Total Synthesis of Spirilloxanthin, Dehydrolycopene, and 1,1′-Dihydroxy-1,2,1′,2′-tetrahydrolycopene[1]

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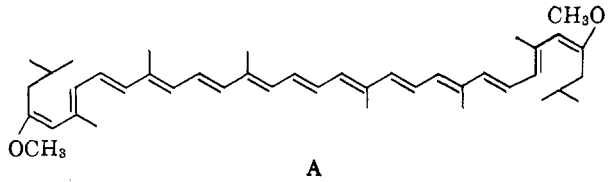
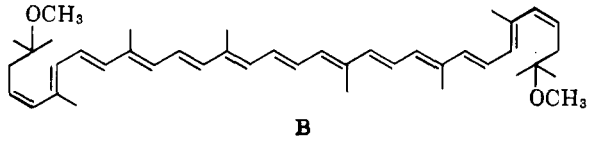
Spirilloxanthin was synthesized by two separate procedures and shown to be identical with an authentic sample of natural spirilloxanthin. Total synthesis for dehydrolycopene and 1,1′-dihydroxy-1,2,1′,2′-tetrahydrolycopene also are reported.

Spirilloxanthin is a dimethoxy carotenoid found in purple bacteria.[2] Zechmeister, *et al.*,[3] found that spirilloxanthin from *Rhodospirillum rubrum* is identical with rhodoviolascin, for which Karrer and Koenig[4] had proposed the tentative structure A.



A

However, this formulation is inconsistent with the compound's resistance to acid hydrolysis. After a study of the n.m.r. spectrum of spirilloxanthin, Barber, *et al.*,[5] proposed the revised structure B.



B

We have now synthesized spirilloxanthin, containing the structure proposed by Barber, by two separate procedures, as outlined in Chart I. The synthetic spirilloxanthin was identical with an authentic sample of spirilloxanthin isolated from *Rhodospirillum rubrum* strain 1,1,1 (from Prof. C. B. von Niel's collection). The n.m.r. spectrum[6] showed the same bands and characteristics cited by Barber, *et al.*,[5] with bands at 6.77 (OCH_3); 7.70 (methylene group with one neighboring proton); 8.03 (methyl group of type $:C(CH_3)\cdot CH:$); and 8.85 μ (methyl groups on a fully substituted carbon). The infrared spectra of the natural and synthetic products were identical, and contained the typical band for methoxyl at 1078 cm.$^{-1}$. The mixture melting point was not lowered.

TABLE I

Spirilloxanthin	Sample	Ultraviolet$_{max}$ mμ
Natural[a]	*trans*	380, 397, 480, 510, 546
	I_2-catalyzed mixture	378, 395, 479, 505, 430
Synthetic[b] spirilloxanthin	*trans*	378, 397, 479, 510, 546
	I_2-catalyzed mixture	378, 397, 478, 506, 431

[a] See ref. 3. [b] See ref. 7.

Iodine catalysis carried out in benzene solution produced the same spectral changes with our synthetic spirilloxanthin as reported for natural spirilloxanthin.

The composition of the iodine-catalyzed equilibrium mixture is presented in Table II.[7] The results were established by paper chromatographic separation on Schleicher and Schüll no. 287 paper with kieselguhr filler.[8] The pigmented zones were eluted with acetone and rapidly determined spectrophotometrically.

TABLE II

Sample	Isomers	Color of zone	Ultraviolet$_{max}$ mμ, in acetone	% of total in I-catalyzed mixture
Natural spirilloxanthin	Neo c	Orange–red	466, 487, 516	5
	Neo b	Red	347, 388, 462, 489, 523	27
	Neo a	Red–purple	374, 388, 462, 489, 523	24
	trans	purple	374, 388, 468, 499, 534	44
Synthetic spirilloxanthin	Neo b (+ c?)	Red–(orange)	373, 389, 465, 490, 523	27
	Neo a	Red–purple	373, 389, 467, 496, 529	29
	trans	Purple	373, 389, 468, 499, 533	44

6-Methoxy-6-methyl-2-heptanone (II) was prepared in 57.6% yield by stirring 6-methylhept-5-en-2-one (I) in a solution of sulfuric acid and methyl alcohol for twenty-four hours at room temperature. Condensation of II with ethyl bromoacetate by the Reformatsky reaction, followed by a dehydration with phosphorus oxychloride in pyridine, yielded 50.2% of 7-methoxy-3,7-dimethyl-2-octenoic acid ethyl ester (III). Reaction of III with N-bromosuccinimide (NBS), followed by dehydrobromination with dimethylaniline, readily formed the unsaturated ester (IV) in 50.0% yield. Compound IV was reduced with lithium aluminum hydride in ethyl ether to give 7-methoxy-3,7-dimethyl-2,4-octadien-1-ol (V) in quantitative yield. The Wittig salt VII was obtained as a white crystalline solid, m.p. 168° (39.6%), by stirring V with VI in methanol for twenty-four hours.

(1) Presented at the 142nd National Meeting of the American Chemical Society, Atlantic City, N. J., September 1962.

