Constituents of *Iva* Species. II. The Structures of Asperilin and Ivasperin, Two New Sesquiterpene Lactones[1]

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The structures of asperilin and ivasperin, two new sesquiterpene lactones from *Iva asperifolia* Less., are shown to be II and X. Asperilin and ivasperin also have been isolated from *Iva texensis* Jackson.

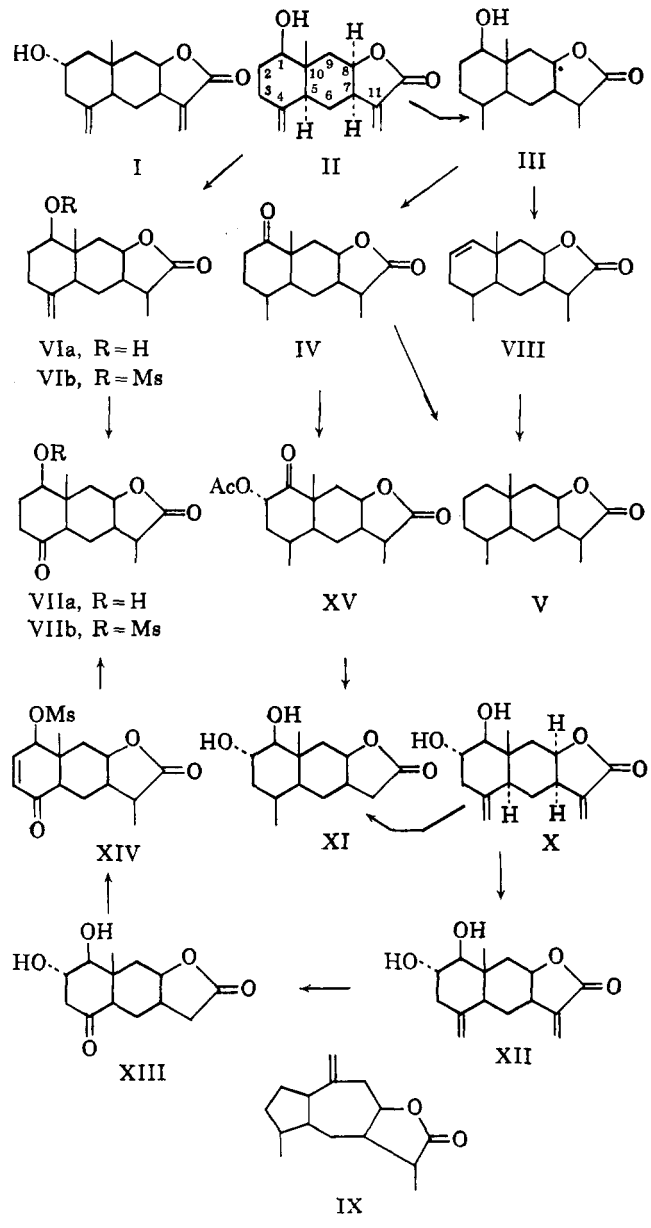
In order to delineate more clearly possible connections between genera related to *Ambrosia* and *Parthenium* suggested by our earlier work,[2,3] we have initiated a systematic phytochemical survey of the genus *Iva*.[4] In the first paper[5] we discussed the structure of ivalin (I), the main sesquiterpene lactone constituent of *I. microcephala* Nutt., and *I. imbricata* Walt. We now report the isolation and structure determination of asperilin and ivasperin, constituents of *I. asperifolia* Less. and *I. texensis* Jackson. Work on other *Iva* species is in progress.

Iva asperifolia is a Mexican species (state of Veracruz) whose distribution in the United States is limited to a small area near the old port of St. Marks, Florida.[4] Material from this source, which may represent an introduction from Mexico, furnished two new sesquiterpene lactones in 0.16 and 0.029% yields, respectively, which we have named asperilin and ivasperin.

Asperilin (II), the less polar material, m.p. 151–152°, $[\alpha]^{23}D$ +149.6°, had the formula $C_{15}H_{20}O_3$ and contained a hydroxyl group (infrared bands at 3700 and 3500 cm.$^{-1}$; formation of an acetate) and two double bonds (infrared bands at 1655 and 1645 cm.$^{-1}$). One of the double bonds was conjugated with a γ-lactone function (ν_{max} 1755 cm.$^{-1}$) as evidenced by the ultraviolet maximum at 211 mμ (ϵ 8730) and the formation of a pyrazoline from asperilin acetate. Hence asperilin is dicarbocyclic.

Catalytic hydrogenation of asperilin in acetic acid using platinum oxide gave tetrahydroasperilin (III) by saturation of both double bonds. Chromic acid oxidation of III gave a ketone (IV) whose infrared spectrum (ν_{max} 1704 cm.$^{-1}$) showed that the carbonyl group was in a six- or higher-membered ring. Hence the hydroxyl group in asperilin is secondary. Desulfurization of the ethylenethioketal of dehydrotetrahydroasperilin (IV) afforded tetrahydroalantolactone[6] (V), identical in all respects with an authentic sample. This establishes the carbon skeleton of asperilin and the stereochemistry at positions 5, 7, 8, and 10.

