2.67 *M* solution of chromic acid in sulfuric acid.[16] The reaction mixture was stirred for 10 min. at room temperature, at the end of which time excess oxidizing agent was still present. Methanol was added until the solution turned green, the mixture was diluted with water, and the product isolated with ether. Evaporation of the ether gave 100 mg. of a colorless oil which crystallized upon trituration with petroleum ether. The crude product was recrystallized from ether–petroleum ether; yield 80 mg. (72%), m.p. 135–137°, $[\alpha]^{20}D$ −61° (*c* 1.11); $\nu_{max}^{CS_2}$ 1730, 1705 cm.$^{-1}$. In contrast to V, this 8-iso-dione crystallized poorly from methanol.

Anal. Calcd. for $C_{26}H_{42}O_2$ (386.58): C, 80.83; H, 10.97. Found: C, 80.76; H, 11.17.

Alkali Isomerization of 8-Iso-B-norcoprostane-3,6-dione (X).—In a nitrogen atmosphere, 2.0 g. of potassium hydroxide was added to a solution of 124 mg. (0.32 mmole) of X in 50 ml. of methanol and the solution was allowed to stand under a nitrogen atmosphere for 18 hr. The excess alkali was neutralized with hydrochloric acid and the product isolated in the usual way with ether. The ether was removed under reduced pressure and the residual yellow crystals chromatographed on Woelm neutral alumina (Act. III). There was obtained 95 mg. (77%) of crystalline dione (m.p. 90–95°) which was recrystallized from petroleum ether to yield B-norcoprostane-3,6-dione, m.p. 104–107°, $[\alpha]^{20}D$ −42° (*c* 1.41). The infrared spectra of this material and of an authentic sample were superimposable.

8-Iso-B-norcoprostane-6-one (XII).—A solution of 121 mg. (0.32 mmole) of B-norcoprostane-6-one[17] in 20 ml. of 5% potassium hydroxide in methanol was heated under reflux for 41 hr. The solution was kept under a nitrogen atmosphere for the entire period. The reaction solution was concentrated under reduced pressure and the oily residue dissolved in ether. The ethereal solution was washed with water, the solution dried, and the solvent removed to yield 114 mg. (94%) of a clear oil. The entire product was chromatographed on Woelm neutral alumina (Act. I) and 70 mg. of a clear oil was obtained by elution with petroleum ether–benzene (9:1 and 4:1). The material could not be obtained crystalline, but it possessed an infrared spectrum grossly different from that of the starting ketone. The material had $[\alpha]^{20}D$ −42.5° (*c* 1.40) as compared[17] to $[\alpha]D$ +34° for the starting material.

Anal. Calcd. for $C_{26}H_{44}O$ (372.61): C, 83.80; H, 11.90. Found: C, 83.35; H, 11.60.

B-Norcholestane-3β,6α-diol (XIV).—A solution of 0.5 g. (1.35 mmoles) of B-norcholesterol and 0.75 g. of boron trifluoride etherate in 20 ml. of dry ether was added dropwise, over a 1-hr. period, to a stirred solution of 0.15 g. of lithium aluminum hydride in 10 ml. of dry ether. The entire reaction was conducted at room temperature under a nitrogen atmosphere. The mixture was stirred for one additional hour, excess saturated magnesium sulfate solution added, the mixture filtered, and the ether evaporated. The residue was dissolved in tetrahydrofuran and stirred for 30 min. with an excess of 10% aqueous potassium hydroxide and 30% hydrogen peroxide. The solution was poured into water and the organic material extracted with ether. Upon removal of the ether the product crystallized, m.p. 178–183°. The product was recrystallized from methanol; yield 450 mg. (86%), m.p. 191–192°, $[\alpha]^{20}D$ +7.4° (*c* 0.89). Upon chromatography on paper, the compound showed one spot and the R_f was different from that shown by B-norcholestane-3β,6β-diol.

Anal. Calcd. for $C_{26}H_{46}O_2$ (390.63): C, 79.94; H, 11.87. Found: C, 79.66; H, 11.66.

B-Norcholestane-3,6-dione (XV).—To a stirred solution of 200 mg. (0.51 mmole) of B-norcholestane-3β,6α-diol in 25 ml. of purified acetone at −25°, under a nitrogen atmosphere, there was added 0.4 ml. of a 2.67 *M* solution of chromic acid in sulfuric acid.[16] After 20 min., excess methanol was added, the mixture was poured into water, and the mixture extracted with ether. The ethereal extract was evaporated and the residual yellow crystals (190 mg., m.p. 110–115°) were recrystallized from methanol; m.p. 138–140°, $[\alpha]^{20}D$ +129° (*c* 1.09); $\nu_{max}^{CHCl_3}$ 1730, 1715 cm.$^{-1}$.

Anal. Calcd. for $C_{26}H_{42}O_2$ (386.58): C, 80.83; H, 10.97. Found: C, 80.63; H, 10.89.

Alkali Isomerization of B-Norcholestane-3,6-dione (XV).—In a nitrogen atmosphere, 2.0 g. of potassium hydroxide was added to a solution of 125 mg. (0.32 mmole) of B-norcholestane-3,6-dione in 50 ml. of methanol and the solution allowed to stand under a nitrogen atmosphere for 18 hr. The solution was poured into dilute hydrochloric acid and the resulting mixture extracted with ether. The ether extract was evaporated and the yellow oily residue was dissolved in methanol and allowed to stand for 2 days in a refrigerator. During this time 17 mg. of $\Delta^{2,4,6}$-B-norcholestatriene-3,6-diol[18] crystallized and was removed. The methanol was removed from the filtrate, the residue dissolved in hexane, and the solution placed on a column of Woelm neutral alumina (Act. III). Elution with hexane–benzene (3:1) gave 17 mg. of an oil. Elution with hexane–benzene (1:1) yielded 42 mg. of an oil which crystallized as yellow needles, m.p. 100–103°. The product was recrystallized from methanol to yield colorless needles, m.p. 112–114°, $[\alpha]^{20}D$ −35° (*c* 0.9). The infrared spectrum was identical with that of B-norcoprostane-3,6-dione, and when the two materials were mixed there was no depression in the melting point.

