Further Studies on the Sesquiterpene Lactones Tulipinolide and Epitulipinolide from *Liriodendron tulipifera* L.[1]

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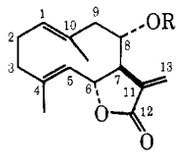
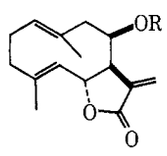
Received February 14, 1972

The dihydro derivatives **3** and **4** of tulipinolide (**1**) and epitulipinolide (**2**) were prepared by $NaBH_4$ reduction since catalytic hydrogenation gave other reduction products. The stereochemistry at the reduced center of these derivatives was determined by cyclization to the already known β-cyclo compounds **6** and **7**. Minor alkaline-hydrolysis products of epitulipinolide (**2**) were established as the methoxy Michael adducts **13** and **15** if methanol was a cosolvent. Without methanol the eudasmanolide diol **16** was formed along with the unusual cadinene lactone **22**, but in no case was the C-8 cis γ-lactone germacranolide (isoeupatolide) detected. A product of alkaline hydrolysis of tulipinolide (**1**) was desacetylisotulipinolide (**23**), a C-8 trans γ-lactone which was also obtained by treatment of eupatolide methanesulfonate (**25**) with hydroxide ion. The acetate of **23** (isotulipinolide) was shown to be identical with the recently isolated germacranolide, laurenobiolide.

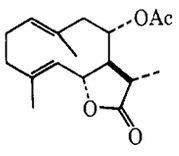
The cytotoxic sesquiterpene lactones, tulipinolide and epitulipinolide, from the root bark of *Liriodendron tulipifera* L., were recently assigned structures **1** and **2**, respectively.[2] We report herein new transformation products of these compounds and show a direct conversion of epitulipinolide to the tulipinolide skeleton by epimerization at the 8 carbon.



1, R = Ac
26, R = H



2, R = Ac
11, R = H
25, R = SO_2Me



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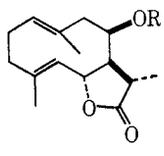
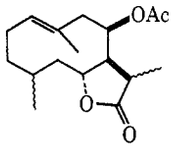


4, R = Ac
12, R = H



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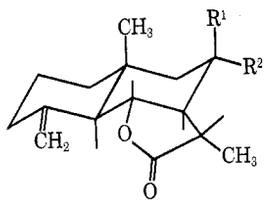


6, R^1 = H, R^2 = OAc
7, R^1 = OAc, R^2 = H

The 11,13-dihydro derivatives **3** and **4** of tulipinolide (**1**) and epitulipinolide (**2**), respectively, were prepared by sodium borohydride reduction, since catalytic hydrogenation gave undesired products. For example, the product of tulipinolide (**1**) and 1 mol of hydrogen still contained the exocyclic methylene protons as seen in the nmr spectrum, while epitulipinolide (**2**), which very rapidly took up 2 mol of hydrogen, gave a substance without the exocyclic methylene but, in addition to the expected secondary C-11 methyl group (δ 1.12, J = 7.1 Hz), the nmr spectrum also showed a three-proton broadened singlet (δ 1.05, $W_{1/2}$ = 7 Hz) reminiscent of a "virtually coupled" methyl group.[3] The physical data support formulation of this substance as **5**.

Dihydrotulipinolide (**3**) has physical properties close to those of acetylbalchanolide,[4] and comparison of their ir spectra[5] indicate that they are most probably identical. The methyl group at C-11 was placed α on the basis that cyclization of dihydrotulipinolide (**3**) produced the known compound dihydrocyclotulipinolide (**6**), for which all of the asymmetric centers were established.[2] Dihydroepitulipinolide (**4**) on cyclization gave the 11 *R* epimer **7** of the two known dihydro-β-cycloepitulipinolides.[2] The cyclization reaction on dihydrotulipinolide and dihydroepitulipinolide also made available the corresponding α-cyclo compounds **8** and **9** as coproducts. To complete the series of dihydrocycloepitulipinolides, the remaining γ isomer **10** was made by sodium borohydride reduction of γ-cycloepitulipinolide.[2] Eupatolide (**11**)[2] was reduced with sodium borohydride to dihydroeupatolide (**12**) with the 11*R* configuration, since on acetylation it gave dihydroepitulipinolide (**4**).

Alkaline hydrolysis of epitulipinolide (**2**) in dilute aqueous methanolic KOH gave mainly eupatolide (**11**) and two additional substances, as observed by thin layer chromatography of the mother liquor. Column chromatography yielded these as crystalline compounds that were characterized as the epimeric Michael addition products of eupatolide and methanol. The major epimer **13** exhibited a methoxy peak at δ 3.34, two olefinic methyls at δ 1.62 and 1.70, and the split AB pattern found typical of H_5 and H_6 protons in the C-6 trans γ-lactone germacranolides.[2] In addi-

(1) Antitumor Agents. VI. Previous paper: R. W. Doskotch, M. Y. Malik, C. D. Hufford, S. N. Malik, J. E. Trent, and W. Kubelka, *J. Pharm. Sci.*, **61**, 570 (1972). This investigation was supported by Public Health Service research grant CA-08133 from the National Cancer Institute and equipment grant FR-00328 from Special Research Resources for purchase of a Varian A-60A nmr spectrometer plus accessories.

