the aqueous layer after cooling to 10 °C, neutralizing (to pH 6) with saturated KHSO$_4$, and extracting with three portions of ether. The combined organic layer was washed with water and brine, dried, and concentrated to give 1.83 g (92%) of the hydroxy acid **19** as an oil: ^{1}H NMR δ 0.83 (d, 3), 3.67 [m, 1, >CHOC(C$_6$H$_5$)$_3$],), 4.33 (m, 1, >CHOH), 5.38 (m, 2), 6.58 (br s, 2, –CO$_2$H, OH).

A mixture of 1.83 g (3.78 mmol) of the carboxylic acid **19**, 0.80 g (5.8 mmol) of K$_2$CO$_3$, and 0.39 mL (4.1 mmol) of dimethyl sulfate in 40 mL of acetone was stirred under reflux for 2 h. The mixture was partitioned between ether and water, and the organic layer was washed with aqueous ammonia, water, and brine, dried, and concentrated to give a thick syrup. After standing at room temperature for 7 days, it was chromatographed on 90 g of Florisil, eluting with 30% ether/hexane, to give 1.1 g (58%) of the isomerized ester **2b** as an oil. ^{1}H NMR δ 1.17 (d, 3), 3.60 (s, 3), 3.75 (m, 1, >CHOC(C$_6$H$_5$)$_3$], 4.15 (m, 1, >CHOH), 5.4 (m, 2); IR (film) 970 (trans CH=CH), 1738 (C=O), 3430 (OH) cm^{-1}. A pure sample of this material was not obtained for combustion analysis. However, treatment with dimsylsodium and chromatography on Florisil (ether) provided an analytical sample of the β-ketosulfoxide **31** (trityl instead of MOM).

Anal. (C$_{34}$H$_{40}$O$_4$S) C, H, S.

***trans*-4-Oxocyclopentane-1,2-dicarboxylic Acid (22).** Trimethyl 4-oxocyclopentane-1,2,3-tricarboxylate[25b] (111 g) was dissolved in 600 mL of concentrated HCl and 100 mL of water and heated on a steam bath for 9 h. The dark solution was cooled, treated with decolorizing charcoal, and filtered, and the major portion of the aqueous HCl was removed at reduced pressure to give 160 g of wet sludge. This material was dissolved in 200 mL of hot water and allowed to crystallize overnight at 0 °C, affording 58.6 g of the trans diacid **22** as colorless crystals. Concentration and further crystallization of the filtrate gave an additional 7.5 g of material (total yield, 88%): mp 188–191 °C (lit.[25b] mp 189 °C).

[1α,2β]-4-Hydroxycyclopentane-1,2-dicarboxylic Acid (23). A so-

lution of 54.0 g (314 mmol) of the keto diacid **22** in 500 mL of methanol was neutralized with 25.6 g (640 mmol) of NaOH in 100 mL of water, cooled to 5 °C, and treated with 6.0 g (160 mmol) of NaBH$_4$. The reaction mixture was kept overnight at room temperature, acidified to pH 3, and concentrated at reduced pressure to give an amorphous solid. This material was extracted with three 50-mL portions of hot THF, and the supernatant was dried and evaporated to give 52.0 g (95%) of the hydroxy acid **23** as a white solid: ^{1}H NMR (D$_2$O) δ 2.0 (m, 4), 3.3 (m, 2), 4.5 (m, 1); IR (mull) 1690, 1710 (C=O), 2700 (br, CO$_2$H), 3440 (OH) cm^{-1}. An analytical sample was obtained by recrystallization from water: mp 164–170 °C (dec).

