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Analogs of Host-Specific Phytotoxin Produced by *Helminthosporium maydis*, Race T

I. Synthesis

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Fourteen analogs of the host-specific corn phytotoxin (T toxin) obtained from cultures of the fungal plant pathogen, *Helminthosporium maydis*, race T, were synthesized. Addition of difunctional Grignard reagents to aldehyde intermediates resulted in shorter versions (C_{15}–C_{26}) of native toxin (C_{35}–C_{45}), containing the β-polyketol functions which appear to account for the specificity and very high toxicity (10^{-8}–10^{-9} *M*) of T toxin toward certain corn varieties.

T Toxin, obtained from the corn pathogen, *Helminthosporium maydis*, race T. exhibits high toxicity (10^{-8}–10^{-9} *M*) toward cells and organelles (*1–3*) of susceptible corn possessing Texas male sterile (Tms) cytoplasm, but not resistant varieties with normal fertile cytoplasm. The toxin is a mixture of several linear polyketols varying from C_{35} to C_{45} in length, with each component possessing apparently identical specific toxicity toward Tms corn (*4–7*). The C_{39} and C_{41} components, comprising 60–90% of native toxin, have been characterized (Fig. 1).

The chemical basis of the unique specificity to Tms corn is of theoretical and practical importance. Previous work has shown that the fully acetylated native toxin (or its methyl ether derivative) is 500- to 1000-fold less active (10^{-5}–10^{-6} *M*) on susceptible corn, indicating that free hydroxyls are essential for high potency, although a question remains as to whether all hydroxyls are necessary. On the other hand, the isolated oxo functions in C_{41} toxins (bands 1, 2 toxins), or the difference in the β,δ-dioxy-oxo (band 2, 1′ toxins) and β-dioxo-oxy (band 1, 3 toxins) functions at the center of the C_{39} and C_{41} toxins seem to have no significant influence on toxicity because all toxins have identical biological activities.

This paper reports the synthesis of 14 shorter (C_{15}–C_{26}) analogs of T toxin via Grignard addition to certain aldehyde intermediates (**4**, **13**, **15**, and **24b**), as shown in Figs. 2 to 4. A companion paper (*24*) describes their biological activities.

The purposes of this work were: (a) to attempt to establish the minimal structural elements required for toxicity and/or specificity; and (b) to provide additional evidence bearing on the purity and proposed structure of native T toxin.

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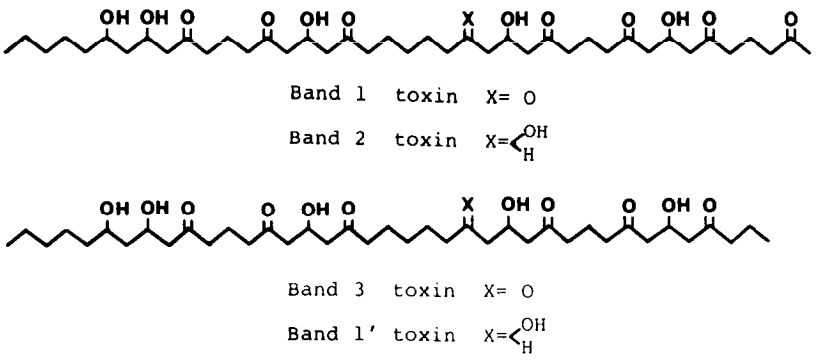

FIG. 1. Proposed structures of host-specific corn phytotoxin produced by *Helminthosporium maydis*, race T.

RESULTS

Preparation of β,δ-Dioxy-oxo Compounds

The aldol condensation (*8*) of hexanal with the dianion of ethyl acetoacetate, generated using NaH and then *n*-BuLi in THF, gave the δ-oxy-β-oxoester **1a** (Fig. 2), which was isolated in 53.0% yield as its silyl ether **1b** by distillation. After the silyl group of **1b** was removed by refluxing in aqueous EtOH, the hyrolysate **1a** was treated with $NaBH_4$ in the presence of NH_4Cl as a buffer, giving a mixture of diastereomers, **2a** and **2b** (2 : 1). This reaction gave more polar substances than **2a**

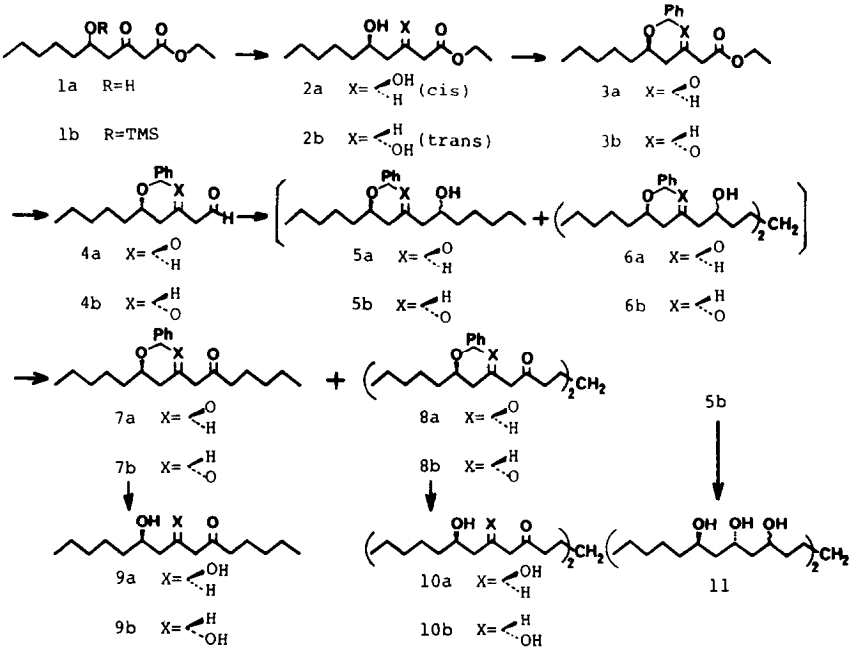

FIG. 2. Synthetic scheme of analogs with β,δ-dioxy-oxo functions of race T toxin.

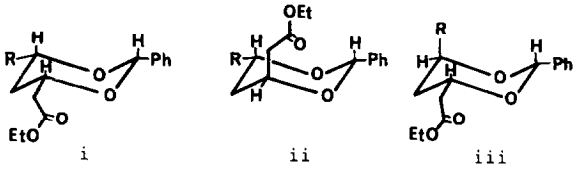

FIG. 3. Relative configurations of benzylidene acetals derived from ethyl 3,5-dihydroxy-decanoates.

and **2b** as main products in the absence of the buffer. Separation of each isomer by column chromatography on silica gel gave **2a** and **2b** in equal yields of 24.0%. Then, exchange reactions of **2a** or **2b** with $PhCH(OEt)_2$ in the presence of *p*-TsOH gave both the benzylidene acetals, **3a** and **3b**, in good yields.