(16) K. Bowden, I. M. Heilbron, E. R. H. Jones and B. C. L. Weedon, *J. Chem. Soc.*, 39 (1946); C. Djerassi, R. R. Engle and A. Bowers, *J. Org. Chem.*, **21**, 1547 (1956).

(17) T. Goto, *J. Am. Chem. Soc.*, **82**, 2005 (1960).

(18) W. G. Dauben, G. A. Boswell, Jr., and W. Templeton, *J. Org. Chem.*, **25**, 1853 (1960).

[CONTRIBUTION FROM THE NATIONAL INSTITUTE OF ARTHRITIS AND METABOLIC DISEASES, NATIONAL INSTITUTES OF HEALTH, BETHESDA 14, MD.]

The Absolute Configuration of Steviol and Isosteviol

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Steviol (I) was degraded to stevane A (XVII) which was identical with (−)-α-dihydrokaurene. A similar transformation was carried out on steviol (I) *via* 19-hydroxystevane B (XXIV) to stevane B (XVIII). Stevane B (XVIII), the C-16 epimer of stevane A (XVII), was found to be identical with the hydrocarbon derived from garryfoline; however, alcohol XXIV and the primary alcohol from garryfoline were different. These chemical interconversions with the phyllocladene-type diterpenes and the diterpene alkaloids of the Garrya, coupled with various optical rotatory dispersion and other physical measurements, have resulted in the establishment of the complete absolute configuration of the diterpene acids steviol (I) and isosteviol (II).

A series of recent communications[2] has described correlations of steviol (I) and isosteviol (II) with other diterpenes and diterpene alkaloids,[3] the results of which have enabled us to assign the absolute configuration of our diterpene acids as depicted in structures I and II.

Stevioside,[4] the principal constituent of *Stevia Rebaudiana* Bertoni, on enzymatic hydrolysis yields steviol (I),[5] while treatment of the glucoside with 48% hydrobromic acid gives isosteviol (II). Isosteviol (II) can also be obtained by acid treatment of steviol (I) *via* a Meerwein rearrangement.

(1) (a) Deceased, May 31, 1962; (b) Visiting Scientist, National Institutes of Health.

(2) (a) F. Dolder, H. Lichti, P. Quitt and E. Mosettig, *J. Am. Chem. Soc.*, **82**, 246 (1960); (b) E. Mosettig, P. Quitt, U. Beglinger, J. A. Waters, H. Vorbrueggen and C. Djerassi, *ibid.*, **83**, 3163 (1961); (c) C. Djerassi, P. Quitt, E. Mosettig, R. C. Cambie, P. S. Rutledge and L. H. Briggs, *ibid.*, **83**, 3720 (1961).

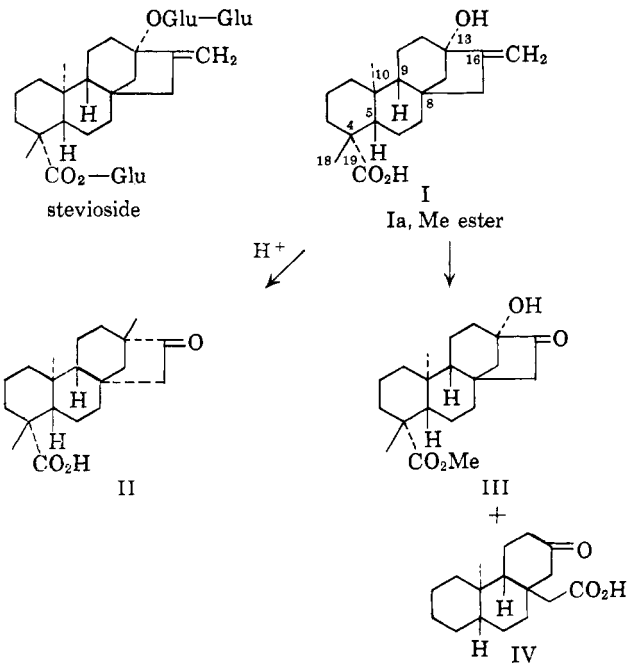
(3) For a complete discussion of the correlations existing between the diterpene alkaloids (Garrya and Atisine) and the diterpenes of the phyllocladene-type (*e.g.*, kaurene and steviol–isosteviol) see H. Vorbrueggen and C. Djerassi, *ibid.*, **84**, 2990 (1962).

(4) H. B. Wood, Jr., R. Allerton, H. W. Diehl and H. G. Fletcher, Jr., *J. Org. Chem.*, **20**, 875 (1955); E. Vis and H. G. Fletcher, *J. Am. Chem. Soc.*, **78**, 4709 (1956).

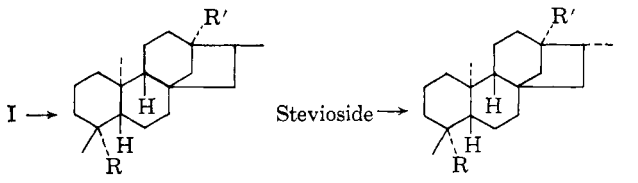
(5) For the initial work on the algucones steviol and isosteviol, see E. Mosettig and W. R. Nes, *J. Org. Chem.*, **20**, 884 (1955).

The stereochemistry of the C/D ring juncture (C-8 and C-13) was initially investigated.[2a] The optical rotatory dispersion curves of isosteviol (II)[6] and gibberic acid[7] were found to be coincident. The report by Stork and Newmann[8] of the absolute configuration of gibberic acid allowed us to assign the configuration at positions 8 and 13 of isosteviol (II) as indicated. Though the conversion of I → II previously mentioned would enable one to formulate the C/D ring stereochemistry of steviol (I), additional evidence for I was obtained. Ozonization of steviol, methyl ester (Ia) gave ketol III plus ketoacid IV. The rotatory dispersion curves of III (positive Cotton effect) and ketonorallogibberic acid[9] were nearly superimposable, thus providing the absolute configuration of the C-8 and C-13 positions of steviol (I).