The nature of the double bonds was indicated by selective reduction of II to dihydroasperilin (VIa), which possessed no ultraviolet maximum. Ozonolysis of its mesylate gave the norketone VIIb. This indicates that asperilin contains one isolated exocyclic methylene group and another methylene group conjugated with a γ-lactone.



The n.m.r. spectra[8] of II, III, VIa, and the acetates of II and III confirmed these conclusions. II had two low-field doublets (6.01 and 5.48 p.p.m., $J \sim 1$ c.p.s.), each representing one proton, characteristic of a methylene group conjugated with a lactone.[2,3,5] These were absent in III and VIa. A doublet at 4.85 p.p.m. ($J \sim 1.5$) represented one proton of the unconjugated exocyclic methylene group, and a multiplet centered

(1) Supported in part by grants from the U. S. Public Health Service (RG-5814) and the National Science Foundation (NSF-G-14396).

(2) W. Herz and G. Högenauer, *J. Org. Chem.*, **26**, 5011 (1961).

(3) W. Herz, H. Watanabe, M. Miyazaki, and Y. Kishida, *J. Am. Chem. Soc.*, **84**, 2601 (1962).

(4) The most recent revision is that of R. C. Jackson, *Univ. Kansas Sci. Bull.*, **41**, 793 (1960).

(5) W. Herz and G. Högenauer, *J. Org. Chem.*, **27**, 905 (1962).

(6) The C-11 methyl group of tetrahydroalantolactone recently has been shown to be β-oriented.[7]

(7) W. Cocker and M. A. Nisbet, *J. Chem. Soc.*, 534 (1963).

(8) N.m.r. spectra were run on an A-60 instrument in deuteriochloroform solution with tetramethylsilane serving as internal reference.

at 4.50 p.p.m., corresponding to two protons, was due to the superposition of the other methylene proton on the signal of the lactonic hydrogen on C-8. A three-proton singlet at 0.80 p.p.m. was due to a tertiary methyl group. A complex set of bands (two doublets centered at 3.4 p.p.m., J_{AX} = 9.5, J_{AY} = 6 c.p.s.) had to be ascribed to the hydrogen on carbon carrying the hydroxyl group since it moved downfield to near 4.5 p.p.m. on acetylation and disappeared on oxidation.

III had no olefinic protons but exhibited, in addition to the methyl singlet at 0.965 p.p.m., two methyl doublets at 1.19 and 1.31 p.p.m. (J = 7 c.p.s.). VIa had two narrowly split doublets centered at 4.84 and 4.55 due to the unconjugated methylene protons,[9] one methyl singlet of 0.79, and one methyl doublet at 1.22 p.p.m. (J = 6 c.p.s.).

The close similarity between asperilin and ivalin (I)[5] suggested that the two compounds differed only in the position of the hydroxyl group on the ring skeleton. That IV contained a $-CO-CH_2-$ system was shown by its positive Zimmermann test and the formation of a monopiperonylidene derivative. Positions 2 and 3, however, were untenable as the locus of the hydroxyl group in asperilin, since IV was different from dehydrotetrahydroivalin (2-ketotetrahydroalantolactone)[5] and dehydrotetrahydroisotelekin (3-ketotetrahydroalantolactone).[10] By elimination, dehydrotetrahydroasperilin (IV) had to be 1-ketotetrahydroalantolactone, and the position of the hydroxyl group in asperilin was hence uniquely fixed as in II. The optical rotatory dispersion of IV supported this conclusion since it exhibited a positive Cotton effect and was very similar to that of 9-methyl-*trans*-1-decalones.[11]

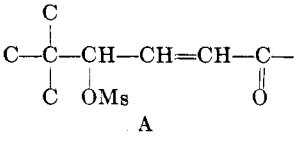
The configuration of this C-1 hydroxyl group remained to be settled. Reduction of IV with sodium borohydride, sodium–ethanol, or hydrogenation with platinum oxide in acetic acid all gave tetrahydroasperilin as the only isolable product. Reduction to the equatorial alcohol is favored on steric grounds as well as on considerations of thermodynamic stability. The hydroxyl group of asperilin must, therefore, be β-oriented.

It is interesting to note that the (noncrystallizable) mesylate of tetrahydroasperilin on treatment with collidine at 200–210° gave VIII in 20% yield, and not IX as was expected in analogy with earlier work[12,13] which would have afforded an entry into the guaianolide series. The structure of VIII was made evident by its reduction to V and by its n.m.r. spectrum (p.p.m.): singlet at 5.5 corresponding to two olefinic protons,[14] two methyl doublets at 0.94 and 1.27, (J = 7 c.p.s.), one methyl singlet at 1.08, and an H-8 multiplet at 4.5. Since much decomposition accompanied the reaction, the formation of IX cannot be completely excluded and further efforts to effect this conversion are contemplated, pending availability of material.

