

# LIPID AND FATTY ACID COMPOSITION OF SOME DEEP-SEA FISHES FROM THE HYDROTHERMAL VENT SITE LUCKY STRIKE

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With 10 tables

*ABSTRACT.* The lipid classes and the fatty acid content and composition of sampled muscles, livers and gonads of some deep-sea fishes (Teleostei: Synbranchidae, Bythitidae, Apogonidae and Selachii: Squalidae) from the hydrothermal vent site “Lucky Strike” were respectively analysed by thin-layer chromatography and gas-liquid chromatography in order to find out eventual lipidic adaptations caused by specific primary producers, the chemosynthetic bacteria of the food chain of thermal vent sites. The distribution of saturated, monounsaturated and polyunsaturated fatty acids and the amounts of eicosapentaenoic acid (C20: 5n-3, EPA) and docosahexaenoic acid (C22: 6n-3, DHA) as well as their ratios were analysed in detail. The results were compared with the fatty acid profile of cod liver oil and with the fatty acid compositions of other deep-sea fishes caught at different depths living away from hydrothermal vent sites. Comparisons with five deep-sea fish species caught at the Portuguese Slope (Algarve, 830 m depth) and at the Madeira Island slope (4000 m depth) were also done.

A quality comparison between the fatty acid profile of the analysed tissues of the hydrothermal vent fishes and cod liver oil showed similar fatty acid compositions. The only major detected feature of lipid composition of deep-sea fishes from the hydrothermal vent site that could be related to their unique food chain was the high content of the monounsaturated fatty acids of the n-7 series (C16: 1n-7 and C18: 1n-7), abundant in bacteria.

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*RESUMO.* As classes de lípidos e o conteúdo e composição em ácidos gordos de amostras de músculos, fígados e gónadas de algumas espécies de peixes profundos (Teleostei: Synbranchidae, Bythitidae, Apogonidae and Selachii: Squalidae) das fontes hidrotermais do campo “Lucky Stricke” foram analisadas respectivamente por cromatografia em camada fina e cromatografia em fase gasosa de modo a procurar eventuais adaptações bioquímicas causadas por produtores primários específicos, as bactérias quimiossintéticas da teia trófica das fontes hidrotermais. A distribuição e quantificação dos ácidos gordos saturados, mono-insaturados e poli-insaturados incluindo os ácidos eicosapentaenoico (EPA, C20: 5n-3) e docosahexaenoico (DHA, C22: 6n-3) foram estudados em detalhe. Os resultados foram comparados com as composições lipídicas de outras espécies de peixes não pertencentes às fontes hidrotermais, nomeadamente o óleo de fígado de bacalhau e outros peixes de diferentes profundidades capturadas nos taludes da costa portuguesa (Algarve, 830 m) e da Ilha da Madeira (4000 m de profundidade).

A comparação qualitativa entre as composições em ácidos gordos dos tecidos analisados das espécies das fontes hidrotermais revelou uma composição lipídica semelhante à do óleo de fígado de bacalhau. A única característica detectada ao nível da composição lipídica dos peixes capturados nas fontes hidrotermais do local “Lucky Strike” que possa resultar das suas singulares cadeias tróficas foram os elevados teores de ácidos gordos mono-insaturados da série n-7 (C16: 1n-7 and C18: 1n-7), abundantes nas bactérias.

PALAVRAS-CHAVE: Ácidos gordos, classes de lípidos, peixes de profundidade, fontes hidrotermais, Crista médio-Atlântica.

## INTRODUCTION

Hydrothermal vent sites have a very specialized biological community. The primary producers are chemosynthetic bacteria that obtain their energy from the synthesis of organic matter by oxidizing hydrogen sulfide contained in the hydrothermal emissions. With reference to the invertebrates, the benthic fauna found at hydrothermal vents is very typical and rich in species. It largely consists of Polychaeta, Bivalvia, Gastropoda, Crustacea Decapoda, Pogonophora and Enteropneusta.

The Lucky Strike vent field, discovered in 1992, is a seamount including active

hydrothermal fields formed between three cones. Venting types range from low-temperature (a few to tens of degrees above ambient) to pooled high-temperature (200° C) water beneath a sulphide flange, to turbulent jets of hot water (292-333° C) from black smoker chimneys (HUMPHRIS *et al.*, 1993). The high temperature fluids are enriched in CH<sub>4</sub> and H<sub>2</sub> and have low sulphide concentrations (< 3.3 mmol/l) (Van DOVER, 1995). Evidence presented by COLODNER *et al.* (1993) and HUMPHRIS *et al.* (1993) supports the hypothesis that Lucky Strike has recently been reactivated. The existence of fresh sulphides on top of older, more weathered and silicified debris were discovered. The biological community is dominated by very rich mussel beds (*Bathymodiolus* sp). Furthermore, bresiliid shrimps and bythograeid crabs were also very abundant, alongside a ubiquitous sea-urchin (diameter: 7 cm) of the genus *Echinus*, ampharetid and free-living polynoid polychaetes, limpets and other gastropods, hydroids, nemertean, tanaids, isopods, ostracods, amphipods and barnacles (Van DOVER *et al.*, 1996).

In contrast to the invertebrates, the fish fauna of the Mid-Atlantic Ridge consists of species which were already known in other abyssal areas (SALDANHA, 1994; SALDANHA & BISCOITO, 1997). Fish species living near those hydrothermal vent sites belong to the end consumers of this special food chain, which differ from those, found in the surface waters where a sufficient supply of light allows a photosynthetic primary productivity.

The objective of this study was to determine of the lipid and fatty acid compositions of muscle, liver and gonad tissues of the deep-sea fishes *Simenchelys parasitica*, *Ilyophis blachei*, *Synaphobranchus kaupi* (Teleostei: Synaphobranchidae); *Cataetyx laticeps* (Teleostei: Bythitidae); *Epigonus telescopus* (Teleostei: Apogonidae) and *Etmopterus princeps* (Selachii: Squalidae). It is of interest to analyse how the unusual primary production of the hot vent community of "Lucky strike" is reflected in the lipid and fatty acid composition of the sampled fish species. The ratios of DHA (C22: 6n-3, docosahexanoic acid) and EPA (C20: 5n-3, eicosapentaenoic acid) fatty acids were determined for all six species in the three tissues (muscle, liver and gonad) and compared among species.

## MATERIALS AND METHODS

### Biological samples

The fish species *Simenchelys parasitica*, *Etmopterus princeps*, *Synaphobranchus kaupi*, *Ilyophis blachei*, *Cataetyx laticeps* and *Epigonus telescopus* were all collected at 1700 m deep in the hydrothermal vent field Lucky Strike on the Mid-Atlantic Ridge (37° 18' N and 32° 17' W) during the Nautilé diving cruise DIVA 2 in June 1994 (Table 1).

**TABLE 1** - List of species analyzed from the Lucky Strike hydrothermal vent field, Mid-Atlantic Ridge.

Species	Family	Catch date	Location	Total Length	Sex	Gonad stage
<i>Simenchelys parasitica</i>	Synaphobranchidae	08.06.1994	37° 17' 42N 32° 16' 74W	350 mm	female	state 3
<i>Etmopterus princeps</i>	Squalidae	08.06.1994	37° 17' 42N 32° 16' 74W	650 mm	female	no sample of gonads
<i>Ilyophis blachei</i>	Synaphobranchidae	10.06.1994	37° 17' 42N 32° 16' 74W	800 mm	female	state 4
<i>Synaphobranchus kaupi</i> (I)	Synaphobranchidae	10.06.1994	37° 17' 42N 32° 16' 74W	630 mm	female	state 3
<i>Cataetyx laticeps</i>	Bythitidae	11.06.1994	37° 17' 42N 32° 16' 74W	675 mm	female	state 4
<i>Epigonus telescopus</i>	Apogonidae	14.06.1994	37° 50' 45N 31° 31' 34W	500 mm	female	no sample of gonads
<i>Synaphobranchus kaupi</i> (II)	Synaphobranchidae	14.06.1994	37° 50' 45N 31° 31' 34W	601 mm	female	state 4

All the species, except *Cataetyx laticeps*, were caught by means of baited traps and dissected immediately after their arrival on board. The organs were frozen in liquid nitrogen until chemical analysis in the laboratory in May 1996. *Cataetyx laticeps* was net caught by the articulated arm of the submersible “Nautilé”. All specimen were female with gonads which were all in an advanced state of development.

For the analyses of samples all the organs were divided in order to have duplicates of each sample. The dorsal muscle, gonad and liver tissues of *S. parasitica*, *I. blachei*, *C. laticeps* and two specimens of *S. kaupi* were analysed. For *E. princeps* there were two muscle samples and one liver sample available for analysis, whereas for *E. telescopus* only one muscle could be analysed. Concerning the analyses of muscle tissue an anticipated distinction between muscles dissected with a fat layer (extant under the skin of the fish) and muscles dissected without a fat layer were done. The distinction was necessary because of clear existing differences in lipid and fatty acid composition between muscles with and without fat layer.

All samples were dried in a vacuum freeze-dryer during 24 h and after stored in an exsicator until lipid extraction.

## **Analysis of the fatty acid composition by GLC**

### **Lipid extraction and trans-methylation of fatty acids**

The extraction of the lipids was done according to the method of BLIGH & DYER (1959). The extraction solvent used was a mixture of methanol, chloroform and water (2: 1: 0.8 (vol/vol)). The homogenization was carried out with a potter (Potter S, B. Braun Company). Chloroform was evaporated by using a rotary evaporator at a temperature of 37° C. The total lipid content was ascertained by gravimetry. The lipid extract which was left behind was dissolved in 2 ml chloroform and 2 ml benzene for gas-liquid-chromatographic analysis or only in 2 ml chloroform for thin-layer chromatographic analysis respectively.

Before storing the samples at -20° C, 100 µl of a standard fatty acid, nonadecanoic acid C19: 0 (10 mg/ml in methanol) was added for later fatty acid quantifications. The tube containing the sample was filled with nitrogen to avoid the oxidation of the fatty acids.