We then decided to convert steviol (I) to the two C-16 epimeric hydrocarbons stevane A (XVII) and stevane B (XVIII)[2a] for comparison with (−)-α-dihydrokaurene,[10] the principal hydrogenation product of kaurene.[11] (−)-Kaurene, at this time, was formulated as having an abnormal A/B *trans* ring fusion and a C/D ring juncture as shown for steviol (I). Hydrogenation of



steviol (I) with palladium-on-charcoal under a pressure of 40° p.s.i. gave dihydrosteviol A (V) in 61% yield. The C-16 epimer of V, designated as dihydrosteviol B (VI), was obtained by hydrogenation of stevioside with platinum oxide in aqueous ethanol. The methyl esters Va and VIa of dihydrosteviol A and B were reduced to dihydroxystevane A (VII) and dihydroxystevane B (VIII) with lithium aluminum hydride in refluxing tetrahydrofuran. Oxidation of VII and VIII with chromic acid–pyridine gave the corresponding aldehydes IX and X, which were immediately converted to the ethylene thioacetals XI and XII because of their autoöxidation to the respective acids. Raney nickel desulfurization in refluxing ethanol gave 13-hydroxystevane A (XIII) and 13-hydroxystevane B (XIV). Replacement of the tertiary hydroxyl groups of XIII and XIV by bromine using freshly prepared phosphorus pentabromide in ether at room temperature gave XV and XVI, respectively. Hydrogenolysis of the bromo compounds with Raney nickel at 100° (1200 p.s.i.) gave the corresponding C-16 epimeric hydrocarbons stevane A (XVII) and stevane B (XVIII). Stevane A, m.p. 87.5–88.5° ($[\alpha]^{20}D$ −31.9°), was identical with (−)-α-dihydrokaurene,[11] m.p. 86–87° ($[\alpha]^{21}D$ −32°), by comparison of the two by mixed melting point, infrared spectra and X-ray powder patterns.[12] Apparently stevane B is identical to (−)-β-dihydrokaurene, a by-product in the hydrogenation of (−) kaurene.[13] The *trans* stereochemistry at positions 5 and 10 was thus confirmed, as well as supporting evidence concerning the nature of the C/D ring juncture.



	R	R′		R	R′
V	COOH	OH	VI	COOH	OH
Va	COOMe	OH	VIa	COOMe	OH
VII	CH_2OH	OH	VIII	CH_2OH	OH
IX	CHO	OH	X	CHO	OH
XI	H S, C—S	OH	XII	H S, C—S	OH
XIII	CH_3	OH	XIV	CH_3	OH
XV	CH_3	Br	XVI	CH_3	Br
XVII	CH_3	H	XVIII	CH_3	H
XIX	COOMe	Br	XX	COOMe	Br
XXI	COOMe	H	XXII	COOMe	H
XXIII	CH_2OH	H	XXIV	CH_2OH	H
XXV	CHO	H	XXVI	CHO	H
XXVII	H S, C—S	H	XXVIII	H S, C—S	H

At this point, the position of the carboxyl group (C-4 or C-10) and the stereochemistry at C-9 were the two remaining problems concerning the absolute configuration of I and II. The report of Vorbrueggen and Djerassi[14] on the conversion of garryfoline *via* a primary alcohol to a saturated hydrocarbon made it desirable to again convert steviol (I) to stevane A (XVII) and stevioside to stevane B (XVIII). A new pathway was chosen so as to proceed through the primary alcohols XXIII and XXIV. The alcohols XXIII and XXIV were needed for comparison with the garryfoline alcohol in which the $-CH_2OH$ was known to be C-10. The newly synthesized sample of stevane B (XVIII) was identical to that of the garryfoline hydrocarbon[14] by mixed melting point and gas chromatographic retention times. Our hydroxystevane B (XXIV), m.p. 142–144° $[\alpha]^{20}D$ −62°, was entirely different from the alcohol[14] derived from garryfoline, m.p. 89–90° $[\alpha]D$ −47°. This comparison thus enabled us to place the carboxyl group of steviol (I) and isosteviol (II) at C-4 rather than C-10. A recent paper concerning the anodic decarboxylation[16] of isostevic acid[5] gave additional evidence of the C-4 carboxyl position.

(6) C. Djerassi, R. Riniker and B. Riniker, *J. Am. Chem. Soc.*, **78**, 6362 (1956).

(7) B. E. Cross, J. F. Gove, J. MacMillan and T. B. C. Mulholland, *J. Chem. Soc.*, 2520 (1958).

(8) G. Stork and H. Newmann, *J. Am. Chem. Soc.*, **81**, 3168 (1959).

(9) T. B. C. Mulholland, *J. Chem. Soc.*, 2693 (1958).

(10) J. Simonsen and D. H. R. Barton, "The Terpenes," Univ. Press, Cambridge, 1952, Vol. III, p. 339.

(11) L. H. Briggs, B. F. Cain, R. C. Cambie and B. R. Davis, *Tetrahedron Letters*, **No. 24**, 18 (1960).

(12) We wish to thank Professor Briggs for these comparisons.

(13) Private information by Professor Briggs.

(14) H. Vorbrueggen and C. Djerassi, *Tetrahedron Letters*, **No. 3**, 119 (1961).