The relative configurations of **3a** and **3b** could be assigned unequivocally on the basis of their pmr spectra. The most stable conformations of **3a** and **3b** can be pictured as a chair form i for the 1,3-cis isomer and forms ii and iii for the trans isomer, as shown in Fig. 3. In fact, **3b** was a mixture of two components; the major one was determined to be the trans conformer ii, because a 1H quartet at δ 4.70 ppm (J = 7.2 Hz) could be assigned to the equatorial proton at C-3 of the conformer ii. Among the signals belonging to the minor component, the *AB*X-type signal at δ 2.32–2.84 ppm was assigned to methylene protons at C-2, resembling closely that of **3a**, which consists of a single component. These data show clearly that **3a** has the cis configuration i and **3b** has the trans configurations ii and iii.

The benzylidene esters, **3a** and **3b**, were converted to the corresponding aldehydes, **4a** and **4b**, in good yields, by reduction with $LiAlH_4$ and then oxidation by CrO_3–pyridine complex (*9*). Aldehydes, **4a** and **4b**, were condensed with pentyl–1,5-dimagnesiumbromide in THF. The respective condensate, without purification, was converted by treatment with the CrO_3–pyridine complex to give a mixture of **7** and **8**, which were separated by silica-gel chromotography yielding **7a** (15.6%), **8a** (13.8%), or **7b** (6.1%), and **8b** (23.0%). Finally, the removal of the benzylidene groups was accomplished by hydrogenation over 10% palladium on carbon in *t*-butanol for 2 to 72 hr, giving the desired β,δ-dioxy-oxo compounds, **9a, 9b, 10a,** and **10b**. Hydrogenation in alcohols other than *t*-butanol caused considerable amounts of cyclic hemiketals to be formed, although the isolated β,δ-dioxy-oxo compounds did not cyclize appreciably in these solvents. Among the synthesized β,δ-dioxy-oxo compounds, the cis isomers **9a** and **10a** were especially unstable and were converted to less polar compounds when dissolved in nonalcoholic solvents, or dried.

The hexahydroxy compound **11** was also obtained by the hydrogenation of **5b** in EtOH.

Preparation of β-Dioxo-oxy Compounds

Ethyl β-hydroxyoctanoate **12a** (Fig. 4), which was prepared by the Reformatsky reaction (*10*) of hexanal with ethyl bromoacetate, was converted to the β-oxo ester

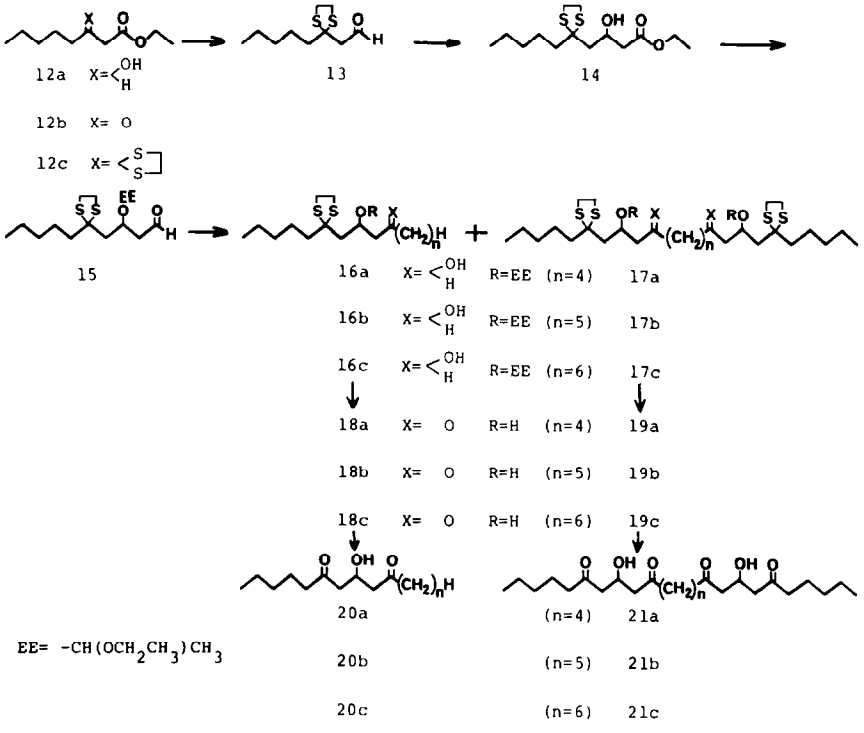

FIG. 4. Synthetic scheme of analogs with β-dioxo-oxy functions of race T toxin.

12b in 65.3% yield by Jones' oxidation. The dithioketalization (*11*) of **12b** using HCl as a catalyst in $CHCl_3$ followed by reduction with $(i\text{-}Bu)_2AlH$ in toluene (*12*) gave the aldehyde **13** in 56.5% yield. Then, **13** was condensed with zinc enolate (*13*) which was prepared from ethyl bromoacetate and zinc in methylal, giving the β-hydroxy ester **14** in 53.8% yield. This reaction did not proceed appreciably under the normal conditions of the Reformatsky reaction (*10*) used above. After the β-hydroxy group was protected as an ethoxyethyl ether (*14*), treatment of **14** with $(i\text{-}Bu)_2AlH$ in toluene gave the aldehyde **15** in a 45.7% yield.

The condensations of **15** with di(bromomagnesium) alkanes ([$MgBr(CH_2)_n$ MgBr], n = 4, 5, and 6) were carried out in the same manner as described above. The condensate mixture of **16** and **17,** without purification, was converted successively by oxidation with CrO_3 in pyridine (*15*) and then hydrolysis (*14*) with dilute acid to the β-hydroxy ketones, **18** and **19**. The yields were 2.2–42.9%. Finally, the ethylenethioketal groups were removed by reaction with $HgCl_2$ in aqueous CH_3CN at room temperature (*16*), and the desired β-dioxo-oxy compounds, **20** and **21,** were obtained in 57.6 to 72.6% yields.