For gas-chromatographic analysis of the fatty acid composition, a trans-methylation was done by adding 14% boro-trifluorid-methanol-complex to the sample and heating in a water bath at 100° C during 45 min (nitrogen atmosphere in the tube). The methylesters were dissolved in 2 ml iso-octane and stored in a nitrogen atmosphere at -20° C until to the injection into the gas chromatograph.

### **Gas chromatographic analysis**

The chromatograph used was a Varian Star 3400 CX (Varian Analytical Instruments; Sugar Land, Texas) with a split/splitless injector (250° C with a split of 10: 1) and a capillary column (0.32 mm ID) with programmed temperature. The carrier gas used was helium (1 ml/min).

The course of the temperature programme was as follows: 180° C during 7 min; heating to 210° C with a velocity of 4° C/min (control velocity: 0.5 cm/min); holding the temperature for 25.5 min and after heating to 220° C (1° C/min) and holding for 10 min. The total running time was 60 min and 30 min for the GLC thermal stabilization.

The injector was a capillary one (Varian 1077 split/splitless). The injection was automatic (Varian autosampler 8100) with the following sandwich method: 0.8 µl lower air gap, 0.5 µl upper air gap, 1 µl solvent plug.

The detection of the methyl fatty acids was done by flame-ionization detection (FID). The chromatograms with the fatty acid composition of several samples were identified by means of comparing a chromatogram of injected cod liver oil in which the composition of fatty acids was known.

**TABLE 2 - Fatty acid composition of the analyzed species. All data are represented as mean ± standard deviation (unit: µg/mg dw). Dash signifies not detected.**