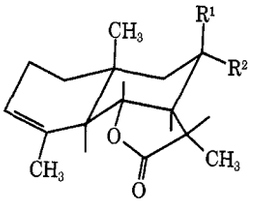
A detailed listing (Table I) of the nmr peaks for the compounds in this publication will appear following these pages in the microfilm edition of this volume of the journal. Single copies may be obtained from the Business Operations Office, Books and Journals Division, American Chemical Society, 1155 Sixteenth St., N.W., Washington, D. C. 20036, by referring to code number JOC-72-2740. Remit check or money order for $3.00 for photocopy or $2.00 for microfiche.

(2) R. W. Doskotch and F. S. El-Feraly, *J. Org. Chem.*, **35**, 1928 (1970).

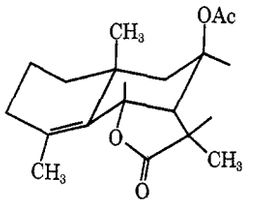
(3) F. A. L. Anet, *Can. J. Chem.*, **39**, 2262 (1961).

(4) J. Hochmannova, V. Herout, and F. Sorm, *Collect. Czech. Chem. Commun.*, **26**, 1826 (1961); V. Herout, M. Suchy, and F. Sorm, *ibid.*, 2612 (1961).

(5) We thank Professor F. Sorm for a copy of the ir spectrum of acetylbalchanolide. Since an authentic sample of the compound was unavailable, our comparison of spectra, one determined on a prism instrument and the other on a grating instrument, precludes a statement about their complete identity. No major differences were noted.



8, $R^1 = H; R^2 = OAc$
9, $R^1 = OAc; R^2 = H$



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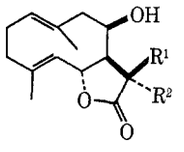
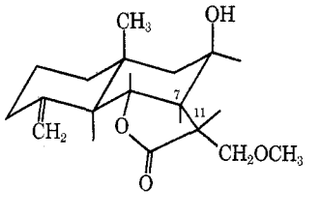


13, $R^1 = H; R^2 = CH_2OCH_3$
15, $R^1 = CH_2OCH_3; R^2 = H$



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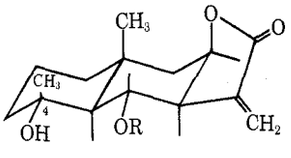
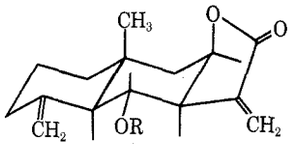


16, R = H
17, R = Ac



18, R = Ac
19, R = H

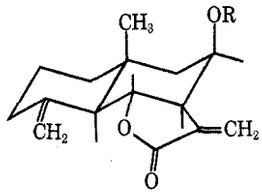
tion, a two-proton, eight-peak pattern, the AB part of an ABX system,[6] was observed for the H_{13} protons, and the X part of this system (H_{11}) was seen as four equal peaks further split by H_7 (J = 12.0 Hz). The large H_7–H_{11} coupling value would require the H_{11} proton to be pseudoaxial, and confirmation of this assignment was obtained from analysis of the cyclized product, 13-methoxydihydro-β-cycloeupatolide (**14**), in which the H_7–H_{11} coupling constant is 12.8 Hz. Values of this order were shown to be indicative of an axial–pseudoaxial interaction for eudesmanolide C-6 trans α-methyl-γ-lactones.[7] Replacement of the α-methyl group with an α-methoxymethylene would not be expected to greatly alter this relationship. The minor dihydro-13-methoxyeupatolide epimer **15** exhibited an nmr spectrum similar to that of the major isomer except that the H_{13} proton pattern appeared as two sharp singlets at δ 3.91 and 3.95,[8] and the H_{11} absorption was hidden in an envelope of peaks, consequently its detailed analysis was not possible.

Elimination of methanol as a cosolvent in the alkaline hydrolysis (followed by acidification) of epitulipinolide eliminated the formation of the methanol adducts **13** and **15**, but instead two other compounds were obtained. One of these analyzed for $C_{15}H_{22}O_4$ and had spectral properties in agreement with structure **16**. Acetylation of the diol **16** to the monoacetate **17** followed by dehydration afforded β-cycloisoepitulipinolide (**18**), identical with the acetate of a product **19** obtained on isomerization by hydrolysis of β-cycloeupatolide (**20**)[2] or of β-cycloepitulipinolide (**21**). This established the

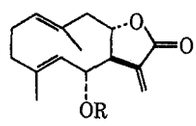
(6) Analysis of this system was carried out as given by R. Bible, "Interpretation of NMR Spectra," Plenum Press, New York, N. Y., 1965, pp 86–92.

(7) C. R. Narayanan and N. K. Venkatasubramanian, *J. Org. Chem.*, **33**, 3156 (1968).

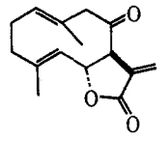
(8) These are undoubtedly the larger inner peaks of an AB quartet for which the smaller outside peaks were too weak to be observed.