The minor and more polar constituent, ivasperin (X), was $C_{15}H_{20}O_4$, m.p. 150–151°, $[\alpha]^{23}D$ +140.5°. Its ultraviolet [λ_{max} 210.5 mμ (ϵ 7750)] and infrared spectra (bands at 3650, 3450, 1760, 1660, and 1650 cm.$^{-1}$) were very similar to those of asperilin. Compound X had two double bonds as shown by hydrogenation with platinum oxide to tetrahydroivasperin (XI). Hydrogenation with palladium–calcium carbonate catalyst gave dihydroivasperin (XII) by reduction of the double bond conjugated with the lactone carbonyl. Ivasperin also contained two hydroxyl groups as shown by the formation of a diacetate. That these were vicinal was indicated by the positive periodic acid test shown by ivasperin and its reduction products.

The n.m.r. spectrum (p.p.m.) of ivasperin had two pairs of doublets at 6.24 and 5.59 (J = 1 c.p.s., $=CH_2$ conjugated with γ-lactone), a doublet at 4.90 (J = 1.5 c.p.s., one of the protons of the unconjugated exocyclic methylene groups), a multiplet centered at 4.59 (two protons, superposition of the other olefinic proton on the lactonic hydrogen at C-8), a poorly defined series of bands near 3.5 (two protons, presumably H on carbon carrying hydroxyl groups), and a methyl singlet at 0.81. Ivasperin diacetate exhibited doublets at 6.06 and 5.54 (conjugated methylene), a complex multiplet centered at 4.86 (three protons, two due to H on carbon carrying acetates and one belonging to the unconjugated methylene), a doublet at 4.63 (J = 1.5 c.p.s., second proton of $=CH_2$), a multiplet at 4.44 (H-8), two acetate singlets at 2.06 and 1.97, and a methyl singlet at 0.91 p.p.m. On the other hand, tetrahydroivasperin (XI) had no peaks in the vinyl proton region, but exhibited two methyl doublets at 1.18 (J = 8 c.p.s.) and 0.945 (J = 6 c.p.s.), and the usual methyl singlet at 1 p.p.m., in addition to the lactone hydrogen multiplet at 4.54, and the two-proton signal centered at 3.5 p.p.m.

Dihydroivasperin (XII) on ozonolysis furnished a norketone (XIII) which exhibited a positive Zimmermann test. The dimesylate of XIII when heated with pyridine afforded an anhydro ketone (XIV) which retained one mesylate function and whose ultraviolet [λ_{max} 222.5 mμ, (ϵ 8150)] and infrared spectra (bands at 1680 and 1630 cm.$^{-1}$) showed that it was an α,β-unsaturated ketone. The n.m.r. spectrum (set of twelve ABX type signals: A at 6.80, B at 6.12, X at 5.42 p.p.m., J_{AB} = 10, J_{AX} = 1.5, J_{BX} = 2.5 c.p.s., lactone hydrogen at 4.5, mesylate at 3.16, methyl doublet at 1.24 p.p.m., J = 7 c.p.s., and methyl singlet at 1.0 p.p.m.) established the presence of partial structure A and confirmed that the two hydroxyl groups of ivasperin were secondary.



A

Catalytic reduction of XIV with palladium on charcoal furnished the ketomesylate VIIb identical in all respects with a mesylate obtained by ozonolysis of dihydroasperilin mesylate. This established the gross

(9) The second of these was again superimposed on the signal of H-8.

(10) V. Benešová, V. Herout, and F. Šorm, *Collection Czech. Chem. Commun.*, **26**, 1350 (1961).

(11) C. Djerassi and W. Klyne, *J. Chem. Soc.*, 4029 (1962). We are indebted to Dr. Ulrich Weiss, Laboratory of Physical Biology, National Institute of Arthritis and Metabolic Diseases, for carrying out this measurement.

(12) R. Hirschmann, C. S. Snoddy, C. F. Hiskey, and N. L. Wendler, *J. Am. Chem. Soc.*, **76**, 4013 (1954).

(13) D. H. R. Barton, O. C. Böckman, and P. de Mayo, *J. Chem. Soc.*, 2263 (1960).

(14) Apparently H-1 and H-2 exhibit the same chemical shift and H-2 is split only slightly by the two adjacent protons at C-3.

structure of ivasperin as well as its stereochemistry, excepting the configuration of the hydroxyl at C-2.

This was clarified as follows. Lead tetraacetate oxidation of dehydrotetrahydroasperilin (IV) under conditions which favor the equatorial isomer[15] gave the 2-α-acetoxy ketone XV. The acetoxy group in this must be equatorial since it is unchanged on prolonged refluxing in acetic acid. Sodium borohydride reduction of XV resulted in simultaneous hydrolysis of the acetoxy group to yield the diol XI identical with tetrahydroivasperin. The hydroxyls of ivasperin must, therefore, be both equatorial which leads to the stereochemistry depicted in X.

Iva texensis Jackson, a newly distinguished species[4] which is very closely related morphologically to *I. asperifolia*, was investigated subsequently and furnished asperilin and ivasperin in 0.22 and 0.18% yield.

