The authors wish to thank professor Otto Bastiansen for his inspiring interest in the project and for valuable discussions. Thanks are also due to cand. real. Arne Almenningen for making all the electron diffraction diagrams, to Miss Anne Reitan for skillful technical assistance, and to *Norges Almenvitenskapelige Forskningsråd* for financial support.

1. Trætteberg, M. *Acta Chem. Scand.* **20** (1966) 1724.
2. Almenningen, A., Bastiansen, O. and Fernholt, L. *Kgl. Norske Videnskab. Selskabs, Skrifter* **1958** No. 3.
3. Trætteberg, M. and Bonham, R. A. *J. Chem. Phys.* **42** (1965) 587.
4. Almenningen, A., Bastiansen, O. and Trætteberg, M. *Acta Chem. Scand.* **12** (1958) 1221.
5. Bastiansen, O., Hedberg, L. and Hedberg, K. *J. Chem. Phys.* **27** (1957) 1311.

Received June 30, 1966.

Algal Carotenoids

V. Iso-fucoxanthin — a Rearrangement Product of Fucoxanthin

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The use of seaweed meal as a feed ingredient in rations to poultry prompted us to investigate the fate of fucoxanthin, the characteristic carotenoid of brown seaweeds, when fed to the laying hen. Seaweed meal feeding was found to increase the yellow colour of the egg yolk.[1] Most of the colour was caused by a transformation product of fucoxanthin and no unchanged fucoxanthin could be detected in the egg yolk.

During studies on the structure of fucoxanthin we observed the formation of a similar, probably identical pigment, upon chromatographic purification of fucoxanthin on alkaline adsorbents. Since studies of the latter rearrangement product might shed some light on the nature of the egg yolk pigment, work on the artefact formed on alkaline adsorbents was undertaken.

Fucoxanthin was adsorbed to a column of magnesium oxide from a petroleum ether:acetone (10:1) solution and left on the column overnight at room temperature. Extensive conversion to more polar pigments took place, and no unchanged fucoxanthin could be eluted from the column. The main product, for which we suggest the name iso-fucoxanthin, was rechromatographed on calcium carbonate and gave, after two crystallisations from diethyl ether-petroleum ether dark red-violet needles which melted at 140°C (uncorr.), λ_{max} (430), 453 ($E^{1\,\%}_{1\,cm}$ = 1600), 482 mμ (petroleum ether); λ_{max} 3450, 2940, 1930, 1730, 1606, 1530, 1450, 1355, 1240, 1150, 1030, 960 cm^{-1} (KBr-disc): τ-values: 8.92, 8.75, 8.64, 8.62, 8.46, 8.18, 8.02, and 7.97. The compound was spectroscopically and chromatographically inseparable from one of the main egg yolk pigments obtained from hens on seaweed meal rations.

By following the acetylation of iso-fucoxanthin on paper chromatograms it was established that the compound formed a monoacetate under standard conditions (acetic anhydride-pyridine at room temperature overnight). The R_F-values of the original pigment (0.24, on Schleicher & Schüll, No. 287 paper using petroleum ether containing 10 % of acetone as solvent) and of the acetate (0.46) indicated the presence of (one) more hydroxy group in these compounds over the corresponding members of the fucoxanthin series (R_F = 0.49 for fucoxanthin and R_F = 0.72 for fucoxanthin acetate). In an attempt to establish the character of the additional hydroxy functions the two acetates were treated with hexamethyldisilazane and trimethylchlorosilane and the course of the ether formation was followed by paper chromatography. Samples were taken from the reaction mixtures at intervals and subjected directly to paper chromatography. The triacetate of fucoxanthol *b* [6] was also included in the experiment. The results are shown in Table 1 and indicate that fucoxanthin acetate and fucoxanthol *b* acetate had only one accessible hydroxy group each, while a diether was formed from iso-fucoxanthin acetate.

The mass spectrum of iso-fucoxanthin showed parent ion peak at mass number 658; further peaks at mass numbers 640 (P−18, water); 622 (P−(2 × 18)); 580

Table 1. Trimethylsilylation of fucoxanthin acetate, fucoxanthol *b* acetate, and iso-fucoxanthin acetate.

Reaction time minutes	Starting material R_F 287A2	Rel.amount	Product 1 R_F	Rel.amount	Product 2 R_F	Rel.amount
Fucoxanthin acetate						
0	21	100	—	0		
5	21	73	71	27		
15	21	60	72	40		
30	18	36	72	64		
60	18	17	72	83		
120	—	0	72	100		
Fucoxanthol acetate	R_F 287A5					
0	22	100	—	0		
7.5	23	46	76	54		
15	23	16	76	84		
30	23	8	74	92		
60	—	trace	75	100		
120	—	0	76	100		
Iso-fucoxanthin acetate	R_F 287A5					
0	8	100	—	0	—	0
5	8	54	40	39	98	7
15	7	12.5	42	43.5	98	44
30	9	5	42	31	100	64
60	8	trace	43	11	100	89
120	—	0	40	3	100	97

R_F-values have been multiplied by 100 and are given for the system: Schleicher & Schüll filter paper No. 287, petroleum ether:acetone (50:1, 287A2) and S.S. filter paper No. 287, petroleum ether:acetone (20:1, 287A5)

(P−18−60, water-acetate; 562 (P−(2 × 18)−60) and 544 (P−(3 × 18)−60). The fragmentation pattern thus supported the assumed presence of three hydroxy and one acetoxy groups.