Component	<i>Simenchelys parasitica</i>			<i>Ilyophis blachei</i>			<i>Cataetys laticeps</i>			<i>Epigonus telescopus</i>			<i>Emtopterus princeps</i>			<i>Synaphobranchus kaupii I</i>			<i>Synaphobranchus kaupii II</i>			Cod liver oil
	muscle	gonad*	liver	muscle	gonad	liver	muscle	gonad	liver	muscle 1	muscle 2	liver	muscle	gonad	liver	muscle	gonad	liver				
C13:0	0.07±0.00	---	0.03±0.03	0.03±0.01	0.03±0.03	0.04±0.01	---	0.01±0.01	0.01±0.01	---	---	0.01±0.00	0.02±0.01	---	---	0.03±0.00	0.06±0.01	0.05±0.01	0.32			
C14:0	11.43±0.78	1.33	4.52±1.43	2.72±0.20	3.26±0.43	7.26±1.32	0.06±0.01	1.63±0.05	13.64±1.19	0.10±0.01	0.08±0.03	0.36±0.02	2.43±0.82	7.87±1.30	3.97±0.88	0.16±0.02	3.15±1.32	8.67±0.33	9.69±1.68	46.08		
C15:0	1.50±0.14	0.44	0.93±0.28	0.24±0.15	0.43±0.06	1.05±0.21	0.02±0.01	0.22±0.01	0.49±0.02	0.05±0.01	0.04±0.01	0.11±0.01	0.33±0.09	1.10±0.22	0.37±0.07	0.05±0.00	0.45±0.18	1.27±0.14	1.34±0.18	5.92		
<b>C16:0</b>	<b>35.51±2.14</b>	<b>36.35</b>	<b>31.79±10.1</b>	<b>11.87±0.09</b>	<b>27.25±3.98</b>	<b>33.41±5.88</b>	<b>1.66±0.08</b>	<b>14.01±0.30</b>	<b>53.85±4.79</b>	<b>2.97±0.31</b>	<b>2.71±0.16</b>	<b>4.78±0.08</b>	<b>10.82±2.86</b>	<b>35.65±6.13</b>	<b>29.04±6.49</b>	<b>4.49±0.31</b>	<b>12.10±4.68</b>	<b>31.45±1.15</b>	<b>38.32±5.21</b>	<b>143.59</b>		
C17:0	1.00±0.04	1.17	0.65±0.16	0.35±0.02	0.42±0.08	0.97±0.17	0.03±0.01	0.34±0.01	0.65±0.04	0.05±0.00	0.09±0.01	0.18±0.01	0.31±0.09	1.03±0.15	0.47±0.11	0.09±0.02	0.43±0.17	0.92±0.04	1.44±0.16	4.02		
C18:0	5.74±0.28	10.95	4.55±1.06	2.21±0.03	5.00±0.75	7.71±1.25	0.43±0.00	4.60±0.04	19.53±1.46	0.75±0.08	0.81±0.06	1.09±0.04	2.24±0.79	8.15±1.36	5.48±1.18	1.14±0.05	2.58±0.91	4.65±0.17	9.12±0.98	23.07		
C20:0	0.95±0.20	0.30	0.17±0.06	0.13±0.03	0.12±0.02	0.56±0.28	0.02±0.00	0.31±0.01	0.57±0.03	---	0.02±0.01	0.06±0.01	0.09±0.01	0.75±0.53	0.63±0.13	0.05±0.01	0.27±0.17	0.46±0.15	0.37±0.11	3.22		
C22:0	0.01±0.01	---	0.27±0.01	0.07±0.03	0.18±0.07	0.05±0.01	0.15±0.01	0.28±0.01	0.05±0.00	0.02±0.02	---	0.07±0.01	0.04±0.05	0.20±0.21	0.54±0.11	0.35±0.02	0.06±0.06	0.02±0.00	0.01±0.01	0.04		
<b>Saturates</b>	<b>56.53±3.57</b>	<b>50.54</b>	<b>42.91±13.1</b>	<b>17.62±0.26</b>	<b>36.72±5.42</b>	<b>51.13±9.14</b>	<b>2.37±0.08</b>	<b>21.43±0.36</b>	<b>88.90±7.55</b>	<b>3.93±0.38</b>	<b>3.73±0.28</b>	<b>6.64±0.18</b>	<b>16.29±4.63</b>	<b>54.85±9.51</b>	<b>40.53±8.97</b>	<b>6.34±0.40</b>	<b>19.08±7.38</b>	<b>47.54±1.57</b>	<b>60.37±8.12</b>	<b>228.00</b>		
C14:1n-5	0.79±0.06	---	0.12±0.05	0.07±0.01	0.11±0.00	0.34±0.05	---	0.02±0.00	0.94±0.06	---	---	0.01±0.00	0.07±0.01	0.36±0.08	0.01±0.01	---	0.11±0.05	0.28±0.01	0.23±0.06	3.28		
C16:1n-9	6.58±0.50	---	2.94±0.98	0.04±0.00	0.11±0.01	0.09±0.01	---	---	0.10±0.00	---	---	0.01±0.00	0.04±0.01	0.10±0.04	---	---	0.07±0.03	0.23±0.01	0.33±0.05	28.72		
<b>n-7</b>	<b>28.18±2.13</b>	<b>10.28</b>	<b>19.25±7.03</b>	<b>6.32±0.35</b>	<b>12.22±1.94</b>	<b>24.36±4.63</b>	<b>0.56±0.18</b>	<b>20.61±0.88</b>	<b>51.46±4.39</b>	<b>0.28±0.04</b>	<b>0.59±0.27</b>	<b>1.23±0.04</b>	<b>6.10±2.15</b>	<b>25.33±5.37</b>	<b>45.28±9.67</b>	<b>1.45±0.41</b>	<b>6.81±2.79</b>	<b>22.60±0.76</b>	<b>21.42±3.51</b>	<b>114.08</b>		
n-5	0.36±0.01	---	0.28±0.07	0.09±0.01	0.12±0.03	0.27±0.05	0.03±0.01	0.26±0.01	0.16±0.01	0.01±0.00	0.01±0.01	0.02±0.00	0.08±0.03	0.28±0.06	0.50±0.11	0.07±0.03	0.13±0.06	0.44±0.03	0.36±0.06	1.52		
C17:1n-8	2.58±0.18	1.35	1.94±0.86	0.75±0.05	1.61±0.26	2.27±0.39	0.05±0.03	0.51±0.01	2.56±0.14	0.07±0.01	0.09±0.00	0.25±0.01	0.87±0.40	2.37±0.49	1.54±0.32	0.13±0.08	0.93±0.35	2.98±0.11	3.48±0.47	10.54		
C18:1n-9	98.39±5.20	27.13	64.18±24.0	23.40±1.22	57.74±7.95	85.56±17.2	0.89±0.10	16.62±0.13	244.18±38.4	1.08±0.14	1.89±0.25	7.80±0.22	21.57±6.67	91.13±17.8	46.55±10.2	2.39±0.33	25.87±11.0	80.42±3.77	117.50±18.50	396.91		
<b>n-7</b>	<b>10.92±1.36</b>	<b>7.67</b>	<b>10.87±3.62</b>	<b>3.26±0.16</b>	<b>10.35±1.71</b>	<b>14.44±2.61</b>	<b>0.91±0.04</b>	<b>15.95±0.29</b>	<b>12.99±18.37</b>	<b>0.20±0.01</b>	<b>0.49±0.16</b>	<b>1.26±0.06</b>	<b>3.25±1.17</b>	<b>15.57±2.90</b>	<b>37.64±7.94</b>	<b>2.41±0.19</b>	<b>2.58±0.98</b>	<b>7.60±0.93</b>	<b>11.66±2.09</b>	<b>45.73</b>		
n-5	0.91±0.04	0.53	0.71±0.22	0.19±0.01	0.41±0.06	0.69±0.13	0.02±0.00	0.54±0.01	0.68±0.05	0.02±0.00	0.04±0.01	0.07±0.01	0.17±0.06	2.57±0.49	1.20±0.25	0.06±0.01	0.40±0.16	1.11±0.12	1.04±0.16	3.75		
C19:1n-10	1.64±0.16	0.31	0.89±0.30	0.29±0.01	0.57±0.08	0.96±0.10	0.01±0.00	0.72±0.01	0.95±0.08	0.01±0.00	0.02±0.01	0.13±0.01	0.26±0.07	0.88±0.04	1.37±0.27	0.04±0.01	0.37±0.14	1.13±0.06	1.72±0.08	6.41		
n-8	0.75±0.07	0.30	0.42±0.13	0.17±0.00	0.47±0.08	0.59±0.00	0.01±0.00	0.16±0.01	0.79±0.07	0.02±0.01	0.02±0.01	0.13±0.00	0.16±0.04	0.83±0.30	0.32±0.07	0.02±0.00	0.20±0.06	0.48±0.02	0.74±0.03	2.89		
<b>C20:1n-9</b>	<b>58.54±6.60</b>	<b>5.19</b>	<b>22.09±5.84</b>	<b>13.81±1.49</b>	<b>14.45±2.19</b>	<b>22.45±4.34</b>	<b>0.19±0.04</b>	<b>3.10±0.01</b>	<b>20.96±0.83</b>	<b>0.16±0.04</b>	<b>0.35±0.07</b>	<b>4.96±0.36</b>	<b>11.62±5.76</b>	<b>24.30±4.66</b>	<b>11.76±2.56</b>	<b>0.50±0.11</b>	<b>15.24±6.19</b>	<b>30.04±1.04</b>	<b>32.86±3.83</b>	<b>216.96</b>		
n-7	1.58±0.05	1.15	0.67±0.21	0.32±0.02	0.45±0.16	0.85±0.57	0.08±0.01	2.59±0.01	1.18±0.10	0.02±0.00	0.07±0.04	1.26±0.06	3.30±0.08	3.23±0.67	6.59±1.41	0.21±0.01	0.41±0.19	0.90±0.06	1.07±0.20	6.61		
n-5	0.66±0.05	0.28	0.43±0.16	0.02±0.00	0.12±0.02	1.13±0.23	0.02±0.00	0.51±0.01	0.70±0.06	0.01±0.01	0.02±0.01	0.07±0.01	0.02±0.01	1.37±0.30	1.53±0.32	0.06±0.01	0.04±0.03	0.10±0.00	0.25±0.07	2.67		
C22:1n-11	14.49±0.15	1.16	1.55±0.32	4.41±0.39	0.78±0.12	8.83±1.79	0.05±0.01	0.16±0.00	1.81±0.08	0.03±0.01	0.05±0.04	2.76±0.17	4.52±2.20	9.98±2.20	1.60±0.34	0.10±0.04	10.93±4.78	15.54±0.77	12.56±2.23	55.10		
n-9	4.54±0.40	0.66	1.77±0.48	1.39±0.13	1.42±0.21	3.50±0.61	0.05±0.00	0.33±0.03	3.40±1.27	0.02±0.01	0.04±0.01	0.87±0.06	1.50±0.74	4.24±0.91	2.26±0.49	0.11±0.02	1.56±0.03	5.18±0.52	5.18±0.69	16.41		
C24:1n-9	2.86±0.19	0.47	0.91±0.09	0.88±0.02	0.89±0.14	3.59±0.92	0.08±0.01	0.55±0.04	1.65±0.15	0.13±0.00	---	0.86±0.06	0.97±0.33	3.72±0.90	1.32±0.28	0.20±0.01	1.02±0.46	1.28±0.06	2.49±0.69	10.32		
n-7	0.64±0.18	---	0.05±0.03	0.06±0.02	0.06±0.01	0.18±0.04	0.01±0.00	0.11±0.01	0.09±0.01	0.01±0.01	---	0.27±0.03	0.07±0.04	0.34±0.28	0.70±0.09	0.04±0.01	0.06±0.03	0.12±0.01	0.18±0.14	0.43		
<b>Monounsaturates</b>	<b>234.4±17.6</b>	<b>56.48</b>	<b>129.0±44.2</b>	<b>55.41±3.85</b>	<b>101.8±14.9</b>	<b>172.1±33.6</b>	<b>2.93±0.08</b>	<b>62.71±1.40</b>	<b>344.55±27.3</b>	<b>2.04±0.21</b>	<b>3.65±0.83</b>	<b>20.74±1.00</b>	<b>51.54±19.57</b>	<b>184.73±7.1</b>	<b>160.1±34.3</b>	<b>7.74±1.03</b>	<b>66.73±27.7</b>	<b>167.15±7.4</b>	<b>213.0±32.4</b>	<b>922.33</b>		
C16:2n-4	2.81±0.12	1.64	2.23±0.69	0.60±0.03	1.20±0.20	1.34±0.30	0.05±0.02	0.47±0.04	1.45±0.06	0.05±0.00	0.10±0.04	0.28±0.02	0.53±0.16	1.43±0.06	0.80±0.37	0.13±0.04	0.84±0.35	2.22±0.09	2.93±0.29	11.30		
C16:3n-4	0.87±0.05	2.39	0.82±0.20	0.26±0.09	0.54±0.09	0.75±0.17	0.04±0.03	0.25±0.00	0.77±0.09	0.05±0.01	0.21±0.06	0.21±0.05	0.49±0.44	0.78±0.16	0.28±0.07	0.12±0.01	0.45±0.13	5.36±0.16	1.72±0.16	3.57		
C16:3n-3	0.03±0.01	0.58	0.07±0.00	0.04±0.01	0.03±0.04	0.05±0.01	0.03±0.02	0.14±0.03	0.04±0.05	0.01±0.01	0.04±0.00	0.06±0.01	0.03±0.04	0.04±0.01	0.13±0.02	0.09±0.01	0.09±0.03	0.02±0.00	0.05±0.01	0.12		
C16:4n-3	0.12±0.01	---	0.11±0.01	0.03±0.01	0.16±0.08	0.08±0.01	---	0.01±0.00	0.11±0.06	0.02±0.03	---	0.01±0.00	0.20±0.21	0.09±0.01	0.07±0.01	---	0.08±0.02	0.10±0.01	0.13±0.04	0.49		
C16:4n-1	0.14±0.00	4.67	0.22±0.03	0.09±0.01	0.10±0.02	0.17±0.02	0.05±0.01	0.18±0.05	0.07±0.02	0.02±0.02	0.31±0.06	0.33±0.06	0.12±0.04	0.18±0.02	0.05±0.01	0.13±0.01	0.14±0.04	0.29±0.03	0.11±0.05	0.58		
C18:2n-6	2.73±0.45	1.27	1.88±0.87	0.89±0.08	0.91±0.15	2.40±0.42	0.08±0.01	0.55±0.00	1.33±0.07	0.09±0.00	0.21±0.01	0.21±0.01	0.96±0.42	2.57±0.49	0.77±0.17	0.19±0.01	1.18±0.46	3.53±0.04	2.76±0.08	10.44		
C18:3n-3	0.32±0.09	---	0.47±0.08	0.04±0.03	---	0.18±0.13	0.01±0.00	0.06±0.00	0.13±0.02	---	0.04±0.04	0.02±0.01	0.02±0.01	0.22±0.21	---	---	0.07±0.06	0.09±0.05	0.36±0.02	1.04		
C18:4n-3	0.59±0.19	---	0.09±0.04	0.03±0.01	0.03±0.01	0.15±0.11	---	0.05±0.01	---	---	0.13±0.14	0.04±0.04	0.02±0.00	0.21±0.21	0.31±0.06	---	0.10±0.10	0.16±0.06	0.13±0.11	1.86		
C20:2n-6	0.97±0.01	0.80	0.35±0.28	0.33±0.02	0.46±0.07	1.45±0.04	0.06±0.01	0.50±0.01	0.70±0.02	0.03±0.01	0.08±0.02	0.18±0.01	0.27±0.08	2.02±0.12	0.88±0.18	0.16±0.02	0.31±0.12	0.79±0.02	0.98±0.06	3.83		
C20:3n-6	0.14±0.03	0.34	0.08±0.02	0.04±0.01	0.05±0.01	0.43±0.01	0.04±0.01	0.14±0.00	0.06±0.01	0.02±0.00	0.02±0.01	0.05±0.02	0.83±0.14	0.38±0.08	0.11±0.04	0.05±0.01	0.07±0.00	0.06±0.01	0.69±0.01	0.59		
C20:4n-6	1.91±0.04	11.62	4.17±1.85	1.92±0.34	2.46±0.43	3.31±0.21	0.33±0.01	3.85±0.01	5.31±0.72	0.63±0.02	1.00±0.11	1.10±0.02	1.87±0.88	4.07±0.66	3.04±0.57							

## Analysis of the lipid class composition by TLC

### Separation of neutral lipids

The solvent system used to separate neutral lipids was petroleum spirit (40-60° C), peroxide-free diethylether and acetic acid in a ratio of 85: 15: 1 (vol/vol). The chamber was lined with filter paper and kept closed during at least 1 h to saturate the atmosphere inside the chamber with solvent.

The pre-coated TLC plates used (Silica gel 60 F-254) had a layer thickness of 0.5 mm and were activated during 1 h at 110° C in an oven (Memmert, Germany). After the plates cooled down the samples were applied (20 µl). The standard used contained cholesterol (20%), cholesteryl oleate (20%), methyl oleate (20%), oleic acid (20%) and triolein (20%), and was also applied at 20 µl per plate.

After about 30 min of running time in the chamber, the plates were charred with 10% phosphomolybdic acid in ethanol. After charring, the plates were heated for 5 min at 120° C and then scanned by a densitometer registering transmittance (Transmittance / Reflectance scanning densitometer, Hoefer Scientific Instruments, San Francisco) for quantification. Finally, the integration was done using the software Varian Star Chromatography Workstation Version 3 (1993).

### Separation of polar lipids

The procedure was the same as for separation of neutral lipids. The choosed mobile phase was a mixture of chloroform, methanol and water (65: 25: 4 (vol/vol)). The standard used for the qualification of the spots contained sphingomyelin, phosphatidyl choline, phosphatidyl serine and phosphatidyl ethanolamine (2 mg/ml in a 1: 1 mixture of chloroform and methanol). The plates were charred with molybdenium blue. The quantification and integration was carried out by the scanning densitometer and the same software as above was used.