Anal. (C$_7$H$_{10}$O$_5$) C, H.

exo-6-Oxo-5-oxabicyclo[2.2.1]heptane-2-carboxylic Acid (21). To a stirred solution of 12.4 g (71 mmol) of hydroxy acid **23** and 10.2 mL (73 mmol) of triethylamine in 250 mL of acetone and 250 mL of THF at −5 °C was added 7.0 mL (73 mmol) of ethyl chloroformate over a period of 10 min. After 30 min at 0 °C, 70 g of Linde 4 Å molecular sieves was added, and the mixture was stirred at room temperature overnight. The mixture was filtered through a pad of celite and concentrated, and the residue was dissolved in 200 mL of ethyl acetate and washed with two 20-mL portions of 2 N H$_2$SO$_4$. The aqueous phase was reextracted with ethyl acetate, and the combined organic layer was dried and evaporated to give 8.4 g (76%) of the bicyclic lactone **21** as a white solid of sufficient purity to be used in the next step. An analytical sample was obtained by recrystallization from chloroform: mp 109–110 °C; ^{1}H NMR δ 1.96 (d, 1, J = 11 Hz, H(5)-exo), 2.25–2.90 [m, 3, H(6)-endo, H(7)], 3.0 (dd, 1, J = 6.5, 7.7 Hz, H(5)], 3.2 [br s, 1, H(4)], 5.0 [br s, 1, H(1)]/ IR (CDCl$_3$) 1720 (CO$_2$H), 1790 (lactone), 3200 (br, CO$_2$H) cm^{-1}.

Anal. (C$_7$H$_8$O$_4$) C, H.

5-Benzyloxyhexyl Phenyl Sulfone (24). A solution of 45.0 g (280 mmol) of sodium benzenesulfinate and 43.0 g (240 mmol) of 6-bromo-2-hexanone^{28} in 200 mL of dimethylformamide was kept at room temperature overnight and then at 70 °C for 1 h. The mixture was poured into 800 mL of ice water, and the solid was collected by filtration, dried under reduced pressure, and recrystallized from hexane to give 34.8 g (60%) of 6-phenylsulfonyl-2-hexanone as colorless needles: mp 66–67 °C; ^{1}H NMR δ 1.68 (m, 4), 2.15 (s, 3), 2.37 (br t, 2), 3.12 (br t, 2), 7.6–8.0 (m, 5); IR (mull) 1153, 1320 (SO$_2$), 1705 (C=O) cm^{-1}.

Anal. (C$_{12}$H$_{16}$O$_3$S) C, H, S.

The ketosulfone was reduced with 2.74 g (73 mmol) of NaBH$_4$ in 430 mL of absolute ethanol at room temperature. After 90 min, the mixture was concentrated at reduced pressure and partitioned between 3 N HCl and ether. The organic layer was washed with saturated NaHCO$_3$ and brine, dried, and evaporated to give a quantitative yield of 6-phenylsulfonyl-2-hexanol as an oil. An analytical sample was obtained by distillation [110 °C (0.035 Torr)]: ^{1}H NMR δ 1.15 (d, 3), 3.12 (t, 2, CH$_2$SO$_2$), 3.72 (m, 1, CHO); IR (film) 1150, 1300 (SO$_2$), 3700 (OH) cm^{-1}.

Anal. (C$_{12}$H$_{18}$O$_3$S) C, H, S.

A 150-mmol portion of sodium hydride was suspended in 80 mL of DMF, and 24.2 g (100 mmol) of the hydroxy sulfone in 70 mL of DMF was introduced. The mixture was briefly warmed to complete hydrogen evolution and 12.5 mL (105 mmol) of benzyl bromide was added slowly, maintaining the temperature of the reaction mixture below 50 °C. After an additional 60 min at room temperature, the mixture was partitioned between ether and water, and the organic layer was washed with 3 N H$_2$SO$_4$, aqueous NaHCO$_3$ and brine, dried, and concentrated to give 24 g of a light yellow oil. After chromatography (ether/hexane, 3:1), 17 g (52%) of pure sulfone **24** was obtained as an oil: ^{1}H NMR δ 1.13 (d, 3), 3.05 (br t, 2), 3.45 (m, 1), 4.45 (AB q, 2, J = 11 Hz, OCH$_2$Ph), 7.3 (s, 5), 7.6–7.9 (m, 5); IR (film) 1150, 1310 (SO$_2$) cm^{-1}. An analytical sample was prepared by distillation [130 °C (0.02 Torr)].