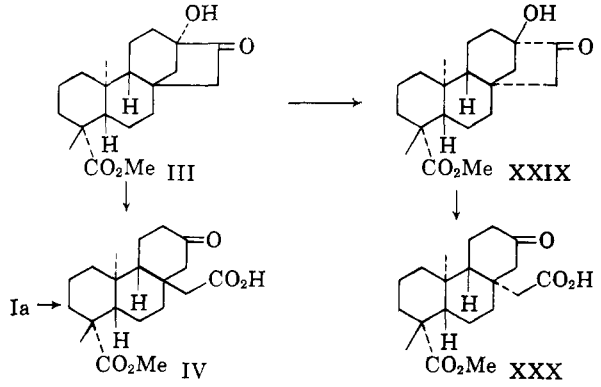
(15) Kindly performed by V. P. Arya; *cf.* P. C. Sommer, V. P. Arya and W. Simon, *ibid.*, **No. 20**, 18 (1960).

(16) J. A. Waters, E. D. Becker and E. Mosettig, *J. Org. Chem.*, **27**, 4689 (1962).

The mass spectrum[17] of stevane A (XVII) (mol. wt. 274) showed an intense peak at mass 189, which can be attributed to the loss of ring A (C_1–C_4 plus the *gem*-dimethyls). The same peak of mass 189 was found in the spectrum of stevic acid A, methyl ester (XXI), (mol. wt. 318). This implies that the ester group in this molecule is lost in this fragmentation and therefore attached to C-4. If the carboxyl were at C-10, one would expect to find a peak due to the loss of 85 mass units as was the case in the stevane A (XVII) spectrum.

Confirmation of a C-4 carboxyl group in the axial configuration was provided by pK^*_{MCS} measurements[15] of five steviol and isosteviol derivatives, ranging from 8.52–8.68 in contrast to the pK^*_{MCS} of the garryfoline carboxylic acid[14] (–COOH at C-10) of 9.49. The pK^*_{MCS} constants of steviol and derivatives are comparable to that of deoxypodocarpic acid (pK^*_{MCS} 8.45), which has an axial carboxyl group at C-4 and an A/B *trans* ring juncture.

The remaining stereochemical feature of C-9 was elucidated as follows. Treatment of norketol III with *n*-butyllithium or potassium *t*-butoxide gave the isonorketol XXIX *via* a D-homosteroid type rearrangement. Periodate cleavage of the norketol III gave the corresponding secoacid IV (also obtained by osmium tetroxide–periodate cleavage of Ia), while similar oxidation of the



isonorketol XXIX gave the isosecoacid XXX. The optical rotatory dispersion curves of III and IV (positive Cotton effects) and XXIX and XXX (negative Cotton effects), discussed in detail in the earlier communication,[2c] allowed us to assign the 9β-H configuration to steviol (I) and isosteviol (II). The establishment of the absolute configuration of steviol and isosteviol as depicted in formulas I and II, respectively, is therefore complete.

Experimental[18]

Isosteviol (II).—This represents an improved method to the one described previously.[5] A solution of 804 mg. of stevioside[4] in 15 ml. of 48% aqueous hydrobromic acid was kept at room temperature for 19 hr. Crystallization began immediately after solution was obtained. The crystals were removed by suction filtration and washed repeatedly with water. The yield of the dry product was 276 mg. (87%), m.p. 228–230°. Recrystallization from ether–hexane gave 226 mg. (71%) of white crystalline product, m.p. 226–228°, identical with authentic isosteviol (infrared, mixture m.p.).

Ozonization of Steviol, Methyl Ester (Ia).—A solution of 511 mg. of methyl ester Ia[5] in 40 ml. of methylene chloride was cooled to −80° and treated with a stream of oxygen containing 0.98% ozone until one equivalent of ozone was consumed. After warming to room temperature the clear, colorless solution was steam distilled. The volatile material was condensed in a gas trap containing a solution of 430 mg. of dimedone in 4 ml. ethanol. Evaporation of solvent and two recrystallizations from dilute ethanol gave 187 mg. (42%) of needles of the dimedone–formaldehyde derivative, m.p. 192–193.5°.

The non-volatile aqueous phase was extracted with ether. The ethereal extracts were washed repeatedly with 2 *N* sodium carbonate. Acidification of the aqueous carbonate washings and then extraction with ether gave 320 mg. of crude acid IV. Crystallization from chloroform–pentane gave 248 mg. (46%) of colorless prisms, m.p. 175–180°. Two additional recrystallizations raised the m.p. of IV to 177–181°, $[\alpha]^{20}D$ −68.4 ± 0.7° (*c* 1.04).

Anal. Calcd. for $C_{20}H_{30}O_5$: C, 68.54; H, 8.63. Found: C, 68.39; H, 8.95.

The neutral non-volatile product (ether extract) yielded 245 mg. of crude III which on crystallization from chloroform–ether–petroleum ether (30–60°) gave 90 mg. (17%) of colorless prisms, m.p. 199–224°. Recrystallization from methylene chloride–hexane gave pure III, m.p. 224–227°, $[\alpha]^{20}D$ −102.5 ± 0.5° (*c* 0.77).

Anal. Calcd. for $C_{20}H_{34}O_4$: C, 71.82; H, 9.04. Found: C, 71.75; H, 9.26.

Oxidation of Steviol, Methyl Ester (Ia) with Osmium Tetroxide and Sodium Periodate.—A solution of 332 mg. of Ia in 10 ml. of dioxane and 2 ml. of water was stirred with 10 mg. of osmium tetroxide at room temperature. After 15 min. the osmium tetroxide was completely dissolved. Then 1.07 g. of sodium periodate was added in portions over 20 min. After 3 hr. of stirring the mixture was colorless and a precipitate of sodium iodate had formed. The mixture was taken up in ether and water. The ether phase was washed with water, saturated with hydrogen sulfide, and filtered. After evaporation of the filtrate, the residue was taken up in ether and extracted with 2 *N* sodium carbonate. The carbonate extract was acidified to yield 197 mg. (56%) of crystalline acid IV, m.p. 172–178°. Repeated recrystallization from methylene chloride–hexane raised the m.p. to 178–181°. A mixture m.p. with secoacid IV obtained from the ozonization reaction showed no depression.

