Pergamon

Comp. Biochem. Physiol. Vol. 107B, No. 2, pp. 331–336, 1994

0305-0491/94 $6.00 + 0.00

Phospholipid and fatty acid compositions of the endemic amphipod crustacean *Issycogammarus bergi* from the brackish mountain lake Issyk-Kul (Tian Shan, Middle Asia)

Valery M. Dembitsky,* Tomas Rezanka† and Andrey G. Kashin‡

*Laboratory of Natural Products, Department of Organic Chemistry, The Hebrew University of Jerusalem, Jerusalem, 91904, Israel; †Institute of Microbiology, Prague, Videnska 1083, 142 20, Czech Republic; and ‡Institute of Ecology of the Volga River Basin, Russian Academy of Sciences, Togliatti 445003, Russia

Lipid and phospholipid compositions of endemic gammarids, belonging to the subphylum Crustacea *Issycogammarus bergi* was studied. The fatty acid composition of total lipids, neutral, glyco- and phospholipid fractions were investigated by capillary gas chromatography–mass spectrometry. Ninety-four fatty acids were identified: 26 saturated (both *iso*-, *anteiso*- and *cyclo*-), 31 monoenoic, 11 dienoic, and 26 polyenoic (including 10 trienoic, eight tetraenoic, six pentaenoic and two hexaenoic). Among polyenoic fatty acids high percentages of three main acids 18:3n-6, 18:3n-3 and 20:4n-6 in phospholipid and glycolipid fractions were identified; high percentages of 16:3n-6 and 16:4n-1 among total lipids were also identified. A number of unusual polyunsaturated fatty acids, such as c-9-17:0, c-9-19:0, 24:4n-6, 24:5n-6 and 24:6n-3 acids, were found.

Key words: Phospholipid; Fatty acid; *Issycogammarus bergi*; Lake Issyk-Kul.

Comp. Biochem. Physiol. 107B, 331–336, 1994.

Introduction

In Central Asia four mountain systems, including Tian Shan, may be distinguished, according to the period during which the most intensive folding took place. The Tian Shan was regarded formerly as a system of folded mountains which were formed during the Tertiary period, and was considered to be of the alpine type. Traces of the glacial epoch are less in evidence in the Tian Shan than in the Alps. The Tian Shan has few lakes. Conspicuous among them is Issyk-Kul, a large and brackish mountain lake which is bordered on the north and south by the snow-clad chains of Tian Shan (Berg, 1950).

Correspondence to: V. M. Dembitsky, Laboratory of Natural Products, Department of Organic Chemistry, The Hebrew University of Jerusalem, Jerusalem, 91904, Israel.
Received 4 June 1993; accepted 14 July 1993.

Issyk-Kul is one of the largest mountain lakes in the world. It lies in a hollow between the Kungei Ala Tau mountain range in the north and the Terskei Ala Tau in the south, rising to more than 5000 m above sea level. Issyk-Kul is one of the deepest lakes in Middle Asia. Its maximum depth is 702 m; more than 63% of the lake bottom is 100 m deep. The surface of the lake itself lies at 1624 m. The lake basin, of tectonic origin, has probably existed since the end of the Paleozoic era. Thus, it is one of the oldest lakes in Middle Asia (Berg, 1950; Matveyev, 1935).

The lake water is well mixed by the strong winds of the Issyk-Kul hollow. Earthquakes, which are frequent in the mountains of Middle Asia, cause temporary but sharp oscillations (seiches) of the surface of the lake.

Temperatures are very stable; in summer, the upper 10–15 m of the pelagic zone are warmed to 15–20°C. From 15 to 25 m, there is a clear-cut thermocline (metalimnion), the temperature dropping sharply to 6–10°C. Since its temperature does not drop below 2.75°C, the lake does not freeze in winter. The water of the lake has a fairly high salt content (up to 5.80 g/k water) (Matveyev, 1935); that is, approximately half as much salt as the waters of the Aral Sea. Despite the sharp temperature stratification and the depth of the lake, the chemical content of the water is remarkably uniform at all depths. The predominating ions, CI (24.1%) and SO_4 (23.8%), make the water undrinkable. In addition, limestone is precipitated in the lake. Its presence in the water is also indicated by the luxuriant stonewort vegetation on the bottom. The bottom fauna are poor. Little is known of the bottom fauna save that they contain the interesting endemic amphipod crustacean *Issycogammarus bergi*.