20, R = H
21, R = Ac



23, R = H
24, R = Ac



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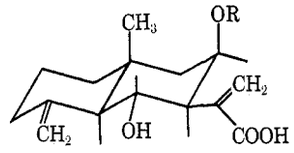

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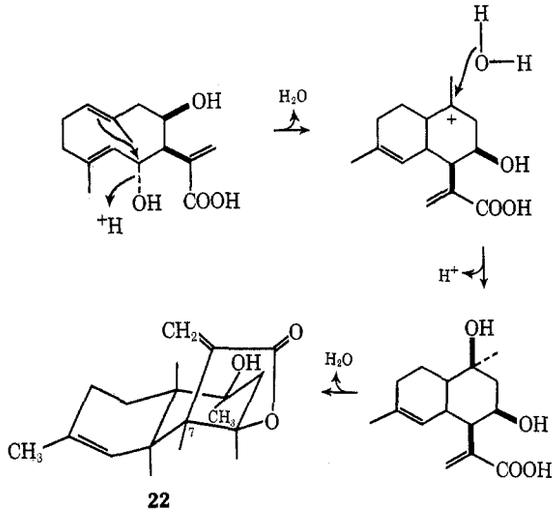
structure and stereochemistry for compound **16** except for the configuration at C-4, which was resolved by utilization of the solvent-induced nmr shift correlations of Demarco, *et al.*,[9] although the dehydrated product itself could be taken as supporting an equatorial hydroxyl. The nmr spectrum of the hydroxy acetate **17** in pyridine-d_5 shows the C-10 methyl at δ 1.01, whereas in $CDCl_3$ it appears at δ 1.09. Since a deshielding effect did not occur, a 1,3-diaxial relationship for the C-4 hydroxyl group and the C-10 methyl does not exist, and the hydroxyl group must be equatorial.

The second minor product from the hydrolysis of epitulipinolide (**2**) analyzed for $C_{15}H_{20}O_3$ and showed from the ir spectrum the presence of a hydroxyl, a γ-lactone, and ethylenic groups. The structure **22** bearing a cadinene-ring system was proposed for the compound from nmr studies, and its formation from the hydroxy acid of eupatolide was rationalized as occurring during the lactone closing phase of the reaction (Scheme I). A pair of doublets typical of the exocyclic methyl-

SCHEME I



22

ene protons was present at δ 6.23 and 5.75 and found, by double-irradiation experiments, to be coupled (J = 3.2 and 3.0 Hz, respectively) to a proton at δ 3.22 which was assigned position 7. Splitting of the H_7 pattern was caused by two additional couplings of J = 4.8 (H_6)

(9) P. V. Demarco, E. Farkas, D. Doddrell, B. L. Mylari, and E. Wenkert, *J. Amer. Chem. Soc.*, **90**, 5480 (1968). Methyl groups 1,3 diaxial to a hydroxyl show a deshielding of 0.2–0.4 ppm in pyridine relative to chloroform.

and 7.2 Hz (H_8), values in accord with two equatorial–axial interactions, if the twist of the relevant dihedral angle by the lactone ring is taken into account. The olefinic methyl at δ 1.75 and the one-proton (H_5) peak ($W_{1/2}$ = 5 Hz) at δ 5.35 are related as shown, since irradiation of one causes marked sharpening of the other. The C-10 tertiary hydroxyl group (D_2O exchangeable sharp singlet at δ 1.93) was placed equatorial because a large deshielding of the H_8 proton was not observed in the nmr spectrum taken in pyridine-d_5.[9] The uncommon cyclization of a germacranolide to a cadinene, as we have observed, has been previously reported for the simple sesquiterpene hydrocarbon, bicyclogermacrene,[10] but not to our knowledge for a germacranolide lactone.

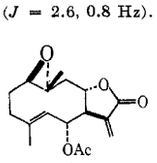
It is of note that none of the products from hydrolysis of epitulipinotide (**2**) is the C-8 cis lactone, isoeupatolide. On the other hand, the hydrolysis of tulipinolide (**1**) does yield deacetylisotulipinolide (**23**), a C-8 trans lactone. An unusual feature of the nmr spectrum of this compound is the presence of a broad singlet ($W_{1/2}$ = 6 Hz) at δ 6.19 for the H_{13} proton situated trans to the lactone and is probably due to long-range coupling to H_6 or H_8, or both. The other H_{13} proton appears at δ 6.38 as a pair of doublets, J = 2.8 and 1 Hz. The latter value is the characteristic geminal coupling commonly noted for the C-6 trans α,β'-unsaturated γ-lactones,[11] which appears to hold in this case for an α-C-8 trans α,β'-unsaturated γ-lactone with C-6 α-OH. Acetylation of **23** to isotulipinolide (**24**) restored the exocyclic methylene proton pattern to a pair of double doublets.[12] After this study was completed, a communication appeared on the isolation of a substance named laurenobiolide from *Laurus nobilis* L.[13] which has the same constitution as isotulipinolide (**24**). A comparison of the nmr and ir spectra and tlc mobility of the two substances showed them to be identical.

Deacetylisotulipinolide (**23**) was obtained in low yields in another way: from eupatolide methanesulfonate (**25**) by inversion of the C-8 center on treatment with potassium hydroxide. The other product of the reaction was eupatolide. Attempts to obtain deacetyltulipinolide (**26**) by sodium borohydride reduction of dehydroeupatolide[2] (**27**) were unsuccessful; the products were eupatolide, 11,13-dihydroeupatolide, and what was tentatively identified from the nmr spectrum as 11,13-dihydrodehydroeupatolide.

(10) K. Nishimura, N. Shinoda, and Y. Hirose, *Tetrahedron Lett.*, 3097 (1969).

(11) H. Yoshioka, T. J. Mabry, M. A. Irwin, T. A. Geissman, and Z. Samek, *Tetrahedron*, **27**, 3317 (1971).