The methyl ester of IV was obtained using excess diazomethane. Recrystallization from hexane gave rectangular prisms, m.p. 114–115°, $[\alpha]^{20}D$ −49.0 ± 1.0° (*c* 1.00).

Anal. Calcd. for $C_{21}H_{32}O_5$: C, 69.20; H, 8.85. Found: C, 68.91; H, 8.82.

Periodate Oxidation of Ketol III.—A solution of 167 mg. of III in 7 ml. of dioxane and 1.5 ml. of water was stirred with 214 mg. of sodium periodate for 18 hr. at room temperature. The mixture was worked up in the usual manner to give 97 mg. of prisms of IV, m.p. 177–181°, from methylene chloride–hexane (no mixture melting point depression with secoacid IV obtained by ozonization).

Treatment of Ketol III with *n*-Butyllithium.—To a solution of 334 mg. of ketol III in 20 ml. of tetrahydrofuran was added 3.2 ml. of 0.6 *N* *n*-butyllithium in ether. The mixture was refluxed under nitrogen for 3 hr. The solvent was evaporated and the residue taken up in ether. The ethereal extract was washed with 2 *N* sodium carbonate, water and then dried. Evaporation of the ether under reduced pressure gave 281 mg. of crude, neutral product. Crystallization from methylene chloride–hexane gave 182 mg. of isoketol XXIX, m.p. 197–199°, $[\alpha]^{20}D$ −62.5 ± 1.6° (*c* 1.03).

Anal. Calcd. for $C_{20}H_{30}O_4$: C, 71.82; H, 9.04. Found: C, 71.76; H, 9.42.

Treatment of Ketol III with Potassium *t*-Butoxide.—To 188 mg. of ketol III was added 10 ml. of 0.87 *N* potassium *t*-butoxide in *t*-butyl alcohol and the mixture refluxed for 13 hr. under nitrogen and then diluted with ether. The ethereal extract was washed with 2 *N* sodium carbonate, water and then dried. Evaporation of solvent under reduced pressure gave only 46 mg. of neutral product, from which only 25 mg. of crystalline isoketol XXIX could be obtained, m.p. 194–196°, from methylene chloride–hexane.

Periodate Oxidation of Isoketol XXIX.—A solution of 150 mg. of XXIX in 7 ml. of dioxane and 1.3 ml. of water was stirred with 214 mg. of sodium periodate for 14 hr. at room temperature. Workup by the usual procedure gave 95 mg. (58%) of product, m.p. 193–196° with trans-crystallization at 150° (from methylene chloride–hexane). Two additional recrystallizations from carbon tetrachloride yielded XXX as prisms which changed to irregular crystals at 141° and melted at 198–200°, $[\alpha]^{20}D$ −46.4 ± 0.9° (*c* 0.93).

Anal. Calcd. for $C_{20}H_{30}O_5$: C, 68.54; H, 8.63. Found: C, 68.60; H, 8.73.

Dihydrosteviol A (V).—To a solution of 794 mg. of steviol (I) in 80 ml. of dioxane and 30 ml. of ethanol was added 2.5 g. of palladium-on-charcoal (10%). The mixture was hydrogenated at 40 p.s.i. at room temperature for 14 hr. using the Parr appara-

(17) We are greatly indebted to Professor K. Biemann for the mass spectra and interpretation of the data obtained.

(18) Analyses and rotations were performed by the Analytical Services Unit, NIAMD, under the direction of Mr. H. G. McCann. All rotations were taken in chloroform. Infrared spectra were obtained on a Perkin-Elmer model 21 spectrophotometer. Vapor phase chromatograms were obtained on a Barber-Coleman model 15 gas chromatograph, using a 9-ft. column (4 mm. i.d.) of 1/4% SE-30 on Gaschrom P (110–120 mesh) at 210° and 14 p.s.i.

tus. The catalyst was then removed by filtration and evaporation of the solvent gave 758 mg. of product which was recrystallized from ethyl acetate to yield 492 mg. (61%) of long, white prisms, m.p. 206–208°. Two additional recrystallizations raised the m.p. to 210–212°, $[\alpha]^{20}D$ −41.5 ± 1.6° (*c* 0.51).

Anal. Calcd. for $C_{20}H_{32}O_3$: C, 74.95; H, 10.07. Found: C, 74.92; H, 10.22.

Dihydrosteviol A, Methyl Ester (Va).—To a stirred solution of 441 mg. of dihydrosteviol A in 8 ml. of methanol was added an excess of diazomethane in ether over a period of 20 min. Stirring was continued for 2 hr. Acetic acid was added to decompose the excess diazomethane and the solvents were removed under vacuum to yield a thick oil. Two recrystallizations from dilute methanol gave 323 mg. (70%) of white needles, m.p. 116–119°, with trans-crystallization at about 90°; $[\alpha]^{20}D$ −69.7° (*c* 0.78).

Anal. Calcd. for $C_{21}H_{34}O_3$: C, 75.41; H, 10.25. Found: C, 75.20; H, 10.55.

Dihydrosteviol B, Methyl Ester (VIa).—A suspension of 597 mg. of dihydrosteviol B in ether was esterified with excess diazomethane for 2 hr. to give 402 mg. (67%) of product, m.p. 130–132°, from dilute methanol. Recrystallization from hexane raised the m.p. to 132–134°, $[\alpha]^{20}D$ −100.5 ± 2.0° (*c* 1.0).

Anal. Calcd. for $C_{21}H_{34}O_3$: C, 75.41; H, 10.25. Found: C, 75.56; H, 10.50.

13,19-Dihydroxystevane A (VII).—To a solution of 290 mg. of ester Va in 15 ml. of tetrahydrofuran was added 84 mg. of lithium aluminum hydride. The stirred mixture was refluxed for 6 hr. On cooling, 2 *N* HCl was added and the solvent evaporated under reduced pressure. The residue was taken up in chloroform and washed with 2 *N* HCl, 2 *N* sodium carbonate and then water. The chloroform extract was dried over sodium sulfate and then evaporated. The residue was recrystallized from chloroform–petroleum ether (30–60°) to yield 236 mg. of product, m.p. 215–216°, $[\alpha]^{20}D$ −40.4 ± 2.0° (*c* 0.48).