Experimental[16]

Extraction of *I. asperifolia* Less.—Ground whole plant, 12 lb., collected in St. Marks, Florida (in September, 1962), was extracted with hot chloroform for 3 days. The chloroform was removed and the residue was dissolved in 1.2 l. of ethanol, warmed to about 50°, and heated with a hot solution of 56 g. of lead acetate and 20 ml. of acetic acid in 1.3 l. of water. The mixture was allowed to stand overnight; the supernatant liquid was filtered, concentrated to about 1.5 l. *in vacuo*, and extracted thoroughly with chloroform; and the chloroform extract was washed, dried, and evaporated. The residual gum, 160 g., was chromatographed over 2 lb. of Alcoa F-20 alumina in benzene solution. The material eluted with benzene was rechromatographed in 3:2 petroleum ether (b.p. 35–60°)–benzene to give 8.5 g. of asperilin, needles from acetone–ether–petroleum ether, m.p. 151–152°. The more polar material in the first chromatogram, eluted with benzene–chloroform and chloroform, gave a slowly solidifying gum, sparingly soluble in benzene and chloroform and very soluble in methanol. Repeated crystallization from ethyl acetate gave prisms of ivasperin, 1.61-g. yield, m.p. 150–151°. The homogeneity of asperilin and ivasperin was checked by thin layer chromatography using silica as adsorbent; R_f values in ether were: asperilin, 0.43; ivasperin, 0.10; in acetone–methanol (97:3): asperilin, 0.98; ivasperin, 0.70.

Asperilin had λ_{max} 211 mμ (ϵ 8730); ν_{max} 3700, 3500, 1755, 1655, and 1645 cm.$^{-1}$; $[\alpha]^{23}D$ +149.6° (*c* 1.35).

Anal. Calcd. for $C_{15}H_{20}O_3$: C, 72.55; H, 8.12; O, 19.33. Found: C, 72.86; H, 8.16; O, 19.25.

Ivasperin had λ_{max} 210 mμ (ϵ 7750); ν_{max} 3650, 3450, 1760, 1660, and 1650 cm.$^{-1}$; $[\alpha]^{23}D$ +140.5° (*c* 2, methanol).

Anal. Calcd. for $C_{15}H_{20}O_4$: C, 68.19; H, 7.58; O, 24.24. Found: C, 67.90; H, 7.56; O, 24.45.

Acetylasperilin.—A mixture of 0.2 g. of asperilin, 2 ml. of acetic anhydride, and 1 ml. of pyridine was allowed to stand for 12 hr. The usual work-up resulted in 0.18 g. of the acetate, m.p. 176–178°, prisms from ethyl acetate–petroleum ether, having ν_{max} 1765 (γ-lactone), 1740 (acetate), 1655, and 1645 cm.$^{-1}$ (double bonds); n.m.r. signals (p.p.m.) at 6.12 and 5.59 (narrowly split doublets, J = 1 c.p.s., conjugated methylene), 4.83 and 4.55 (narrowly split doublets, unconjugated methylene) superimposed on multiplets of H-1 centered at 4.7, and H-8 centered at 4.55, 2.06 (acetate), and 0.89 (C-10 methyl).

Anal. Calcd. for $C_{17}H_{22}O_4$: C, 70.32; H, 7.64; O, 22.04. Found: C, 70.70; H, 7.59; O, 21.94.

The pyrazoline was prepared by allowing 0.12 g. of the acetate in 10 ml. of ether to stand with 20 ml. of ethereal diazomethane for 3 days at ice-box temperature. Evaporation followed by several crystallizations from acetone–petroleum ether furnished the derivative, m.p. 170° dec.

Anal. Calcd. for $C_{18}H_{24}N_2O_4$: C, 65.04; H, 7.28. Found: C, 65.48; H, 6.96.

Tetrahydroasperilin (III).—A solution of 1 g. of II in 50 ml. of acetic acid was hydrogenated with 0.2 g. of platinum oxide at 30 lb./in.^{2} for 2 hr. The catalyst was filtered, the solvent was evaporated, and the residue was crystallized twice from ethyl acetate–petroleum ether, yielding 0.71 g., m.p. 150–151°; $[\alpha]^{23}D$ +16.6° (*c*, 3.3); ν_{max} 3700, 3500 (–OH), and 1770 cm.$^{-1}$ (γ-lactone).

Anal. Calcd. for $C_{15}H_{24}O_3$: C, 71.39; H, 9.59; O, 19.02. Found: C, 71.46; H, 9.56; O, 18.85.

The acetate was prepared in the usual manner and was crystallized from ethyl acetate–petroleum ether, m.p. 104–105°. The n.m.r. spectrum (p.p.m.) had signals at 4.52 (two protons, complex multiplet, superposition of H-1 and H-8), 2.02 (acetate), 1.18 and 0.90 (two doublets, C-4 and C-11 methyls), and 1.02 (singlet, C-10 methyl).

Anal. Calcd. for $C_{17}H_{26}O_4$: C, 69.39; H, 8.85; O, 21.77. Found: C, 69.37; H, 9.15; O, 21.53.