(2) P. Karrer and U. Solmssen, *Helv. Chim. Acta*, **18**, 1306 (1935).

(3) L. Zechmeister, *et al.*, *Arch. Biochim.*, **5**, 243 (1944).

(4) P. Karrer and H. Koenig, *Helv. Chim. Acta*, **23**, 460 (1940).

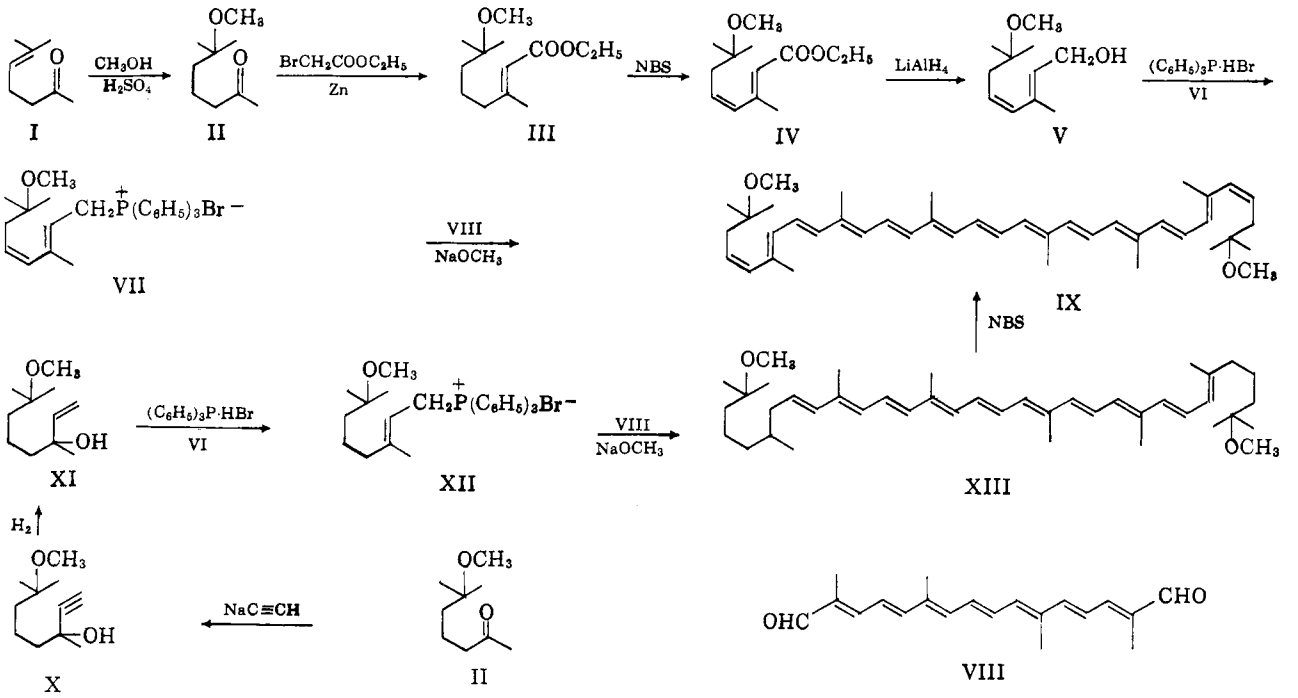
(5) M. S. Barber, L. M. Jackman, and B. C. L. Weedon, *Proc. Chem. Soc.*, 96 (1959).

(6) The nuclear magnetic resonance spectra were determined on deuteriochloroform solutions, and the chemical shifts relative to tetramethylsilane are given in τ units.

(7) Data supplied by S. L. Jensen (Institute of Organic Chemistry, Norges tekniske høgskole, Trondheim, Norway) who kindly assisted in comparing our synthetic spirilloxanthin with natural spirilloxanthin.

(8) A. Jensen and S. L. Jensen, *Acta Chem. Scand.*, **13**, 1863 (1959).

CHART I



The Wittig condensation[9] with crocetin dialdehyde (VIII)[10,11] was carried out in a freshly prepared solution of sodium methylate in methanol. The ylid formed from VII reacted with the dialdehyde VIII on stirring at the reflux temperature of methanol, giving spirilloxanthin (IX) as spindle-shaped violet colored crystals, m.p. 217°, in 30% yield.

For the second synthesis of spirilloxanthin, II was ethynylated with sodium acetylide in liquid ammonia to yield 85% of 7-methoxy-3,7-dimethyl-1-octyn-3-ol (X). Selective hydrogenation of X resulted in 7-methoxy-3,7-dimethyl-1-octen-3-ol (XI) (90%). The Wittig salt XII was prepared in 80% yield by stirring XI and VI in methanol for forty-eight hours.