Anal. Calcd. for $C_{20}H_{34}O_2$: C, 78.38; H, 11.18. Found: C, 78.31; H, 11.00.

13,19-Dihydroxystevane B (VIII) was prepared in a manner similar to that described for VII. From 2.11 g. of VI was obtained 1.81 g. crude white solid, m.p. 185–187°. Crystallization from chloroform–petroleum ether (30–60°) gave prisms, m.p. 187–188.5°, $[\alpha]^{20}D$ −65.6 ± 1.4° (*c* 0.83).

Anal. Calcd. for $C_{20}H_{34}O_2$: C, 78.38; H, 11.18. Found: C, 78.24; H, 11.37.

13-Hydroxy-19-stevanal A (IX).—To a solution of 963 mg. of dihydroxystevane A (VII) was added an ice-cold suspension of 1 g. of chromic acid in 7 ml. of pyridine. The mixture was stirred at room temperature for 2.5 hr. and was then taken up in ether and 2 *N* HCl. The ether layer was washed with 2 *N* HCl, 2 *N* Na_2CO_3 and water. The ethereal extract was dried over sodium sulfate and then evaporated under reduced pressure to give an oil, which crystallized on standing. The product was immediately used in the next step, due to the lability of the aldehyde toward air oxidation.

13-Hydroxy-19-stevanal B (X) was prepared in a manner similar to that described for IX and was used immediately in the subsequent thioacetal preparation, without attempting a purification. The semicarbazone of X, recrystallized from methanol, melted at 244–248°.

Anal. Calcd. for $C_{21}H_{35}N_3O_2$: C, 69.77; H, 9.76; N, 11.62. Found: C, 69.93; H, 10.01; N, 11.55.

13-Hydroxy-19-stevanal A, Ethylene Thioacetal (XI).—A mixture of 400 mg. of crude aldehyde IX in 3 ml. of ethanedithiol and 3 ml. of BF_3 etherate was stirred for 1 hr. at room temperature. The mixture was dissolved in ether, the ethereal extract washed 5 times with 2 *N* NaOH, 3 times with water and then dried over sodium sulfate. Evaporation of the solvent gave a residue which crystallized from benzene–petroleum ether (30–60°) to give 382 mg. of product, m.p. 156–158°. Further recrystallization raised the m.p. to 166–168°, $[\alpha]^{20}D$ −35.4 ± 1.7° (*c* 1.06).

Anal. Calcd. for $C_{22}H_{36}OS_2$: C, 69.42; H, 9.53. Found: C, 69.83; H, 9.37.

13-Hydroxy-19-stevanal B, ethylene thioacetal (XII) was prepared from 800 mg. of X in a manner described for XI. The residue on evaporation of the solvent was chromatographed on 30 g. of alumina (grade III). Benzene–ether and ether fractions gave 620 mg. of crude XII, recrystallized from ether–hexane to give white prisms, m.p. 170–171°, $[\alpha]^{20}D$ −59.3 ± 1.8° (*c* 1.25).

Anal. Calcd. for $C_{22}H_{36}OS_2$: C, 69.42; H, 9.53. Found: C, 69.21; H, 9.45.

13-Hydroxystevane A (XIII).—To a solution of 210 mg. of thioacetal XI in 60 ml. of ethanol was added 5 g. of Raney nickel. The mixture was refluxed for 6 hr. The catalyst was removed by filtration and the solvent evaporated. Crystallization of the product from dilute methanol gave 75 mg. of white solid, m.p. 135–142°. Sublimation gave a product melting at 147–148°, $[\alpha]^{20}D$ −24.4 ± 1.4° (*c* 1.11).

Anal. Calcd. for $C_{20}H_{34}O$: C, 82.69; H, 11.80. Found: C, 82.63; H, 11.63.

13-Hydroxystevane B (XIV) was prepared in a manner similar to that described for XIII. From 600 mg. of thioacetal XII was obtained 246 mg. of product, m.p. 151–153°, after two recrystallizations from dilute methanol, followed by sublimation; $[\alpha]^{20}D$ −51.7 ± 1.3° (*c* 0.97).

Anal. Calcd. for $C_{20}H_{34}O$: C, 82.69; H, 11.80. Found: C, 82.80; H, 11.53.

13-Bromostevane A (XV).—To a stirred solution of 117 mg. of XIII in 10 ml. of ether cooled to 0° was added 500 mg. of freshly prepared phosphorus pentabromide.[19] The mixture was then allowed to come to room temperature and stirring was continued for 18 hr. Ice was then cautiously added and the mixture diluted with ether. The ether layer was washed twice with sodium carbonate solution, followed by water. After drying the ethereal extract with sodium sulfate, the solvent was removed under reduced pressure. The thick oil was chromatographed on 4 g. of alumina (grade I). Elution with hexane gave a crystalline product which was recrystallized twice from ether–methanol to yield 60 mg. (42%) of white crystals, m.p. 107–108°. Further recrystallization raised the m.p. to 110–112°, $[\alpha]^{20}D$ −17.0 ± 1.2° (*c* 1.03).

Anal. Calcd. for $C_{20}H_{33}Br$: C, 67.97; H, 9.41. Found: C, 67.85; H, 9.63.

13-Bromostevane B (XVI) was prepared in a manner similar to that described for XV. From 129 mg. of XIV was obtained 95 mg. of product after chromatography on grade I alumina (elution with hexane). Recrystallization from ether–methanol gave 61 mg. of product, m.p. 113–116°. Further recrystallization raised the m.p. to 117–119°, $[\alpha]^{20}D$ −66.1 ± 0.9° (*c* 1.52).