Anal. (C$_{19}$H$_{24}$O$_3$S) C, H, S.

[1α,2β,4β]-2-(6-Benzyloxy-1-oxo-2-phenylsulfonylheptyl)-4-hydroxycyclopentanecarboxylic Acid (25). To a solution of 4.82 g (14.5 mmol) of the sulfone **24** and 4.7 mL (33.4 mmol) of diisopropylamine in 90 mL of THF stirred under nitrogen at −78 °C was added 45.7 mmol of an *n*-butyllithium/hexane solution. After stirring the yellow solution for 10 min, 2.22 g (14.2 mmol) of lactone **21** in 90 mL of THF was added over a 5-min period. The mixture was stirred at −78 °C for 15 min, warmed gradually to room temperature, and partitioned between ether and ice water. The organic layer was extracted with 5% NaCO$_3$, and the combined aqueous phase was washed with ether,

acidified (pH 2), and extracted with ether. The ether layer was washed with water and brine, dried, and concentrated to give 6.32 g (91%) of analytically pure β-ketosulfone **25** as a thick syrup: ^{1}H NMR δ 1.10 (d, 3, J = 6 Hz), 0.95–2.28 (m, 10), 2.78–4.00 (m, 4), 4.32–4.42 (m, 4, >CHO), 7.25 (s, 5, Ph), 7.4–7.9 (m, 5, PhSO$_2$); IR (film) 1150, 1315 (SO$_2$), 1450 (CH$_2$), 1710 (C=O), 3400 (OH) cm^{-1}.

Anal. (C$_{26}$H$_{32}$O$_7$S) C, H, S.

Methoxymethyl [1α,2β,4β]-2-(6-Benzyloxy-1-oxo-2-phenylsulfonylheptyl)-4-methoxymethoxycyclopentanecarboxylic Acid (26). A solution of 17.7 g (36.2 mmol) of hydroxy acid **25** in 150 mL of dimethoxymethane and 150 mL of chloroform was stirred at 2 °C and treated with 54 g (190 mmol) of P$_4$O$_{10}$ in several portions.^{29} After stirring for 60 min at 2–3 °C, the supernatant was decanted into a mixture of ether and saturated aqueous NaHCO$_3$, the residue was washed with ether, and the combined organic layer was washed with aqueous NaHCO$_3$ and brine, dried, and concentrated to give 18.4 g (88%) of the bis(methoxymethyl) derivative **26** as an oil: ^{1}H NMR (of diastereomeric mixture) δ 1.08 (d, 3), 3.23 and 3.30 (s, 3), 3.38 and 3.47 (s, 3), 4.17 [quintet, 1, J = 5 Hz, H(4)], 4.38 (AB q, 2, J = 11 Hz, OCH$_2$Ph), 4.48 and 4.57 (s, 2), 5.13 (s) and 5.23 (AB q, 2, J = 6 Hz, CO$_2$CH$_2$O); IR (film) 1150 and 1300 (SO$_2$), 1720 and 1740 (C=O) cm^{-1}. An analytical sample was obtained by chromatography (ether).

Anal. (C$_{30}$H$_{40}$O$_9$S) C, H, S.

Methoxymethyl [1α,2β,4β]-2-(6-Benzyloxy-1-[diphenoxyphosphinyl]oxy-2-phenylsulfonyl-1-heptenyl)-4-methoxymethoxycyclopentanecarboxylate (27). The addition of 1.0 g (8.2 mmol) of 4-dimethylaminopyridine to a solution of 18.0 g (31.2 mmol) of the protected β-keto sulfone **26**, 5.8 mL (41.3 mmol) of triethylamine, and 7.8 mL (37.5 mmol) of diphenylphosphorochloridate in 100 mL of dry acetonitrile resulted in a mildly exothermic reaction and immediate precipitation of triethylammonium chloride. After stirring for 1 h at room temperature, the mixture was partitioned between ether and dilute NaHCO$_3$, and the organic layer was washed with 3 N H$_2$SO$_4$, aqueous NaHCO$_3$, and brine, dried, and concentrated to give 22.7 g (90%) of the enol phosphate **27** as an oil: ^{1}H NMR δ 1.02 (d, 3), 3.27 (s, 3), 3.37 (s, 3), 4.27 (m, 1), 4.35 s, 2, CH$_2$Ph), 4.47 (s, 2, OCH$_2$O), 5.13 (AB q, 2, CO$_2$CH$_2$O), 7.17 (s, 5, PhCH$_2$), 7.22 (s, 10, PhO), 7.45–7.90 (m, 5, PhSO$_2$); IR (film) 1160 (P=O), 1050 and 1308 (SO$_2$), 1597 and 1613 (C=C), 1740 (C=O) cm^{-1}. A sample was purified for analysis by chromatography (ether).