Considering the above details it may be of interest to investigate the lipid compositions of the endemic gammarids which inhabit such brackish water. The lipid composition of *Issycogammarus bergi* has not been studied.

At present, marine amphipod lipids have been studied rather intensively with respect to some interesting components such as polyunsaturated fatty acids (Lee, 1975; Morris *et al.*, 1982; Paradis and Ackman, 1966a, b; Takahashi and Yamada, 1976; Clarke *et al.*, 1985), phospholipids (Dembitsky, 1979; Dembitsky and Vaskovsky, 1976; Vaskovsky, 1989), wax esters (Kayama *et al.*, 1976; Lee, 1974, 1975; Sargent and Lee, 1975; Takahashi and Yamada, 1976), sterols and fatty acids (Fricke and Oehlenschlager, 1988) which are abundant in this order.

Although the lipid composition of marine crustaceans (Joseph, 1989) is well documented, there is only a limited amount of information on the lipid composition of the amphipods living in salty water. Recently, we studied lipid composition of the Baikalian endemic freshwater amphipods (Dembitsky *et al.*, 1994).

The present paper deals with the detailed study of fatty acid and lipid compositions of endemic gammarids living in the brackish mountain Lake Issyk-Kul.

Table 1. Lipid composition of gammarid

Lipid fractions	*Issycogammarus bergi*
Neutral lipids (NL)	68.4
Glycolipids (GL)	12.4
Phospholipids (PL)	19.2
Total lipids (TL) % dry mass	9.5

NL, GL and PL as percentages of total lipids.

Table 2. Phospholipid composition of *I. bergi*

Phospholipid	(% of total lipid phosphorus)
Phosphatidylethanolamine (PE)	11.9 ± 0.6
Phosphatidalethanolamine (PEP) (plasmalogen form)	10.8 ± 0.7
Phosphatidylserine (PS)	8.5 ± 0.4
The sum of amino-containing phospholipids	31.2
Phosphatidylcholine (PC)	38.0 ± 0.9
Phosphatidalcholine (PCP) (plasmalogen form)	7.1 ± 0.4
Sphingomyelin (SPH)	7.8 ± 0.5
The sum of choline-containing phospholipids	52.9
Phosphatidylinositol (PI)	7.4 ± 0.3
Diphosphatidylglycerol (DPG)	8.5 ± 0.5

Material and Methods

Collection of samples

Issycogammarus bergi M., belonging to the phylum Arthropoda, subphylum Crustacea, class Malacostraca, order Amphipoda, were sampled in the south-western part of Lake Issyk-Kul near Kadzhi Baj village (Kirgizstan Republic) in August 1992, at depths of 0.5–1.0 m. The animals were put in flasks in a

Table 3. Saturated fatty acids of *I. bergi*

Fatty acids	TL	NL	GL	PL
13:0	0.03	0.19	0.03	0.00
14:0	1.01	6.68	1.22	0.82
15:0	0.08	0.44	0.15	0.12
16:0	8.51	16.22	4.08	3.27
17:0	0.14	0.99	0.09	0.05
18:0	4.92	8.46	0.72	0.60
19:0	0.30	1.46	0.15	0.22
20:0	0.06	1.98	0.37	0.19
22:0	0.13	4.79	0.20	0.16
24:0	1.14	0.24	0.13	0.40
i-13:0	0.04	0.01	0.00	0.00
i-14:0	0.03	0.16	0.02	0.01
i-15:0	0.03	0.34	0.04	0.10
i-16:0	0.06	0.35	0.00	0.00
i-17:0	0.01	0.05	0.00	0.00
i-18:0	0.36	0.63	0.07	0.05
i-19:0	0.05	0.48	0.04	0.06
ai-13:0	0.03	0.06	0.01	0.00
ai-15:0	0.02	0.48	0.03	0.05
ai-17:0	0.07	0.08	0.01	0.36
ai-19:0	0.16	0.59	0.07	0.07
c-9-17:0	0.02	0.76	0.17	0.04
c-9-19:0	0.01	0.22	0.01	0.44
4,8,12-triMe-13:0	0.42	0.83	0.14	0.09
Pristanic	0.40	0.24	0.02	0.07
Phytanic	0.26	0.69	0.02	0.25
The sum of saturated	18.29	47.42	7.79	7.42
The sum of *iso*-	0.58	2.02	0.17	0.22
The sum of *anteiso*-	0.28	1.21	0.12	0.48
The sum of *cyclo*-	0.03	0.98	0.18	0.48