Dehydrotetrahydroasperilin (IV).—A solution of 1 g. of III in 15 ml. of acetic acid was treated dropwise with a solution of 0.75 g. of chromic acid in 15 ml. of acetic acid and 2 ml. of water. The resulting mixture was allowed to stand overnight at room temperature, concentrated at reduced pressure, diluted with water, and extracted with chloroform. The extract was washed, dried, and evaporated. The residue on crystallization from ether–petroleum ether gave 0.76 g. of IV, m.p. 131–132°; $[\alpha]^{23}D$ +64.85° (*c*, 2.39); infrared bands at 1770 (γ-lactone) and 1704 cm.$^{-1}$ (ketone); positive Zimmermann test; optical rotatory dispersion curve in methanol (*c* 0.081): $[\alpha]_{589}$ +64.8°, $[\alpha]_{305}$ +821°, $[\alpha]_{275}$ −117°. The n.m.r. spectrum (p.p.m.) exhibited signals at 4.5 (multiplet, H-8), 1.25 (singlet, C-10 methyl), and 1.20 and 1.15 (doublets, J = 7 c.p.s., C-4 and C-11 methyls).

Anal. Calcd. for $C_{15}H_{22}O_3$: C, 71.97; H, 8.86. Found: C, 71.55; H, 8.66.

The piperonylidene derivative was prepared by allowing a solution of 0.135 g. of IV and 0.22 g. of piperonal in 5 ml. of ethanol to stand overnight with 5 ml. of ethanol saturated with hydrogen chloride. The solution was diluted with ice–water and extracted with methylene chloride. The extract was washed, dried, and concentrated, and the residue was taken up in benzene and chromatographed over acid-washed alumina. Benzene–chloroform (4:1) eluted the material which was crystallized from acetone–petroleum ether. The silky needles melted at 204–205°.

Anal. Calcd. for $C_{23}H_{26}O_5$: C, 72.24; H, 6.81; O, 20.94. Found: C, 71.93; H, 6.81; O, 21.62.

Tetrahydroalantolactone (V).—A suspension of 0.22 g. of IV in 0.45 ml. of ethanedithiol was cooled, mixed with 1.1 ml. of boron trifluoride etherate, left overnight at room temperature, diluted with water, and extracted thoroughly with ether. The ether extract was washed, dried, and evaporated, and the residue was crystallized from benzene–petroleum ether, yielding 0.19 g. of thioketal, m.p. 214–215°.

Anal. Calcd. for $C_{17}H_{26}O_2S_2$: C, 62.56; H, 8.03; S, 19.61. Found: C, 62.01; H, 8.00; S, 20.12.

A solution of 0.32 g. of the thioketal in 25 ml. of absolute ethanol was refluxed for 24 hr. with 2 teaspoonfuls of Raney nickel. Evaporation of the filtrate and crystallization of the residue from ethanol afforded 0.13 g. of tetrahydroalantolactone, m.p. 143–144°, undepressed on admixture of an authentic sample. The infrared spectra and the rotations of the two samples were identical.

Reductions of Dehydrotetrahydroasperilin. A. Sodium Borohydride.—A solution of 0.32 g. of IV in 25 ml. of methanol was left overnight with 0.15 g. of sodium borohydride. Excess reagent was decomposed with a few drops of acetic acid, the solution was concentrated *in vacuo*, and the residue was diluted with water and extracted with chloroform. The extract was washed, dried, and evaporated; crystallization of the residue afforded 0.25 g. (78%) of tetrahydroasperilin, m.p. and m.m.p. (with an authentic sample) 149–150°. The infrared spectra and rotations of the two samples were identical.

B. Catalytic Reduction.—A solution of 0.1 g. of IV in 15 ml. of acetic acid was shaken with 0.03 g. of platinum oxide at a hydrogen pressure of 35 lb./in.^{2}. The solution was filtered and concentrated, and the residue was recrystallized from ethyl acetate–petroleum ether, yielding 0.075 g. (74%) of tetrahydroasperilin, m.p. and m.m.p. 150°.

C. Sodium–Ethanol.—A solution of 0.15 g. of IV in 40 ml. of ethanol was warmed on a water bath and treated, during 20 min., with 2.2 g. of sodium. After 45 min. at reflux, the solution was concentrated, acidified to congo red, and extracted with chloro-

(15) K. Yamakawa, *J. Org. Chem.*, **24**, 897 (1959).

(16) Melting points are uncorrected; analyses are by Dr. F. Pascher, Bonn Germany. Infrared spectra and rotations were run in chloroform solution unless otherwise specified, ultraviolet spectra in 95% ethanol.

form; the extract was washed and dried. Chromatography over acid-washed alumina and recrystallization afforded 0.065 g. (43%) of tetrahydroasperilin, m.p. and m.m.p. 150°. No other fraction was isolated.

Dihydroasperilin (VIa).—A solution of 0.6 g. of II in 30 ml. of ethanol was hydrogenated at atmospheric pressure with 0.09 g. of 5% palladium on calcium carbonate. The reduction was stopped when 60 ml. of hydrogen (at NTP) had been absorbed. The catalyst was filtered, the solution was evaporated, and the solid residue was recrystallized twice from ethyl acetate–petroleum ether, yielding 0.375 g. of needles, m.p. 184–185°. The product had infrared bands at 3700 and 3450 (—OH), 1770 (γ-lactone), and 1650 cm.$^{-1}$ (double bond); no ultraviolet maximum; and n.m.r. signals at 4.84 d and 4.55 d (2, exocyclic methylene), 4.55 m (H-8), 1.22 d (C-11 methyl), and 0.79 p.p.m. (C-10 methyl).