(12) Although geminal coupling does not appear to be common for C-6 α-oxygenated, C-8 α-germacranolides (ref 11), it does occur in some cases, *e.g.*, pyrethrosin (i), where the exocyclic methylenes are found at δ_{CDCl_3} 6.37 (J = 3.0, 0.8 Hz) and 5.93 (J = 2.6, 0.8 Hz).



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X-Ray studies on pyrethrosin were reported by E. J. Gabe, S. Neidle, D. Rogers, and C. E. Nordman, *Chem. Commun.*, 559 (1971), and chemical studies by S. Iriuchijima and S. Tamura, *Agr. Biol. Chem.* (*Tokyo*), **34**, 204 (1970); R. W. Doskotch, F. S. El-Feraly and C. D. Hufford, *Can. J. Chem.*, **49**, 2103 (1971), and references cited therein.

(13) H. Tada and K. Takeda, *Chem. Commun.*, 1391 (1971). We thank Dr. Tada for the copies of the ir and nmr spectra and the sample of laurenobiolide.

Experimental Section[14]

Dihydrotulipinolide (**3**).—An 80-mg sample of tulipinolide (**1**) suspended in 4 ml of absolute EtOH was treated with 20 mg of $NaBH_4$. When the suspension cleared (10 min) the acidified (10% HOAc) solution was evaporated at reduced pressure and the residue was dissolved in chloroform. The chloroform solution was washed with H_2O and dried (Na_2SO_4), and the crystalline residue remaining after solvent removal crystallized from *n*-hexane to give **3** as needles (57 mg): mp 120–122°; $[\alpha]^{22}D$ +45° (*c* 0.056, MeOH); CD (*c* 0.056, MeOH), 22°, $[\theta]_{220}$ + 141,000; uv end absorption 210 nm (log ϵ 3.93); ir 1770 (γ-lactone), 1740 (acetate), 1660 (olefin) and 1240 cm^{-1} (CO stretching); mass spectrum *m/e* (rel intensity) M^+ 292 (0.4), 250 (1.2), 232 (33), 121 (68), 93 (68), and 43 (100).[15]

Anal. Calcd for $C_{17}H_{24}O_4$: C, 69.83; H, 8.27. Found: C, 69.61; H, 8.16.

The physical properties of **3** were very close to those of acetylbalchanolide [lit.[4] mp 125°, $[\alpha]^{20}D$ +128.1° (*c* 3.38, $CHCl_3$)], and the ir spectra were the same.[5]

Dihydroepitulipinolide (**4**).—A 100-mg sample of epitulipinolide (**2**) in 5 ml of absolute EtOH was treated with 30 mg of $NaBH_4$. After 15 min the acidified (10% HOAc) solution was evaporated at reduced pressure to remove the EtOH; the residue was taken up in $CHCl_3$, washed with H_2O, dried (Na_2SO_4), and evaporated to dryness. The colorless residue was chromatographed on 7 g of silica gel G to give an oil (85 mg) that crystallized from diethyl ether–pentane as needles (59 mg): mp 102–103°; $[\alpha]^{22}D$ +135° (*c* 0.048, MeOH); CD (*c* 0.048, MeOH), 22°, $[\theta]_{220}$ + 162,000; uv end absorption 220 nm (log ϵ 3.80); ir 1770, 1740, 1670, 1240 and 965 cm^{-1}; R_f 0.5 on tlc (silica gel G) with $CHCl_3$–Et_2O (1:1); mass spectrum *m/e* (rel intensity) M^+ 292 (0.5), 250 (0.9), 232 (36), 121 (42), 93 (46) and 43 (100).

Anal. Calcd for $C_{17}H_{24}O_4$: C, 69.83; H, 8.27. Found: C, 69.62; H, 8.30.

Catalytic Hydrogenation of Epitulipinolide (**2**).—A sample (100 mg) of **2** dissolved in 20 ml of absolute EtOH was reduced over 20 mg of 5% Pd/C presaturated with hydrogen at ambient temperature and atmospheric pressure. Two moles of hydrogen was rapidly absorbed and uptake ceased. The residue, after removal of catalyst and solvent, crystallized (33 mg) from ether: mp 147–148°; $[\alpha]^{22}D$ −218° (*c* 0.070, MeOH); CD (*c* 0.070, MeOH) 22°, $[\theta]_{225}$ +1030; ir 1770 (γ-lactone), 1735 (acetate), and 1235 cm^{-1} (C–O stretching); mass spectrum *m/e* (rel intensity) M^+ 294 (2), 252 (11), 234 (16), 161 (38), and 43 (100).

Anal. Calcd for $C_{17}H_{26}O_4$: C, 69.36 H, 8.90. Found: C, 69.29 H, 8.70.

Structure **5** is proposed on the basis of the physical data, in particular the nmr spectrum.

Cyclization of Dihydroepitulipinolide (**4**).—A sample (70 mg) of **4** in 4 ml of $CHCl_3$ was treated with 0.2 ml of $SOCl_2$ for 30 min at room temperature. The residue remaining, after evaporation of the reaction mixture, was chromatographed on 5 g of silica gel G containing 10% $AgNO_3$ with $CHCl_3$–Et_2O (5:1) as eluting solvent. Fractions (4 ml) were collected. The α-cyclo isomer **9** emerged first and was crystallized from *n*-hexane to give colorless plates (15 mg): mp 102–104°; $[\alpha]^{22}D$ −36° (*c* 0.19, MeOH); ir 1775, 1740, and 1245 cm^{-1}; mass spectrum *m/e* (rel intensity) M^+ 292 (35), 232 (58), 217 (81), 108 (77), and 43 (100).