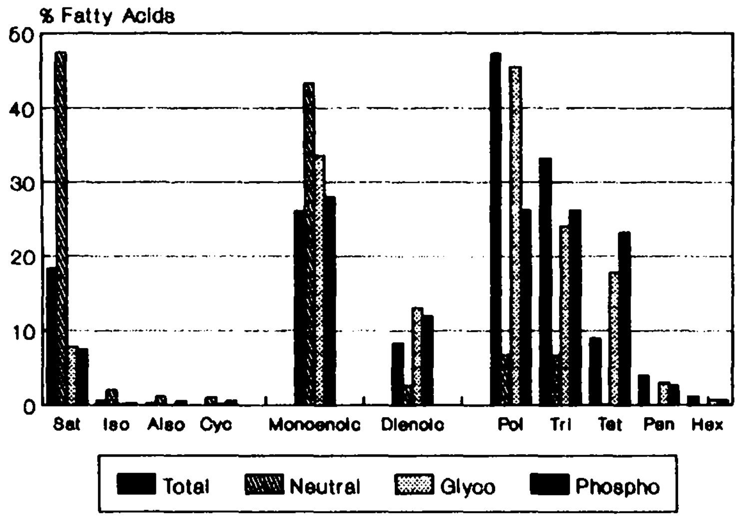

Fig. 1. Distribution of saturated (Sat) [including both *iso-* (Iso), *anteiso-* (Aiso) and *cyclo-* (Cyc)], monoenoic, dienoic and polyenoic [including trienoic (Tri), tetraenoic (Tet), pentaenoic (Pen) and hexaenoic (Hex)] fatty acids into different lipid fractions (neutral, glyco- and phospho-) and into total lipids isolated from endemic amphipod *Issycogammarus bergi*.

mixture of chloroform and methanol at the site of collection and transported to the laboratory.

Lipid analysis

The species were treated immediately in the laboratory, as described previously (Dembitsky *et al.*, 1993). Lipids were extracted, separated by column and thin-layer chromatography, as described elsewhere (Dembitsky *et al.*, 1993). A plasmalogen form of phospholipids was detected by method of Dembitsky (1988).

Fatty acids ME esters were analysed by capillary gas–liquid chromagraphy–mass spectrometry using a Shimadzu QP-1000 instrument as described previously (Dembitsky *et al.*, 1993).

Results and Discussion

The zooplankton of the open lake is monotonous, with *Diaptomus salinus* markedly predominating. The Issyk-Kulian faunae are poor. They include oligochaetes, small midge larvae and, nearer the shores, *Valvata*. The amphipods *Rivulogammarus ocellatus* and *Issycogammarus bergi* (living in Issyk-Kul only) are endemic to the mountain lakes of Middle Asia. Numerous bottom crustaceans (ostracods and cyclopods) are also encountered. The species predominating in their zooplankton are the rotifers *Synchaeta oblonga* and *Polyarthre trigla*. Crustaceans include *Cyclopus vicinus* and a very small number of *Daphnia hyalina* (Zhadin and Gerd, 1963).

The general lipid compositions from the *I. bergi* are given in Table 1. According to our results, TL in *I. bergi* amount to 9.5% of dry mass, phospholipids amount to 9.2%, and the major lipids, NL, make up 68.4%. In the examined gammarids, the major phospholipid classes