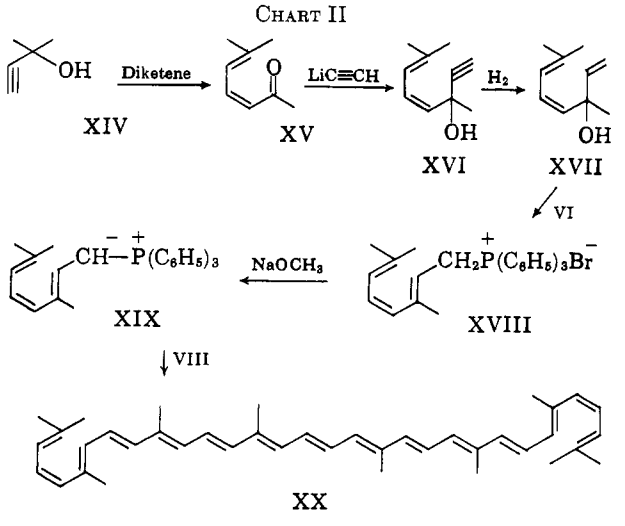
When XII was condensed with VIII, 1,1′-dimethoxy-1,2,1′,2′-tetrahydrolycopene (XIII) was obtained in 60% yield, crystallizing as red needles from pyridine, m.p. 168°. The ultraviolet spectrum had maxima at 503 mμ, $E^{1\%}_{1\,cm}$ 2700; 471 mμ, $E^{1\%}_{1\,cm}$ 2990; 443 mμ, $E^{1\%}_{1\,cm}$ 1968 (petroleum ether).

The reaction of XIII with N-bromosuccinimide, according to the procedure of Entschel and Karrer,[12] resulted in 24% yield of spirilloxanthin (IX).

The structure of dehydrolycopene[13] was established by Karrer in 1945 by a dehydrogenation of natural lycopene. It contains fifteen conjugated double bonds in its system, as compared to eleven conjugated double bonds for lycopene. In 1959 Winterstein[14] showed that dehydrolycopene exists in nature. We have now prepared dehydrolycopene by total synthesis according to the sequence in Chart II.

XIV was condensed with diketene,[15] and the resulting acetoacetate on pyrolysis gave methylheptadienone

CHART II



(XV) in 60% yield. Condensation of XV with lithium acetylide in liquid ammonia resulted in 75% of 3,7-dimethyl-4,6-octadien-1-yn-3-ol (XVI). Hydrogenation of XVI with a selective catalyst[16] gave 3,7-dimethyl-1,4,6-octatrien-3-ol (XVII) (90%).

The Wittig salt XVIII was prepared by condensing XVII and VI in methylene chloride, then recrystallizing from acetone, yield 45%, m.p. 186°. It was converted to the ylid XIX with sodium methylate in methyl alcohol and condensed with VIII to give dehydrolycopene (XX) as a black crystalline solid in 70% yield. The dehydrolycopene was purified by recrystallization from pyridine, in which it formed a brilliant violet-red solution, to give a product melting at 240° dec. The ultraviolet spectrum had maxima at 480, 510, and 540 mμ in petroleum ether, 492, 528, and 568 mμ in pyridine. The synthetic dehydrolycopene was identical with a sample of dehydrolycopene, prepared according to Karrer,[13] with respect to infrared and ultraviolet spectra and mixture melting point.

(9) G. Wittig, *Ber.*, **87**, 1318 (1954); G. Wittig and G. Geissler, *Ann.*, **580**, 44 (1953).

(10) H. H. Inhoffen and G. Raspé, *ibid.*, **592**, 211 (1955).

(11) O. Isler, H. Gutmann, H. Lindlar, M. Montavon, R. Rüegg, G. Ryser, and P. Zeller, *Helv. Chim. Acta*, **54**, 463 (1956).

(12) R. Entschel and P. Karrer, *ibid.*, **41**, 402 (1958).

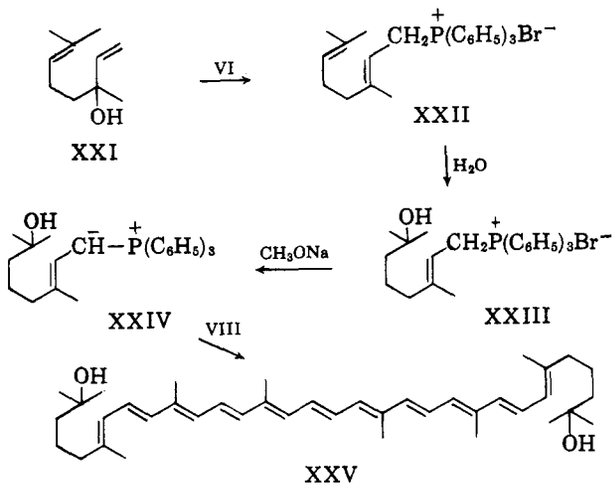
(13) P. Karrer and J. Rutschmann, *ibid.*, **28**, 793 (1945).

(14) A. Winterstein, *et al.*, *Ber.*, **93**, 2951 (1960).

(15) W. Kimel, J. D. Surmatis, J. Weber, G. O. Chase, N. W. Sax, and A. Ofner, *J. Org. Chem.*, **22**, 1611 (1957).

(16) H. Lindlar, *Helv. Chim. Acta*, **35**, 445 (1952).

