Experimental[9,10]

Isolation of Citranaxanthin (I).—The *Sinton citrangequat* fruit was collected when the flavedo had attained its deepest color (bright orange-red). Eight kilograms of peel was separated from the endocarp and extracted with acetone. The carotenoids were partitioned between petroleum ether (b.p. 30–60°) and 90% methanol. The epiphase was submitted to column chromatography on magnesium oxide–Hyflo Supercel (1:1 w./w.). The isolated ketone crystallized from petroleum ether, yielding 90 mg.: m.p. 156–156°; λ_{max} in petroleum ether 463 ($E^{1\%}_{1\,cm}$ 2145) and 495 mμ, in ethanol 475 mμ (shoulder at 489 mμ); infrared bands at 2900, 1662 (conjugated carbonyl), 1590, 1505, 1440, 1280, 1190, 1160, 1025, 960, 900, and 832 cm.$^{-1}$; n.m.r. signals[11] at τ 2.50 (J = 16 c.p.s.), 7.72, 8.02, 8.25, and 8.92.

Anal. Calcd. for $C_{33}H_{44}O$: C, 86.76; H, 9.74. Found: C, 86.6; H, 9.77.

Reaction of I with hydroxylamine hydrochloride–pyridine in ethanol and recrystallization from benzene–petroleum ether afforded the oxime derivative, m.p. 196–197°.

Alkali Cleavage of Citranaxanthin (I).—A solution of 50 mg. of I in 10 ml. of ethanol and 0.5 ml. of 1 *N* potassium hydroxide was distilled (heated at 55–65°) for 20 min. with vigorous stirring in a constant stream of nitrogen into a receiver containing a solution of 2,4-dinitrophenylhydrazine in ethanol. The precipitate was recrystallized from ethanol, furnishing the 2,4-dinitrophenylhydrazone of acetone, m.p. 125–126° (melting point of an authentic sample 125°). Admixture of authentic sample did not depress the melting point.

The nonvolatile mixture was extracted with petroleum ether and chromatographed on a column of magnesium oxide–Hyflo Supercel. Pure II was isolated and crystallized from petroleum ether; m.p. 137–138°. The substance did not depress the melting point of an authentic sample of β-apo-8′-carotenal (m.p. 138–139°), kindly furnished by Hoffmann-La Roche, and was identical with authentic β-apo-8′-carotenal by thin layer chromatography and infrared spectroscopy. The oxime had m.p. 178–179° (melting point of an authentic sample, 178–179°).

Reduction of Citranaxanthin.—To 0.5 mg. of citranaxanthin in 5 ml. of methanol was added 10 mg. of sodium borohydride under nitrogen. The mixture was shaken at *ca.* 10° for 60 min. whereupon a hypsochromic shift was observed. The carotenoids were transferred to petroleum ether. The petroleum ether extract was washed carefully with water and dried over anhydrous sodium sulfate. Chromatography on Microcel C furnished citranaxanthol, λ_{max} in petroleum ether 418, (sh), 442, and 469 mμ.

Citranaxanthin (I).—A solution of 0.5 g. of β-apo-8′-carotenal (II) in 5 ml. of acetone and 5 ml. of ethanol was added drop by drop in an atmosphere of nitrogen to a well-stirred mixture of 0.5 ml. of 1 *N* potassium hydroxide and 5 ml. of ethanol, and the reaction mixture was stirred at room temperature for 5 hr. The petroleum ether extract of the reaction mixture was chromatographed on a column of magnesium oxide–Hyflo Supercel. I was isolated and crystallized from petroleum ether, yielding 0.4 g., m.p. 155–156°, undepressed on admixture of natural citranaxanthin; both samples exhibited the same thin layer chromatographic behavior: λ_{max} in petroleum ether 463 and 495 mμ, in ethanol 475 mμ (shoulder at 488 mμ). The n.m.r. spectrum [singlets at 7.72, 8.02, 8.25, and 8.92; doublet at 2.50 (J = 16 c.p.s.)] is in full accord with structure I. Further proof of identity with natural citranaxanthin was established by infrared spectroscopy.

Anal. Calcd. for $C_{33}H_{44}O$: C, 86.76; H, 9.74. Found: C, 86.6; H, 9.70.

The oxime had a melting point of 196–197°. The substance did not depress the melting point of the oxime of natural citranaxanthin. Retroaldol cleavage with alkali yielded the same products, acetone and β-apo-8′-carotenal (II), as described above for natural citranaxanthin.