Anal. (C$_{42}$H$_{49}$O$_{12}$SP) C, H, P, S.

[1α,2β,4β]-2-(6-Benzyloxy-1-[diphenoxyphosphinyl]oxy-2-phenylsulfonyl-1-heptenyl)-4-methoxymethoxycyclopentanecarboxylic Acid (28). A solution of 22.7 g (27.2 mmol) of the enol phosphate **27** in 185 mL of glacial acetic acid with 5.5 mL of trifluoroacetic acid was kept at 35 °C for 12 h and at 45 °C for 2 h. The acetic acid was removed by evaporation at reduced pressure and by dissolving the residue in toluene and evaporating again. The residue was partitioned between ethyl acetate and a 1:1 mixture of saturated NaHCO$_3$ and brine, and the organic layer was dried and concentrated to give 19.9 g (95%) of the carboxylic acid **28** as an oil: ^{1}H NMR δ 0.98 (d, 3), 3.23 (s, 3), 4.33 (s, 2, OCH$_2$Ph), 4.47 (s, 2, OCH$_2$O), 7.17 (s, 5, PhCH$_2$), 7.23 (s, 10, PhO), 7.5–7.8 (m, 5, PhSO$_2$); IR (film) 1160 (P=O), 1050, 1300 (SO$_2$), 1595, 1630 (C=C), 1738 (C=O), 2700 (br, CO$_2$H) cm^{-1}. An analytical sample was purified by chromatography (ether).

Anal. (C$_{40}$H$_{45}$O$_{11}$SP) C, H, P, S.

[1α,2β(E),4β]-2-(6-Hydroxy-1-heptenyl)-4-methoxymethoxycyclopentanecarboxylic Acid (29). To a solution of 3.0 g (0.13 mol) of sodium in 65 mL of ammonia at −78 °C was added a solution of 5.0 g (6.5 mmol) of the enol phosphate **28** in 18 mL of THF over a period of 10 min. The mixture was stirred under reflux for 5 h, quenched with NH$_4$Cl, and diluted with ether, and the ammonia was allowed to evaporate. The residue was extracted with water and with 1 N NaOH, and the combined aqueous layer was brought to pH 9 with H$_3$PO$_4$, washed twice with ether, and finally acidified (to pH 3.5) with H$_3$PO$_4$ and extracted with ethyl acetate. The ethyl acetate phase was washed with brine, dried, and evaporated to give 1.39 g (74%) of olefinic acid **29** as an oil: ^{1}H NMR δ 1.15 (d, 3), 3.35 (s, 3), 4.62 (s, 2, OCH$_2$O), 5.43 (m, 2, CH=CH), 7.0 (br s, 2, CO$_2$H and OH); IR (film) 1705 (C=O), 2650 (br, CO$_2$H), 3415 (OH) cm^{-1}. A sample was purified for analysis by chromatography (ether).

Anal. (C$_{15}$H$_{26}$O$_5$) C, H.