CHART III



1,1′-Dihydroxy-1,2,1′,2′-tetrahydrolycopene (XXV) was prepared by the scheme in Chart III.

The addition of water to the Wittig salt XXII to form XXIII went with surprising ease in 90% yield. 7-Hydroxy-3,7-dimethyl-2-octenyltriphenylphosphonium bromide (XXIII) was obtained as a stable white crystalline salt, m.p. 194°, after recrystallization from methylene chloride–ethyl acetate.

Condensation of XXIV and VIII in boiling methyl alcohol resulted in 55% of 1,1 -dihydroxy-1,2,1′,2′-tetrahydrolycopene (XXV). as a red crystalline solid, m.p. 190°. The absorption spectra had maxima at 515, 482, and 456 mμ (chloroform); 503, 470, and 444 mμ (petroleum ether); $E^{1\%}_{1\ cm}$ (470 mμ) 3150. A solution of XXV and palmitoyl chloride in pyridine, heated under an atmosphere of nitrogen, resulted in a dipalmitate ester XXVI, m.p. 118°, after recrystallization from acetone; absorption maxima at 499, 468, and 440 mμ (petroleum ether); $E^{1\%}_{1\ cm}$ (468 mμ) 1690. The dihydroxytetrahydrolycopene (XXV) dehydrated on heating with phosphorus oxychloride in pyridine to form *trans*-lycopene (XXVII) in 64% yield, m.p. 171°. The absorption spectrum had maxima at 507 mμ, $E^{1\%}_{1\ cm}$ 3010; 475 mμ, $E^{1\%}_{1\ cm}$ 3370; 447 mμ, $E^{1\%}_{1\ cm}$ 2230. The identity of the lycopene was further confirmed by mixture melting point and infrared absorption spectra.

Experimental[17]

6-Methoxy-6-methyl-2-heptanone (II).—Methyl alcohol (4 l.) was cooled to 10° with an ice bath, and concentrated sulfuric acid (800 ml.) was dropped into the flask with stirring. 6-Methylhept-5-en-2-one (I) (2 kg.) was added to the cold solution and stirred for 8 hr., while the temperature was allowed to rise to room temperature. The contents of the reaction flask were poured onto water (10 l.) in a glass separator provided with an air stirrer. After stirring for several minutes, the oil layer was separated, and the aqueous portion was extracted with benzene. The benzene extract was combined with the oil, and the mixture was washed, dried over Drierite, and fractionated in a packed column with an efficiency of approximately 25 theoretical plates. The yield was 1447 g. (57.6%) of 6-methoxy-6-methyl-2-heptanone (II); b.p. 94° (14 mm.); $n^{25}D$ 1.4285.

Anal. Calcd. for $C_9H_{18}O_2$: C, 68.31; H, 11.46; methoxyl, 19.61. Found: C, 68.45; H, 11.50; methoxyl, 19.86.

Ethyl Ester of 7-Methoxy-3,7-dimethyl-2-octenoic Acid (III).—6-Methoxy-6-methyl-2-heptanone (II) (400 g.), benzene (1,500 ml.), ethyl bromoacetate (600 g.), and potassium bromide (30 g.) were placed in a 5-l., round-bottom flask fitted with a mechanical stirrer, thermometer, and a reflux condenser. The solution was heated to boiling, then granular zinc (300 g.) was added slowly to the reaction mixture, maintaining a vigorous reflux without further external heating. The mixture was stirred until it had cooled to room temperature. It was then transferred to a glass separator, which was provided with an air stirrer, and diluted with water (2 l.). Aqueous 5% sulfuric acid was added to the stirred mixture until the solid, which was formed on dilution with water, was dissolved. The upper oil layer was separated, and the aqueous portion was extracted with benzene. The benzene extract was combined with the oil, and the mixture washed first with water, then with saturated sodium bicarbonate solution. On distillation in a Vigreux column *in vacuo*, there was obtained 448 g. (72.3%) of 3-hydroxy-7-methoxy-3,7-dimethyloctanoic acid ethyl ester; b.p. 117° (0.5 mm.); $n^{25}D$ 1.4439–1.4448.

Dehydration.—Phosphorus oxychloride (172 ml.), pyridine (640 ml.), and toluene (640 ml.) were placed in a 5-l. flask fitted with a mechanical stirrer, thermometer, and a condenser. The previously described hydroxy ester was dropped into the stirred solution in 30 min. Stirring was continued at 95–100° for 2 hr. The reaction mixture was cooled and poured onto ice–water (3 l.). The oil layer was separated and washed with 5% sulfuric acid, then with water, and finally with saturated sodium bicarbonate solution. Fractionation through a packed column, with an efficiency of 10 theoretical plates, afforded 288 g. (50.2%) of ester III; b.p. 75° (0.1 mm.); $n^{25}D$ 1.4480.

Anal. Calcd. for $C_{13}H_{24}O_3$: C, 68.38; H, 10.59; alkoxyl, 33.33. Found: C, 68.28; H, 10.88; alkoxyl, 33.22.