Acknowledgment.—The authors are indebted to Dr. David Dreyer of this laboratory for his helpful discussion and assistance in the n.m.r. analysis, and to Dr. J. Furr of the U. S. Department of Agriculture Date and Citrus Experiment Station, Indio, California, and Dr. E. Olson of the U. S. Department of Agriculture Crops Research Division, Weslaco, Texas, for fruit collections.

(9) All melting point determinations were carried out in evacuated capillary tubes on an Electrothermal melting point apparatus and are uncorrected. Infrared spectra were recorded in a KBr disk on Perkin-Elmer Models 137 and 521 spectrophotometers. Visible spectra were measured with a Cary Model 14 spectrophotometer. The n.m.r. spectra were determined in carbon tetrachloride on a Varian A-60 n.m.r. spectrometer, with tetramethylsilane as an internal standard. Analyses were provided by Mr. L. M. White.

(10) Use of trade names of specific materials or equipment does not constitute a recommendation by the U. S. Department of Agriculture to the exclusion of others which may also be available.

(11) Relative areas of n.m.r. peaks were consistent with assignments.

Citrus Carotenoids. III. The Structure of Reticulataxanthin

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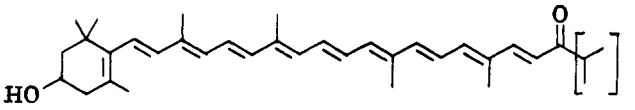
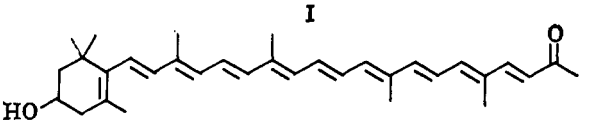
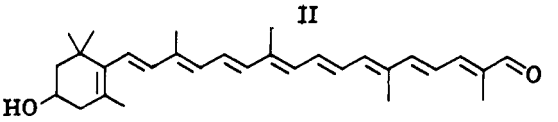
Curl[2] described the isolation and proposed a tentative structure (I) of a carotenoid pigment, reticulataxanthin from the peel of tangerine fruit (*Citrus reticulata*). He deduced structure I through chemical and visible



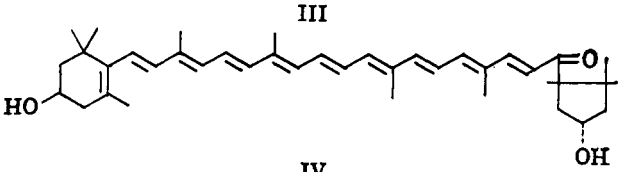
I



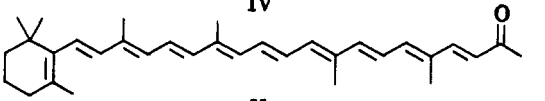
II



III



IV



V

spectral investigations. The nature of the terminal group attached to the carbonyl was uncertain. This ambiguity invited further investigation, and we report herein a more complete structural study of reticulataxanthin.

The pigment used in our study was extracted from the peel of the citrus hybrid, *Minneola tangor* (*Citrus reticulata* × *Citrus sinensis*) which proved to be a much richer source than tangerine. Column chromatography on magnesium oxide–Hyflo Supercel isolated and separated the pigment. Crystallization from peroxide-free ether–petroleum ether (b.p. 30–60°) furnished reticulataxanthin, $C_{33}H_{44}O_2$.

(1) A laboratory of the Western Utilization Research and Development Division, Agricultural Research Service, U. S. Department of Agriculture.

(2) A. L. Curl, *J. Food Sci.*, **27**, 537 (1962).

The infrared spectrum of the isolated pigment exhibited bands at 3450 (hydroxyl) and 1662 cm.$^{-1}$ (conjugated carbonyl). The visible spectrum (Figure 1) of reticulataxanthin indicated a decaenone chromophore similar to that of capsanthin (IV)[3-5] and citranaxanthin (V).[6]

The n.m.r. spectrum revealed a singlet at τ 7.72 which can be assigned to an end-of-chain methyl group α to a carbonyl group. Additionally, no signal could be detected in the τ 0.3–0.6 region characteristic of aldehydic protons with α,β-unsaturation.[7] The doublet at τ 2.50 (J = 16 c.p.s.) indicated that the double bond to which the vinyl proton β to the carbonyl is attached has the *trans* configuration.[8,9]

On treatment with aqueous alcoholic potassium hydroxide reticulataxanthin underwent a retroaldol cleavage to yield acetone and a compound identical with β-citraurin (III).[10,11]

On the basis of results described above, reticulataxanthin can be represented by structure II, but not I. This compound is a 3-hydroxy derivative of citranaxanthin.