## RESULTS

### Fatty acid composition

The fatty acid composition of muscle, gonad and liver of *S. parasitica*, *E. princeps*, *I. blachei*, *S. kaupi* (I) + (II), *C. laticeps* and *E. telescopus* is shown in Table 2 (expressed in µg/mg dw).

In *Simenchelys parasitica* the total identified fatty acid content was 318.7 µg/mg dw in the muscle (with fat layer), 209.1 µg/mg dw in the gonad and 201.1 µg/mg dw in the liver. Monounsaturated fatty acids dominated in muscle (73.5%) and liver (64.2%)

while the content of polyunsaturated fatty acids was relatively low (muscle: 5.7%; liver: 11.2%). These values differed from the gonad tissue where the polyunsaturated fatty acids constituted the main part (45.4%) beside 27.0% of monounsaturated fatty acids and 24.2% of saturated fatty acids.

The concentration of high unsaturated fatty acids (HUFA) was clearly greater in gonad (42.1%) when compared to muscle (5.4%) and liver (10.4%) tissues.

The most abundant fatty acid in muscle and liver was oleic acid C18: 1n-9 (muscle: 30.9%; liver: 31.9%) while C18: 1n-9 took up the third place in abundance in gonad tissue (13.0%). C22: 6n-3 showed the highest concentration in the gonad of *S. parasitica*, followed by C16: 0 (17.4%). C16: 0 and C20: 1n-9 were also very abundant in muscle and liver tissues besides oleic acid (C16: 0 – muscle, 11.1%; liver – 15.8%; C20: 1n-9 - muscle: 18.4%, liver: 11.0%). C18: 2n-6 and C20: 4n-6 existed only in small amounts in the analysed tissues (between 0.6 to 0.9%) but C20: 4n-6 showed higher amounts in gonad (5.6%) and liver (2.1%).

The concentration of eicosapentaenoic acid C20: 5n-3 was very low in muscle (0.6%) and liver (0.9%) while the gonad contained 2.8% of this fatty acid. This relation fitted also to the content of docosahexanoic acid C22: 6n-3. The amount in muscle (3.8%) and liver (4.9%) was clearly lower than in the gonad (28.3%). These results are reflected in the ratio of DHA/EPA. In muscle this value is about 6.5, in liver 5.5 and in gonad 10.1.

In *Ilyophis blachei* the total identified fatty acid content amounted to 95.3 µg/mg dw in muscle, 159.2 µg/mg dw in gonad and 274.43 µg/mg dw in liver. This species showed in all three tissues a clear domination of monounsaturated fatty acids (muscle: 58.1%; gonad: 64.0%; liver: 62.7%) whereas the concentration of polyunsaturated and saturated fatty acids were similar, in a range of 10.5 to 23.1%. The value of HUFA was greater in muscle (20.4%) followed by 16.1% in liver and 10.2% in gonad. The most abundant fatty acid in all three tissues was C18: 1n-9 (muscle: 24.6%; liver: 31.2%; gonad: 36.3%). C16: 0 was also very abundant in gonad (17.1%) and liver (12.2%), C22: 6n-3 in liver (9.2%) and muscle (12.8%) and C20: 1n-9 in muscle (14.5%) and gonad (9.1%). For C18: 2n-6 and C20: 4n-6 all tissues showed similar values (C18: 2n-6: 0.6 to 0.9% (see *S. parasitica*); C20: 4n-6: 1.2 to 2.0%). Also the concentrations for eicosapentaenoic acid were on the same level (muscle: 2.5%; gonad: 1.2%; liver: 2.6%). In contrast to *S. parasitica* the muscle (12.8%) and the liver (9.2%) of *I. blachei* showed a high amount of C22: 6n-3. The gonad contained 5.9%. The DHA/EPA ratio was 5.2 in muscle, 4.8 in gonad and 3.5 in liver.

Two specimen of *Synaphobranchus kaupi* were analysed. The total identified fatty acid content in *S. kaupi* I was 298.2 µg/mg dw in muscle, 228.2 µg/mg dw in gonad and 26.1 µg/mg dw in the liver whereas in *S. kaupi* II 108.9 µg/mg dw in muscle, 283.0 µg/mg dw in gonad and 307.5 µg/mg dw in liver could be identified.

In both *S. kaupi* I and II monounsaturated fatty acids (content ranged from 59.1

to 70.2%) dominated in all tissues with the exception of the liver of *S. kaupi* I where the highest amount belonged to the polyunsaturated fatty acids (43.5%). Thus, the HUFA of this liver tissue was high (42.7%). In contrast to this value the liver of *S. kaupi* II showed only a HUFA concentration of 7.4%. The content of HUFA in the remaining gonad and muscle tissues of *S. kaupi* I and II ranged from 10.2 to 18.9%.

The most abundant fatty acid was C18: 1n-9. In muscle, gonad and liver tissue of *S. kaupi* II, and in muscle and gonad tissue of *S. kaupi* I the concentration of this monounsaturated fatty acid ranged from 20.4 to 38.2%. In the liver of *S. kaupi* I the dominating fatty acid was C22: 6n-3 (31.3%). In the gonad of *S. kaupi* I a high content of C16: 1n-7 (19.8%) and C18: 1n-7 (16.9%) was found. C18: 1n-7 was also very abundant in the liver tissue of *S. kaupi* I (9.2%).

In agreement with a high concentration of polyunsaturated fatty acids in the liver of *S. kaupi* (I), this tissue showed also a high concentration of eicosapentaenoic acid (5.4%) and docosahexaenoic acid (31.3%), whereas the liver of *S. kaupi* II had lower amounts of eicosapentaenoic acid (0.6%) and docosahexaenoic acid (4.4%). In the gonad and muscle of both specimens the amount of eicosapentaenoic acid ranged from 1.9 to 3.9% and of docosahexaenoic acid from 9.1 to 11.5%. The ratio DHA/EPA had, for the different tissues in both specimens, comparable mean values (muscle: 3.5; gonad: 2.7; liver: 6.4), while liver showed the highest value and gonad the lowest.

The total amount of identified fatty acids in *Cataetx laticeps* was 9.7 µg/mg dw in the muscle, 113.8 µg/mg dw in the gonad, and 476.37 µg/mg dw in the liver. In the gonad and liver tissues the monounsaturated fatty acids dominated (gonad: 55.1%; liver: 72.3%), whereas the muscle had 43.1% polyunsaturated fatty acids (30.0% monounsaturated; 24.3% saturated). The quantity of HUFA was high in the muscle (42.4%) followed by the gonad with 24.0% and the liver with 7.8%. *C. laticeps* was in the same way similar to *S. kaupi* I in having a high abundance of monounsaturated fatty acids of the n-7 series. C16: 1n-7 was the most abundant fatty acid in the gonad of this species (18.1%) and also very abundant in the liver (10.8%). C18: 1n-7 was 14.0% in the gonad and 9.3% in the muscle. The most abundant fatty acid in muscle was docosahexaenoic acid (31.3%) and in the liver C18: 1n-9 (51.3%). The concentration of eicosapentaenoic acid in the liver was low (1.0%) compared to the gonad (5.6%) and the muscle (4.8%). The highest DHA/EPA ratio was found in the muscle tissue (6.6). The value was 4.5 for the liver and 2.3 for the gonad.

From the squalid shark *Etmopterus princeps* two muscle samples and one liver were available. Muscle 1 had a total identified fatty acid content of 14.8 µg/mg dw, muscle 2 amounted to 39.4 µg/mg dw, and the liver 89.5 µg/mg dw. The muscles showed a high variation in their fatty acid profile. While in muscle 2 and in liver then was a clear domination of monounsaturated fatty acids and muscle 1 had 46.9% of polyunsaturated fatty acids. The high content of HUFA (42.5%) corresponds to this value and is comparable with the amount in the muscle of *Cataetx laticeps*. The most

abundant fatty acids in muscle 2 and liver were C18: 1n-9 (muscle 2: 19.8%; liver: 24.1%), C22: 6n-3 (muscle 2: 15.8%; liver: 12.1%) and C20: 1n-9 (muscle 2: 12.6%; liver: 13.0%). The most abundant fatty acids in muscle 1 were C22: 6n-3 (30.6%), C16: 0 (18.4%) and C18: 1n-9 (12.8%). The concentration of eicosapentaenoic acid (muscle 1: 3.1%; m<sup>2</sup>: 2.0%; liver: 3.1%) was in both muscles and in the liver low compared to the concentration of docosahexaenoic acid (muscle 1: 30.6%; m<sup>2</sup>: 15.8%; liver: 12.1%). That led to relatively high DHA/EPA ratio in muscle 1 with 10.2, a ratio of 7.9 in muscle 2 and of 4.2 in the liver.

The only available sample of *Epigonus telescopus* was the dorsal muscle, which had an identified fatty acid amount of 12.9 µg/mg dw. The dominating fatty acids were polyunsaturated (51.8%) accompanied by a relatively high (compared to the amount in the tissues of the other species) content of saturated fatty acids (30.5%). The concentration of monounsaturated fatty acids amounted to 15.9%. The HUFA of the muscle was high with 51.8%. Docosahexaenoic acid was the most abundant fatty acid in this tissue sample (37.3%). C16: 0 (23.1%) and C18: 1n-9 (8.4%) were also very abundant. Eicosapentaenoic acid was present with an amount of 5.4%, leading to a DHA/EPA ratio of 6.9.

### Comparison of fatty acid compositions of muscle tissues

The muscle tissue total fatty acid concentration ranged between 10.69 µg/mg dw to 348.62 µg/mg dw, whereas the total identified fatty acid concentration was between 9.77 µg/mg dw to 318.67 µg/mg dw. Thus, the percentage of not identified (unknown) fatty acids ranged between 4.37% to 14.27%.