Methyl [1α,2β(E),4β]-2-(6-Hydroxy-1-heptenyl)-4-methoxymethoxycyclopentanecarboxylate (2c). The hydroxy acid **29** (1.39 g, 4.86 mmol) was dissolved in 40 mL of methylene chloride and treated

with ethereal diazomethane until a yellow color persisted. The excess diazomethane was quenched with acetic acid, and the mixture was diluted with ether, washed with saturated NaHCO₃ and brine, dried, evaporated, and distilled [100 °C (0.04 Torr)] to give 1.0 g (71%) of the ester **2c** as an oil: ¹H NMR δ 1.17 (d, 3), 3.35 (s, 3), 3.67 (s, 3), 3.70 (m, 1, >CHOH), 4.17 (quintet, 1, *J* = 6 Hz, >CHOCH₂), 4.60 (s, 2), 5.38 (m, 2); IR (firm) 1738 (C=O), 3500 (OH) cm⁻¹.

Anal. (C₁₆H₁₈O₅) C, H.

1-([1α,2β(*E*),4β]-2-[6-Hydroxy-1-heptenyl]-4-methoxymethoxy-cyclopentyl)-2-methylsulfinylethanone (31). A solution of 21.0 mmol of dimsylsodium in 10 mL of Me₂SO was prepared at 75 °C, diluted with 30 mL of THF, and cooled to −10 °C. To this solution was added 1.58 g (5.26 mmol) of the ester **2c** in 15 mL of THF over a 20-min period. After an additional 30 min at −10 °C, the mixture was partitioned between ether and water, and the organic layer was extracted with 1 N NaOH. The combined aqueous phase was washed with ether, saturated with NH₄Cl, and extracted with ethyl acetate, and the ethyl acetate layer was washed with water and brine, dried, and evaporated to give 1.59 g (87%) of the β-keto sulfoxide **31** as a thick syrup: ¹H NMR δ 1.13 (d, 3), 2.67 [s, 3, CH₃S(=O)], 3.33 (s, 3, CH₃O), 3.67 (m, 1, >CHOH), 3.80 [br s, 2, (O=)CCH₂S(=O)], 4.07 (quintet, 1, *J* = 6 Hz, >CHOCH₂), 4.57 (s, 2, OCH₂O), 5.43 (m, 2); IR (film) 1040 (S=O), 1710 (C=O), 3430 (OH) cm⁻¹. A sample for analysis was purified by chromatography (acetone).

Anal. (C₁₇H₃₀O₅S) C, H, S.

Methyl (2*E*)-4-([1α,2β(*E*),4β]-2-[6-Hydroxy-1-heptenyl]-4-methoxymethoxycyclopentyl)-4-oxo-2-butenoate (33). To a solution of 271 mg (0.78 mmol) of the β-keto sulfoxide **31** in 1.37 mL of a 0.60 M solution of potassium *tert*-butoxide in THF (0.82 mmol) at 0 °C was added 108 μL (1.17 mmol) of methyl bromoacetate. After 1 h at 0 °C, the resulting slurry was partitioned between ethyl acetate and saturated NH₄Cl, and the organic layer was washed with water and brine, dried, and concentrated to give the crude, alkylated product as a yellow oil. This material was dissolved in 3.3 mL of dioxane, with 90 mg (1.1 mmol) of NaHCO₃ and 72 μL (0.78 mmol) of methyl bromoacetate, and stirred at reflux for 90 min.¹² After diluting with ether, the mixture was washed with 1 N NaOH, water and brine, dried, and concentrated to give 196 mg (71%) of the γ-keto acrylate **33** as an oil: ¹H NMR δ 3.35 (s, 3), 3.78 (s, 3), 4.60 (s, 2), 5.38 (m, 2), 6.55 and 7.07 (AB q, 2, *J* = 16 Hz); IR (film) 1640 (C=C), 1703, 1738 (C=O), 3520 (OH) cm⁻¹; UV (ethanol) λ_max (log ε) 224 nm (4.09). An analytical sample was prepared by distillation [110 °C (0.025 Torr)].

Anal. (C₁₉H₃₀O₆) C, H.