Preparation of δ-Dioxo and β-Oxy-oxo Compounds

3-Hydroxy-octene **22a** (Fig. 5), prepared by the Grignard addition of hexanal with vinylmagnesium bromide in 66.0% yield, was converted to its enone **22b** in 49.6% yield by treatment with the CrO_3–3,5-dimethylpyrazole complex (*17*). The

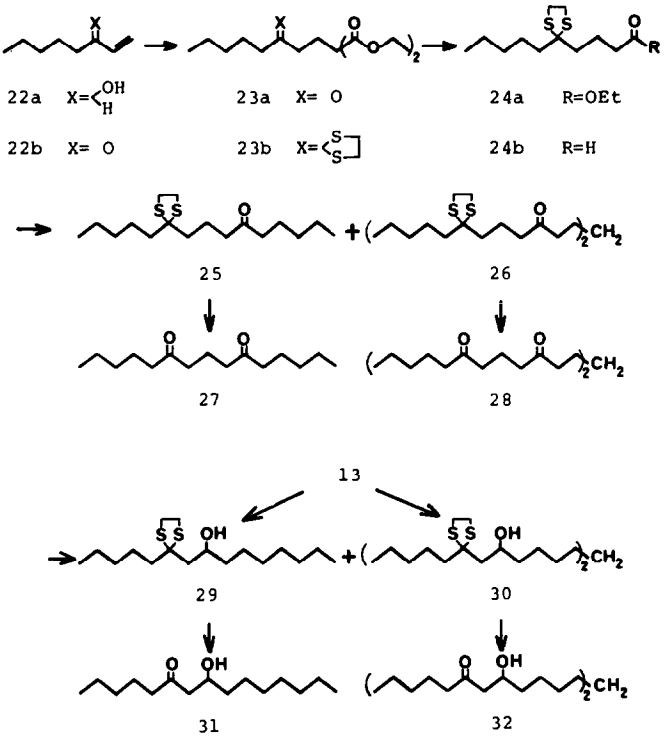

FIG. 5. Synthetic scheme of analogs with partial functional groups of race T toxin.

Michael addition (*18*) of **22b** with diethyl malonate using KOH as a catalyst gave **23a** in good yield. The dithioketalization of **23a** followed by decarboxylation (*19*) using NaCl in aqueous dimethyl sulfoxide gave **24a** in 82.7% yield. The conversions of **24b** to the δ-dioxo compounds, **27** and **28**, were carried out in a similar manner as described above for β-dioxo-oxy compounds.

Similarly, the β-oxy-oxo compounds, **31** and **32**, were obtained via the condensation of **13** with hexylmagnesiumbromide or nonyl-1, 9-dimagnesiumbromide and then hydrolysis.

EXPERIMENTAL

Analytical procedure. All melting points (mp) were recorded on a Yanaco microscope hot plate and are uncorrected. The infrared spectra (ir) were recorded on a Beckman Acculab 6 and a Shimadzu IR-27 with films on NaCl plates or with KBr disks. The pmr spectra were recorded on a Varian EM360 instrument (60 MHz), a Varian XL-100 (100 MHz), or a JEOL FX-90Q (90 MHz) and the chemical shifts are reported in δ units downfield from tetramethylsilane. Low-resolution mass spectra (MS) were recorded on an AEI-M5902 or a Hitachi RMU-6M(G) with a direct inlet system. Field desorption–mass spectra (FD–MS) were recorded on a Hitachi M-80.

Reaction condition. All dry solvents except pyridine were distilled immediately before use, from $LiAlH_4$ (THF and dioxane), Na metal (toluene and methyal), and P_2O_5 (CH_2Cl_2 and $CHCl_3$) and reactions using these dry solvents were run under at an atmosphere of nitrogen.

Ethyl 5-trimethylsilyloxy-3-oxodecanoate (**1b**). The aldol condensation of hexanal with the dianion of ethyl acetoacetate (200 m*M*) was carried out using the procedure of Weiler (*8*). The condensate decomposed completely on distillation, so it was converted to its silylether. To a stirred solution of the crude condensate **1a** in 180 ml of dry pyridine was added 31 g of trimethylsilyl chloride (286 m*M*). The reaction mixture was filtered after 30 min, the residue washed with ether, the combined filtrate washed successively with water and sat. brine solution, and dried over $MgSO_4$. Evaporation of the solvent and subsequent distillation gave 32.0 g of **1b** (53.0%), bp 108–110°C/0.2 mmHg; pmr ($CDCl_3$): δ 2.50–2.88 (2H, *AB*X, *J* = 6.5, 5.4, 15.0 Hz, 4-H), 3.47 (2H, s, 2-H), 4.16 (1H, m, 5-H), and small peaks due to vinyl and allylic protons of the enolic forms [δ 5.00 and 5.12 (each s, 2-H) and 2.00–2.40 (m, 4-H), ir (film): 1735, 1715 cm^{-1}.

Ethyl 3,5-dihydroxy-decanoates (**2a** *and* **2b**). A solution of 18.9 g of **1b** (62.5 m*M*) in 500 ml of 80% EtOH was refluxed for 50 min. Then, to this solution cooled to −20°C was added 3.37 g of NH_4Cl (63 m*M*) and 2.39 g of $NaBH_4$ (63 m*M*). The reaction mixture diluted with water after 70 min of stirring was extracted with ether, the combined extracts were washed with sat. brine and dried, and the solvent evaporated. Purification by column chromatography on silica gel with EtOAc–*n*-hexane (20 : 80 v/v) gave 3.0 g of **2a** as an oil and subsequently 3.0 g of **2b** as crystals from *n*-hexane (mp 46–47°C), both in 24.0% yields.

2a pmr ($CDCl_3$): δ 2.48 (2H, d, *J* = 6.2 Hz, 2-H), 3.25 and 3.80 (each 1H, s, O*H*), 3.86 (1H, br quint, 5-H), 4.26 (1H, quint, *J* = 6.2 Hz, 3-H); ir (film): 3400, 1735 cm^{-1}; MS (rel. int. %): *m*/*z* 214 (M^+-18, 0.5), 143 (100).

2b pmr ($CDCl_3$): δ 2.50 and 2.51 (each 1H, d, *J* = 5.0 and 7.2 Hz, 2-H), ~2.50 (1H, O*H*), 3.50 (1H, d, *J* = 3.6 Hz, O*H*), 3.88 (1H, br quint, 5-H), 4.32 (1H, quint, *J* = 6.0 Hz, 3-H); ir (KBr): 3500, 3450, 3350, 1730, 1705 cm^{-1}; MS (rel. int. %): *m*/*z* 214 (M^+-18, 0.5), 143 (100).

Ethyl 3,5-O-benzylidene decanoates (**3a** *and* **3b**). To a stirred mixture of 2.0 g of **2a** (8.65 m*M*), 10 ml each of benzaldehyde and its diethyl acetal, and 20 ml of dry dioxane was added 125 mg of *p*-TsOH at 20°C. The reaction mixture was poured into 5% $NaHCO_3$ solution after 60 min and the contents extracted with ether. The combined extracts was washed with sat. brine and dried. Evaporation of the solvent gave 2.6 g of **3a** (94.0%) as an oil which was almost exclusively assignable to **3a** by pmr analysis. Similar treatment of **2b** afforded **3b** (94.0%) as a mixture of the conformational isomers ii and iii.