(14) Melting points were taken in capillaries on a Thomas-Hoover apparatus or on a Fisher-Johns hot stage, and are uncorrected. Elemental analyses were by Dr. Alfred Bernhardt, Germany, or the Scandinavian Microanalytical Laboratory, Denmark. Infrared spectra were taken in $CHCl_3$ or in KBr pellets on a Perkin-Elmer Model 237 or 257 spectrophotometer and ultraviolet spectra were obtained in CH_3OH on a Cary Model 15 spectrophotometer. The nmr spectra were measured in $CDCl_3$ or as stated otherwise on a Varian A-60A or T-60 instrument with $(CH_3)_4Si$ as internal standard, and chemical shifts are reported in δ (parts per million) units. The ORD, CD, and optical rotation values were determined on a Jasco ORD/UV-5 spectropolarimeter with CD attachment. Mass spectra were obtained on an AEI MS-9 double focusing instrument and samples were introduced *via* the direct inlet probe. Thin layer chromatography (tlc) was performed on silica gel G (Merck) with detection by iodine vapor or spraying with 0.3% $KMnO_4$ solution. Plates incorporating $AgNO_3$ were poured as a slurry with the per cent (w/w) of complexing agent indicated. Columns poured with such adsorbents were made from the powdered (through 100 mesh), dried (110°) slurries prepared for the plates and continuously protected from light.

(15) For the mass spectral data we are grateful to Dr. R. L. Foltz of Battelle Memorial Institute, which was made possible by the National Institutes of Health Contract No. NIH-71-2483, and to Mr. R. Weisenberger of our Chemistry Department for results from their instrument.

Dihydro-β-cycloepitulipinolide (**7**) was eluted next and crystallized from *n*-pentane as needles (12 mg), mp 84–85°, and showed identical ir and nmr spectra with a sample of the same substance produced by another route.[2]

Cyclization of Dihydrotulipinolide (3).—A 77-mg sample of **3** was cyclized and the products were separated in a manner given for dihydroepitulipinolide (**4**). Dihydro-α-cyclotulipinolide (**8**, 11 mg) was crystallized from *n*-hexane: mp 95–97°; $[\alpha]^{22}D$ +552° (*c* 0.038, MeOH); ir 1775, 1740, and 1240 cm^{-1}; mass spectrum *m/e* (rel intensity) M^+ 292 (8), 232 (100), 217 (39), 136 (50), and 43 (49).

Dihydro-β-cyclotulipinolide (**6**, 17 mg), mp 139–141°, crystallized from isopropyl ether–*n*-hexane and was identical (melting point, ir, and nmr) with the same compound produced by a different route.[2]

Dihydro-γ-cycloepitulipinolide (10).—γ-Cyclotulipinolide[2] (46 mg) dissolved in 2 ml of absolute EtOH was treated with 12 mg of $NaBH_4$ for 10 min at room temperature. After acidification with 10% HOAc and evaporation of solvent, the $CHCl_3$ solution of the residue was washed with water and evaporated to dryness. The solid crystallized from EtOH–H_2O as fine white needles (33 mg): mp 114–115°; $[\alpha]^{22}D$ −14° (*c* 0.036, MeOH); ir 1775, 1740, and 1245 cm^{-1}; mass spectrum *m/e* (rel intensity) M^+ 292 (28), 232 (68), 217 (94), 188 (86), and 173 (96).

Anal. Calcd for $C_{17}H_{24}O_4$: C, 69.83; H, 8.27. Found: C, 70.18; H, 8.35.

Dihydroeupatolide (Deacetyldihydroepitulipinolide) (12).—Eupatolide (deacetylepitulipinolide) (**11**, 60 mg) was suspended in 2 ml of absolute EtOH and treated, while stirring, with 20 mg of $NaBH_4$. After 10 min, the clear solution was worked up as reported for dihydrotulipinolide. Crystallization from benzene gave 42 mg of dihydroeupatolide (**12**) as colorless needles: mp 184–187°; $[\alpha]^{22}D$ +215° (*c* 0.070, MeOH); ir 3605, 3460, 1760, and 1665 cm^{-1}.

Anal. Calcd for $C_{15}H_{22}O_3$: C, 71.97 H, 8.86. Found: C, 72.24 H, 9.01.

Catalytic hydrogenation (Pd on C) of eupatolide gave the same dihydroeupatolide but in much lower yield.

The acetate of dihydroeupatolide made by Ac_2O and pyridine treatment was identical (melting point, ir, and nmr) with dihydroepitulipinolide (**4**).

Hydrolysis of Epitulipinolide (11) in Aqueous MeOH.—A 3-g sample of epitulipinolide dissolved in 120 ml of MeOH was stirred and treated with 480 ml (3 g) of aqueous KOH. After 2 days at room temperature, the acidified solution was evaporated to remove MeOH, saturated with NaCl and extracted with 3 × 300 ml of $CHCl_3$. The $CHCl_3$ solution was extracted with 1% $NaHCO_3$ and H_2O, dried (Na_2SO_4), and evaporated to dryness. The residue (2.6 g) gave from isopropyl ether 1.47 g of eupatolide,[2] mp 186–188°.

