Specific Fatty Acid Status in the White Sea Herring From Different Bays of the White Sea in Regard to Ecological Factors

Role of Fatty Acids in Ecological and Biochemical Adaptations of Fishes in Sub-Arctic

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***Abstract*—Fatty acid status of the White Sea herring from Dvina, Kandalaksha and Onega Bays of the White Sea in autumn was investigated. The fatty acid profile of the herring groups from studied sites was characterized by high amount of monounsaturated fatty acids, among which 18:1(n-9) and 16:1(n-7) dominated. The herring from Kandalaksha Bay had higher ($p \leq 0.05$) level of 18:1(n-9) fatty acid than the fish from Onega Bay, whereas the last one was characterized by low ($p \leq 0.05$) level of 16:1(n-7). Polyunsaturated fatty acids (PUFA) were the second largest group in the herrings, mainly because of (n-3) fatty acids. In the herring from Onega Bay, the level of 22:6(n-3) was significantly ($p \leq 0.05$) higher than in other fishes. Besides that, this fishes were characterized by high level of (n-3)/(n-6) PUFA, 18:3(n-3)/18:3(n-6) and 16:0/18:1(n-9) ratios. Detected lipid status reflects ecological and biochemical features of fish adaptation in relation to specific ecological factors of the inhabitation areas of the White Sea and choice of optimal and adequate survival strategy in sub-Arctic. The results of this work present information about some features of mechanisms of adaptations on the level of lipid spectra and the role of fatty acid constituents in ecological and biochemical adaptions of the White Sea herring.**

Keywords-biochemical adaptation; fatty acids; lipids; the White Sea herring; Sub-Arctic

I. INTRODUCTION

Lipids and fatty acids have important functions in cell metabolism and play a significant role in biochemical adaptations of organisms to changing environmental conditions (abiotic, biotic, anthropogenic factors) [1-4]. Fish lipids, mainly their fatty acids (FA), are the basic source of metabolic energy and structural elements for their growth, development, reproduction, and survival [1-3]. Especially, it concerns fishes that have high level of lipids in their bodies as herring, salmon, trout etc. [5-8]. Fatty acids are one of the most sensitive lipid components that take active part in compensatory mechanisms of adaptation of organisms in normal and in stress [1-2]. Quick change of lipid and fatty-acid profile of fish tissues and organs contributes to optimal process of life activity and their adaptation, including thermal adaptation, to changing environment [3].

The population of the White Sea herring *Clupea pallasii marisalbi* Berg (Clupeiformes, Clupeidae) is one of the similar forms of Pacific herring and one of the main and the most commercial fish in the White Sea. The local groups of herring are subject to particular hydrological conditions in different bays of the sea [9]. The biochemical studies of the White Sea herring are not numerous. Biochemical adaptations to specific ecological conditions of the White Sea (specific temperature regime, high speed of currents, water exchange with the Barents Sea, small depths, low concentration of mineral substances and organic compounds, low level of salinity, relatively short vegetation period, etc.) cannot ignore changes in lipid status of the fish.

Fatty acid status of the White Sea herring from different bays (Dvina, Kandalaksha and Onega bays) in autumn season was investigated.

The results of this research will contribute to the current knowledge about the role and functions of lipids that take part in development and formation of biochemical adaptations of other fish species. They also present new information about the features of mechanisms of adaptations on the level of lipid status of the studied fish in the sub-Arctic region.

The presented paper consists of next sections: material and methods, results of fatty acid analysis of the White Sea herring from different Bays, discussion, conclusion and acknowledgement.

II. MATERIAL AND METHODS

The White Sea herring adults were collected in October, 2010 in Dvina ($64^{o}57'$ $38^{o}23'$), Kandalaksha ($67^{o}02'$ $32^{o}23'$) and Onega Bays ($64^{o}59'$ $36^{o}37'$) of the White Sea at depth 50, 25 and 38 meters and at temperature 6.5^{o}, 2.9^{o} and 6.7^{o}C, respectively, according the method described by [10]. In total 66 individual specimens (n) were analyzed.

Flesh of the fish was homogenized in 10 volumes (10 ml each) of 96% ethyl alcohol mixed with 0.001% of the antioxidant. Sample homogenates were placed in glass vials and stored onboard the ship in a cold room until delivery to the laboratory. The material was then fixed in a solvent system of chloroform:methanol (2:1, volume/volume), and

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total lipids (TL) were extracted following the method [11]. The residues recovered after lipid extraction of the flesh were dried over phosphoric anhydride until the samples had reached a constant weight. The residues were weighted to determine the approximate percentage of total lipids (T) on a dry-weight basis (1), where β – lipids extracted (g) and α – residue weight (g):

$$T = \beta * 100/(\alpha + \beta). \quad (1)$$

The fatty acids composition of the total lipids was analyzed by gas-liquid chromatography. Fatty acid methyl esters (FAME) were identified with a "Chromatek-Crystall-5000.1" (Russia) gas chromatograph with flame-ionization detector on a Zebron Capillary Gas Chromatographic Column (Phenomenex, USA). The isothermic column regime was used (225^0C), temperature of detector – 250^0C; temperature of evaporator – 240^0C. For recording and integration "Chromatek-Analytik-5000.1" software (Russia) was applied.