3a pmr ($CDCl_3$): δ 2.34–2.86 (2H, *AB*X, *J* = 7.2, 6.3, 15.5 Hz, 2-H), 3.80 (1H, br m, 5-H), 4.24 (1H, br m, 3-H), 5.54 (1H, s, benzylic-H); MS (rel. int. %): *m*/*z* 320 (M^+, 100).

3b pmr ($CDCl_3$): δ 1.84–2.20 (1H, m, 4-H), 2.58–3.20 (2H, *AB*X, *J* = 8.4, 7.2, 14.4 Hz, 2-H), 3.92 (1H, br m, 5-H), 4.70 (1H, q, *J* = 7.2 Hz, 3-H), 5.79 (1H, s, benzylic-H), and some small peaks due to the conformational isomer iii [δ 2.32–2.84 (*ABX*, 2-H); MS (rel. int. %): *m*/*z* 320 (M^+, 100).

3,5-O-Benzylidene-decanals (**4a** *and* **4b**). To a stirred and cooled solution of 1.73 g of **3a** (5.4 m*M*) in 10 ml of dry THF at −70°C was added a solution of 205 mg of $LiAlH_4$ (5.4 m*M*) in 5 ml of dry THF. After the reaction mixture was kept for 60 min at −70°C and a further 30 min at room temperature, it was worked up according to the directions of Mícovíc and Mihailovíc (*20*). The crude alcohol (1.4 g), without purification, was converted into its aldehyde **4a** by treatment with CrO_3–pyridine complex in CH_2Cl_2. The procedure followed that of Ratcliffe and Rodehorst (*9*), giving 1.04 g of **4a** (78.0%) as an oil after filtration on silica gel with EtOAc–*n*-hexane (20:80 v/v). Similarly, **4b** was obtained in 66.0% yield.

4a pmr ($CDCl_3$): δ 2.40–2.98 (2H, m, 2-H), 3.80 (1H, br m, 5-H), 4.34 (1H, br m, 3-H), 5.55 (1H, s, benzylic-H), 9.83 (1H, t, J = 1.8 Hz); ir (film): 2720, 1725 cm^{-1}; MS (rel. int. %): *m/z* 286 (M^+, 50), 107 (100).

4b pmr ($CDCl_3$): δ 1.84–2.28 (1H, m, 4-H), 2.63–3.40 (2H, m, 2-H), 3.78 (1H, br m, 5-H), 4.88 (1H, q, J = 7.5 Hz, 3-H), 5.74 (1H, s, benzylic-H), and some minor peaks due to the isomer iii [δ 2.40–2.96 (m, 2-H), 4.20 (m), 4.56 (m), 5.80 (s, benzylic-H)]; ir (film): 2720, 1725 cm^{-1}; MS (rel. int. %): *m/z* 286 (M^+, 35), 107 (100).

6,8-O-Benzylidene-pentadecane-10-ones (**7a** *and* **7b**) *and 6,8,18,20-di-O-benzylidene-eicosane-10,16-diones* (**8a** *and* **8b**). A mixture of 99 mg of Mg turnings (4.1 mg atoms), 446 mg of 1,5-dibromopentane (1.94 m*M*), and a crystal of I_2 in 4 ml of dry THF was refluxed for 20 min. A solution of 1.07 g of **4a** (3.88 m*M*) in 2 ml of dry THF was added dropwise at −10°C for 10 min. It was warmed to room temperature over 45 min and then poured into sat. NH_4Cl solution. The organic phase was separated, the aqueous phase extracted with ether, and the extracts washed with sat. brine and dried, and the solvent evaporated. The residual oil was oxidized with the CrO_3–pyridine complex used above, giving a mixture of **7a** and **8a**. Separation by column chromatography on silica gel afforded 210 mg of **7a** (15.6%) and 268 mg of **8a** (13.8%) as oils. Similar treatments of **4b** gave **7b** and **8b** (each 18.1 and 17.1%) as oils.

7a pmr ($CDCl_3$): δ 2.46 (2H, 5, J = 7.2 Hz, 11-H) 2.38–3.00 (2H, *AB*X, J = 7.2, 6.3, 15.5 Hz, 9-H), 3.80 (1H, br m, 6-H), 4.30 (1H, br m, 8-H), 5.53 (1H, s, benzylic-H); MS (rel. int. %): *m/z* 346 (M^+, 9), 99 (100).

7b pmr ($CDCl_3$): δ 1.80–1.30 (1H, m, 7-H), 2.49 (2H, t, J = 7.2 Hz, 11-H), 2.68–3.50 (2H, *AB*X, J = 8.3, 7.2, 15.5 Hz, 9-H), 3.93 (1H, br m, 6-H), 4.76 (1H, q, J = 7.2 Hz, 8-H), 5.75 (1H, s, benzylic-H), and some minor peaks due to the conformational isomer iii [δ 2.48 (t), 2.72–2.98 (*AB*X), 4.18 (m), 4.50 (m), 5.77 (s)]; MS (rel. int. %): *m/z* 346 (M^+, 7), 99 (100).

8a pmr ($CDCl_3$): δ 2.43 (4H, t, J = 7.2 Hz, 11,15-H), 2.35–2.96 (4H, *AB*X, J = 7.2, 6.0, 16.0 Hz, 9,17-H), 3.80 (2H, br m, 6,20-H), 4.30 (2H, br m, 8,18-H), 5.52 (2H, s, benzylic-H); MS (rel. int. %): *m/z* 620 (M^+, 32), 105 (100).

8b pmr ($CDCl_3$): δ 1.80–2.30 (2H, m, 7,19-H), 2.46 (4H, t, J = 7.2 Hz, 11,15-H), 2.64–3.28 (4H, *AB*X, J = 8.3, 7.2, 15.5 Hz, 9,17-H), 3.90 (2H, br m, 6,20-H), 4.75 (2H, q, J = 7.2 Hz, 8,18-H), 5.74 (2H, s, benzylic-H), and some minor peaks due to the conformational isomer iii [δ 2.43 (t), 2.72–2.95 (*AB*X), 4.16 (m), 4.50 (m), 5.76 (s)]; MS (rel. int. %): *m/z* 620 (M^+, 40), 105 (100).