Two structural formulas have recently been suggested for fucoxanthin, namely structure II (see Fig. 1) by Weedon and co-workers [2] and structure I by us,[3] differing only in the substitution pattern of ring B. Mass spectrometry and NMR-data led Weedon's group to suggest the presence of an epoxy function in the 5′,6′-position, while partition and chromatographic properties of fucoxanthin derivatives together with the ready formation of yellow rearrangement products were taken by us to indicate the presence of a tertiary hydroxy group in the 5′-position. The course of the trimethylsilylation of fucoxanthin acetate and of fucoxanthol *b* acetate clearly showed

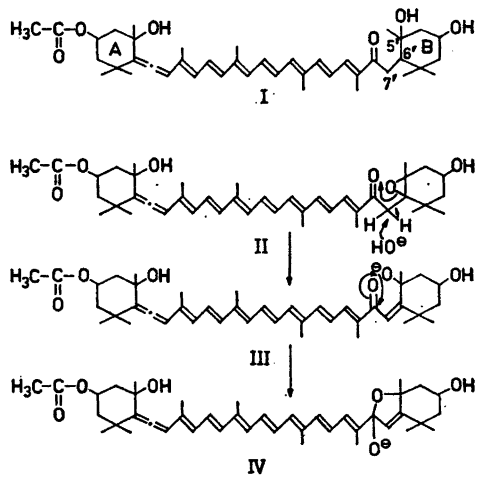

Fig. 1.

that these compounds had only one accessible hydroxy group, and thus ruled out the presence of such a function at C-5′. It was not expected that the short-time treatment of fucoxanthin with lithium aluminium hydride to yield fucoxanthol *b* should open the epoxy ring, since Grob and Siekmann [4] had to reflux β-carotene monoepoxide for 18 h to obtain reasonable quantities of 5-hydroxy-5,6-dihydro-β-carotene.

Structure II thus seems to be a more likely structure for fucoxanthin. We therefore suggest that iso-fucoxanthin can be represented by formula III after the addition of a proton. Its ready formation from fucoxanthin can be explained as an elimination of one acidic proton from position C-7′ by the alkaline adsorbent, followed by opening of the epoxy group and simultaneous rearrangement to the unsaturated alcoholate III, which would add one proton to give iso-fucoxanthin. Further rearrangement of III consisting of a nucleophilic attack by the alcoholate oxygen on the carbonyl-bearing carbon atom would lead to a semi-acetal slightly different from the one suggested by us previously.[3] Both acetals have the same chromophore active in visible light and may equally well explain the formation of pigments adsorbing at 425 mμ reported by Liaaen and Sørensen.[5]

The structure suggested above for iso-fucoxanthin seems to be in good accord with the chemical and physical properties of the compound. Compared with fucoxanthin the chromophore active in visible light has been extended by cross-conjugation to one more carbon-carbon double bond, which is known to give a small increment only in λ_{max}. However, the rearrangement clearly influenced the conjugated carbonyl absorption and the hydroxylic frequency in the infrared region. The increase in polarity and decrease of R_F-values were in good agreement with the postulated transformation of an epoxy function into an unsaturated alcohol. The formation of a di-trimethylsilylether demonstrated that iso-fucoxanthin contained one tertiary hydroxy group more than fucoxanthin and fucoxanthol *b* did, and the mass spectrometric data confirmed the postulated presence of three hydroxy groups in iso-fucoxanthin. In addition the AB-system (doublets at 7.41 and 6.37 τ in the NMR-spectrum) formed by the methylene protons at position 7′ in fucoxanthin could not be detected in the NMR-spectrum of iso-fucoxanthin. Instead a new signal at 3.70 τ indicative of the appearence of a new olefinic proton could be seen. Other small differences in the NMR-spectrum of iso-fucoxanthin when compared with that of fucoxanthin were also consistent with the above structure proposed for the isomerisation product. The formation of iso-fucoxanthin may therefore be regarded as additional evidence in favour of the structure II for fucoxanthin.

Acknowledgements. The author is very grateful to Dr. J. Feeney of Varian Associates Ltd., London, for NMR-spectrum of iso-fucoxanthin and to Drs. O. Isler and U. Schwieter of F. Hoffman-LaRoche, Basle, Switzerland, for the mass spectrum of the same compound.

1. Jensen, A. *Poultry Sci.* **42** (1963) 913.
2. Bonnett, R., Spark, A. A., Tee, J. L. and Weedon, B. C. L. *Proc. Chem. Soc.* **1964** 419.
3. Jensen, A. *Acta Chem. Scand.* **18** (1964) 2005.
4. Grob, E. C. and Siekmann, W. *Helv. Chim. Acta* **48** (1965) 1399.
5. Liaaen, S. and Sørensen, N. A. In Braarud, T. and Sørensen, N. A. *2nd Internat. Seaweed Symp.*, Pergamon, London 1956, p. 25.
6. Jensen, A. *Acta Chem. Scand.* **15** (1961) 1605.

Received June 30, 1966.