Separation and Identification of Alkoxyglycerols

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In the liver oils of several Elasmobranch fishes a relatively large proportion of the unsaponifiable matter consists of alkoxyglycerols [1]. Hitherto three such compounds have been isolated, *viz.* chimyl, batyl and selachyl alcohols. Through the use of the technique described below we were able to discover several new alkoxyglycerols.

We separated the unsaponifiable matter of marine oils or of other biological materials by adsorption on a column of activated aluminium oxide. After elution of the hydrocarbons by light petroleum and the cholesterol by methylene dichloride, the alkoxyglycerols were eluted by 10 % methanol in methylene dichloride. The quantity of alkoxyglycerols was then determined gravimetrically. In order to be able to study the composition of the mixtures of the alkoxyglycerols by gas chromatography we converted the free hydroxy groups into methyl ether groups by the method described by Müller and Rundel [2], treating the hydroxy compound with diazomethane and boron trifluoride. Next the dimethyl ethers of the alkoxyglycerols were separated from monomethyl ethers and unchanged alkoxyglycerols by chromatography on aluminium oxide. The dimethyl ethers were eluted by 25 % diethyl ether in light petroleum and afterwards submitted to gas chromatography.

For the gas chromatographic analyses we used a Perkin Elmer apparatus (model 116) with an outlet system modified to permit collecting of fractions of high-boiling substances. Silicone grease and a polyester Reoplex 400 (Geigy) [3] were used as stationary phases and helium as the carrier gas. The samples were analyzed before and after hydrogenation. The components were identified by mass spectrometry and in the case of batyl alcohol also by comparing the retention time with that of synthetic batyl alcohol. The mass spectra of the dimethyl ethers of the alkoxyglycerols will be published later.

Besides glyceryl ethers with hexadecanyl, octadecanyl or octadecenyl as the long-chain component, we found compounds with tetradecanyl, with monounsaturated C_{16}, C_{20} and C_{22} chains, and with a C_{18} chain with two double bonds. There were also small amounts of glyceryl ethers with a long chain containing an odd number of carbon atoms. In Table 1 the composition of the alkoxyglycerols from the liver oils of three marine species of Elasmobranch fish are presented. The chimyl, batyl and selachyl alcohols to-

Table 1. The percentage composition (wt) of the alkoxyglycerols from liver oils. The alkoxyglycerols are represented by the long-chain part of the molecule.

Alkoxyglycerol	Grey dogfish *Squalus acanthias*	Greenland shark *Somniosus microcephalus*	Ratfish *Chimaera monstrosa*
C_{12}	trace	trace	trace
C_{14}, *n*-tetradecanyl	5.7	2.0	1.7
C_{15}	1.9	0.7	1.1
C_{16}, *n*-hexadecanyl (chimyl)	13.2	9.1	10.4
C_{16}, *n*-hexadecenyl	10.6	10.8	9.1
C_{17}	3.0	3.6	4.7
C_{18}, *n*-octadecanyl (batyl)	3.4	2.8	6.7
C_{18}, *n*-octadecenyl (selachyl)	47.8	59.4	53.6
C_{18}, octadecadienyl	2.4	1.6	2.5
C_{18}, octadecatrienyl	trace	?	?
C_{19}, *n*-nonadecenyl	1.2	1.5	2.4
C_{20}, *n*-eicosenyl	8.0	6.2	6.4
C_{21}	trace	?	?
C_{22}, *n*-docosenyl	2.7	2.2	1.0

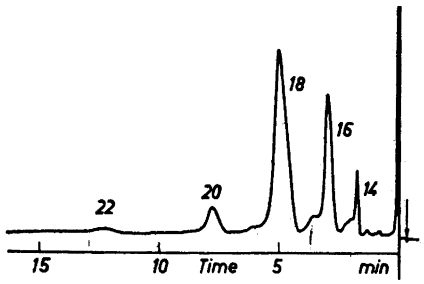

Fig. 1. Gas chromatogram of the dimethyl ethers of the alkoxyglycerols in the liver oil of *Squalus acanthias*. Stationary phase: Silicone grease. Temperature: 265°C. Column length: 2 m. The figures beside the peaks denote the number of carbon atoms in the long-chain part of the molecule.

gether constitute only 64 % of the alkoxyglycerols in the liver of the grey dogfish studied and 71 % in the Greenland shark and ratfish. The rest is composed of alkoxyglycerols not previously identified.

A typical gas chromatogram on silicone of the dimethyl ethers of the alkoxyglycerols in the liver oil from *Squalus acanthias* is shown in Fig. 1. and the corresponding

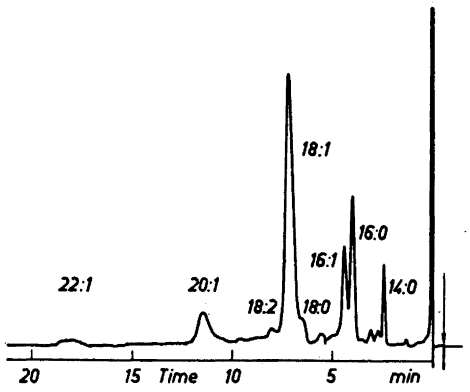

Fig. 2. Gas chromatogram of the dimethyl ethers of the alkoxyglycerols in the liver oil of *Squalus acanthias*. Stationary phase: Reoplex 400. Temperature: 247°C. Column length: 3 m. The figures beside the peaks refer to the long-chain part of the molecule. The figures to the left of the colons denote the number of carbon atoms and the figures after the colon the number of double bonds.

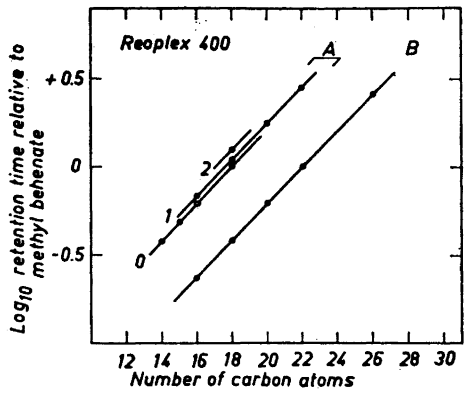

Fig. 3. Relationships between $\log_{10}$ retention times relative to methyl behenate in Reoplex 400 at 247°C and the number of carbon atoms for the alkoxyglycerols (A) and for the saturated straight chain fatty acids (B). For the alkoxyglycerols the number of carbon atoms refer to the long chain part of the molecule. The number of double bonds in the long chains are denoted by 0, 1 or 2.

chromatogram on Reoplex 400 in Fig. 2. The retention time for the component with the highest molecular weight (1,2-dimethoxy-3-docosenoxy-propan) was about 12 min on the silicone and 18 min on the Reoplex column. The logarithms of the retention times on Reoplex 400 relative to methyl behenate were calculated and plotted against the number of carbon atoms (Fig. 3). The saturated ethers fell on a straight line and the mono-unsaturated ones on a line parallell to it.

Oxidation of 1,2-dimethoxy-3-hexadecenoxy-propan by chromium trioxide gave heptanoic acid. This could be established by gas chromatography. Thus the double bond in the hexadecenyl chain is situated between carbon atoms 9 and 10.

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