Table 4. Monoenoic fatty acids of *I. bergi*

Fatty acids	TL	NL	GL	PL
14:1n-7	0.16	0.23	0.44	0.49
14:1n-5	0.01	0.35	0.53	0.63
16:1n-9	5.30	14.43	12.84	12.69
16:1n-7	2.61	1.11	0.93	1.03
16:1n-5	0.16	0.32	0.38	0.45
17:1n-8	0.22	0.14	0.15	0.19
17:1b	0.03	0.03	0.03	0.29
18:1n-13	0.28	0.07	0.19	0.16
18:1n-11	2.86	0.33	0.57	0.43
18:1n-9	10.34	21.87	11.15	4.21
18:1n-7	0.23	0.92	0.78	0.71
18:1n-5	0.26	0.16	0.13	0.09
19:1b	0.08	0.01	0.01	0.17
19:1b	0.03	0.00	0.01	0.14
19:1n-8	0.17	0.00	0.09	0.05
19:1c	0.03	0.13	0.12	0.11
20:1n-15	0.24	0.22	0.23	0.25
20:1n-13	1.00	0.08	0.63	0.55
20:1n-11	0.33	0.12	0.21	0.26
20:1n-9	0.07	0.35	0.77	1.72
20:1n-7	0.16	0.00	0.05	0.04
21:1b	0.11	0.00	0.00	0.60
21:1n-9	0.35	0.08	0.58	0.37
22:1n-15	0.24	0.11	0.19	0.25
22:1n-13	0.08	0.07	0.34	0.29
22:1n-11	0.31	0.03	0.23	0.51
22:1n-9	0.31	0.04	0.27	0.33
22:1n-7	0.07	0.79	0.41	0.29
23:1c	0.00	0.00	0.17	0.01
23:1n-9	0.02	0.11	0.19	0.10
24:1n-9	0.06	1.17	1.05	0.54
The sum of monoenoic	26.12	43.27	33.67	27.95

a. Identified only on the basis retention time and mass spectrum of methyl ester.
b. Fatty acids probably with cyclopropanoic ring(s).
c. Unknown position of double bond(s), identified by retention time and value of molecular ion of methyl ester.

Table 5. Dienoic fatty acids of *I. bergi*

Fatty acids	TL	NL	GL	PL
16:2n-4	0.77	0.00	0.28	0.38
18:2n-9	4.51	2.01	11.04	8.94
18:2n-6	1.59	0.11	0.16	0.80
18:2b	0.12	0.00	0.00	0.12
18:2b	0.35	0.01	0.02	0.38
5,11-20:2	0.13	0.00	0.21	0.32
5,13-20:2	0.08	0.00	0.25	0.09
20:2n-9	0.20	0.00	0.46	0.53
20:2n-6	0.38	0.00	0.19	0.14
7,13-22:2	0.19	0.00	0.10	0.12
7,15-22:2	0.00	0.45	0.38	0.18
The sum of dienoic	8.32	2.58	13.09	12.00

b. See footnote in Table 4 for explanation.

were choline-containing lipids which constituted, in gammarids, 52.9% of the total phospholipids (Table 2). PC, the major phospholipid class, was found to form 38%, and their plasmalogens to form 7.1% of total phospholipids. PE is second major phospholipid at 22.7%, inclusive of the plasmalogen form. The studied species were found to possess SPH in amounts up to 7.8%. SPH is widespread among marine (Vaskovsky, 1989) and freshwater amphipods (Dembitsky *et al.*, 1994). A high percentage of plasmalogen was found for freshwater amphipods (47.6% in PE and 5.7% in PC).

Table 6. Polyenoic fatty acids of *I. bergi*

Fatty acids	TL	NL	GL	PL
16:3n-4	0.98	0.00	0.24	0.57
16:3n-6	2.22	0.00	1.19	2.19
18:3n-6	17.17	2.93	10.18	8.94
18:3n-3	10.14	3.72	10.28	12.01
20:3n-9	0.59	0.00	0.41	0.41
5,11,14-20:3	0.23	0.00	0.54	0.66
20:3n-6	0.42	0.00	0.09	0.12
20:3n-3	1.11	0.00	0.33	0.51
22:3n-9	0.09	0.00	0.07	0.55
22:3n-6	0.27	0.00	0.76	0.24
16:4n-1	1.22	0.00	1.13	0.92
18:4n-3	0.81	0.00	0.14	0.19
20:4n-6	4.22	0.08	14.99	19.92
5,11,14,17-20:4	0.52	0.00	0.60	0.72
20:4n-3	0.54	0.00	0.19	0.24
22:4n-6	0.24	0.00	0.13	0.10
22:4n-3	0.77	0.00	0.22	0.57
24:4n-6	0.55	0.00	0.39	0.58
18:5n-3	0.30	0.00	0.35	0.52
20:5n-3	0.26	0.00	0.08	0.14
21:5n-3	0.16	0.00	0.01	0.01
22:5n-6	0.21	0.00	0.11	0.15
22:5n-3	1.47	0.00	2.07	1.47
24:5n-6	1.61	0.00	0.31	0.24
22:6n-3	0.38	0.00	0.26	0.15
24:6n-3	0.79	0.00	0.38	0.51
The sum of polyenoic	47.27	6.73	45.45	52.63
The sum of trienoic	33.22	6.65	24.09	26.20
The sum of tetraenoic	8.87	0.08	17.79	23.24
The sum of pentaenoic	4.01	—	2.93	2.53
The sum of hexaenoic	1.17	—	0.64	0.66