Chromatography of 470 mg of the mother liquor residue, which showed two spots (R_f 0.8 and 0.5) on tlc [silica gel G, EtOH—Et_2O (1:100)], on 21 g of silica gel G with 1% EtOH in Et_2O as solvent gave 123 mg of (11*R*)-13-methoxydihydroeupatolide (**13**), identical with the product obtained when epitulipinolide (**2**) was treated with $NaOCH_3$.[2] Later column fractions gave an oil (90 mg), which yielded from isopropyl ether–EtOH 46 mg of (11*S*)-13-methoxydihydroeupatolide (**15**): mp 98–99°; mass spectrum *m/e* (rel intensity) M^+ 280 (6), 262 (14), 235 (16), 217 (100), and 45 (36).

Cyclization of (11*R*)-13-Methoxydihydroeupatolide (13).—A 400-mg sample of **13** in 50 ml of $CHCl_3$ containing 0.2 ml of $SOCl_2$ was stirred for 30 min at room temperature. Evaporation of the solvent left a residue from which 240 mg of a crystalline mixture was deposited from isopropyl ether. Chromatography of the mixture on 14 g of silica gel G containing 5% $AgNO_3$ with Et_2O as eluent gave from the first eluted fraction (66 mg) (11*R*)-13-methoxydihydro-α-cycloeupatolide (44 mg, isopropyl ether–$CHCl_3$): mp 146–148°; mass spectrum *m/e* (rel intensity) M^+ 280 (25), 217 (50), and 45 (100); nmr δ 5.35 (br m, H_3) and 1.84 (br, C-4 Me). The second column fraction (135 mg) yielded from the same solvent system the β isomer **14** (111 mg): mp 173–174°; $[\alpha]^{22}D$ +94° (*c* 0.072, MeOH); ir 3610, 1770, and 1655 cm^{-1}.

Anal. Calcd for $C_{16}H_{24}O_4$: C, 68.54; H, 8.63. Found: C, 68.53; H, 8.65.

Hydrolysis of Epitulipinolide (2) in H_2O.—A 5-g sample of **2** was stirred in 1 l. of 0.28 *N* KOH for 48 hr at room temperature. After acidification to pH 3 with 1 *N* H_2SO_4, saturation with NaCl, and stirring for 2 hr, the solution was extracted with four 900-ml portions of $CHCl_3$. The combined $CHCl_3$ extract was washed with 5% $NaHCO_3$ and H_2O, dried (Na_2SO_4), and evaporated to dryness. The 4.2 g residue was crystallized from isopropyl ether to give 2.34 g of eupatolide (**11**).[2]

Chromatography of the mother liquor residue on 64 g of silica gel G with ether as eluent gave a fraction (211 mg) that was still a mixture (nmr). Elution with EtOAc gave a fraction (174 mg) that crystallized from $CHCl_3$–Et_2O to give the lactone diol **16** (100 mg): mp 196–197°; $[\alpha]^{22}D$ +15° (*c* 0.092, MeOH); ir 3580, 3400, 1760, and 1665 cm^{-1}.

Anal. Calcd for $C_{15}H_{22}O_4$: C, 67.64; H, 8.33. Found: C, 67.45; H, 8.29.

Rechromatography of the first-eluted fraction (211 mg) on 15 g of silica gel G impregnated with 10% $AgNO_3$ and elution with EtOAc gave a crystalline fraction that on recrystallization from Et_2O–benzene yielded the cadinene **22** (80 mg): mp 137–138°; $[\alpha]^{22}D$ +36° (*c* 0.078, MeOH); ir 3680, 3450, 1760 and 1660 cm^{-1}; mass spectrum *m/e* (rel intensity) M^+ 248 (2), 230 (8), 139 (70), 94 (100), and 95 (91).

Anal. Calcd for $C_{15}H_{20}O_3$: C, 72.55; H, 8.12. Found: C, 72.57 H, 7.95.

Acetylation of Lactone Diol 16.—Compound **16** (50 mg) was treated with 1 ml of pyridine and 0.4 ml of Ac_2O at 40° for 40 hr. Work-up of the reaction in the usual manner gave a residue that yielded 29 mg of the monoacetate **17** from EtOH–hexane: mp 133–134°; $[\alpha]^{22}D$ +46° (*c* 0.076, MeOH); ir 3590 (sharp), 1770, 1735 (sh, d), and 1670.

Anal. Calcd for $C_{17}H_{24}O_5$: C, 66.21; H, 7.85. Found: C, 65.84; H, 7.84.

Dehydration of Acetate 17.—The monoacetate **17** (12 mg) was dissolved in 1 ml of dry pyridine, cooled at 5°, and treated with 0.1 ml of $SOCl_2$ for 10 min. The solution was diluted with water, $CHCl_3$ was added, and the organic layer was washed with dilute acid, base, and H_2O. The crystalline residue (13 mg) from the $CHCl_3$ solution gave 4 mg of β-cycloisoepitulipinolide (**18**) from benzene–pentane as colorless needles, mp 193–194°, identical (mixture melting point, ir, and nmr) with a sample of the same compound prepared from β-cycloepitulipinolide (**21**) or β-cycloeupatolide (**20**).