Anal. Calcd. for $C_{14}H_{22}O_3$: C, 71.97; H, 8.86; O, 19.17. Found: C, 72.14; H, 8.76; O, 19.11.

Ozonolysis of Dihydroasperilin.—A solution of 0.4 g. of VIa in 15 ml. of methanol was ozonized at −70°. The resulting solution was shaken with hydrogen at 20 lb./in.^{2} for 0.5 hr. in the presence of 0.05 g. of 5% palladium on charcoal, filtered, and evaporated. The residue was recrystallized from ethyl acetate, yielding 0.15 g. of VIIa, m.p. 202–203°, infrared bands at 1780 (γ-lactone) and 1718 cm.$^{-1}$ (ketone).

Anal. Calcd. for $C_{14}H_{20}O_4$: C, 66.64; H, 7.99; O, 25.37. Found: C, 66.85; H, 7.72; O, 25.61.

Ozonolysis of VIb.—The mesylate of VIa (VIb) melted at 128° dec. after recrystallization from ethyl acetate–petroleum ether.

Anal. Calcd. for $C_{16}H_{24}O_5S$: C, 58.41; H, 7.37; S, 9.77. Found: C, 57.65; H, 7.18; S, 10.19.

A solution of 0.2 g. of the mesylate in 20 ml. of methanol was ozonized at −70° for 1 hr. The solution was shaken with 0.06 g. of 5% palladium-on-charcoal catalyst at a hydrogen pressure of 20 lb./in.^{2} for 0.5 hr., filtered, and evaporated. The residue was taken up in chloroform and filtered by passage through a short column of acid-washed alumina. Elution with chloroform and crystallization of the product from chloroform–petroleum ether gave 0.085 g. of the ketomesylate VIIb, m.p. 158–159°; infrared bands at 1775 (γ-lactone), 1720 (ketone), 1360, and 1170 cm.$^{-1}$ (mesylate group).

Anal. Calcd. for $C_{15}H_{22}O_6S$: C, 54.54; H, 6.71. Found: C, 54.30; H, 6.97.

Anhydrotetrahydroasperilin (VIII).—A solution of 0.33 g. of III in 3 ml. of pyridine was treated at 5° with 0.4 ml. of mesyl chloride and left overnight at 5°. The solution was diluted with water and extracted with ether. The ether extract was washed, dried, and evaporated to yield the mesylate as an uncrystallizable gum. The mesylate was heated in a sealed tube at 200–210° with 7 ml. of collidine for 15 hr., the base was removed at reduced pressure, and the residue was poured on ice–hydrochloric acid. Extraction with chloroform gave a gum which was chromatographed over acid-washed alumina, with benzene–petroleum ether (2:1) as solvent and eluent. The solid material was recrystallized from ether–petroleum ether, yielding 0.045 g. of VIII, m.p. 140–142°, infrared bands at 1770 (γ-lactone) and 1645 cm.$^{-1}$ (weak, double bond).

Anal. Calcd. for $C_{15}H_{22}O_2$: C, 76.88; H, 9.46; O, 13.66. Found: C, 76.82; H, 9.37; O, 13.79.

A solution of 0.06 g. of VIII in 10 ml. of ethanol was hydrogenated catalytically with platinum oxide. After removal of solvent and crystallization from ethanol there was obtained 0.04 g. of tetrahydroalantolactone, m.p. and m.m.p. 142–143°.

Diacetylivasperin.—A mixture of 0.18 g. of ivasperin, 2 ml. of acetic anhydride, and 1 ml. of pyridine was warmed, left overnight at room temperature, poured on water, and filtered. The solid was recrystallized from benzene–petroleum ether, yielding 0.15 g., m.p. 170–172°; infrared bands at 1770 (γ-lactone), 1745 (double strength, acetates), 1665, and 1655 cm.$^{-1}$ (double bonds).

Anal. Calcd. for $C_{19}H_{24}O_6$: C, 65.51; H, 6.90; O, 27.58. Found: C, 65.64; H, 6.87; O, 27.14.

Tetrahydroivasperin (XI).—A solution of 0.31 g. of X in 5 ml. of acetic acid was added to 0.07 g. of prereduced platinum oxide in 25 ml. of acetic acid and shaken at atmospheric pressure until hydrogen absorption ceased. The catalyst was filtered, the solution was concentrated, and the residual solid was recrystallized from ethyl acetate–petroleum ether, yielding 0.24 g. of prisms, m.p. 163–165°, $[\alpha]^{22}D$ +30° (*c* 2, methanol), infrared bands at 3700 and 3500 (—OH) and 1770 cm.$^{-1}$ (γ-lactone).

Anal. Calcd. for $C_{15}H_{24}O_4$: C, 67.17; H, 8.96; O, 23.89. Found: C, 66.88; H, 8.73; O, 24.03.