The fatty acid composition of the freshwater amphipod species examined by capillary GC–MS is shown in Tables 3–6. We identified 94 fatty acids, including 26 saturated. The proportion of saturated fatty acids in the examined species varied from 7.4 in phospholipid fractions, to 47.42% in neutral fractions, of total fatty acids (Table 3). The major saturated acids were 14:0, 16:0, 18:0 and 24:0 of TL. Among saturated fatty acids were high percentages of cycloacids (0.98%), *iso*- (2.02%), and *anteiso*- (1.21%) in neutral fractions. Distribution of the different fatty acids among lipid fractions are shown in Fig. 1.

Most of the acids identified in the course of the present study were monoenoic, ranging from 14:1 to 24:1n-9. C_{18}, C_{20} and C_{22} acids were represented by five isomers of each; 19:1 acids were also represented by four isomers, and 16:1 was represented by three isomers (n-5), (n-7) and (n-9). A wide range (31 acids from C_{14} to C_{24}) of monoenoic acids was detected in amounts of generally less than 1%. Several acids, 16:1n-9, 16:1n-7, 18:1n-11, 18:1n-9 and 20:1n-13, were present in amounts exceeding 1%, and were characteristic of the studied species (Table 4).

Eleven fatty acids were identified as dienoic acids, among which were non-methylene-interrupted (NMI) acids: 5,11-20:2, 7,13-20:2, 7,13-22:2 and 7,15-22:2, in amounts less than 0.2% (Table 5).

Trienoic, tetra-, penta- and hexaenoic compounds are the most abundant fatty acids in the examined species (26 acids), including 10 trienoic (33.22%), eight tetraenoic (8.87%), pentaenoic (4.01%) and two hexaenoic (1.17%) acids in total lipids. A high percentage of arachidonic acid was found in phospholipid (19.92%) and glycolipid (14.99%) fractions (Table 6). Among polyenoic fatty acids in neutral fraction were found three acids only, namely: 18:3n-6 (2.93%), 18:3n-3 (3.72%) and 20:4n-6 (0.08%). The contents of major fatty acids in neutral, glyco- and phospholipid fractions are shown in Fig. 2.

The invertebrates live in brackish water and have an interesting composition of fatty acids, which differs from those of freshwater or marine species. Thus, we recently examined some Caspian invertebrates, including two crustacean species living in brackish water, namely the crab *Rhithropanopeus harrissi* and the crayfish *Astacus leptodactylus eichwaldi* (Dembitsky *et al.*, 1993). The crab and the crayfish both contained a small amount of 20:5n-3 (0.23 and 0.34%), and 3.02 and 4.06% arachidonic acids, respectively. Marine amphipods were usually found to contain a high level of

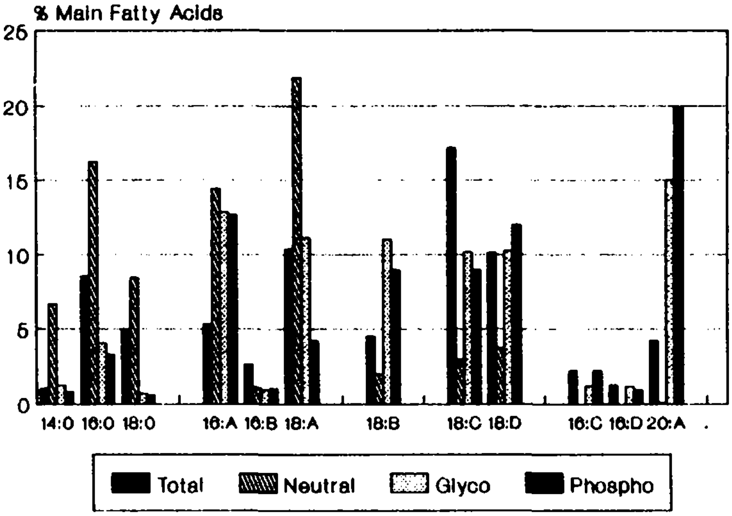

Fig. 2. The quantitative content of main fatty acids identified from different lipid fractions and total lipids of endemic amphipod *Issycogammarus bergi*. Saturated fatty acids: 14:0, 16:0 and 18:0. Monoenoic fatty acids: 16:1n-9 (16:A), 16:1n-7 (16:B), and 18:1n-9 (18:A). Dienoic fatty acids: 18:2n-9 (18:B). Polyenoic fatty acids: 16:3n-6 (16:C), 16:4n-1 (16:D), 18:3n-6 (18:C), 18:3n-3 (18:D), and 20:4n-6 (20:A).