Ethyl 7-Methoxy-3,7-dimethylocta-2,4-dienoate (IV).—Ester III (430 g.), carbon tetrachloride (2,500 ml.), sodium bicarbonate (200 g.), calcium oxide (160 g.), and N-bromosuccinimide (500 g.) were placed, in the order named, in a 5-l. round-bottom flask fitted with an efficient coil condenser, mechanical stirrer, thermometer, and a nitrogen inlet tube. The reaction mixture was heated, while stirring under a blanket of nitrogen, until refluxing started. The external heating was removed, while vigorous boiling continued for about 10 min. When the reaction subsided, external heating was resumed, and the mixture was refluxed for an additional 20 min.

The dehydrobromination of the bromination product obtained was carried out by adding quinoline (400 ml.) to the cooled contents of the flask. The solids present in the mixture were removed by filtration through a fritted glass funnel and washed with carbon tetrachloride. The solvent was distilled, and the residue was heated under an atmosphere of nitrogen for 2 hr. at 95–100°, by means of a water bath. Pyridine (200 ml.) was added, and the heating was continued for 1 hr. longer. The cooled reaction mixture was poured onto water (4 l.) and extracted with petroleum ether. The combined extracts were washed with 5% sulfuric acid, water, and sodium bicarbonate solution. Distillation under vacuum yielded 212 g. (50.0%) of ester IV; b.p. 75–115° (0.3 mm.); $n^{25}D$ 1.4695. A fractionated analytical sample had b.p. 80° (0.15 mm.), $n^{25}D$ 1.4620.

Anal. Calcd. for $C_{13}H_{22}O_3$: C, 68.99; H, 9.79; alkoxyl, 33.62. Found: C, 68.65; H, 9.65; alkoxyl, 33.55.

7-Methoxy-3,7-dimethyl-2,4-octadien-1-ol (V).—Ethyl ether (2 l.) and lithium aluminum hydride (40 g.) were placed in a 5-l. flask and stirred at room temperature for 1 hr. The partial solution was cooled to −10°, and 150 g. of ester IV was dropped into the stirred mixture in 2 hr., while the temperature was maintained at 0° to −10°. The cooling bath was removed and stirring was continued all night.

On the following morning, the reaction mixture was again cooled to −10°, and 5% sulfuric acid was dropped in from a separatory funnel to decompose excess lithium aluminum hydride. The contents of the reaction flask were transferred to a glass separator provided with an air stirrer, and mixed with water (4 l.). Five per cent sulfuric acid was added, while stirring, until the precipitated solid had dissolved. The oil layer was removed and the water layer was extracted with ether. The oil layer was combined with the ether extracts; the solution was washed with water and dried over anhydrous calcium sulfate (Drierite). The ether was removed by distillation under reduced pressure. There was obtained 126 g. (100%) of the crude carbinol V, $n^{25}D$ 1.4730. Fractionation of an analytical sample through a packed column, with an efficiency of approximately 10 theoretical plates, gave material boiling at 78° (0.1 mm.), $n^{25}D$ 1.4682.

(17) The boiling and melting points are uncorrected. The melting points were determined in vacuum capillaries.

Anal. Calcd. for $C_{11}H_{20}O_2$: C, 71.69; H, 10.94; methoxyl, 16.84. Found: 71.55; H, 10.95; methoxyl, 16.89.

Triphenylphosphonium Bromide (VI).—Triphenylphosphine (524 g.) and ethyl acetate (2.5 l.) were placed in a 5-l., three-neck flask fitted with an efficient stirrer. Hydrogen bromide was bubbled into the stirred reaction mixture until 160 g. were consumed. The crystalline product was filtered off by suction, washed with warm (40°) ethyl acetate (2 l.), and dried *in vacuo* at 50°; yield, 550 g. (80%); m.p. 195°. Recrystallization from methylene chloride–ethyl acetate afforded a product of m.p. 202°.

Anal. Calcd. for $C_{18}H_{16}PBr$: C, 62.99; H, 4.70. Found: C, 62.90; H, 4.65.

(7-Methoxy-3,7-dimethyl-2,4-octadien-1-yl)triphenylphosphonium Bromide (VII).—Triphenylphosphonium bromide (VI) (178 g.), methyl alcohol (1 l.) and dienol V (96 g.) were placed in a 2-l. flask provided with a stirrer and nitrogen inlet tube. The mixture was stirred at room temperature, under a blanket of nitrogen, for 48 hr. The reaction mixture was mixed with water (2 l.) and extracted with methylene chloride. After the solvent was removed under reduced pressure, the thick sirup was washed with ethyl ether (500 ml.). Ethyl acetate (1 l.) was added and stirring was continued at room temperature until crystallization started. The mixture was then cooled overnight in a refrigerator. Upon filtering and drying in a vacuum oven (50°) there was afforded 105 g. (39.6%) of VII. An analytical sample, after recrystallization from methylene chloride–ethyl acetate, melted at 168°.

Anal. Calcd. for $C_{29}H_{34}BrOP$: methoxyl, 6.09. Found: methoxyl, 6.14.