In the analysed muscles of *S. parasitica*, *I. blachei*, *S. kaupi* I + II, and *E. princeps* (muscle 2) monounsaturated fatty acids dominated (52.6 to 73.5%), it should be noted that all those muscles were dissected with traces of fat layer located directly under the skin. In the muscles of *C. laticeps*, *E. telescopus* and *E. princeps* (muscle 1, without fat layer) polyunsaturated fatty acids presented the highest amount (43.1 to 51.8%) even though not as clearly as the dominant MUFA in the other five muscles.

The concentration of the saturated fatty acids ranged in the muscles with fat between 16.9 to 18.5% and in the muscles without fat layer between 24.3 to 30.5%. Furthermore, the dominating fatty acid in the 'fatty' muscles was the MUFA C18: 1n-9 (19.8-30.8%) whereas in the muscles of *C. laticeps*, *E. telescopus* and *E. princeps* (muscle 1), without fat layer, the PUFA docosahexaenoic acid C22: 6n-3 dominated (30.6 to 37.3%). This fatty acid was also abundant in the muscles of *I. blachei*, *S. kaupi* I + II and *E. princeps* (muscle 2) (9.1 to 15.8%). In the muscles of *S. parasitica*, *I. blachei*, *S. kaupi* II and *E. princeps* (muscle 2) the MUFA C20: 1n-9 was present in a high concentration (12.6 to 18.4%). In the muscle of *C. laticeps* the MUFA C18: 1n-7 was the third highest abundant fatty acid (9.3%).

The most abundant saturated fatty acid in the muscle samples with fat layer was palmitic acid 16: 0 (17.0 to 23.1%). C18: 0 and C14: 0 were also abundant.

The amount of HUFA was according to the values of the amount of saturated fatty acids in the samples with additional fat (of the fat layer) lower (5.4% in *S. parasitica* to 26.8% in *E. princeps* (muscle 2) than in the samples without fat layer (42.4 to 51.8%).

The ratio DHA/EPA showed no remarkable difference between samples with or without fat layer. It was relatively high in all samples and ranged between 3.22 in *S. kaupi* I to 10.15 in *E. princeps* (muscle 1).

### Comparison of fatty acid compositions of liver tissues

From all the caught species except *E. telescopus*, liver samples for fatty acid analysis were available. The identified fatty acid concentration ranged from 26.1 µg/mg dw in *S. kaupi* I to 476.4 µg/mg dw in *C. laticeps*, the non-identified percentage between 1.9 to 12.7%.

In all species, except in *S. kaupi* I, the MUFA concentration dominated in the range of 57.6 to 72.3% of the total identified fatty acid amount. *S. kaupi* I showed a higher concentration in PUFA (43.5%). The concentration of saturated fatty acids ranged from 18.2% in *E. princeps* to 24.3% in *S. kaupi* I.

The most abundant fatty acids were C18: 1n-9 in *C. laticeps* (51.3%), *S. kaupi* II (38.2%), *S. parasitica* (31.9%), *I. blachei* (31.2%) and *E. princeps* (24.1%) and C22: 6n-3 in *S. kaupi* I (31.3%). C22: 6n-3 was also very abundant in the liver of *E. princeps* (12.1%) and *I. blachei* (9.2%). The MUFA C20: 1n-9 was again very abundant in *E. princeps* (13.0%), *S. parasitica* (11.0%) and *S. kaupi* II (10.7%). The vacenic acid C18: 1n-7 was one of the most abundant fatty acids in *S. kaupi* I (9.2%) and in the liver of *C. laticeps*, palmitoleic acid C16: 1n-7 had the highest concentration (10.8%).

The most abundant saturated fatty acid was palmitic acid (C16: 0) with a content of 10.8% in *E. princeps* to 17.2% in *S. kaupi* I.

The concentration of HUFA ranged from 7.4% in *S. kaupi* II to 42.7% in *S. kaupi* I. The content was clearly higher in *S. kaupi* I (42.7%), *E. princeps* (20.7%), *I. blachei* (16.1%) and *S. parasitica* (10.4%).

The ratio DHA/EPA in the liver samples ranged from 3.5 (*I. blachei*) to 6.9 (*S. kaupi* II) and was lower than in the muscles comparing the averages (6.29 in muscle; 5.27 in liver).

### Comparison of fatty acid compositions of gonad tissues

For the analysis of the gonad fatty acid composition in hot vent fishes only the gonads of *S. parasitica*, *I. blachei*, *C. laticeps* and both *S. kaupi* specimens were available.

The results of this examination showed also a clear domination of monounsaturated fatty acids in the range of 55.1% in *C. laticeps* to 70.2% in *S. kaupi* I. The exception was *S. parasitica* with a monounsaturated fatty acid content of 27.0% and a dominating polyunsaturated amount of 45.4%.

The most abundant fatty acid in the gonad was C18: 1n-9 in *I. blachei* (36.3%), *S. kaupi* I (20.4%) and *S. kaupi* II (28.4%), whereas in *S. parasitica* C22: 6n-3 was found as the most abundant (28.3%). In *C. laticeps* C16: 1n-7 was the dominated fatty acid (18.1%) followed by C18: 1n-9 (14.6%) and C18: 1n-7 (14.0%). Also *S. kaupi* I showed a high abundance of n-7 fatty acids with C16: 1n-7 (19.8%) and C18: 1n-7 (16.9%). C16: 0 was in all gonads the most abundant saturated fatty acid (17.4% in *S. parasitica*, 17.1% in *I. blachei*, 12.7% in *S. kaupi* I, 11.1% in *S. kaupi* II and 12.3% in *C. laticeps*).

The content of HUFA amounted to 42.1% in *S. parasitica*, whereas the content was clearly lower in *I. blachei* (10.2%), *S. kaupi* I (10.2%), *S. kaupi* II (18.9%) and *C. laticeps* (24.0%).

*I. blachei*, *C. laticeps* and the two *S. kaupi* specimens ranged from 2.3 to 4.8 concerning the DHA/EPA ratio, whereas *S. parasitica* showed a higher DHA/EPA ratio (10.1).

## Lipid class composition

### Neutral lipids

The dominant lipid classes in all three organs of these six species were triglycerides, sterols and polar lipids (Tables 3 to 6). Only the gonad of *S. parasitica*, the muscle 1 of *E. princeps*, and the muscle of *C. laticeps* deviated from this result. In these samples no triglycerides could be detected. Furthermore, free fatty acids were registered and in some samples sterolesters or alkyl-diacyl-glycerides were found.

The content of triglycerides was very high in the muscle of *S. parasitica* (50.7%), whereas in the liver only 18.9% of the lipids are triglycerides and in the gonad no triglycerides could be detected. The concentration of sterols and polar lipids were equivalent in amount and relation in the three different tissues. The muscle showed 17.4% polar lipids and 18.9% sterols. The amount of both classes were higher in the liver, 27.9% polar lipids and 32.5% sterols. Finally, in the gonad 29.2% polar lipids and 27.2% sterols were found. The amount of free fatty acids were similar in muscle, liver and gonad (8.7 to 9.9%). Besides, 3.4% of lipid in the muscle of *S. parasitica* were sterolester; in the liver 7.7% of sterolester were detected.

*I. blachei* showed a high concentration of triglycerides in muscle (42.2%) and gonad (40.9%), whereas in liver the amount of triglycerides were about 29.0%. In the liver sterols dominated with a percentage of 34.9 (in muscle: 11.0%; in gonad: 10.1%).

The highest amount of polar lipids were also found in the muscle tissue with 34.4%, followed by 20.1% in the liver and 17.0% in the gonad. Free fatty acids made out 4.0% of the lipids in muscle, 10.1% in liver, and 7.4% in gonad. In liver and gonad sterolester and alkyl-diacyl-glyceride could be detected, in gonad even a high amount of sterolester about 15.7%.

**TABLE 3** - Lipid class composition and content (%) in muscle tissues (with fat layer) (nd = not detected).

Lipid Class	<i>S. parasitica</i>	<i>E. princeps</i> muscle 2	<i>I. blachei</i>	<i>S. kaupi</i> I	<i>S. kaupi</i> II
Polar lipids	17.4	10.4	34.4	11.7	18.3
Free fatty acids	8.7	3.8	4.0	6.8	5.6
Sterols	18.9	9.4	11.0	13.8	10.3
Triglycerides	50.7	21.5	42.2	64.1	63.0
Sterolester	3.4	0.6	nd	nd	nd
Alkyl-diacyl-glycerides	nd	39.0	nd	nd	nd

**TABLE 4** - Lipid class composition and content (%) of muscle tissues (without fat layer) (nd = not detected).

Lipid Class	<i>E. princeps</i> muscle 1	<i>C. laticeps</i>	<i>E. telescopus</i>
Polar lipids	37.0	44.4	32.0
Free fatty acids	9.5	8.7	6.8
Sterols	26.1	28.7	51.6
Triglycerides	nd	nd	nd
Sterolester	nd	nd	8.4
Alkyl-diacyl-glycerides	nd	nd	nd

The analysed neutral lipid composition of the two *S. kaupi* specimens corresponded to each other. Only in the liver were deviations registered which are worth mentioning. The content of polar lipids in liver of both *S. kaupi* I and II was similar (13.2% (I) / 19.7% (II)). That applied also for the concentration of free fatty acids in the liver of both specimen (6.8% (I) / 5.6% (II)).