Hydrolysis of β-Cycloepitulipinolide (20).—A 60-mg sample of β-cycloepitulipinolide (**21**)[2] was stirred in 10 ml of 0.33 *N* KOH for 25 hr at room temperature. The clear solution was acidified (1 *N* HCl) and extracted with 3 × 10 ml of $CHCl_3$. The extract was washed with 1% $NaHCO_3$ and H_2O, dried (Na_2SO_4), and evaporated to leave a residue (14 mg) of deacetyl-β-cycloisoepitulipinolide (**19**). Acetylation with Ac_2O–pyridine at room temperature for 50 hr gave β-cycloisoepitulipinolide (**18**), which was crystallized from benzene–pentane to give 6 mg of product: mp 193–194°; $[\alpha]^{22}D$ +168° (*c* 0.086, MeOH); ir 1765, 1740, and 1650 cm^{-1}.

Anal. Calcd for $C_{17}H_{22}O_4$: C, 70.32; H, 7.64. Found: C, 70.49; H, 7.72.

From the bicarbonate extract after acidification was isolated a crystalline residue (45 mg) that on recrystallization from EtOH–pentane yielded 21 mg of hydroxy acid **28**: mp 174°; $[\alpha]^{22}D$ −49° (*c* 0.036, MeOH); ir (KBr) 3290, 2600 (bonded OH of COOH), 1740 (acetate), 1675 (unsaturated acid), 1650, and 1630 cm^{-1}; mass spectrum *m/e* (rel intensity) M^+ 308 (absent), 290 (4), 248 (8), 230 (23), 161 (24), and 43 (100).

Anal. Calcd for $C_{17}H_{24}O_5$: C, 66.21; H, 7.85. Found: C, 65.72; H, 7.80.

Isomerization of β-Cycloeupatolide (20).—A 317-mg sample of lactone **20** was stirred in 48 ml of aqueous KOH (640 mg) solution for 2 hr at room temperature. The clear solution was acidified (1 *N* HCl) and extracted with 3 × 10 ml of $CHCl_3$. The crystalline residue from $CHCl_3$ was dissolved in 3 ml of absolute EtOH, 1 drop of 1 *N* HCl was added, and the contents were left overnight at room temperature for relactonization. The residue on removal of EtOH was recrystallized twice from Et_2O–hexane to give 202 mg of β-cycloisoeupatolide (**19**): mp 147–148°; $[\alpha]^{22}D$ +143° (*c* 0.15, MeOH); ir 3590, 3500, 1765, 1670, and 1648 cm^{-1}.

Anal. Calcd for $C_{15}H_{20}O_3$: C, 72.55; H, 8.12. Found: C, 72.22; H, 8.17.

Acetylation of β-Cycloisoeupatolide (19).—A 50-mg sample of **19** in 2 ml of dry pyridine was treated with 0.2 ml of Ac_2O at 40° for 43 hr. Evaporation of the mixture at reduced pressure left a crystalline residue that gave 38 mg of β-cycloisoepitulipinolide (**18**) from benzene–hexane, that had identical properties [ir,

nmr, melting point, mixture melting point, and tlc mobility (R_f 0.36, silica gel G, ether)] as the previously prepared sample.

Isotulipinolide (24).—Tulipinolide (1, 175 mg) was suspended in 1 *N* KOH and warmed for 2 hr on a steam bath. The solution was quickly evaporated, treated with 3 ml HOAc, and evaporated, and the process repeated again. The residue was dissolved in $CHCl_3$, extracted with 5% $NaHCO_3$ and H_2O, dried (Na_2SO_4), and evaporated to leave a 50-mg residue. Chromatography of the residue over 4 g of silica gel G with $CHCl_3$–Et_2O (1:1) as eluting solvent gave 27 mg of an oil that was one spot on tlc and was formulated as desacetylisotulipinolide (**23**): $[\alpha]^{22}D$ +130° (*c* 0.054, MeOH); ir 3610, 3460, 1760, and 1660 cm^{-1}; mass spectrum *m/e* (rel intensity) M^+ 248.1417 (3.5) ($C_{15}H_{20}O_3$ calcd 248.1412), 230 (3), 108 (13), 84 (36), and 18 (100).

Acetylation of desacetylisotulipinolide (**23**, 27 mg) with Ac_2O–pyridine at room temperature for 20 hr gave a residue that after chromatography on silica gel G using $CHCl_3$–Et_2O (20:1) as eluting solvent gave 15 mg of an oil that was one spot on tlc and was formulated as isotulipinolide (**24**): $[\alpha]^{22}D$ +36 (*c* 0.056, MeOH), ir 1760, 1735, 1660, and 1250 cm^{-1}; mass spectrum *m/e* (rel intensity) M^+ 290.1507 (0.6) ($C_{17}H_{22}O_4$ calcd 290.1518), 230 (5), 107 (14), 84 (50), and 43 (100). A comparison (ir, nmr, and tlc) of this material with laurenobiolide[13] showed them to be the same.

Eupatolide Methanesulfonate (25).—A 500-mg sample of eupatolide (**11**) dissolved in 4 ml of pyridine and cooled in an ice bath was treated with 0.3 ml of CH_3SO_2Cl. After 17 hr the solution was diluted with H_2O and extracted with $CHCl_3$ and the $CHCl_3$ extract was washed with 1% HCl and H_2O. The $CHCl_3$-soluble residue was crystallized from EtOH–isopropyl ether to give 412 mg of eupatolide methanesulfonate (**25**): mp 112–113°; ir 1770, 1670, 1350, and 1175 cm^{-1}.