eicosapentaenoic acid. The Antarctic amphipod *Thermisto gaudichaudii* contains an extreme content of 20:5n-3 (21.1%) and 22:6n-3 (15.9%) in total fatty acids (Fricke and Oehlenschlager, 1988). Some mid-water oceanic crustaceans *Gnathophausia* sp., *Acanthepyra purpurea* and *Nematobrachion sexspinosus* contain a very high content of 22:6 (32.5, 51.4 and 32.4%, respectively) in phospholipids (Morris and Sargent, 1973). Other freshwater (Morris, 1984) and marine crustaceans also contain high levels of eicosapentaenoic acid (Lee, 1974, 1975; Castell, 1981; Clarke *et al.*, 1985; Joseph, 1989; Webster and Lovell, 1991), and docosahexaenoic acid (Morris, 1984; Taghon, 1988; Morris and Sargent, 1973; Lee, 1975; Fricke and Oehlenschalager, 1988).

Morris *et al.* (1982) reported interesting data for the acclimated freshwater amphipod *Gammarus duebeni* to seawater. The level of eicosapentaenoic acid increases from 6% (field controls) to 23% (100% seawater, 60 days acclimated) in the gill phospholipids; and decreases to 6.7% (100% seawater, 66 days) after acclimation of the animals to different salinities. Changes in the apparent permeability to water occur in a number of euryhaline species of crustaceans on transfer from one salinity to another (Smith, 1967; Cornell, 1973; Bolt *et al.*, 1980; Dawson *et al.*, 1984).

Based on the present results, it may be concluded that endemic gammarids living in brackish water have interesting compositions of fatty acids which differ from those of both their freshwater and marine relatives.

References

Berg L. S. (1950) *Natural Regions of the USSR* (Edited by Morrison J. A. and Nikiforoff C. C.). MacMillan, New York.

Bolt S. R. L., Dawson M. E., Inman C. B. E. and Lockwood A. P. M. (1980) Variation of apparent permeability to water and sodium transport in *Gammarus duebeni* exposed to fluctuating salinities. *Comp. Biochem. Physiol.* **67B,** 465–473.

Castell J. D. (1981) Fatty acid metabolism in crustaceans. In *Proceedings of the Second International Conference on Aquaculture Nutrition: Biochemical and Physiological Approaches to Shellfish Nutrition* (Edited by Pruder G., Langdon C. J. and Conklin D. E.), Special publication No. 2, pp. 124–145. World Mariculture Society, Baton Rouge, LA.

Clarke A., Skadsheim A. and Holmes L. J. (1985) Lipid biochemistry and reproductive biology in two species of Gammaridae (Crustacea: Amphipoda). *Mar. Biol.* **88,** 247–263.

Cornell J. C. (1973) A reduction in water permeability in response to a dilute medium in the stenohaline crab *Libinia emarginata* (Brachyura, Majidae). *Biol. Bull. mar. biol. Lab., Woods Hole* **145,** 430–431.

Dawson M. E., Morris R. J. and Lockwood A. P. M. (1984) Some combined effects of temperature and salinity on water permeability and gill lipid composition in the amphipod, *Gammarus duebeni*. *Comp. Biochem. Physiol.* **78A,** 729–735.

Dembitsky V. M. (1979) Plasmalogens of phospholipids of marine invertebrates. *Biol. Morya, Vladiv.* **5,** 86–90.

Dembitsky V. M. (1988) Quantification of plasmalogen, alkylacyl and diacyl glycerophospholipids by micro-thin-layer chromatography. *J. Chromat.* **436,** 467–473.

Dembitsky V. M., Rezanka T. and Kashin A. G. (1993) Comparative examination of phospholipids and fatty acids from some Caspian invertebrates. *Comp. Biochem. Physiol.* **104B,** 617–622.