**TABLE 5** - Lipid class composition and content (%) of gonad tissues (nd = not detected).

Lipid Class	<i>S. parasitica</i>	<i>I. blachei</i>	<i>C. laticeps</i>	<i>S. kaupi</i> I	<i>S. kaupi</i> II
Polar lipids	29.2	17.0	13.8	9.0	11.1
Free fatty acids	9.9	7.4	13.7	5.9	7.2
Sterols	27.2	10.1	15.2	13.7	8.8
Triglycerides	nd	40.9	40.6	54.7	48.7
Sterolester	nd	15.7	9.9	6.7	7.9
Alkyl-diacyl-glycerides	nd	2.3	3.3	1.9	2.7

**TABLE 6** - Lipid class composition and content (%) of liver tissues (nd = not detected).

Lipid Class	<i>S. parasitica</i>	<i>E. princeps</i>	<i>I. blachei</i>	<i>C. laticeps</i>	<i>S. kaupi</i> I	<i>S. kaupi</i> II
Polar lipid	27.9	23.6	20.1	11.5	13.2	19.7
Free fatty acids	9.3	5.3	10.1	10.2	9.1	5.1
Sterol	32.5	10.7	34.9	15.9	38.7	16.1
Triglycerides	18.9	26.4	29	50.6	22.4	48
Sterolester	7.7	nd	4.4	11.5	9	nd
Alkyl-diacyl-glycerides	nd	31.2	0.1	0.7	2.3	nd

Marked differences of concentration of sterols and triglycerides were observed. In the liver of *S. kaupi* I 38.7% of the total lipid content were sterols, but in *S. kaupi* II sterols amounted to 16.1%. On the other hand, *S. kaupi* II showed a high amount of triglycerides (48.0%), whereas in the liver of *S. kaupi* I only 22.4% of the total lipid content were triglycerides. Furthermore, in the liver of *S. kaupi* I sterolesters and alkyl-diacyl-glycerides could be registered but there was no detection of these two lipid classes in the liver of the other specimen.

The muscles of *S. kaupi* I and II showed also no sterolesters and alkyl-diacyl-glycerides. The content of triglycerides amounted 64.1% or 63.0%, respectively. The concentration of the sterols was about 13.8% in *S. kaupi* I and 10.3% in *S. kaupi* II. In specimen I, 11.7% of the lipids were polar, whereas in specimen II 18.3% were polar.

In the gonad the content of polar lipids were slightly lower (9.0% or 11.1%, respectively). Triglycerides content amounted to 54.7% in *S. kaupi* I and of 48.7% in *S. kaupi* II, whereas the percentage of the sterols were 13.7 and 8.8 respectively. In both gonads sterolesters (specimen I: 6.7% / specimen II: 7.9%) and alkyl-diacyl-glycerides (specimen I: 1.9% / specimen II: 2.7%) were found.

The muscle of *C. laticeps* contained 44.4% polar lipids, 8.7% free fatty acids, and 28.7% sterols. No triglycerides, sterolesters or alkyl-diacyl-glycerides were detected. In the liver and the gonad of *C. laticeps* triglycerides (50.6% in liver; 40.6% in gonad) were the dominant lipid class, followed by the sterols. The percentage of polar lipids in liver (11.5%) and gonad (13.8%) was clearly lower than in the muscle of this species.

Beside this, in the liver and gonad a high percentage of sterolesters was registered, with 11.5% sterolesters in the liver and 9.9% in gonad. Furthermore, 3.3% alkyl-diacyl-glycerides in gonad could be detected and also a trace of 0.7% in the liver.

In the muscle 1 of *E. princeps* a similar lipid composition to that in *C. laticeps* was found. Of the total lipid content, 37.0% were polar lipids, 26.1% sterols, and 9.5% free fatty acids. The other classes were not registered, while in muscle 2, 21.5% triglycerides occurred and even 39.0% alkyl-diacyl-glycerides, which were, in that muscle, the main part of the total lipid content: 10.4% were polar lipids, 3.8% free fatty acids and 9.4% sterols. A small trace of sterolester was detected (0.6%), while in the liver of *E. princeps* as well as in muscle 1, no sterolesters were found.

The liver also contained a high amount of alkyl-diacyl-glycerides (31.2%), followed by 26.4% triglycerides and 23.6% polar lipids. The free fatty acids were represented by 5.3% and the sterols by 10.7%.

The muscle, the only organ analysed from *E. telescopus* showed a clear dominance of sterols (51.6%), while the polar lipids made up 32.0% of the total lipid content: 6.8% were free fatty acids and 8.4% sterolester. No triglycerides or alkyl-diacyl-glycerides were detected.

### **Comparison of neutral lipid composition of muscles (Tables 3 and 4)**

The three muscles without fat (Table 4) were muscle 1 of *E. princeps*, the muscle of *C. laticeps* and of *E. telescopus*. In all muscles we could find a relatively high content of polar lipids (*E. telescopus*: 32.0%), whereas in *E. princeps* (37.0%) and *C. laticeps* (44.4%) this lipid class was even the most abundant one. The second most abundant class were the sterols, represented in *E. princeps* by 26.1%, in *C. laticeps* by 28.7%, and in *E. telescopus* by 51.6%. In *E. princeps* and *C. laticeps* only the further lipid class of free fatty acids was found (9.5% in *E. princeps*; 8.7% in *C. laticeps*), while *E. telescopus* contained 6.8% free fatty acids and also 8.4% sterolester. In these muscles neither triglycerides nor alkyl-diacyl-glycerides were registered.

The results in the muscles containing fat layer (Table 3) were different. In these muscles the main lipid class were the triglycerides (*S. parasitica*: 50.7%; *I. blachei*: 42.2%; *S. kaupi* I: 64.1%; *S. kaupi* II: 63.0%; *E. princeps*: 21.5%) with an exception in muscle 2 of *E. princeps*. Here, the most abundant class were the alkyl-diacyl-glycerides (39.0%).

The concentration of polar lipids were lower than in the muscles without fat. They ranged from 10.4% in *E. princeps* to 18.3% in *S. kaupi* II (except *I. blachei* with 34.4% of polar lipids). The sterols ranged between 9.4% in *E. princeps* to 18.9% in *S. parasitica*.

In addition, 3.4% of sterolesters in *S. parasitica* and 0.6% in *E. princeps* (muscle 2) were found, while in the remaining muscles with fat neither sterolester nor alkyl-diacyl-glycerides could be detected.

### **Comparison of neutral lipid composition of gonads (Table 5)**

The gonads of four of the five analysed species showed a dominance in triglycerides (40.6 to 54.7%), while in the gonad of *S. parasitica* no triglycerides could be registered, even neither sterolester nor alkyl-diacyl-glycerides. In *S. parasitica* 29.2% of polar lipids, 27.2% of sterols and 9.9% of free fatty acids were detected. The other four gonads of *I. blachei*, *C. laticeps* and the two *S. kaupi* specimens were more similar to each other. Polar lipid concentrations between 9.0 to 17.0% were found, the sterol content ranged between 8.8% in *S. kaupi* II to 15.2% in *C. laticeps*, and the percentages of sterolesters were between 6.7% in *S. kaupi* I to 15.7% in *I. blachei*. The lower concentration of alkyl-diacyl-glycerides ranged between 1.9 to 3.3%.

### **Comparison of neutral lipid composition of livers (Table 6)**

Comparing the lipid composition in the liver of *S. parasitica*, *I. blachei*, *C. laticeps*, *S. kaupi* I and II and *E. princeps* no clear dominance of one lipid class was detected. In *S. parasitica* (32.5%), *I. blachei* (34.9%) and *S. kaupi* I (38.7%), the sterols were the most abundant group, while in *C. laticeps* and *S. kaupi* II triglycerides were dominant and in *E. princeps* the class of the alkyl-diacyl-glycerides. In *I. blachei*, *C. laticeps* and *S. kaupi* I small traces of alkyl-diacyl-glycerides were found (0.1 to 2.3%). Sterolester was detected in *S. parasitica* (7.7%), *I. blachei* (4.4%), *C. laticeps* (11.5%) and *S. kaupi* I (9.0%). The content of polar lipids ranged between 11.5% in *C. laticeps* to 27.9% in *S. parasitica*.

### **Polar lipids (Tables 7-10)**

As shown in Tables 7, 9 and 10, a dominance of phosphatidylinositol in muscle, liver and gonad of *S. parasitica* was found. The muscle contained 50.3% phosphatidylinositol of the total polar lipid amount, the liver 44.5% and the gonad 44.7%. Concerning the concentrations of further polar compounds like lysophosphatidylcholine, phosphatidylserine, phosphatidylcholine and phosphatidylethanolamin the different organs showed considerable variability. In muscle and liver of *S. parasitica* no lysophosphatidylcholine was detected, but in gonad 7.4%

of this phospholipid was found. The liver showed 27.3% of phosphatidylserine, while the gonad contained only 4.0% and the muscle only 1.9%. The liver was the organ mostly rich in phosphatidylcholine (22.5%) (muscle: 11.8%; gonad: 8.8%). Finally, phosphatidylethanolamine was present in a high amount (32.5%) in the muscle of this species; the liver showed a low content of 1.1% and the gonad contained 17.5%.