The diacetate was prepared using pyridine–acetic anhydride and recrystallized from ethyl acetate–petroleum ether, m.p. 186–187°, infrared bands at 1760 (γ-lactone) and 1740 cm.$^{-1}$ (double strength, acetates).

Anal. Calcd. for $C_{19}H_{28}O_6$: C, 64.75; H, 8.01; O, 27.24. Found: C, 64.80; H, 8.14; O, 27.39.

Dihydroivasperin (XII).—A solution of 0.7 g. of X in 15 ml. of ethanol was hydrogenated at atmospheric pressure with 0.14 g. of 5% palladium-on-calcium carbonate catalyst. The solution was evaporated and the residue was recrystallized from ethyl acetate; yielding 0.58 g., m.p. 180–181°, no ultraviolet maximum.

Anal. Calcd. for $C_{15}H_{22}O_4 \cdot H_2O$: C, 63.36; H, 8.51; O, 28.14. Found: C, 63.48; H, 8.71; O, 27.88.

Ozonolysis of Dihydroivasperin.—A solution of 0.2 g. of XII in 15 ml. of methanol was ozonized at −70° for 1 hr. and worked up reductively as described for previous ozonolyses. Recrystallization of the solid residue from methanol–ether furnished 0.11 g. of the norketone XIII, m.p. 194–195°; infrared bands at 3700 and 3500 (—OH), 1770 (γ-lactone), and 1725 cm.$^{-1}$ (ketone); positive Zimmermann test.

Anal. Calcd. for $C_{14}H_{20}O_5$: C, 62.67; H, 7.51; O, 29.82. Found: C, 62.34; H, 7.64; O, 30.27.

A solution of 0.09 g. of XIII in 2 ml. of pyridine was mixed with 1 ml. of methanesulfonyl chloride at 5° and allowed to stand at 10–15° overnight. The solution was poured on ice and extracted with chloroform. The extract on washing, drying, and evaporating furnished a gummy residue which was a mixture of dimesylate and anhydromonomesylate XIV (infrared and n.m.r. spectrum). To complete the conversion, it was heated on the steam bath with 3 ml. of pyridine for 3 hr., poured on ice–hydrochloric acid, and extracted with chloroform. The product XIV was crystallized from chloroform–petroleum ether as needles, m.p. 124°, in 0.05-g. yield; infrared bands at 1775 (γ-lactone), 1680 (conjugated ketone), 1630 (weak, double bond), 1365, and 1170 cm.$^{-1}$ (mesylate); λ_{max} 222.5 mμ (ε 8150). Since the material decomposed fairly rapidly on standing, it was not analyzed.

A solution of 0.09 g. of XIV in 15 ml. of methanol was hydrogenated at 20 lb./in.^{2} with 0.03 g. of 5% palladium on charcoal. The solution was filtered and evaporated, the residue was taken up in chloroform and chromatographed over acid-alumina. Chloroform eluted material which was recrystallized from chloroform–petroleum ether had m.p. 157–158°, undepressed on admixture of the mesylate VIIb. Infrared spectra of the samples were identical.

2-α-Acetoxy-1-ketotetrahydroalantolactone (XV).—A solution of 0.5 g. of IV in 100 ml. of acetic acid was refluxed for 5 hr. with 1 g. of lead tetraacetate. The solvent was removed *in vacuo*, and the residue was heated with aqueous sodium bicarbonate solution and extracted with chloroform. The chloroform extract was washed, dried, and concentrated. Repeated crystallization of the residue from ethyl acetate–petroleum ether gave 0.14 g. of needles, m.p. 203–204°; infrared bands at 1775 (γ-lactone), 1745 (acetate), and 1725 cm.$^{-1}$ (ketone). The n.m.r. spectrum (p.p.m.) exhibited signals at 5.72 (quadruplet, X of ABX spectrum, J_{AX} = 12, J_{BX} = 7 c.p.s., H-2), 4.55 (multiplet, H-8), 2.78 (quadruplet, J = 7 c.p.s., H-11), 2.10 (acetate), 1.36 (C-10 methyl), and 1.26 and 1.24 (two doublets, J = 7 c.p.s., C-4 and C-11 methyls).

Anal. Calcd. for $C_{17}H_{28}O_5$: C, 65.80; H, 8.40; O, 25.80. Found: C, 65.60; H, 8.19; O, 26.47.

Reduction of 0.1 g. of XV in 5 ml. of methanol with 0.1 g. of sodium borohydride for 1 hr. at reflux and overnight at room temperature, dilution with water, and extraction with chloroform gave, after recrystallization from ethyl acetate–petroleum ether, 0.035 g. of needles, m.p. 163–164°, undepressed on admixture of an authentic sample of tetrahydroivasperin (XI). The infrared spectra of the two samples were identical.