Dembitsky V. M., Rezanka T. and Kashin A. G. (1994) Comparative study of the endemic freshwater fauna of Lake Baikal—III. Phospholipid and fatty acid composition of amphipod crustacean of genus *Eulimnogammarus*. *Comp. Biochem. Physiol.* **107B,** 317–323.

Dembitsky V. M. and Vaskovsky V. E. (1976) Distribution of plasmalogens in various phospholipids classes of the marine invertebrates. *Biol. Morya, USSR* **5,** 68–72.

Fricke H. and Oelenschlager J. (1988) Fatty acid and sterol composition of the Antarctic amphipod *Themisto gaudichaudii* Guern 1828. *Comp. Biochem. Physiol.* **89B,** 39–42.

Joseph J. D. (1989) Distribution and composition of lipids in marine invertebrates. In *Marine Biogenic Lipids, Fats and Oils* (Edited by Ackman R. G.), Vol. 2, pp. 49–144. CRC Press, Boca Raton, FL.

Kayama M., Ikeda Y. and Komaki Y. (1976) Studies on the lipids of marine zooplankton with special reference to wax ester distribution in crustaceans and its *in vivo* formation. *Yukagaku* **25,** 329–338.

Lee R. F. (1974) Lipids of zooplankton from Bute Inlet, British Columbia. *J. Fish. Res. Bd Can.* **31,** 1577–1581.

Lee R. F. (1975) Lipids of Arctic zooplankton. *Comp. Biochem. Physiol.* **51B,** 263–266.

Matveyev V. P. (1935) Hydrological explorations on Issyk-Kul in 1928. In *Lake Issyk-Kul. Council for Study of Natural Resources.* pp. 35–124. Academy of Sciences of the USSR.

Morris R. J. (1984) The endemic faunae of Lake Baikal: their general biochemistry and detailed lipid composition. *Proc. R. Soc. Lond. B* **222,** 51–78.

Morris R. J., Lockwood A. P. M. and Dawson M. E. (1982) An effect of acclimation salinity on the fatty acid composition of the gill phospholipids and water flux of the amphipod crustacean *Gammarus dueberi*. *Comp. Biochem. Physiol.* **72A,** 497–503.

Morris R. J. and Sargest J. R. (1973) Studies on the lipid metabolism of some oceanic crustaceans. *Mar. Biol.* **22,** 77–83.

Paradis M. and Ackman R. G. (1966a) Localization of a marine source of odd chain-length fatty acids. 1. The amphipod, *Pontoporeia femorata* (Kroyer). *Lipids* **11,** 863–870.

Paradis M. and Ackman R. G. (1966b) Localization of a marine source of odd chain-length fatty acids. 2. Seasonal propagation of odd chain-length monoethylenic fatty acids in a marine food chain. *Lipids* **11,** 871–879.

Sargent J. R. and Lee R. F. (1975) Biosynthesis of lipids in zooplankton from Saanich Inlet, British Columbia, Canada. *Mar. Biol.* **31,** 15–19.

Smith R. I. (1967) Osmotic regulation and adaptive reduction of water permeability in a brackish water crab, *Rhithropanopaes harrisii* (Brachyura, Xanthidae). *Biol. Bull. mar. biol. Lab., Woods Hole* **133,** 643–658.

Taghon G. L. (1988) Phospholipid fatty acid composition of the deep-sea hydrothermal vent polycheate *Paralvinella palmiformis* (Polychaeta–Amphareidae): effects of thermal regime and comparison with two shallow-water confamilial species. *Comp. Biochem. Physiol.* **91B,** 593–596.

Takahashi H. and Yamada M. (1976) Lipid composition of seven species of crustacean plankton. *Bull. Jap. Soc. Sci. Fish.* **42,** 769–773.

Vaskovsky V. E. (1989) Phospholipids. In *Marine Biogenic Lipids, Fats and Oils* (Edited by Ackman R. G.), Vol. 1, pp. 199–242. CRC Press, Boca Raton, FL.

Webster C. D. and Lovell R. T. (1991) Lipid composition of three geographical sources of brine shrimp nauplii (*Artemia* sp.). *Comp. Biochem. Physiol.* **100B,** 555–559.

Zhadin V. I. and Gerd S. V. (1963) *Fauna and Flora of the Rivers, Lakes and Reservoirs of the USSR*, pp. 476. Israel Program for Scientific Translations, Jerusalem.