**TABLE 7** - Polar lipid composition and content (%) of muscles (with fat layer) (nd = not detected).

<b>Lipid Class</b>	<i>S. parasitica</i>	<i>I. blachei</i>	<i>S. kaupi</i> <b>I</b>	<i>S. kaupi</i> <b>II</b>	<i>E. princeps</i> <b>muscle 2</b>
Lysophosphatidyl choline	nd	1.9	7.2	nd	4.6
Phosphatidylserine	1.9	5.8	7.9	2.2	12.0
Phosphatidylcholine	11.8	28.3	20.5	15.2	46.7
Phosphatidylinositol	50.3	19.2	44.3	52.9	nd
Phosphatidylethanol-amine	32.5	31.0	11.1	26.6	34.0

**TABLE 8** - Polar lipid composition and content (%) of muscles (without fat layer).

<b>Lipid Class</b>	<i>C. laticeps</i>	<i>E. telescopus</i>	<i>E. princeps</i> <b>muscle 1</b>
Lysophosphatidylcholine	3.4	1.8	3.0
Phosphatidylserine	4.1	7.8	2.5
Phosphatidylcholine	8.3	25.0	13.1
Phosphatidylinositol	36.6	35.5	48.1
Phosphatidylethanolamine	17.4	13.2	21.4

**TABLE 9** - Polar lipid composition and content (%) of gonads (nd = not detected).

<b>Lipid Class</b>	<i>S. parasitica</i>	<i>I. blachei</i>	<i>S. kaupi</i> <b>I</b>	<i>S. kaupi</i> <b>II</b>	<i>C. laticeps</i>
Lysophosphatidylcholine	7.4	2.8	3.4	nd	nd
Phosphatidylserine	4.0	11.1	4.1	nd	3.1
Phosphatidylcholine	8.8	9.5	15.3	21.8	23.4
Phosphatidylinositol	44.7	53.1	55.5	47.1	44.8
Phosphatidylethanolamine	17.5	19.6	17.6	30.1	26.3

**TABLE 10** - Polar lipid composition (%) of livers (nd = not detected).

Lipid Class	<i>S. parasitica</i>	<i>I. blachei</i>	<i>S. kaupi</i> I	<i>S. kaupi</i> II	<i>C. laticeps</i>	<i>E.princeps</i>
Lysophosphatidylcholine	nd	14.1	4.8	nd	11.5	7.1
Phosphatidylserine	27.3	nd	7.3	25.6	9.3	11.7
Phosphatidylcholine	22.5	15.4	16.3	20.9	10.9	43.4
Phosphatidylinositol	44.5	33.1	49.6	48.1	58.2	12.8
Phosphatidylethanolamine	1.1	13.5	12.6	nd	3.8	18.0

In the tissues of *I. blachei* (Tables 7, 9 and 10), the dominance of phosphatidylinositol was only found in liver (33.1%) and gonad (53.1%), whereas the muscle contained 19.2% and 31.0% of phosphatidylethanolamine and 28.3% of phosphatidylcholine. In the muscle, lysophosphatidylcholine and phosphatidylserine were found in lower amounts (1.9% or 5.8%, respectively). In the gonad a corresponding concentration of lysophosphatidylcholine (2.8%) was detected. However, the liver contained 14.1% lysophosphatidylcholine, but no phosphatidylcholine (content in gonad: 11.1%). Concerning the concentrations of phosphatidylcholine and phosphatidylethanolamine liver and gonad had corresponding values. In the liver 15.4% of the total polar lipid content is phosphatidylcholine and in the gonad 9.5%. Phosphatidylethanolamine was 13.5% in the liver and 19.6% in the gonad.

The two specimen of *S. kaupi* (Tables 7, 9 and 10) had only a few concentrations in common. In both specimens there was a clear dominance of phosphatidylinositol in muscle, liver and gonad. This amount ranged from 44.3 to 55.5%. Also, the values of the content of phosphatidylcholine were similar. All of them ranged from 15.2 to 21.8% in the three different tissues.

In muscle, liver and gonad of *S. kaupi* II no amounts of lysophosphatidylcholine were detected, whereas in the muscle of *S. kaupi* I 7.2%, in the liver 4.8%, and in the gonad 3.4% were found. *S. kaupi* II showed an irregular distribution of phosphatidylserine in the different organs: in the liver 25.6% of the polar lipids were phosphatidylserine; in the muscle 2.2%, and in gonad no phosphatidylserine could be detected; whereas *S. kaupi* I contained in all three organs a concentration ranging from 4.1 to 7.9%. *S. kaupi* I showed a content of phosphatidylethanolamine ranging from 11.1% in the muscle, 12.6% in the liver to 17.6% in the gonad. However, *S. kaupi* II contained 26.6% in the muscle, 30.1% in the gonad, but no amount in the liver.

Also in the tissues of *C. laticeps* (Tables 8-10), phosphatidylinositol was the dominant component: in the muscle 36.6%, in the liver 58.2% and in the gonad 44.8%. Furthermore, the gonad had high amounts of phosphatidylcholine (23.4%) and

phosphatidylethanolamine (26.3%). In the liver 10.9% of phosphatidylcholine and 3.8% of phosphatidylethanolamine were detected and in the muscle 8.3% and 17.4% respectively. The highest amount of lysophosphatidylcholine was found in the liver (11.5%), while 3.4% were detected in the muscle but nothing in the gonad tissue. Phosphatidylserine was represented with 9.3% in liver, 4.1% in muscle, and 3.1% in gonad.

The two muscles of *E. princeps* showed a different pattern of polar lipid composition. Muscle 1 (without fat layer) was rich in phosphatidylinositol (48.1%), whereas in muscle 2 (with fat layer) no phosphatidylinositol could be detected. In muscle 2 phosphatidylcholine was the dominant component (46.7%), while muscle 1 contained only 13.1% (Tables 7 and 8).

The concentration of lysophosphatidylcholine was similar (muscle 1: 3.0%; muscle 2: 4.6%); also the concentration of phosphatidylethanolamine (muscle 1: 21.4%; muscle 2: 34.0%). The content of phosphatidylserine amounted to 2.5% in muscle 1 and 12.0% in muscle 2 (Tables 7 and 8).

The liver showed, like the muscle with fat layer, a dominance of phosphatidylcholine, followed by 18.0% of phosphatidylethanolamine, 12.8% of phosphatidylinositol and 11.7% phosphatidylserine. The amount of lysophosphatidylcholine was a little higher than in the muscles (7.1%) (Table 10).

Finally, the muscle sample of *E. telescopus* showed a dominance of phosphatidylinositol (35.5%), followed by 25.0% content of phosphatidylcholine. Phosphatidylethanolamine was represented by 13.2%, phosphatidylserine by 7.8%, and lysophosphatidylcholine by 1.8% (Table 8).

### **Comparison of polar lipid composition of muscles**

Regarding the muscle with fat layer (Table 7) phosphatidylinositol was dominant (44.3 to 52.9%), except in the muscle of *I. blachei* (19.2%) and muscle 2 of *E. princeps* (not detected). In *I. blachei*, the main amount of polar lipids belongs to phosphatidylethanolamine (31.0%) and phosphatidylcholine (28.3%), while in *E. princeps* no phosphatidylinositol was found, but a high amount of phosphatidylcholine (46.7%) and phosphatidylethanolamine (34.0%). Also in *S. parasitica* (32.5%) and *S. kaupi* II (26.6%) a high amount of phosphatidylethanolamine was detected (*S. kaupi* I: 11.1%), but the concentrations of phosphatidylcholine in both *S. kaupi* specimen and *S. parasitica* ranged from 11.8 to 20.5%. Lysophosphatidylcholine was not detected in the muscle of *S. parasitica* and *S. kaupi* II and ranged in the remaining species from 1.9% in *I. blachei*, 4.6% in *E. princeps* to 7.2% in *S. kaupi* I (Table 7).

The muscles without fat layer (Table 8) of *C. laticeps*, *E. telescopus* and *E. princeps* all showed a dominance of phosphatidylinositol ranging from 35.5% in *E. telescopus* to 48.1% in *E. princeps*. Furthermore, phosphatidylethanolamine (13.2 to 21.4%) and phosphatidylcholine (8.3 to 25.0%) was abundant.

### Comparison of polar lipid composition of gonads

In the analysed gonads (Table 9) a clear dominance of phosphatidylinositol was found (44.7 to 55.5%). Also phosphatidylethanolamine (17.5 to 30.1%) and phosphatidylcholine (8.8 to 23.4%) were very abundant. Some differences existed in the content of lysophosphatidylcholine and phosphatidylserine. In *S. kaupi* II and *C. laticeps* no lysophosphatidylcholine was detected. *I. blachei* showed an amount of 2.8%, *S. kaupi* I 3.4%, and *S. parasitica* 7.4%. The amount of phosphatidylserine ranged from 3.1% in *C. laticeps* to 11.1% in *I. blachei*, but in *S. kaupi* II no phosphatidylserine was detected.

### Comparison of polar lipid composition of livers

As shown in Table 10, phosphatidylinositol was the most abundant polar component in the liver of *S. parasitica* (44.5%), *I. blachei* (33.1%), *S. kaupi* I (49.6%), *S. kaupi* II (48.1%) and *C. laticeps* (58.2%), while in *E. princeps* only 12.8% were phosphatidylinositol, but 43.3% phosphatidylcholine. Also in the liver of the other species an abundance of phosphatidylcholine was observed (10.9 to 22.5%), but in the remaining polar components there was a high variation. The content of lysophosphatidylcholine ranged from 0 to 14.1%, the content of phosphatidylserine from 0 to 27.3% and that of phosphatidylethanolamine from 0 to 18.0%.

## DISCUSSION

### Fatty acid composition

Monounsaturated fatty acids were the dominant components in muscle, gonad and liver tissue of the species examined. The degree of monounsaturation ranged from 53 to 74% in these tissues. Those values concur with the amount of monounsaturated fatty acids (40-70%) found in the same tissues of some deep-sea fish species caught at the Madeira Island Slope (n = 5) at a depth of 4000 m. In fish species caught at the Portuguese Slope (n = 5) at a depth of 830 m monounsaturated fatty acids also dominated but not as obvious as in the species examined in this work and in the species from the 4000 m depth. The monounsaturated amount of fatty acids in the species at the Portuguese Slope ranged from 28% (average in muscle) to 49% (average in liver). Cod liver oil showed a percentage of 71.5% monounsaturated fatty acids.

Exceptions concerning the clear predominance of monounsaturated fatty acids were found in the muscles of *C. laticeps*, *E. telescopus* and muscle 1 of *E. princeps* ([PUFA] = 43-52%); furthermore in the liver of *S. kaupi* I ([PUFA] = 44%) and the gonad of *S. parasitica* ([PUFA] = 45%). In these tissues the polyunsaturated fatty acids dominated.

Thus, this high degree of monounsaturation in the fatty acid profile found in the hydrothermal vent fishes and in fishes living in less deep waters or caught at a depth at 4000 m (non-hydrothermal vent fishes) does not reflect the fact of hydrothermal vent species being consumers of alternative food chains of which chemoautolithotrophic bacteria are the primary producers. Results suggest that in marine vertebrates, at least in the class of fishes, the predominance of monounsaturated fatty acids is common. The opposite case was found in surface waters-dwelling marine invertebrates (BEN-MLIH *et al.*, 1992). Littoral mussels were characterized by a predominance of polyunsaturated fatty acids (C20: 5n-3, C22: 6n-3), reflecting the planktonic origin of their food. On the other hand, deep hydrothermal vent symbiotic bivalves fatty acid distribution was dominated by an abundance of monounsaturated acids (double bond in the n-7 position) reflecting their bacterial origin.