(2*E*)-4-([1α,2β(*E*),4β]-2-[6-Hydroxy-1-heptenyl]-4-methoxymethoxycyclopentyl)-4-oxo-2-butenoic Acid (34). The keto ester **33** (377 mg, 1.06 mmol) was dissolved in 12 mL of a 0.2 M solution of lithium hydroxide in 50% aqueous THF and kept at 0 °C for 30 min. The mixture was diluted with ethyl acetate, the aqueous layer was brought to pH 3 with 2 N H₃PO₄ and extracted, and the organic layer was washed with brine, dried, and concentrated to give 360 mg (100%) of the carboxylic acid **34** as a thick syrup: ¹H NMR δ 3.35 (s, 3), 5.37 (m, 2), 6.35 (br s, 2, CO₂H and OH), 6.52 and 7.05 (AB q, 2, *J* = 16 Hz); IR (film) 1695, 1730 (C=O), 2670 (br, CO₂H), 3470 (OH) cm⁻¹.

(2*E*,6*R,10*E*,11a*R**,13*R**,14a*S**)-13-Methoxymethoxy-6-methyl-7,8,9,11a,12,13,14,14a-octahydro-6*H*-cyclopent[*f*]oxacyclotridecin-1,4-dione (4-Dehydrobrefeldin A 7-Methoxymethyl Ether) (35), and the 6*S** Stereoisomer (15-*epi*-4-Dehydrobrefeldin A 7-Methoxymethyl Ether) (36).** A solution of 173 mg (0.51 mmol) of hydroxy acid **34** and 0.56 mL (4.1 mmol) of triethylamine in 40 mL of methylene chloride was added via syringe pump to a refluxing solution of 508 mg (2.36 mmol) of 2-chloro-1-methylpyridinium tetrafluoroborate in 37 mL of CH₂Cl₂ and 12 mL of acetonitrile over an 8-h period.³¹ The solvent was removed under reduced pressure and the residue was dissolved in ethyl acetate, washed with water, dilute H₃PO₄, saturated NaHCO₃ and brine, dried, and concentrated to give 170 mg of a dark oil. This material was chromatographed (ether/pentane, 1:1) to give 60 mg (37%) of a mixture of C-15 epimers **35** and **36**.

Anal. (C₁₈H₂₆O₅) C, H.

The two components were separated by preparative thin-layer chromatography (silica gel; eluted with ether/pentane, 1:1) or high-pressure column chromatography (Partisil M-9; eluted with ether/hexane, 1:2).

4-Dehydrobrefeldin A 7-methoxymethyl ether, **35:** R_f 0.40; ¹H

NMR (180 MHz) δ 1.33 (d, 3, *J* = 6.1 Hz), 2.26 [m, 2, H(12)], 2.57 [m, 1, H(9)], 2.89 [q, 1, *J* = 9 Hz, H(5)], 3.36 (s, 3, CH₃O), 4.10 [quintet, 1, *J* = 5.3 Hz, H(7)], 4.63 (s, 2, OCH₂O), 4.68 [m, 1, H(15)], 5.53 [dd, 1, *J* = 10.3 and 15.2 Hz, H(10)], 5.87 [ddd, 1, *J* = 4.4, 10.2, 15.2 Hz, H(11)], 6.45 [d, 1, *J* = 15.9 Hz, H(3)], 7.77 [d, 1, *J* = 15.9 Hz, H(2)]; UV (ethanol) λ_max (log ε) 220 nm (3.67).

15-*epi*-4-Dehydrobrefeldin A 7-methoxymethyl ether, **36:** R_f 0.45; ¹H NMR (180 MHz) δ 1.26 (d, 3, *J* = 6.5 Hz), 3.05 [q, 1, *J* = 8.4 Hz, H(5)], 3.36 (s, 3, CH₃O), 4.08 [m, 1, H(7)], 4.64 (s, 2, OCH₂O), 5.23 [m, 1, H(15)], 5.37 (m, 2, H(10), H(11)], 6.44 [d, 1, *J* = 15.8 Hz, H(3)], 7.44 [d, 1, *J* = 15.8 Hz, H(2)]; UV (ethanol) λ_max (log ε) 224 nm (3.72).