Removal of the benzylidene acetals by hydrogenation. A mixture of 55 mg of **7b**

(0.159 m*M*) and 40 mg of 10% Pd–C in 8 ml of *t*-BuOH was hydrogenated for 2 hr at room temperature and atmospheric pressure. The reaction mixture was passed on a small column of Sephadex LH-20 with *t*-BuOH and the filtrate concentrated at room temperature, crystallization from *n*-hexane, affording 31.1 mg of **9b** (56.4%), mp 67–72°C.

Similarly, **9a, 10a, 10b,** and **11** were obtained as follows. The cis isomer **9a** was purified by column chromatography of silica gel with $CHCl_3$ after 10 hr of the reaction. **9a** decomposed to the dehydrated compounds when dissolved in *n*-hexane and stored at −20°C for precipitation or when dried. Both **10a** and **10b** were purified by precipitation from *n*-hexane–CH_2Cl_2 after each 72 and 24 hr of the reaction. The hexahydroxy compound **11** was isolated also by precipitation from *n*-hexane–CH_2Cl_2 after 2.5 hr of the reaction in EtOH solution.

9a A colorless oil (58.0% yield), pmr ($CDCl_3$): δ 2.41 (2H, t, J = 7.2 Hz, 11-H), 2.56 (2H, d, J = 6.0 Hz, 9-H), 3.0–3.6 (2H, br s, O*H*), 3.85 (1H, br quint, 6-H), 4.29 (1H, quint, J = 6.0 Hz, 8-H); MS of the phenylborate of **9a** (rel. int. %): m/z 344 (M^+, 6), 99 (100).

9b pmr ($CDCl_3$): δ 2.43 (2H, t, J = 7.2 Hz, 11-H), 2.60 (2H, d, J = 5.5 Hz, 9-H), 2.50 and 3.50 (each 1H, O*H*), 3.88 (1H, br m, 6-H), 4.36 (1H, quint, J = 6.0 Hz, 8-H); ir (KBr): 3400, 1700 cm^{-1}; MS of the phenylborate of **9b** (rel. int. %): m/z 344 (M^+, 8), 229 (100).

10a A colorless powder (25.0%), pmr ($CDCl_3$): δ 2.43 (4H, t, J = 7.2 Hz, 11,15-H), 2.57 (4H, d, J = 6.3 Hz, 9,17-H), 3.2–4.1 (4H, br m, O*H*), 3.86 (2H, br quint, 6,21-H), 4.30 (2H, quint, J = 6.3 Hz, 8,18-H); ir (KBr): 3370, 1710 cm^{-1}; MS of the diphenylborate of **10a**: m/z 616 (M^+, 6), 312 (100).

10b A colorless powder (55.1% yield), pmr ($CDCl_3$): δ 2.44 (2H, t, J = 7.2 Hz, 11,15-H), 2.60 and 2.61 (each 2H, J = 6.8, 5.4 Hz, 9,17-H), 2.50 and 3.48 (each 2H, O*H*), 3.88 (2H, br m, 6,21-H), 4.36 (2H, quint, J = 5.7 Hz, 8,18-H); ir (KBr): 3400, 1700 cm^{-1}; MS of the diphenylborate of **10b** (rel. int. %): m/z 616 (M^+, 7), 312 (100).

11 A colorless powder (48.4%), pmr (d_5-pyridine): δ 3.90–5.00 (6H, br m, 6,8,10,16,18,20-H); ir (KBr): 3300 cm^{-1}; FD–MS (rel. int. %): m/z 471 [$(M + Na)^+$, 2], 449 [$(M + H)^+$, 100].

Ethyl 3-oxooctanoate (**12a**). Ethyl 3-hydroxyoctanoate **12a** (100 m*M*) prepared according to the directions of Rathke and Lindert (*10*), without purification, was treated with 29 ml of Jones' reagent in 800 ml of acetone at 0°C for 10 min. The reaction mixture, concentrated after neutralization by adding 16 ml of conc. NH_4OH, was extracted with ether. The combined extracts were washed with sat. brine and dried. Evaporation of the solvent followed by distillation gave 12.12 g of **12b** (65.3%), bp 72–72°C/0.5 mm Hg [lit. 86–91°C/6 mm Hg (*21*)].

Octylaldehyde-3-ethylenethioketal (**13**). A solution of 12.0 g of **12b** (64.5 m*M*) and 9.1 g of ethanedithiol (97 m*M*) in 65 ml of dry $CHCl_3$ was stirred for 60 min at room temperature and then cooled to −20°C. Dry HCl gas was slowly passed through the solution for 10 min. The solution was allowed to warm to room temperature. After 24 hr, the reaction mixture was worked up by successively washing with water, 10% KOH solution, and water and drying. Evaporation of the solvent followed by distillation gave 12.52 g of **12c** (74.0%), bp 90–97°C/0.07 mm

Hg. Then, **12c** was reduced to give **13**. To a stirred and cooled solution of 12.0 g of **12c** (45.8 m*M*) in 150 ml of dry toluene at −70°C was added dropwise 48.1 ml of 1 *M* $(i\text{-Bu})_2AlH$ in *n*-hexane solution (48.1 m*M*). The reaction mixture was poured into 5% NaOH solution after 60 min. The organic phase was separated, the aqueous layer extracted with ether, the combined extracts washed with sat. brine and dried, and the solvent evaporated. Purification by short-column chromatography on silica gel followed by distillation afforded 7.63 g of **13** (76.5%), bp 90–96°C/0.07 mm Hg.

12c pmr ($CDCl_3$): δ 1.8–2.4 (2H, m, 4-H), 3.05 (2H, s, 2-H), 3.30 (4H, s); ir (film): 1735 cm^{-1}; MS (rel. int. %): *m/z* 262 (M^+, 30), 191 (100).

13 pmr ($CDCl_3$): δ 1.8–2.2 (2H, m, 4-H), 2.92 (2H, d, J = 2.0 Hz, 2-H), 3.34 (4H, s), 9.80 (1H, t, J = 2.0 Hz, 1-H); ir (film): 2720, 1725 cm^{-1}; MS (rel. int. %): *m/z* 218 (M^+, 20), 147 (100).

Ethyl 3-hydroxy-octanoate-5-ethylenethioketal (**14**). A solution of 7.6 g of **13** (34.9 m*M*) in 4 ml of dry methylal was added dropwise for 5 min at 4°C to the zinc enolate solution which was prepared by treatment of 8.26 ml of ethyl bromoacetate (70 m*M*) with 4.56 g of zinc metal (70-mg atoms) in 40 ml of dry methylal at 40°C for 3 hr, according to the direction of Cure and Gaudemar (*13*). The reaction mixture was poured into sat. NH_4Cl solution after stirring for 18 hr at room temperature and the contents extracted with ether. The combined extracts were washed with sat. brine and dried. Evaporation of the solvent followed by chromatography on silica gel with MeOH-CH_2Cl_2 (1 : 99 v/v) gave 6.24 g of **14** (53.8%); pmr ($CDCl_3$): δ 1.28 (3H, t, J = 7.0 Hz, —OCH_2CH_3), 1.8–2.3 (4H, m, 4,6-H), 2.53 (2H, d, J = 6.0 Hz, 2-H), 3.30 (4H, s), 4.13 (2H, q, J = 7.0 Hz, —OCH_2CH_3), 4.30 (1H, m, 3-H); ir (film): 3450, 1735 cm^{-1}; MS (rel. int. %): *m/z* 306 (M^+, 19), 217 (100).