A quality comparison between the fatty acid profile of the analysed tissues of the hot vent fishes and cod liver oil showed an identical fatty acid composition. In all analysed tissues and in cod liver oil the fatty acids C18: 3n-6 and Iso C18: 0 were not detected. Furthermore, the polyunsaturated fatty acid C16: 3n-1 was absent in cod liver oil but extant in the gonad of *I. blachei* and *S. kaupi* I and in the liver of *I. blachei* and *S. kaupi* II.

The assumption that different food chains at hydrothermal vent sites are reflected in the fatty acid profile of the consumers was tested looking at the major fatty acids detected in the hydrothermal vent deep-sea species. In *S. parasitica*, *I. blachei*, *E. princeps*, *E. telescopus* and one *S. kaupi* species (II) the major fatty acids were C18: 1n-9, C20: 1n-9, C22: 6n-3, C16: 0 similar to those found in the deep-sea fishes from the Madeira Island Slope, the fishes from the Portuguese Slope and for the oreo and orange roughy species analysed by BAKES *et al.* (1995). Besides, in muscle, gonad and liver of *Cataetyx laticeps* and in gonad and liver tissue of *Synaphobranchus kaupi* I considerable amounts of monounsaturated fatty acids of the n-7 series were detected. The muscle of *C. laticeps* showed a content of 9.3% for the MUFA C18: 1n-7. Sixteen per cent of the total fatty acid content in this muscle belongs to monounsaturated fatty acids having the double bond in n-7 position and 53.2% of the monounsaturated fatty acid amounts in the muscle are n-7 fatty acids. In the gonad of *C. laticeps* even 34.5% of the total fatty acid content were monounsaturated n-7 fatty acids and made up 62.6% of the monounsaturated amount. Here, the concentration of C18: 1n-7 amounted to 14% and the concentration of C16: 1n-7 to 18.1%. The liver contained 10.8% of C16: 1n-7. In *S. kaupi* I the gonad had 39.5% of the total fatty acid content monounsaturated n-7 fatty acids. C16: 1n-7 made up 19.8% and C18: 1n-7 16.9% of the total amount. In the liver of *S. kaupi* I 9.2% were C18: 1n-7, 53.1% of the monounsaturated fatty acids were n-7 fatty acids, and this part made up 15.8% of the total fatty acid amount. The high content of the monounsaturated fatty acids of the n-7 series can reflect the effects of the different energy source at the hydrothermal vent site, because they are characteristic for bacteria.

The concentration of branched *iso* and *ante-iso* fatty acids in the gonad, muscle and liver of the seven examined species was low (gonad: 0.7-1.6%; muscle: 0.6-1.4%; liver: 0.4-1.2%) compared to the values found by BEN-MLIH *et al.* (1992) in deep hydrothermal vent symbiotic bivalves (15.6%). Branched fatty acids from C14 to C19 in *iso* and *ante-iso* positions were reported in most bacteria accounting for as much as 70% of the total fatty acids (BEN-MLIH *et al.*, 1992). The small content of those branched fatty acids found in the analysed tissues of the hydrothermal vent fishes does not reflect the participation at the chemosynthetic bacteria-based food chain. The amounts do not differ from that detected in fishes from the Portuguese Slope (830 m) or Madeira Island Slope (4000 m), respectively.

In all three tissues of the species a high amount of HUFA was detected. In the liver the amount of HUFA ranged from 7 to 43%, in the gonad from 10 to 42%, in the muscle with fat layer from 5 to 27% and in the muscle without fat layer from 42 to 53%. Thus, all three tissues are good sources of the high-unsaturated fatty acids, which are important for animal and human nutrition.

The ratio of docosahexaenoic acid (DHA) to eicosapentaenoic acid (EPA) ranged from 4.5 (average in gonad), 5.1 (average in liver) to 6.3 (average in muscle). There were no remarkable differences in the ratio between muscle with or without fat layer, respectively. In all these tissues the concentration of DHA (muscle: 3.8 to 37.3%; gonad: 5.9 to 28.3%; liver: 4.4 to 31.3%) was clearly higher than the concentration of EPA (muscle: 0.6 to 5.4%; gonad: 1.2 to 5.6%; liver: 0.6 to 5.4%). Thus, the gonad is a source from which high amounts of both EPA and DHA are available. Finally, muscle, gonad and liver tissue of the seven hot vent species are rich sources of the docosahexaenoic acid.

To summarise, only the high content of fatty acids of the n-7 series leads to the conclusion that the fatty acid profile of the examined hydrothermal vent fishes compared to other deep-sea fishes is caused by the different primary production in the form of chemosynthetic bacteria.

### **Lipid class composition**

In general, the lipid composition of muscle, gonad and liver was dominated by three classes: polar lipid, triglycerides and sterols. BAKES *et al.* (1995) found that the dominant lipid classes in the muscle of oreo from Australian waters and orange roughy from Northern Atlantic waters were polar lipid, wax ester and triacylglycerides. Free fatty acids, sterolesters and alkyl-diacyl-glycerides were minor components, but between the species and also between the different tissues of one species existed high variations. For instance, in the muscle of *C. laticeps*, *E. telescopus* and muscle 1 of *E. princeps* no triglycerides were detected whereas in muscle 2 of *E. princeps* the major lipid class was alkyl-diacyl-glyceride. Similarly, in the liver of *E. princeps*: alkyl-diacyl-

glycerides dominated as well as triglycerides and polar lipids. The liver of the other six species showed the clear dominance of triglycerides, polar lipids and sterols. In the liver of *C. laticeps* also sterolesters were abundant. The gonad tissue of *C. laticeps* and *S. kaupi* I and II showed the general abundance of the three lipid classes named above, but *C. laticeps* had also a higher abundance of free fatty acids and sterolesters. Also *I. blachei* had, besides the major classes, a higher abundance of sterolesters than of sterols. In *S. parasitica* only polar lipid, sterol and free fatty acid occurred.

For further comparison with the corresponding tissues of deep-sea fishes, which do not live near a hydrothermal vent no material was available for analyses.

The major polar lipid component in all species was phosphatidylinosite, followed by phosphatidylcholine and phosphatidylethanolamine, the minor components were phosphatidylserine and lysophosphatidylcholine. In all gonad tissues, which were available of *S. parasitica*, *I. blachei*, *C. laticeps* and *S. kaupi* I and II the clear predominance of phosphatidylinositol (45 to 56%) was obvious, as well as abundant components phosphatidylethanolamine (18 to 30%) and phosphatidylcholine (9 to 23%). Equivalent values were found in the muscle without fat layer of *C. laticeps*, *E. telescopus* and *E. princeps* (muscle 1), also in the muscles with fat layer of *S. parasitica*, and *S. kaupi* I and II (P.-inositol: 44 to 53%; P.-ethanolamine: 11 to 33%; P.-choline: 12 to 21%). On the other hand, in *E. princeps* no phosphatidylinositol was detected and phosphatidylcholine (47%) was the dominating polar component (P.-ethanolamine: 34%; P.-serine: 12%). In the muscle of *I. blachei* dominated phosphatidylethanolamine (31%).

All three major polar lipid components are found in animals, higher plants and micro-organisms, thus, the abundance of one component allows no conclusion about the food source of the fishes. Further investigations would be necessary analysing the polar lipid composition and content of 'non-hydrothermal vent' deep-sea fishes.

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## REFERENCES

BAKES, M. J., N. G. ELLIOTT, G. J. GREEN & P. D. NICHOLS:

1995. Variation in lipid composition of some deep-sea fish (Teleostei: Oreosomatidae and Trachichthyidae). *Comp. Biochem. Physiol.*, **111** B (4): 633-642.

BEN-MLIH, F., J.-C. MARTY & A. FIALA-MEDIONI:

1992. Fatty acid composition in deep hydrothermal vent symbiotic bivalves. *Journal of Lipid Research*, **33**: 1797-1806.

BLIGH, E. G. & W. J. DYER:

1959. A rapid method of total lipid extraction and purification. *Can. J. Biochem. Physiol.*, **37**: 911-917.

COLODNER, D., J. LIN, K. Von DAMM, L. BUTTERMORE, R. KOZLOWSKY, J.-L.

CHARLOU, J.-P. DONVAL, C. WILSON & THE LUCKY STRIKE TEAM:

1993. Chemistry of Lucky Strike hydrothermal fluids: initial results. *Transactions of the American Geophysical Union*, **74**: 99.

HUMPHRIS, S. E., M. K. TIVEY & Y. FOUQUET:

1993. Comparison of hydrothermal deposits at the Lucky Strike Vent Field with other Mid-Ocean Ridge vent sites. *Transactions of the American Geophysical Union*, **74**: 100.

SALDANHA, L.:

1994. Fishes observed and collected during the ALVIN dives at the Lucky Strike thermal vent site (Mid-Atlantic Ridge-1993). *Cybium*, **18** (4): 460-462.

SALDANHA, L. & M. BISCOITO:

1997. Fishes from the Lucky Strike and Menez Gwen hydrothermal vent sites (Mid-Atlantic Ridge). *Boletim do Museu Municipal do Funchal (História Natural)*, **49**: 189-206.

Van DOVER, C. L.:

1995. Ecology of Mid-Atlantic Ridge hydrothermal vents. In: *Hydrothermal Vents and Processes* (eds.: L. M. Parson & D. R. Dixon). Geological Society Special Publication, **87**: 257-294.

Van DOVER, C. L., D. DESBRUYÈRES, M. SEGONZAC, T. COMTET, L. SALDANHA,  
A. FIALA-MÉDIONI & C. LANGMUIR:

1996. Biology of the Lucky Strike hydrothermal field. *Deep-Sea Research*, **43** (9):  
1509-1529.