(1*R,2*E*,6*R**,10*E*,11a*R**,13*R**,14a*S**)-1,6,7,8,9,11a,12,13,14,-14a-Decahydro-1-hydroxy-13-methoxymethoxy-6-methyl-4*H*-cyclopent[*f*]oxacyclotridecin-4-one (Brefeldin A 7-Methoxymethyl Ether) (37).** Sodium borohydride (2 mg, 50 μmol) was added to a solution of 8.9 mg (28 μmol) of the ketone **35** in 0.35 mL of methanol at −78 °C. After 90 min, 0.2 mL of acetone was added and the solution was allowed to warm to room temperature. The mixture was diluted with ethyl acetate, washed with water, saturated NaHCO₃ and brine, dried, and evaporated to give 7.7 mg (87%) of brefeldin A 7-methoxymethyl ether **37** and its 4-epimer in a ratio of greater than 10:1: ¹H NMR (180 MHz) δ 1.26 (d, 3, *J* = 6.2 Hz), 3.36 (s, 2, CH₃O), 4.11 [m, 2, H(4) and H(7)], 4.63 (s, 2, OCH₂O), 4.84 [m, 1, H(15)], 5.26 [dd, 1, *J* = 8.9 and 15.0 Hz, H(10)], 5.70 [ddd, 1, *J* = 4.8, 9.8, and 15.0 Hz, H(11)], 5.90 [dd, 1, *J* = 2.0 and 15.7 Hz, H(2)], 7.35 [dd, 1, *J* = 3.1 and 15.7 Hz, H(3)].

(1*R,2*E*,6*R**,10*E*,11a*R**,13*R**,14a*S**)-1,6,7,8,9,11a,12,13,-14,14a-Decahydro-1,13-dihydroxy-6-methyl-4*H*-cyclopent[*f*]oxacyclotridecin-4-one [(±)-Brefeldin A] (1).** A solution of 6 mg (19 μmol) of the methoxymethyl ether **37** and 10 μL of trifluoroacetic acid in 0.3 mL of a 2:1 mixture of ethanol and acetic acid was kept at 50 °C for 12 h. The mixture was diluted with ethyl acetate, washed with saturated NaHCO₃ and brine, dried, and concentrated to give an oil from which 2 mg of racemic brefeldin A (mp 169–173 °C) was obtained on trituration with ether. Chromatography of the mother liquor residue (preparative TLC, ethyl acetate) afforded an additional 1 mg of pure material (total yield, 55%). Recrystallization from ethyl acetate afforded material having mp 175–175.5 °C: ¹H NMR (180 MHz) δ 1.26 (d, 2, *J* = 6.3 Hz), 4.1 [br d, 1, H(4)], 4.33 [~quintet, 1, *J* ≃ 4.7, H(7)], 4.8 [m, 1, H(15)], 5.27 [dd, 1, *J* = 9.2 and 15.0 Hz, H(10)], 5.70 [ddd, 1, *J* = 4.9, 10.0, and 15.0 Hz, H(11)], 5.90 [dd, 1, *J* = 2.0 and 15.7 Hz, H(2)], 7.35 [dd, 1, *J* = 3.1 and 15.7 Hz, H(3)].

(1*R,2*E*,6*R**,10*E*,11a*S**,13*S**,14a*R**)-1,6,7,8,9,11a,12,13,-14,14a-Decahydro-1-hydroxy-13-methoxymethoxy-6-methyl-4*H*-cyclopent[*f*]oxacyclotridecin-4-one (4-*epi*-15-*epi*-Brefeldin A 7-Methoxymethyl Ether) (39).** Sodium borohydride reduction of isomer **36** was performed as described above for epimer **35**, and afforded an 84% yield of the alcohol **39:** ¹H NMR (180 MHz) δ 1.21 (d, 3, *J* = 6.6 Hz), 3.36 (s, 3, CH₃O), 4.1 [m, 2, H(4), H(7)], 4.5 [m, 1, H(15)], 4.64 (s, 2, OCH₂O), 5.2–5.3 [m, 2, H(10), H(11)], 6.01 [dd, 1, *J* = 2.3 and 15.7 Hz, H(2)], 6.94 [dd, 1, *J* = 2.0 and 15.7 Hz, H(3)]. No resonances attributable to epimeric material were visible in the NMR spectrum.

