XCVI. THE COMPOSITION OF THE FATTY ACIDS PRESENT AS GLYCERIDES IN THE LIVER OIL OF THE THRESHER SHARK (*ALOPOECIA VULPES*).

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In a recent communication [Guha, Hilditch and Lovern, 1930] an account was given of quantitative analyses of the fatty acids of a series of fish-liver oils. When these were subsequently arranged according to their biological families, the oils of the elasmobranch group, including shark, dogfish and skate, lent support to the view that there was a definite and close relation between the amount of squalene present in the non-glyceride portion of the oil and the nature of the fatty acid constituents. When the amount of squalene was high, as in *Scymnorhinus lichia*, the acids were almost exclusively monoethylenic, and contained, in contrast to other fish-liver oils examined, such as those of the Teleostomi group, appreciable amounts of an unsaturated acid containing twenty-four carbon atoms (selacholeic acid). As the percentage of squalene present decreased there was a steady increase in the degree of unsaturation of the fatty acids, together with a steady diminution in the amount of C_{24} acid present, *e.g. Squalus acanthias*, with *ca.* 4 % of squalene (10·5 % unsaponifiable). In the liver oil of the skate (*Raia maculata*) studied, squalene was practically entirely absent, the unsaturation of the C_{20} and C_{22} groups of acids was exceedingly high, and acids of the C_{24} group were not present. It is highly desirable that further information regarding the liver oils of members of this group should be obtained, since the correlation outlined above rests at present on but a few instances.

In this connection the author was fortunate to obtain access to the fatty acid portion of the liver oil of a thresher shark (*Alopoecia vulpes*), the liver of which had been secured by Prof. J. C. Drummond of University College, London, to whom, with Mr L. C. Baker, the writer desires to express his thanks for the information given regarding the crude oil. This particular shark was caught off the Dorset coast on the 18th of June, 1928, and landed at Abbotsbury. The weight of the fish was 4 cwt., and the liver, procured on the 20th of June, weighed 4 kg. The final yield of oil (prepared by steaming) was 650 g. This was of a golden brown colour, with some separation of crystalline matter on standing. The characteristics of the oil were as follows:

% free fatty acids (as oleic)	0·20
Saponification value	181
Iodine value (Dam)	176
Refractive index 20°	1·4741
% unsaponifiable	1·83

The iodine value of the unsaponifiable matter was 102 and it contained 21·8 % of cholesterol; this corresponds with an iodine value for the rest of the unsaponifiable matter of about 112, as against 370 for squalene, so that the amount of squalene (if any) in thresher shark-liver oil must be very low. From this standpoint it is therefore in the same category as skate-liver oil (*Raia maculata*), and, as the following analysis shows, the fatty acid composition is also remarkably similar to that of skate-liver oil. The method of analysis was precisely the same as that given for the other oils [Guha *et al.*, 1930] and only the actual data are given here. It may be noted in passing that in both skate- and thresher shark-liver oils there was very little tendency to polymerisation, although both oils had been kept for over a year before analysis, and both contained a large proportion of acids of a very high degree of unsaturation.

Experimental.

Fatty acids; I.V. 181·4, sap. eq. 289·6.

Lead salt separation.

	g.	%	I.V. (of corresponding methyl esters)
"Solid" acids S	51·0	24·5	38·64
"Liquid" acids L	149·0	74·5	236·3

Fractionation of methyl esters.