Extraction of *I. texensis* Jackson.—Extraction of 17.5 lb. of *I. texensis* (above-ground part), collected by Dr. N. C. Henderson on the north end of Galveston Island, Texas, on September 29, 1962, in the usual manner, furnished a gum, approximately 200 g., which was chromatographed over 3 lb. of Alcoa alumina F-20. Elution with benzene, benzene–chloroform, and chloroform gave oils and gums. Elution with chloroform–methanol (19:1) gave solid material which was crystallized from ethyl acetate–petroleum ether, 7.4 g., m.p. 150–151°, identified as ivasperin by mixture melting point and comparison of the n.m.r. and infrared spectra. The identity was confirmed by conversion to diacetylivasperin, m.p. 170–172°, undepressed on admixture with an authentic sample.

A 40-g. aliquot of the less polar gummy fractions (total wt. 240 g. from 35 lb. of plant) was dissolved in benzene–petroleum ether (1:2) and rechromatographed over 600 g. of alumina. Benzene–petroleum ether and benzene eluted oils. Benzene–chloroform (3:1, 2:1, 1:2) eluted semisolid material which was triturated with ether–petroleum ether, filtered, and recrystallized from ether–petroleum ether–acetone, yielding 4.0 g. of asperilin, m.p. 150–151°, undepressed on admixture with an authentic sample, infrared spectra superimposable. Chloroform eluted a noncrystallizable gum and chloroform–methanol (19:1) eluted an additional 1.3 g. of ivasperin. Altogether, 35 g. (0.22%) of asperilin and 29 g. (0.18%) of ivasperin were obtained from 35 lb. of plant material.

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Configurational Studies with 2,3-Dihydroxyoctadecanoic Acids

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The configurations of 2,3-dihydroxyoctadecanoic acids have been proved unambiguously by conversion to 2,3-octadecanediols. The properties of the intermediate cyclic ketals as well as of some other derivatives are compared and discussed. An attempt is made to explain the unusual melting points of long-chain 2,3-dihydroxyalkanoic acids in terms of unusual preferred conformations.

The interrelationship between 2,3-dihydroxyoctadecanoic acids was studied by Myers[1] and was based on Swern's previous work with 9,10-dihydroxyoctadecanoic acids.[2] Using stereospecific methods, Myers could transform the higher melting acid to the lower melting one, but their unusual melting points were probably the reason why he was unable to ascertain which of them had the *erythro* and which the *threo* configuration. In a recent review,[3] it was stated again that the unusual melting points are due to the proximity of the carboxyl group, but no explanation of this effect was given. The problem was, therefore, to determine unambiguously the configurations of 2,3-dihydroxyoctadecanoic acids, and, in addition, to explain some "anomalous" physical properties on the basis of preferred conformations.

The first question was settled by elimination of the bulky carboxyl group which could influence the configuration of the adjacent carbon atom.[4] This was done even though it is known[5] that the attack of a positively charged agent in hydroxylation reactions occurs preferentially on the adjacent carbon atom which is the most electronegative site of the double bond, and that it is preferentially the C-3 atom, the configuration of which might be influenced, and which is electron deficient. In spite of this evidence it seemed desirable to introduce at the end of the molecule a group which by itself would be unable to disturb the normal hydroxylation reaction. For preliminary studies, *trans*-2-octadecenol, prepared by the method of Grob and Jenny,[6] was subjected to the Woodward *cis* hydroxylation method. The product was 1,2,3-octadecanetriol (m.p. 91–94°) identical in all respects with the triol obtained by lithium aluminum hydride reduction of the 2,3-dihydroxyoctadecanoic acid, melting at 127°.[7] Thus it was confirmed that the *trans* double bond is hydroxylated in the same manner when vicinal to a primary hydroxyl or to a carboxyl group.

The most rigorous proof for the *threo* and *erythro* configurations of 2,3-dihydroxyoctadecanoic acids (I),[8] melting at 127 and 108°, respectively, was based on Swern's theoretical considerations for 9,10-dihydroxyoctadecanoic acids[9] and consisted in comparing steric relationships between hydroxyl groups of the two isomeric 2,3-octadecanediols (VII). The configurations of such vicinal diols are known with certainty as a result of lead tetraacetate cleavage experiments made by Criegee, *et al.*[10] Each acid (I) was esterified separately, and the free hydroxyl groups of these esters (II) were protected in the form of cyclic ketals. The ketal esters (III) were then reduced by lithium aluminum hydride to give ketal alcohols (IV), treated with *p*-toluenesulfonyl chloride, and the resulting tosyl esters (V) reduced with lithium aluminum hydride to VI from which the diols (VII) were obtained by acid hydrolysis. (See Scheme I.)

In the most favorable staggered conformation of *erythro*-2,3-octadecanediol (VIII) the hydroxyl substituents are on opposite sides of the axis between the C-2 and C-3 atoms and are, therefore, almost or wholly incapable of intramolecular hydrogen bonding, while *threo*-2,3-octadecanediol (IX), because of the proximity of hydroxyl substituents, allows a higher degree of intramolecular hydrogen bonding in the crystal lattice. Even in very dilute solutions *threo* diols show a greater tendency to form intramolecular hydrogen bonds than *erythro* diols,[11] and the anti-*trans* conformation is less abundant in the *threo* than in the *erythro* derivatives.[12] The diol having the lower melting point (70°) was de-

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