Anal. Calcd for $C_{16}H_{22}O_5S$: C, 58.88 H, 6.80 S, 9.81. Found: C, 58.57 H, 6.93 S, 9.68.

Treatment of Eupatolide Methanesulfonate (25) with KOH.—A 400-mg sample of **25** was stirred with 8 ml of EtOH, and 72 ml of 0.4 *N* KOH was added. After 24 hr at room temperature, the reaction solution was acidified with dilute HOAc, saturated with NaCl, and extracted with ether. The ether extract was washed with water, dried (Na_2SO_4), and evaporated to leave 243 mg of an oily residue. Chromatography of the oil was on 15 g of silica gel G with $CHCl_3$–Et_2O (1:1) as eluting agent. Early fractions gave 37 mg of deacetylisotulipinolide (**23**) identical (tlc, ir, and nmr) with the product of alkaline hydrolysis of tulipinolide (**1**). Later fractions contained eupatolide (**11**).

Reduction of Dehydroeupatolide (27).—To a solution of dehydroeupatolide[2] (100 mg) in 7 ml of *i*-PrOH at 40° was added 6 mg of $NaBH_4$. After 10 min the solution was acidified with dilute HOAc, diluted with H_2O, and extracted with ether. The ether-soluble residue (95 mg) showed three spots on tlc. Separation of these substances was accomplished on 5 g of silica gel G using $CHCl_3$–Et_2O (1:1) as solvent system. Two products were identified as eupatolide (**11**) and dihydroeupatolide (**12**), while the third from spectral evidence appeared to be 11,13-dihydrodehydroeupatolide, but deacetyltulipinolide (**26**) was not detected.

Registry No.—**1**, 24164-12-3; **2**, 24164-13-4; **3**, 35001-07-1; **4**, 35001-08-2; **5**, 35001-09-3; **6**, 24164-20-3; **7**, 24165-31-9; **8**, 35001-12-8; **9**, 35001-13-9; **10**, 35001-14-0; **12**, 35001-16-2; **13**, 35001-15-1; **14**, 35001-17-3; **15**, 35001-18-4; **16**, 35001-19-5; **17**, 35001-20-8; **18**, 35001-21-9; **19**, 35001-22-0; **22**, 35001-23-1; **23**, 35001-24-2; **24**, 35001-25-3; **25**, 35001-26-4; **28**, 35001-27-5.

1,4 Addition of Organometallic Reagents to α,β-Unsaturated Ketones in the Presence of (−)-Sparteine

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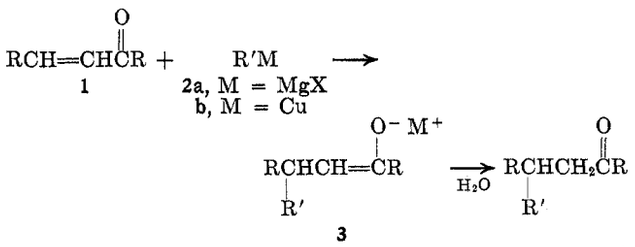
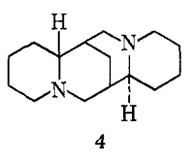
Received February 22, 1972

The reaction of 2-cyclohexenone, 3-penten-2-one, and 1,3-diphenyl-2-propen-1-one with a series of Grignard reagents has been studied in the presence of (−)-sparteine (**4**) and other additives. The resulting conjugate addition products possess an optical purity of 3–6% and represent the first examples of asymmetric 1,4 addition of achiral organometallic reagents to prochiral α,β-unsaturated ketones. Subsequent reactions of enolate anions initially produced by conjugate addition of the organometallic reagents are discussed. (−)-Sparteine is shown to reduce the reactivity of methylmagnesium iodide toward α,β-unsaturated ketones.

The ability of α,β-unsaturated ketones (**1**) to add Grignard reagents (**2a**)[1] and organocopper(I) compounds (**2b**)[2] in a 1,4 manner is well documented in



the literature. Recently, it has been shown that the course of this reaction can be influenced to some extent by solvent or the ligands attached to the organometallic reagent.[2b,c] In view of this, we have examined the reaction of some α,β-unsaturated ketones with Grignard reagents in the presence of (−)-sparteine (**4**). The



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results, indicated in Tables I and II, represent the first examples of asymmetric 1,4 addition of achiral organometallic reagents to prochiral α,β-unsaturated ketones.

The most apparent effect of an equimolar amount of (−)-sparteine (**4**) on an ether solution of methylmagnesium iodide is a drastic reduction of reactivity toward the enone substrates (Table I). Both 2-cyclohexenone and 1,3-diphenyl-2-propen-1-one were recovered unchanged after exposure to this reagent system for over 1 hr at room temperature. Enolization

(1) (a) M. S. Kharasch and O. Reinmuth, "Grignard Reactions of Nonmetallic Substances," Prentice-Hall, New York, N. Y., 1954, pp 196–234; (b) J. Munch-Petersen, *Bull. Soc. Chim. Fr.*, 471 (1966).

(2) (a) H. Gilman, R. G. Jones, and L. A. Woods, *J. Org. Chem.*, **17**, 1630 (1952); (b) H. O. House, W. L. Respess, and G. M. Whitesides, *ibid.*, **31**, 3128 (1966); (c) H. O. House and W. F. Fischer, *ibid.*, **33**, 949 (1968).