Spirilloxanthin (IX).—To a freshly prepared 0.35 *M* solution of sodium methylate in methyl alcohol (600 ml.), the phosphonium bromide VII (50 g.) was added and the solution stirred for 10 min. Crocetin dialdehyde (VIII) (10.0 g.) was dropped in, and the mixture was stirred while heating at reflux temperature for 4 hr. It was cooled overnight in a refrigerator and the reaction product filtered off by suction over a fritted glass funnel. Spirilloxanthin (IX) was obtained as glistening dark violet crystals, m.p. 180–191°. After repeated recrystallization from hot benzene, an analytical sample was obtained; 6.5 g. (30%); m.p. 217°, in a vacuum capillary.

Anal. Calcd. for $C_{42}H_{60}O_2$: C, 84.51; H, 10.12; methoxyl, 10.40. Found: C, 84.81; H, 10.17; methoxyl, 10.37.

7-Methoxy-3,7-dimethyl-1-octyn-3-ol (X).—By the procedure described in an earlier communication,[15] 6-methoxy-6-methyl-2-heptanone (II) reacted with sodium acetylide in liquid ammonia to afford X in 85% yield; b.p. 112° (8 mm.); $n^{25}D$ 1.4544.

Anal. Calcd. for $C_{11}H_{20}O_2$: C, 71.69; H, 10.94; methoxyl, 16.84. Found: C, 72.04; H, 10.73; methoxyl, 17.24.

7-Methoxy-3,7-dimethyl-1-octen-3-ol (XI).—A solution of X (184.3 g.) in petroleum ether (150 ml.) was hydrogenated in the presence of Lindlar catalyst (15 g.) at room temperature until one molar equivalent of hydrogen was consumed. Fractional distillation gave XI, 168 g. (90%); b.p. 107° (8 mm.); $n^{25}D$ 1.4512.

Anal. Calcd. for $C_{11}H_{22}O_2$: C, 70.92; H, 11.91; methoxyl, 16.66. Found C, 71.21; H, 12.06; methoxyl, 16.89.

(7-Methoxy-3,7-dimethyl-2-octen-1-yl)triphenylphosphonium Bromide (XII).—7-Methoxy-3,7-dimethyl-1-octen-3-ol (XI) (186.3 g.), triphenylphosphonium bromide (312.2 g.), and methyl alcohol (1860 ml.) were placed in a 3-l. flask and stirred under a blanket of nitrogen for 48 hr. The solution was poured onto 3 l. of water and extracted with methylene chloride. After the solvent was removed, XII was crystallized from ethyl acetate. There was afforded 384 g. (80%) of (7-methoxy-3,7-dimethyl-2-octen-1-yl)triphenylphosphonium bromide as a white crystalline solid. The analytical sample melted at 168–171°.

Anal. Calcd. for $C_{29}H_{36}BrOP$: methoxyl, 6.07. Found: methoxyl, 5.94.

1,1′-Dimethoxy-1,2,1′,2′-tetrahydrolycopene (XIII).—Lithium (14.0 g.) reacted with bromobenzene (173 g.) in ethyl ether (1 l.). The phosphonium bromide XII (436 g.) was added in several portions in 30 min. Crocetin dialdehyde (80 g.) was added as a fine powder, and the reaction mixture was refluxed for 2 hr. The mixture was stirred into 2 l. of ice-water and the precipitate filtered off. The red solid on the filter was washed with water, methyl alcohol, and petroleum ether. After recrystallization from pyridine and then from benzene, there was afforded 97 g. (60%) of XIII, m.p. 168°.

Anal. Calcd. for $C_{42}H_{64}O_2$: C, 83.94; H, 10.74; methoxyl, 10.33. Found: C, 83.88; H, 10.81; methoxyl, 10.60.

Spirilloxanthin from XIII.—A solution of XIII (10.0 g.) in chloroform (1 l.) was cooled to −15°. *N*-Bromosuccinimide (6.6 g.) and glacial acetic acid (30 ml.) were dissolved in chloroform (800 ml.) and poured at once into the vigorously stirred solution. The stirring was continued for 30 sec. and diethylaniline (80 ml.) was then added. The solution was stirred for 1 hr. at room temperature, under a blanket of nitrogen. After washing with 5% sulfuric acid, water, and sodium bicarbonate solution, the solvent was removed under reduced pressure. Benzene (100 ml.), potassium hydroxide (10 g.), and methyl alcohol (200 ml.) were added to the residue, and this was stirred and refluxed for 1 hr. The solvent was again removed *in vacuo*, and the residue was crystallized from benzene–petroleum ether. After recrystallization from hot benzene, there was afforded 2.4 g. (24%) of spirilloxanthin (IX), m.p. 216–217°. This material was identical with an authentic sample of spirilloxanthin according to mixture melting point, the ultraviolet and visible absorption spectra, and the infrared spectrum.

6-Methyl-3,5-heptadien-2-one (XV).—2-Methyl-3-butyn-2-ol (XIV) (8411 g.) was treated with diketene according to a procedure described in an earlier publication[15] to give the corresponding acetoacetate. Pyrolysis afforded 6-methyl-3,5-heptadien-2-one in yield of 7450 g. (60%); b.p. 63° (3.0 mm.); $n^{25}D$ 1.5332.