3-O-(1-Ethoxy)ethyl-decanal-5-ethylenethioketal (**15**). To a stirred solution of 6.0 g of 14 (19.6 m*M*) and 272 mg of *p*-TsOH (1.47 m*M*) in 50 ml of dry dioxane at 20°C was added dropwise 3.75 g of ethyl vinyl ether (52 m*M*) for 5 min. The reaction mixture was poured into sat. $NaHCO_3$ solution after 10 min, the contents extracted with EtOAc, and the combined extracts washed with sat. brine and dried. Evaporation of the solvent gave a slightly red oil which, without purification, was converted to 3.0 g of **15** (45.7%) as an oil, by reduction with $(i\text{-Bu})_2AlH$, as described above, pmr ($CDCl_3$): δ 1.7–2.5 (4H, m, 4,6-H), 2.80 (2H, m, 2-H), 3.27 and 3.38 (each 2H, s), 3.56 (2H, m), 4.32 (1H, m, 3-H), 4.72 (1H, m, —OC*H*O—), 9.81 (1H, t, J = 2.0 Hz, 1-H); ir (film): 2720, 1725 cm^{-1}; MS (rel. int. %): *m/z* 334 (M^+, 4), 175 (100).

Condensation of 15 with di(bromamagnesium)alkanes, oxidation, and hydrolysis. The condensation of **15** (6 m*M*) with butyl-1,4-dimagnesiumbromide (3 m*M*) was carried out by the method described above. The condensate mixture, without purification, was subjected to oxidation by CrO_3 in pyridine. A solution of the condensate mixture in 6 ml of dry pyridine was added to a slurry of 2.96 g of CrO_3 (29.6 m*M*) in 200 ml of dry pyridine. The reaction mixture was stirred for 18 hr, about 100 ml of ether added, the insoluble material filtered off, the filtrate washed with water and then sat. brine and dried, and the solvent evaporated. The residue was taken up in a mixture of 6 ml of 0.1 *N* HCl and 54 ml of THF. The reaction

mixture was worked up in the usual manner after 70 min of the reaction at 15°C. Separation by column chromatography on silica gel gave **18a** and **19a** (each 2.2 and 29.0%) as oils. Their longer homologs, **18b, 19b,** and **19c,** were also prepared similarly.

18a pmr ($CDCl_3$): δ 1.8–2.2 (4H, m, 8,10-H), 2.45 (2H, t, J = 7.2 Hz, 4-H), 2.28–2.82 (2H, *AB*X, J = 7.2, 5.0, 16.0 Hz, 6-H), 3.30 (4H, s), 3.64 (d, J = 2.4 Hz, O*H*), 4.40 (1H, br m, 7-H); ir (film): 3450, 1715 cm^{-1}; MS (rel. int. %): m/z 318 (M^+, 13), 175 (100).

18b A colorless oil (5.9% yield), pmr ($CDCl_3$): δ 1.8–2.2 (4H, m, 9,11-H), 2.45 (2H, t, J = 7.2 Hz, 5-H), 2.28–2.82 (2H, *ABX*, J = 7.2, 5.0, 16.0 *Hz*, 7-H), 3.30 (4H, s), 3.65 (1H, d, J = 2.4 Hz, O*H*), 4.40 (1H, br m, 8-H); ir (film): 3450, 1715 cm^{-1}; MS (rel. int. %): m/z 332 (M^+, 12), 175 (100).

19a pmr ($CDCl_3$): δ 1.8–2.2 (8H, m, 5,7,18,20-H), 2.48 (4H, t, J = 5.1 Hz, 11,14-H), 2.40–2.82 (4H, *AB*X, J = 7.2, 5.0, 16.0 Hz, 9,16-H), 3.30 (8H, s), 3.64 (2H, d, J = 2.4 Hz, O*H*), 4.40 (2H, br m, 8,17-H); ir (film): 3450, 1710 cm^{-1}; MS (rel. int. %): m/z 578 (M^+, 0.5), 175 (100).

19b A colorless oil (39.8%), pmr ($CDCl_3$): δ 1.8–2.2 (8H, m, 5,7,19,21-H), 2.46 (4H, t, J = 7.2 Hz, 11,15-H), 2.36–2.82 (4H, *AB*X, J = 7.2, 5.0, 16.0 Hz, 9,17-H), 3.30 (8H, s), 3.64 (2H, d, J = 2.4 Hz, O*H*), 4.40 (2H, br m, 8,18-H); ir (film): 3450, 1710 cm^{-1}; MS (rel. int. %): m/z 592 (M^+, 0.5), 175 (100).

19c A colorless oil (42.9%), pmr ($CDCl_3$): δ 1.8–2.2 (8H, m, 5,7,20,22-H), 2.44 (4H, t, J = 7.2 Hz, 11,16-H), 2.36–2.80 (4H, *ABX*, J = 7.2, 5.0, 16.0 Hz, 9,18-H), 3.30 (8H, s), 3.64 (2H, d, J = 2.0 Hz, O*H*), 4.40 (2H, br m, 8,19-H); ir (film): 3450, 1710 cm^{-1}; MS (rel. int. %): m/z 606 (M^+, 0.4), 175 (100).

Hydrolysis of the ethylenethioketal groups. A mixture of 495 mg of **19a** (0.856 m*M*), 1.0 g of $HgCl_2$, 308 mg of HgO, 1.5 ml of water, and 23.5 ml of acetonitrile was stirred for 4 hr at room temperature. The cold mixture was filtered and the residue washed with $CHCl_3$. The filtrate was diluted with about 75 ml of water extracted with $CHCl_3$. The combined extracts were washed twice with sat. NH_4Cl solution and dried over $MgSO_4$. Evaporation of the solvent followed by precipitation from *n*-hexane–CH_2Cl_2 gave a colorless powder **21a** (72.6%). Similarly, **20b, 21b,** and **21c** were also obtained (each 65.2, 61.1, and 57.6%), after precipitation from *n*-hexane (**20b**) or *n*-hexane–CH_2Cl_2 (**21b** and **21c**).