Experimental[12,13]

Isolation of Reticulataxanthin.—The fruit of *Minneola tangor* was collected in Riverside, Calif., in Feb. 1964, when in the most highly pigmented stage. The peel (7 kg.) was separated from the endocarp and extracted with methanol. The methanol extract, covered with nitrogen, was then saponified overnight at room temperature. The nonsaponifiable mixture was transferred to ether, washed free of alkali, and evaporated to dryness *in vacuo*. The residue was taken up in methanol, and the pigment mixture was partitioned between 99% methanol and petroleum ether (b.p. 30–60°). Chromatography of the hypophase on a column of magnesium oxide–Hyflo Supercel (1:2 w./w.) isolated the ketone. Crystallization from peroxide-free ether–petroleum ether afforded 160 mg. of the pure ketone: m.p. 171–172°; infrared bands at 3450 (hydroxyl), 1662 (conjugated carbonyl), 1550, 1440, 1380, 1260, 1180, 1020, 970, 890, and 822 cm.$^{-1}$; λ_{max} in petroleum ether 463 and 490 mμ; n.m.r. signals[14] at τ 2.50 (J = 16 c.p.s.), 7.72, 8.02 (in-chain olefinic methyl group), 8.25 (methyl group on C=C in the cyclohexene ring), and 8.92 (*gem*-dimethyl group).

Anal. Calcd. for $C_{33}H_{44}O_2$: C, 83.82; H, 9.41. Found: C, 83.6; H, 9.45.

The oxime, prepared in the usual manner, had m.p. 202–203°. The substance was identical by chromatographic (on both magnesium oxide and deactivated alumina) and visible spectral criteria with reticulataxanthin kindly furnished by Dr. A. L. Curl.

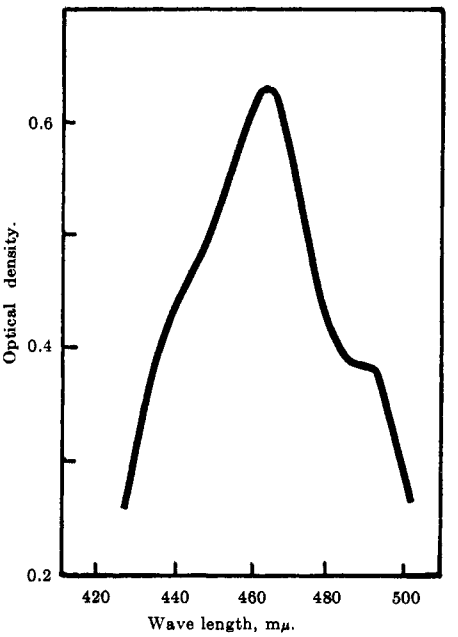


Figure 1.—Visible spectrum of reticulataxanthin in petroleum ether.

Alkali Cleavage of Reticulataxanthin.—A solution of 50 mg. of reticulataxanthin in 10 ml. of ethanol and 0.5 ml. of 1 N potassium hydroxide was heated at 55–65° with vigorous stirring in a stream of nitrogen for 20 min., and the distillate was collected in a receiver containing a solution of 2,4-dinitrophenylhydrazine in ethanol. The precipitate was recrystallized from ethanol to yield the 2,4-dinitrophenylhydrazone of acetone, m.p. 125–126° (melting point of an authentic sample 125°). Admixture of authentic sample did not depress the melting point.

The nonvolatile portion was extracted from the reaction mixture with peroxide-free ether and chromatographed on a column of magnesium oxide–Hyflo Supercel (1:2 w./w.). Pure β-citraurin (III) was isolated and crystallized from peroxide-free ether and petroleum ether, m.p. 145–146° (lit.[9] m.p. 147°). The oxime had m.p. 188–189° (lit.[10] m.p. 188°). Only about 0.1 mg. of β-citraurin, isolated from tangerine peel, was available for comparison. By chromatographic and visible spectral criteria the nonvolatile product obtained from the retroaldol cleavage of reticulataxanthin was identical with natural β-citraurin.

Acknowledgment.—The authors are grateful to Dr. David Dreyer of this laboratory for his helpful discussion and assistance in the n.m.r. analysis and to Dr. Walter Reuther and Mr. Ed Nauer of the University of California at Riverside for the fruit collections.

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(13) Use of trade names of specific materials or equipment does not constitute a recommendation by the U. S. Department of Agriculture to the exclusion of others which may also be suitable.
(14) Relative areas of n.m.r. peaks were consistent with assignments.

Formation of N-Methyl from Reduction of the N-Carbobenzoxy Group with Lithium Aluminum Hydride[1]

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It was anticipated that the $LiAlH_4$ reduction of the N-carbobenzoxy group, which consists of an adjacent

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