Anal. Calcd. for $C_8H_{12}O$; C, 77.37; H, 9.74. Found: C, 77.30; H, 9.65.

3,7-Dimethyl-4,6-octadien-1-yn-3-ol (XVI).—By the procedure described for the preparation of ethynylcarbinols,[15] except that lithium was substituted for sodium, XV was converted to XVI in 75% yield; b.p. 76° (1.5 mm.); $n^{25}D$ 1.5052.

Anal. Calcd. for $C_{10}H_{14}O$; C, 79.96; H, 9.39. Found: C, 79.70; H, 9.30.

3,7-Dimethyl-1,4,6-octatrien-3-ol (XVII).—A solution of XVI (393 g.) in benzene (1 l.) was hydrogenated in the presence of Lindlar catalyst[16] until one molar equivalent of hydrogen was consumed. After removal of the catalyst by filtration and fractionation of the product, there was obtained 356 g. (90%) of XVII; b.p. 56° (0.3 mm.); $n^{25}D$ 1.5024.

Anal. Calcd. for $C_{10}H_{16}O$: C, 78.89; H, 10.59. Found: C, 78.48; H, 10.46.

(3,7-Dimethyl-2,4,6-octatrienyl)triphenylphosphonium Bromide (XVIII).—Triphenylphosphonium bromide (VI) (343 g.) and methylene chloride (1 l.) were placed in a 2-l. flask provided with a stirrer, condenser, thermometer, and a dropping funnel. 3,7-Dimethyl-1,4,6-octatrien-3-ol (XVII) (152.2 g.) was added from a dropping funnel in 30 min. The heat of reaction caused the methylene chloride to reflux. Stirring was continued for an additional 2 hr. The reaction mixture was then stored at room temperature overnight. After the solvent was removed under reduced pressure, the thick brown residue was dissolved in acetone (2 l.) and crystallized at −40°. Recrystallization from acetone afforded 223 g. (45%) of XVIII, m.p. 184°.

Anal. Calcd. for $C_{28}H_{30}PBr$: C, 70.44; H, 6.33. Found: C, 70.20; H, 6.23.

Dehydrolycopene (XX).—To a 0.35 *M* solution of freshly prepared sodium methylate in methyl alcohol (2,400 ml.), there was added 200 g. of the phosphonium bromide XVIII with vigorous stirring. Crocetin dialdehyde (VIII) (40 g.) was dropped in as a solid, and the reaction was stirred under an atmosphere of nitrogen while heating at reflux for 4 hr. It was then cooled overnight in a refrigerator and the reaction product filtered off. After recrystallizing from pyridine, there resulted 50.2 g. (70%) of dehydrolycopene (XX) as a black crystalline solid which decomposed without melting at 240° in an evacuated tube. The absorption spectrum had maxima at 480, 510, and 540 mμ (in petroleum ether); 492, 528, and 568 mμ (in pyridine).

Anal. Calcd. for $C_{40}H_{52}$: C, 90.16; H, 9.83. Found: C, 90.05; H, 9.80.

(3,7-Dimethyl-2,6-octadienyl)triphenylphosphonium Bromide (XXII).—Triphenylphosphonium bromide (VI) (2 kg.) and methyl alcohol (5 l.) were placed in a 12-l. flask provided with an efficient stirrer. Fractionated linalool (XXI) (1 kg.) was dropped into the stirred reaction mixture in 1 hr. Stirring was continued for an additional 20 hr. The reaction mixture was mixed with water (10 l.) and extracted with methylene chloride. After the solvent was removed under reduced pressure, the residue was stirred in ethyl acetate (5 l.), whereby the product crystallized. After filtering and drying *in vacuo*, there was afforded 2,004 g. of XXII, m.p. 170°. The yield was 64.5% based on linalool. An analytical sample, after recrystallization from methylene chloride and ethyl acetate, melted at 189°.

Anal. Calcd. for $C_{28}H_{32}PBr$: C, 70.14; H, 6.73. Found: C, 69.85; H, 6.70.

(7-Hydroxy-3,7-dimethyl-2-octenyl)triphenylphosphonium Bromide (XXIII).—(3,7-Dimethyl-2,6-octadienyl)triphenylphosphonium bromide (XXII) (479 g.) was placed in water (5 l.) and stirred at reflux temperature for 20 hr. The reaction mixture was allowed to cool overnight to room temperature and the product was extracted with methylene chloride. The solution was concentrated *in vacuo* to a small volume, and the residue was induced to crystallize by the addition of ethyl acetate. After cooling in a refrigerator overnight, the product was filtered off and recrystallized from methylene chloride and ethyl acetate. There was obtained 447 g. (90%) of XXIII, m.p. 194°.

Anal. Calcd. for $C_{28}H_{34}OPBr$: C, 67.60; H, 6.89. Found: C, 67.84; H, 6.84.