20b A colorless powder, mp 67.5–69.0°C, pmr ($CDCl_3$): δ 2.43 (4H, t, J = 7.2 Hz, 5,11-H), 2.60 (4H, d, J = 6.2 Hz, 7,9-H), 3.46 (1H, d, J = 3.6 Hz, O*H*), 4.45 (1H, d of quint, J = 6.2 and 3.6 Hz, 8-H); ir (KBr): 3400, 1700 cm^{-1}; MS (rel. int. %): m/z 256 (M^+, 4), 99 (100).

21a pmr ($CDCl_3$): δ 2.42 (8H, t, J = 7.2 Hz, 5,11,14,20-H), 2.58 (8H, d, J = 6.2 Hz, 7,9,16,18-H), 3.44 (2H, d, J = 3.6 Hz, O*H*), 4.45 (2H, d of quint, J = 6.2, 3.6 Hz, 8,17-H); ir (KBr): 3320, 3230, 1705 cm^{-1}; FD–MS (rel. int. %): m/z 449 [$(M + Na)^+$, 50], 427 [$(M + H)^+$, 100].

21b A colorless powder, pmr ($CDCl_3$): δ 2.43 (8H, t, J = 7.2 Hz, 5,11,15,21-H), 2.59 (8H, d, J = 6.2 Hz, 7,9,17,19-H), 3.48 (2H, d, J = 3.6 Hz, O*H*), 4.45 (2H, d of quint, J = 6.2, 3.6 Hz, 8,18-H); ir (KBr): 3400, 1700 cm^{-1}; FD–MS (rel. int. %): m/z [$(M + K)^+$, 5], [$(M + Na)^+$, 100], [$(M + H)^+$, 13].

21c A colorless powder, pmr ($CDCl_3$): δ 2.43 (8H, t, J = 7.2 Hz, 5,11,16,22-H),

2.60 (8H, d, J = 6.2 Hz, 7,9,18,20-H), 3.46 (2H, d, J = 3.6 Hz, O*H*), 4.45 (2H, d of quint, J = 6.2, 3.6 Hz, 8,19-H); ir (KBr): 3400, 1700 cm^{-1}; FD–MS (rel. int. %): m/z 477 [$(M + Na)^+$, 4], 455 [$(M + H)^+$, 100].

2-Ethoxycarbonyl-5-oxodecanoate (**23a**). The Grignard reaction of hexanal with 1.1 *M* vinyl magnesiumbromide in THF solution was carried out as described above, giving **22a**, bp 63–65°C/8 mm Hg [lit. 70–74°C/15 mm Hg (*22*)], in 66.0% yield. Then **22a** was subjected to oxidation by CrO_3–3,5-dimethylpyrazole complex. A solution of 8.49 g of **22a** (66 m*M*) in 40 ml of dry CH_2Cl_2 was added in one portion to the complex solution prepared from 17.1 g of CrO_3 (171 m*M*) and 17.1 g of 3,5-dimethylpyrazole (171 m*M*) in 150 ml of dry CH_2Cl_2 according to the directions of Corey and Fleet (*17*). After stirring for 4 hr, the solvent was removed at room temperature, the residue extracted with ether, and the combined extracts filtered. Evaporation of the solvent followed by distillation gave 4.13 g of **22b** (49.6%), bp 53°C/8 mm Hg [lit. 59–60°C/16 mm Hg (*23*)]. Then, a solution of 4.13 g of **22b** (32.7 m*M*) in 16.4 ml of ether was added dropwise for 90 min to a mixture of 1.05 g of diethylmalonate (65.5 m*M*) and 852 mg of KOH (16.3 m*M*) in 25 ml of ether and 3.3 ml of EtOH at 15°C. The reaction mixture was stirred for an additional 2 hr at room temperature, quenched by pouring it over ice, and extracted with ether. The dried ether extracts were distilled to remove the solvent and then excess of diethylmalonate, giving 8.97 g of **23a** (96.0%); pmr ($CDCl_3$): δ 1.27 (6H, t, J = 7.0 Hz, —OCH_2CH_3), 2.0–2.6 (6H, m, 3,4,6-H), 3.38 (1H, t, J = 7.0 Hz, 2-H), 4.18 (4H, q, J = 7.0 Hz, —OCH_2CH_3); ir (film): 1750, 1730, 1715 cm^{-1}; MS (rel. int. %): m/z 286 (M^+, 3), 169 (100).

Ethyl decanoate-5-ethylenethioketal (**24a**). The dithioketalization of 7.15 g of **23a** (25 m*M*), as described above, gave 8.39 g of **23b** (93.3%) which, without purification, was subjected to decarboxylation. The procedure followed that of Krapcho and Lovey (*19*). A mixture of 7.68 g of **23b** (23 m*M*), 590 mg of water (32.8 m*M*), 16.4 ml of dimethyl sulfoxide, and 1.15 g of NaCl (19.7 m*M*) was heated at 155–160°C for 4 hr. The reaction mixture was worked up in the usual manner. Purification by column chromatography on silica gel with EtOAc–*n*-hexane (20 : 80 v/v) gave 5.25 g of **24a** (82.7%) as an oil.

23b pmr ($CDCl_3$): δ 1.7–2.3 (6H, m, 3,4,6-H), 3.25 (4H, s), 3.33 (1H, t, J = 7.0 Hz, 2-H); ir (film): 1750, 1730 cm^{-1}; MS (rel. int. %): m/z 362 (M^+, 18), 175 (100).

24a pmr ($CDCl_3$): δ 1.7–2.3 (6H, 3,4,6-H), 2.30 (2H, t, J = 5.7 Hz, 2-H), 3.24 (4H, s), 4.10 (2H, q, J = 7.0 Hz, —OCH_2CH_3); ir (film): 1730 cm^{-1}; MS (rel. int. %): m/z 290 (M^+, 22), 175 (100).

Pentadecane-6,10-dione (*27*) *and pentaeicosane-6,10,16,20-tetraone* (**28**). The reduction by $(i\text{-}Bu)_2AlH$, Grignard condensation, oxidation by CrO_3 in pyridine, and hydrolysis were carried out as described above.

24b A colorless oil (72.4%); pmr ($CDCl_3$): δ 2.45 (2H, br t, 2-H), 9.80 (1H, t, 1-H); ir (film): 2720, 1730 cm^{-1}; MS (rel. int. %): 246 (M^+, 12), 175 (100).

25 A colorless oil (23.0%); pmr ($CDCl_3$): δ 1.7–2.0 (6H, m, 8,9,11-H), 2.40 (4H, m, 5,7-H), 3.26 (4H, s); ir (film): 1715 cm^{-1}; MS (rel. int. %): m/z 316 (M^+, 8), 175 (100).