Anal. Calcd. for $C_{20}H_{33}Br$: C, 67.97; H, 9.41. Found: C, 68.07; H, 9.29.

Stevane A (XVII). Method 1.—A solution of 160 mg. of 13-bromostevane A (XV) in 25 ml. of ethyl acetate and 0.75 ml. of triethylamine was added to a slurry of 1.5 g. of Raney nickel in 25 ml. of ethyl acetate. The mixture was hydrogenated at 100° (1200 p.s.i.) for 15 hr. The catalyst was removed by filtration and the solution concentrated to *ca.* one-third the original volume. The solution was then washed with water and dried over sodium sulfate. Evaporation of the solvent under reduced pressure gave an oil which was chromatographed on 3 g. of alumina (grade I). Elution with pentane gave 86 mg. of crystalline product. Recrystallization from ether–acetone gave 50 mg., m.p. 83–86°. Further recrystallization and then sublimation raised the m.p. to 87.5–88.5°, $[\alpha]^{20}D$ −31.9 ± 0.8° (*c* 1.14). The compound was identical with (−)-α-dihydrokaurene (m.p. 86–87°, $[\alpha]^{21}D$ −32°) by mixture melting point, infrared spectrum, and X-ray powder patterns.[12]

Anal. Calcd. for $C_{20}H_{34}$: C, 87.51; H, 12.49. Found: C, 87.38; H, 12.61.

Stevane B (XVIII) was prepared in a manner similar to that described for XVII. From 148 mg. of XVI was obtained 53 mg. of compound, m.p. 40–44° after sublimation and then recrystallization from methanol–acetone. Further recrystallization produced crystals melting at either 47° or 54–55°. Both modifications could be interconverted on seeding. The higher melting form often trans-crystallized at 46–47°, $[\alpha]^{20}D$ −66.8 ± 1.3° (*c* 1.03).

Anal. Calcd. for $C_{20}H_{34}$: C, 87.51; H, 12.49. Found: C, 87.20; H, 12.62.

13-Bromostevic acid A, methyl ester (XIX) was prepared in a manner similar to that described for XV. From 368 mg. of V was obtained 239 mg. of white solid, m.p. 115–123°, obtained by chromatography of the crude oil on 12 g. of grade II alumina (elution with petroleum ether, 30–60°). Recrystallization from acetone–water gave 208 mg. (48%) of fine, white needles, m.p. 122–124°. An analytical sample melted at 124–124.5°, $[\alpha]^{20}D$ −52.0° (*c* 0.87).

Anal. Calcd. for $C_{21}H_{33}O_2Br$: C, 63.47; H, 8.37. Found: C, 63.21; H, 8.68.

13-Bromostevic acid B, methyl ester (XX) was prepared in a manner similar to that described for XV. From 430 mg. of VI was obtained 300 mg. of white prisms, m.p. 120–124°, from methanol. Recrystallization from acetone–water raised the m.p. to 121–124°, $[\alpha]^{20}D$ 104.0 ± 1.5° (*c* 0.97).

Anal. Calcd. for $C_{21}H_{33}O_2Br$: C, 63.47; H, 8.37; Br, 20.11. Found: C, 63.72; H, 8.67; Br, 20.23.

Stevic Acid A, Methyl Ester (XXI).—A mixture of 208 mg. of XIX in 100 ml. of dioxane and 1.5 teaspoonfuls of Raney nickel[20] was refluxed for 22 hr. The catalyst was removed by filtration and washed repeatedly with dioxane. The solvent was removed

(19) Prepared in ether at 0° by adding an equivalent of bromine to PBr_3. The ether was then evaporated under reduced pressure at −10°.

(20) A. Ruff and T. Reichstein, *Helv. Chim. Acta*, **34**, 79 (1951); *cf.* footnote 1.

under reduced pressure to yield a colorless oil. Crystallization from acetone–water gave fine, white needles, 131 mg. (78%), m.p. 83–84°. An analytical sample melted at 84–85°, $[\alpha]^{20}D$ −68.0 ± 1° (*c* 1.1).

Anal. Calcd. for $C_{21}H_{34}O_2$: C, 79.19; H, 10.76. Found: C, 79.44; H, 10.90.

Stevic acid B, methyl ester (XXII) was prepared in a manner similar to that described for XXI. From 300 mg. of XX was obtained 200 mg. (83%) of crystals from methanol; m.p. 105–115°. Three recrystallizations from dilute methanol gave white prisms, m.p. 118–120°, $[\alpha]^{20}D$ −93.7 ± 1° (*c* 1.08).

Anal. Calcd. for $C_{21}H_{34}O_2$: C, 79.19; H, 10.76. Found: C, 79.42; H, 10.83.

19-Hydroxystevane A (XXIII) was prepared in a manner similar to that described for VII. From 131 mg. of methyl ester XXI was obtained a crude white solid, m.p. 147.5–153°, which on recrystallization from acetonitrile gave long, white needles, 87 mg. (73%), m.p. 153–154°, $[\alpha]^{20}D$ −29.3 ± 2° (*c* 1.09).

Anal. Calcd. for $C_{20}H_{34}O$: C, 82.69; H, 11.80. Found: C, 82.55; H, 11.84.

19-Hydroxystevane B (XXIV) was prepared in a manner similar to that described for VII. From 200 mg. of stevic acid B, methyl ester (XXII) and 140 mg. of lithium aluminum hydride in refluxing tetrahydrofuran (6 hr.) was obtained 133 mg. (72%) of white solid, m.p. 133–136° (trans-crystallization at 125–130° from prisms to needles) from methanol. The resolidified compound melted at 142–144°, $[\alpha]^{20}D$ −61.7° ± 2.2° (*c* 0.87).

Anal. Calcd. for $C_{20}H_{34}O$: C, 82.69; H, 11.80. Found: C, 82.57; H, 11.82.