Methyl (2*E*,4*R)-4-Hydroxy-4-([1*R**,2*S**(*E*),4*S**]-2-(6-hydroxy-1-heptenyl)-4-methoxymethoxycyclopentyl)-2-butenoate (41).** A mixture of 4 mg of lactone **39** and 2 mg of K₂CO₃ in 0.2 mL of methanol was kept at room temperature for 16 h, diluted with ethyl acetate, washed with NaHCO₃ and brine, dried, and concentrated to give the monocyclic ester **41** as an oil: ¹H NMR (180 MHz) δ 6.05 [dd, 1, *J* = 1.6 and 15.6 Hz, H(2)], 6.92 (dd, 1, *J* = 5.1 and 15.6 Hz).

Methyl (2*E*,4*R)-4-Hydroxy-4-([1*S**,2*R**(*E*),4*R**]-2-(6-hydroxy-1-heptenyl)-4-methoxymethoxycyclopentyl)-2-butenoate (42).** Sodium borohydride reduction of **33** was performed as described above for compound **35** and gave an 80% yield of a 1:5 mixture of two isomers, **42** and **41**. The major isomer corresponded to that obtained from methanolysis of **39** by ¹H NMR and TLC comparison. The minor isomer corresponded to the material obtained on methanolysis of brefeldin A 7-methoxymethyl ether (**37**) by the same criteria and was therefore identified as **42:** ¹H NMR (180 MHz) δ 6.05 [dd, 1, *J* = 1.6 and 15.7 Hz, H(2)], 6.97 (dd, 1, *J* = 4.3 and 15.7 Hz).

Note Added in Proof. Another approach to the total synthesis of brefeldin A has been reported: D. P. Curran and D.

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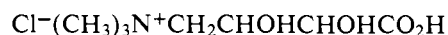# Synthesis of Anthopleurine, the Alarm Pheromone from *Anthopleura elegantissima*

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Abstract: Three of the four possible stereoisomers, **6, 7,** and **8,** of (3-carboxy-2,3-dihydroxy-*N,N,N*-trimethyl)-1-propanaminium chloride, the proposed structure of anthopleurine, the alarm pheromone from the sea anemone *Anthopleura elegantissima,* were prepared from the corresponding 4-amino-2,3-dihydroxybutyric acids, **3, 4,** and **5,** by betaine formation with *O*-methyl-*N,N'*-diisopropylisourea (**2**). Amino acids **3, 4,** and **5** were prepared from D-glucose, L-tartaric acid, and D-tartaric acid, respectively. Biological assay of betaine hydrochlorides **6, 7,** and **8** on the sea anemone, and comparison of physical and spectral data, establishes the structure of anthopleurine as 4-amino-4-deoxy-L-threonic acid betaine hydrochloride (**7**).

Anthopleurine, the alarm pheromone of the sea anemone *Anthopleura elegantissima,* has been identified by chemical and spectroscopic methods as the quaternary ammonium ion, (3-carboxy-2,3-dihydroxy-*N,N,N*-trimethyl)-1-propanaminium, isolated and characterized as the crystalline chloride **1.** Released from wounded anemones, the pheromone evokes a characteristic contraction in nearby conspecifics at a median effective concentration, for the crystalline pheromone, of 3.5 × 10⁻¹⁰ M.[1] The response of the anemone to anthopleurine is mediated by the through-conducting system[2] and is com-

petitively inhibited by L-proline and certain analogues thereof.[3] Anthopleurine has been subsequently shown to evoke an alarm response, visually indistinguishable from that of *A. elegantissima,* in the sea anemone *A. xanthogrammica.*[2]

$$Cl^-(CH_3)_3N^+CH_2CHOHCHOHCO_2H$$

1

To further investigate the utility of *O*-methyl-*N,N'*-diisopropylisourea (**2**) in the conversion of amino acids to their corresponding betaines[4] and to confirm the structure and as-