26 A colorless oil (28.7%); pmr ($CDCl_3$): δ 1.7–2.0 (12H, m, 5,7,8,18,19,21-H), 2.40 (8H, m, 9,11,15,17-H), 3.25 (8H, s); ir (film): 1715 cm^{-1}; MS (rel. int. %): m/z

560 (M^+, 2), 175 (100).

27 A colorless powder (*n*-hexane–CH_2Cl_2), 74.4%; pmr ($CDCl_3$): δ 0.89 (6H, t, J = 6.0 Hz), 1.7–2.0 (2H, m, 8-H), 2.38 (4H, t, J = 7.2 Hz, 5,11-H), 2.43 (4H, t, J = 6.6 Hz, 7,9-H); ir (KBr): 1710, 1700 cm^{-1}; MS (rel. int. %): m/z 240 (M^+, 4), 128 (100).

28 A colorless powder (*n*-hexane–CH_2Cl_2), 65.0%; pmr ($CDCl_3$): δ 0.89 (6H, t, J = 6.0 Hz), 1.7–2.0 (4H, m, 8,18-H), 2.38 (8H, t, J = 7.2 Hz, 5,11,15,21-H), 2.42 (8H, t, J = 6.7 Hz, 7,9,17,19-H); ir (KBr): 1710, 1700 cm^{-1}; MS (rel. int. %): m/z 408 (M^+, 0.2), 169 (100).

8-Hydroxy-pentadecane-6-one (**31**) *and 8,18-dihydroxy-pentaeicosane-6,20-dione* (**32**). The condensation of **13** with the Grignard reagents (heptylmagnesium bromide or nonyl-1,9-dimagnesiumbromide) followed by deprotection were carried out as described above.

29 A colorless oil (68.5%); pmr ($CDCl_3$): δ 1.8–2.1 (4H, m, 5,7-H), 3.30 (4H, s), 3.34 (1H, d, J = 2.7 Hz, O*H*), 4.00 (1H, m, 8-H); ir (film): 3450 cm^{-1}; MS (rel. int. %): m/z 318 (M^+, 28), 247 (100).

30 A colorless oil (61.5%); pmr ($CDCl_3$): δ 1.8–2.1 (8H, m, 5,7,19,21-H), 3.30 (8H, s), 3.33 (2H, d, J = 2.7 Hz, O*H*), 4.00 (2H, m, 8,18-H); ir (film): 3450 cm^{-1}; MS (rel. int. %): m/z 564 (M^+, 2), 175 (100).

31 mp 49–49.5°C (*n*-hexane–CH_2Cl_2), 61.0%; pmr ($CDCl_3$): δ 0.89 (6H, t, J = 5.7 Hz), 2.42 (2H, t, J = 7.2 Hz, 5-H), 2.52 and 2.55 (each 1H, d, J = 7.2, 3.6 Hz, 7-H), 3.04 (1H, d, J = 3.6 Hz, O*H*), 4.00 (1H, m, 8-H); ir (KBr): 3350, 1705 cm^{-1}; MS (rel. int. %): 242 (M^+, 1), 99 (100).

32 A colorless powder (acetone–CH_2Cl_2), 63.9%; pmr ($CDCl_3$): δ 0.89 (6H, t, J = 6.3 Hz), 2.42 (4H, t, J = 7.2 Hz, 5,21-H), 2.52 and 2.55 (each 2H, J = 7.2, 3.6 Hz, 7,19-H), 3.03 (2H, d, J = 3.6 Hz, O*H*), 4.00 (2H, m, 8,20-H); ir (KBr): 3350, 1705 cm^{-1}; MS (rel. int. %): m/z 413 [M + H)$^+$, 0.1], 99 (100).

REFERENCES

1. G. Payne, Y. Kono, and J. M. Daly, *Plant Physiol.* **65,** 785 (1980).
2. G. Payne, H. W. Knoche, Y. Kono, and J. M. Daly, *Physiol. Plant Pathol.* **16,** 227 (1980).
3. J. M. Daly and B. Barna, *Plant Physiol.* **66,** 580 (1980).
4. Y. Kono and J. M. Daly, *Bioorg. Chem.* **8,** 391 (1979).
5. Y. Kono, S. Takeuchi, A. Kawarada, J. M. Daly, and H. W. Knoche, *Tetrahedron Lett.* 1537 (1980).
6. Y. Kono, S. Takeuchi, A. Kawarada, J. M. Daly, and H. W. Knoche, *Agric. Biol. Chem.* **44,** 2613 (1980).
7. Y. Kono, S. Takeuchi, A. Kawarada, J. M. Daly, and H. W. Knoche, *Bioorg. Chem.* **10,** 206 (1981).
8. S. N. Hucking and L. Weiler, *Tetrahedron Lett.* 4835 (1971).
9. R. Ratcliffe and R. Rodehorst, *J. Org. Chem.* **35,** 4000 (1970).
10. M. W. Rathke and A. Lindert, *J. Org. Chem.* **35,** 3966 (1970).
11. D. Seebach and E. J. Corey, *J. Org. Chem.* **40,** 231 (1975).
12. L. I. Zakharkin and I. M. Khorlina, *Tetrahedron Lett.* 619 (1962).
13. J. Cure and M. Gaudemar, *Bull. Soc. Chem. Fr.* 2471 (1969).
14. J. H. van Boom and J. D. M. Hershied, *Synthesis* 169 (1973).

15. N. PAPPAS AND H. R. NACE, *J. Amer. Chem. Soc.* **81,** 4556 (1959).
16. D. SEEBACH, N. R. JONES, AND E. J. COREY, *J. Org. Chem.* **33,** 300 (1968).
17. E. J. COREY AND G. W. J. FLEET, *Tetrahedron Lett.* 4499 (1973).
18. N. C. ROSS AND R. LEVINE, *J. Org. Chem.* **29,** 2346 (1964).
19. A. P. KRAPCHO AND A. J. LOVEY, *Tetrahedron Lett.* 957 (1973).
20. V. M. MÍCOVÍC AND M. L. J. MIHAILOVÍC, *J. Org. Chem.* **18,** 1190 (1953).
21. K. KOJIMA AND R. OSAWA, *J. Pharm. Soc. Japan* **72,** 916 (1952).
22. M. B. GREDY, *Bull. Soc. Chim. Fr.* 1093 (1936).
23. M. KOULKES, *Bull. Soc. Chim. Fr.* 39 (1954).
24. Y. SUZUKI, K. J. TEGTMEIER, J. M. DALY, AND H. W. KNOCHE, *Bioorg. Chem.* **11,** 313 (1982).