Primary fractions					Refractionations				
No.	g.	B.P./1 mm. °C.	S.E.	I.V.	No.	g.	B.P./1 mm. °C.	S.E.	I.V.
(i) *Esters of "solid" acids S* (49·5 g.).									
S 1	35·37	100–130	261·0	10·69					
S 2	5·73	130–145	289·6	73·99					
S 3	8·00	Residue	325·8	143·2					
(Fraction S 1 (16·72 g.) gave 15·00 g. of saturated esters (S.E. 259·4) on oxidation.)									
(ii) *Esters of "liquid" acids L* (145 g.).									
L 1	39·79	127–140	272·1	113·6	L 11	8·49	105–122	249·7	68·0
					L 12	18·92	122–128	270·4	104·4
					L 13	8·07	Residue	292·1	188·8
L 2	34·36	140–158	306·5	218·6	L 21	4·42	120–139	290·6	148·5
					L 22	19·09	139–143	304·5	204·4
					L 23	6·27	Residue	319·6	286·9
L 3	56·44	158–175	329·6	320·9	L 31	4·20	132–165	307·3	276·4
					L 32	29·54	165–170	327·3	320·3
					L 33	10·15	170–175	331·8	342·2
					L 34	5·66	Residue	331·1	326·5
L 4	13·83	Residue	346·3*	290·2					

* Saponification equivalent of residual esters, freed from unsaponifiable matter, L 4 : 337·7.

Estimated composition of fatty acids.

	"Solid" acids S (24·5 %)	"Liquid" acids L (74·5 %)	Total %	Fatty acids %	Fatty acids Mean unsaturation
Saturated					
Myristic	5·8	1·6	7·4	7·4	
Palmitic	11·3	—	11·3	11·3	
Stearic	0·2	—	0·2	0·2	
Unsaturated					
C_{14} group	—	1·6	1·6	1·6	
C_{16} ,,	—	12·0	12·0	12·0	(− 2·0 H)
C_{18} ,,	4·0	15·1	19·1	19·2	(− 3·4 H)
C_{20} ,,	3·7	27·2	30·9	31·0	(− 6·6 H)
C_{22} ,,	0·5	16·8	17·3	17·3	(− 10·5 H)
Unsaponifiable	—	0·2	0·2	—	

It will be observed that the results for thresher shark-liver oil are in entire agreement with the theory already tentatively advanced, but it should be reiterated that the number of cases examined up to the present is insufficient for this generalisation to be regarded as proved. More data are evidently very desirable before any deductions are seriously advanced, but some reference to the many problems suggested by such a relation (if proved) may perhaps be permitted. For instance, is the relation between the squalene content and the fatty acids definite for any particular species? Only one sample of oil from each species has been fully examined, whereas it is known that the unsaponifiable content of particular members of the Elasmobranchi may vary between wide limits. Thus, unsaponifiable matter between the limits of 4 and 32·9 % has been found in the liver oil of the dogfish (*Squalus acanthias*), a mixture of squalene and cholesterol being present in many cases [Grün and Halden, 1929]; liver oils of the basking shark (*Cetorhinus maxima*) have been observed [Grün and Halden, 1929] to contain from 20 % to as much as 56 % of unsaponifiable matter (of which approximately one-half is frequently squalene); whilst, similarly, in oils from livers of *Scymnorhinus lichia* [Channon, 1928] the unsaponifiable content varied from 48·5 % to 81·5 %.

There is thus no reason to assume that skate-liver oil, for instance, will always be found to contain negligible amounts of squalene. Channon [1928] has given evidence which suggests that the squalene present in these fish-liver oils is not taken in the food, but is synthesised by the fish. To mention two results: (1) there is no squalene present in the various samples of plankton examined; (2) the feeding habits of all members of the Squalidae family appear to be similar, and yet squalene is present in large amount in some, and entirely absent from others.

One has also to take into account, if accepting the theory of synthetic origin, the fact that squalene is by no means the only unsaponifiable matter present in elasmobranch oils. Other substances frequently found (in varying amounts, sometimes alone and sometimes with squalene) include hydrocarbons (*e.g. iso*octadecane, decane, etc.), cholesterol, and ethers such as batyl, chimyl and selachyl alcohols. It has yet to be determined whether there is any relation between the presence or absence of these substances and the composition of the accompanying fatty acids.

In conclusion, the author wishes to express his thanks and appreciation to Prof. T. P. Hilditch for his kind interest and advice throughout the course of this work.

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