19-Stevanal A (XXV) was prepared in a manner similar to that described for IX. From 85 mg. of XXIII was obtained 76 mg. (90%) of white flakes, m.p. 109.5–118°. The crude compound was used immediately for thioacetal formation and no attempt was made to recrystallize it.

19-Stevanol B (XXVI) was prepared in a manner similar to that described for IX. From 174 mg. of XXIV was obtained 158 mg. of crude oil, used directly for the preparation of the thioacetal.

19-Stevanal A, ethylene thioacetal (XXVII) was prepared in a manner similar to that described for XI. From 76 mg. of crude aldehyde XXV was obtained 74 mg. of crude thioacetal by chromatography on 3 g. of grade II alumina (elution with petroleum ether, 30–60°). Attempts to crystallize this compound failed.

19-Stevanal B, ethylene thioacetal (XXVIII) was prepared in a manner similar to that described for XI. From 158 mg. of crude aldehyde XXVI was obtained 105 mg. (41%) of crystals, m.p. 128–134°. Recrystallization from ether–methanol raised the m.p. to 131–132.5°, $[\alpha]^{20}D$ −75.4 ± 3.2°.

Anal. Calcd. for $C_{22}H_{36}S_2$: C, 72.46; H, 9.95. Found: C, 72.27; H, 10.15.

Stevane A (XVII). Method 2.—To a solution of 74 mg. of crude thioacetal XXVII in 40 ml. of dioxane was added 0.5 teaspoonful of Raney nickel and the mixture was refluxed for 18 hr. The catalyst was removed by filtration and the solvent removed under reduced pressure. The product was chromatographed on 1.8 g. of grade II alumina. Elution with petroleum ether (30–60°) gave a white solid, which on recrystallization from acetone gave 38.9 mg. of white needles (66%), m.p. 87.5–89°, $[\alpha]^{20}D$ −30.3 ± 1.5° (*c* 1.09). A mixture m.p. with stevane A from method 1 showed no depression. Infrared and gas chromatographic retention times of both stevane A samples were identical.

Stevane B (XVIII) was prepared in a manner similar to that described for XVII (method 2). From 80 mg. of thioacetal XXVIII was obtained 25.6 mg. (42%) of white solid, m.p. 44.5–47° $[\alpha]^{20}D$ −51.9 ± 1.5°. Recrystallization from acetone–methanol raised the m.p. to 53.5–55°, $[\alpha]^{20}D$ −56.3 ± 1.5°. The compound was identical (mixture m.p., infrared spectra, gas chromatography) with stevane B from method 1. It was identical[21] with the hydrocarbon obtained from garryfoline[14] (gas chromatography, infrared spectra).

(21) We wish to thank Professor C. Djerassi for making these comparisons.

[CONTRIBUTION FROM THE DEPARTMENT OF CHEMISTRY, CORNELL UNIVERSITY, ITHACA, NEW YORK]

Keto–Enol Transformation of 1,2-Cyclohexanedione. I. Hydration and Keto–Enol Equilibria[1-3]

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Although 1,2-cyclohexanedione is normally available only as its mono enol, the ketone can be prepared in almost pure form through its bisulfite complex. The monohydrate of the ketone forms rapidly and almost completely in aqueous solution. Studies of the hydration reaction in aqueous dioxane at 25° lead to an equilibrium constant for hydrate formation of 182 ± 5. The keto–enol transformation is slow in aqueous solution but is catalyzed by both acids and bases. The stoichiometric concentration equilibrium constant, $K_e = [K]/[E]$, is close to unity in aqueous solutions which are dilute in both substrate and electrolyte but varies almost 10-fold from dilute acidic solutions to solutions which are 6 *M* in mineral acids. The explanation is shown to be that the activity coefficient of the ketone increases much more rapidly with electrolyte concentration than does that of the enol. Studies of the keto–enol equilibrium as a function of temperature lead, for formation of the ketone, to values of $\Delta H^0 = -6$ kcal./mole and $\Delta S^0 = -21$ e.u. This large entropy change is about the expected value for transformation of an unhydrated enol to a monohydrated ketone.

Only a few keto–enol equilibria have been studied in aqueous solution; most of these have been between β-diketones and their enols, a reason being that this is one important class of ketones for which significant amounts of enol are to be found in aqueous solutions.[4-6] Cyclic α-diketones represent an interestingly different class of compounds to study. The cyclic character of these molecules can clearly contribute strain to the system and it is of interest to see the effect of this. A different but perhaps related reason is that cyclic α-diketones tend to be strongly hydrated and this should have a large effect on the keto–enol equilibrium.

The present study considers both the hydration and keto–enol equilibria for a typical cyclic species, 1,2-cyclohexanedione. Although this compound is customarily known only as its enol, Schwarzenbach and Wittwer some time ago presented data which indicated that it reached an equilibrium in aqueous solution in which the relative amounts of keto and enol were approximately one to one.[7] This result was uncertain in the sense that no direct evidence for the presence of ketone was obtained, but it did suggest the desirability of further studies of this system. Another incentive is the fact that closely related cyclic diketones have frequently been found to be hydrated. Thus it has been shown that one of the carbonyl groups of 3,3,6,6-tetramethyl-1,2-cyclohexanedione (a compound which cannot enolize) is almost completely hydrated.[8] Similarly, 3,4-diketotetrahydrofuran has been shown to be extensively hydrated.[9]

Experimental

The mono-enol of 1,2-cyclohexanedione melts at about 38°. It forms colorless solutions in water and in anhydrous organic solvents such as chloroform and dioxane. It absorbs strongly in the ultraviolet, the main absorption being a broad peak with a

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(2) Presented in part at the 139th National Meeting of the American Chemical Society, St. Louis, Mo., April, 1961.

(3) Original data in thesis of Ronald Bakule, Cornell University, 1962, available from University Microfilms, Ann Arbor, Mich.

(4) For early references see C. K. Ingold, "Structure and Mechanism in Organic Chemistry," Cornell University, Ithaca, N. Y., 1953, p. 555.

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