1,1'-Dihydroxy-1,2,1',2'-tetrahydrolycopene XXV.—By the same procedure described for the preparation of XX, there was obtained from XXIII (1091 g.) and crocetin dialdehyde (VIII) (296 g.), 315 g. (55%) of XXV, m.p. 190°, after recrystallization from pyridine. The absorption spectrum had maxima at 515, 482 and 456 mμ (in chloroform); 503, 470, and 444 mμ (in petroleum ether); $E^{1\%}_{1\,cm}$ (470 mμ) 3150.

Anal. Calcd. for $C_{40}H_{60}O_2$: C, 83.86; H, 10.56. Found: C, 83.94; H, 10.68.

Dipalmitate XXVI.—1,1'-Dihydroxy-1,2,1',2'-tetrahydrolycopene (XXV) (28.6 g.), pyridine (200 ml.), and palmitoyl chloride (50 ml.) were placed in a 1-l. flask and stirred under an atmosphere of nitrogen at 70–75° for 4 hr. The reaction mixture was diluted with methyl alcohol (500 ml.) and cooled for 24 hr. in a refrigerator. The product, which crystallized as a dark red solid, was filtered off under an atmosphere of nitrogen, and washed successively with water and cold methyl alcohol. After recrystallization from acetone, there was obtained 36.8 g. (70%) of XXVI, m.p. 118°; absorption maxima at 499, 468, and 440 mμ (in petroleum ether); $E^{1\%}_{1\,cm}$ (468 mμ) 1690.

Anal. Calcd. for $C_{72}H_{120}O_4$: C, 82.38; H, 11.52. Found: C, 82.44; H, 11.85.

***trans*-Lycopene (XXVII).**—A mixture of pyridine (50 ml.), phosphorus oxychloride (5.0 ml.), and XXV (2.0 g.) was stirred for 1 hr. under an atmosphere of nitrogen at 90–95°. The cooled reaction mixture was diluted with ethyl alcohol (100 ml.) and stored overnight in a refrigerator. The product was filtered off and recrystallized from benzene, giving 1.2 g. (64%) of lycopene (XXVII), m.p. 171°. The absorption spectrum had maxima at 507 mμ, $E^{1\%}_{1\,cm}$ 3010; 475 mμ, $E^{1\%}_{1\,cm}$ 3370; 447 mμ, $E^{1\%}_{1\,cm}$ 2230.

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The Microbiological Hydroxylation of Solasodine[1]

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The steroidal alkaloid solasodine has been hydroxylated by the fungus *Helicostylum piriforme* to yield 9α-hydroxysolasodine, 11α-hydroxysolasodine, 7β-hydroxysolasodine, and probably 7ξ,11α-dihydroxysolasodine. The proofs for their structures are discussed.

The microbial hydroxylation of steroids has been a most powerful tool for the introduction of hydroxyl groups into the steroid molecule, and it has been extensively applied to the C_{19} and C_{21} steroids of adrenal and sex hormonal origin.[3,4] More recently the technique has been extended to the cardiac lactones.[5,6]

We wish now to report the successful hydroxylation of the steroidal alkaloid, solasodine,[7] one of the more important members of this family of alkaloids. The incubation of solasodine (I) with the fungus *Helicostylum piriforme* (A.T.C.C. 8992) resulted in the formation of 9α-hydroxysolasodine (IIa), in amounts ranging from 30 to 35%. Two other monohydroxysolasodines, the 11α-hydroxy, IIIa, and the 7β-hydroxy compound IV were isolated in lesser yields (*ca.* 1% each). A smaller amount (*ca.* 0.5%) of a fourth component with analysis for a dihydroxysolasodine V also was isolated and is tentatively ascribed a 7ξ,11α-dihydroxy structure.

The identity of IIa as 9α-hydroxysolasodine was established by its conversion to 9α-hydroxyprogesterone (IX) by utilizing the degradative procedure previously reported from our laboratory.[8] Careful acetylation of IIa afforded the hydroxy O,N-diacetyl derivative IIb, which was submitted to isomerization by brief treatment with glacial acetic acid, and, without isolation of the unsaturated pseudoderivative, oxidized with chromium trioxide in acetic acid and hydrolyzed with potassium hydroxide in *t*-butyl alcohol[9] to afford a dihydroxy-5,16-pregnadien-20-one (VIa). Catalytic reduction (10% palladium on barium sulfate in ethyl acetate) of the 16-dehydroacetyl derivative VIb to the 16,17-dihydro compound VIIIb, and Oppenauer oxidation of the free alcohol VIIIa afforded the crude hydroxyprogesterone, the purified product of which proved to be identical (melting point, mixture melting point, and infrared spectrum) with an authentic specimen of 9α-hydroxyprogesterone (IX).[10]

During the course of these transformations the pregnadiendiolone acetate (VIb) was also converted into 3β,9α-dihydroxy-5α-pregnan-20-one acetate (VII) by catalytic reduction with Adams' catalyst and oxidation of the resulting C-20 hydroxyl moiety with Kiliani's oxidant.[11]

Compound IIIa was established as 11α-hydroxysolasodine by a similar series of transformations which involved acetylation to the O,O,N-triacetyl derivative

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