The research was carried out using the facilities of the Equipment Sharing Centre of the Institute of Biology, KarRC of RAS

Data were tested for normal distribution. Differences between means of fatty acids of total lipids in herrings from studied areas of the sea were analyzed by analysis of variance ANOVA ($p \leq 0.05$).

III. RESULTS

The biochemical analysis showed that the herring from Kandalaksha Bay had higher level of TL (41.7% dw) compared to the herring from Dvinsky and Onega Bays (34.5%; 31.8% dw, respectively) (Table I). In Table 1, Values in the same row with the same superscripted letter are not significantly different ($p>0.05$) among fishes from different areas of the Sea and values in the same row with the different superscripted letter are significantly different ($p \leq 0.05$) among fishes.

TABLE I. TOTAL LIPIDS (% DRY WEIGHT) AND FATTY ACID SPECTRUM (% OF TOTAL FA) IN THE WHITE SEA HERRING FROM DVINA, KANDALAKSHA AND ONEGA BAYS IN AUTUMN. DATA ARE M±M.

FA/Bay	Dvina (A)	Kandalaksha (B)	Onega (C)
n	20	21	25
TL	34.5±0.6 ^{BC}	41.7±1.4 ^{AC}	31.8±0.8 ^{AB}
12:0	0.30±0.21 ^{B}	0.04±0.01 ^{AC}	0.18±0.01 ^{B}
14:0	10.53±0.35 ^{BC}	12.40±0.28 ^{AC}	8.80±0.43 ^{AB}
15:0	0.89±0.03 ^{BC}	0.57±0.02 ^{AC}	0.72±0.03 ^{AB}
16:0	21.66±0.47	21.40±0.35	22.26±0.62
17:0	0.47±0.06 ^{C}	0.37±0.06 ^{C}	0.90±0.02 ^{AB}
18:0	2.25±0.06 ^{BC}	1.66±0.07 ^{AC}	3.21±0.17 ^{AB}
20:0	0.87±0.03 ^{B}	0.66±0.05 ^{AC}	0.94±0.05 ^{B}
24:0	0.16±0.01 ^{BC}	0.12±0.01 ^{AC}	0.25±0.02 ^{AB}
Sum SFA	37.12±0.80	37.23±0.66	37.26±0.81
14:1(n-5)	0.39±0.01 ^{C}	0.37±0.01 ^{C}	0.34±0.01 ^{AB}
16:1(n-9)	0.73±0.06	0.40±0.04 ^{C}	0.74±0.10 ^{B}
16:1(n-7)	11.31±0.29 ^{C}	11.45±0.22 ^{C}	7.73±0.24 ^{AB}
16:1(n-5)	0.25±0.02	0.25±0.02	0.29±0.03
17:1(n-7)	0.35±0.04 ^{C}	0.34±0.04 ^{C}	0.59±0.03 ^{AB}
18:1(n-9)	19.80±0.35	20.14±0.33 ^{C}	18.40±0.73 ^{B}
18:1(n-7)	4.05±0.28 ^{BC}	3.19±0.26 ^{A}	3.39±0.10 ^{A}
18:1(n-5)	0.82±0.05 ^{BC}	1.09±0.05 ^{AC}	0.69±0.03 ^{AB}
20:1(n-11)	0.24±0.05	0.17±0.02	0.58±0.24
20:1(n-9)	1.46±0.12 ^{B}	2.10±0.21 ^{A}	1.93±0.56
20:1(n-7)	0.35±0.01	0.33±0.01	0.33±0.02
22:1(n-11)	0.88±0.08 ^{B}	1.44±0.17 ^{A}	1.40±0.46
22:1(n-9)	0.32±0.02	0.32±0.02	0.79±0.42
22:1(n-7)	0.23±0.01 ^{C}	0.21±0.01 ^{C}	0.43±0.03 ^{AB}
24:1(n-9)	0.22±0.08 ^{C}	0.27±0.08 ^{C}	1.18±0.04 ^{AB}
Sum MUFA	41.40±0.75	42.06±0.51	38.81±1.57
16:2(n-9)	0.23±0.02 ^{BC}	0.16±0.02 ^{A}	0.13±0.02 ^{A}
18:2(n-9)	0.08±0.01	0.06±0.01	0.06±0.01
20:2(n-9)	0.09±0.01	0.10±0.01 ^{C}	0.07±0.01 ^{B}
22:2(n-9)	0.05±0.01 ^{C}	0.05±0.01 ^{C}	0.02±0.01 ^{AB}
Sum (n-9) PUFA	0.44±0.04 ^{C}	0.37±0.02 ^{C}	0.29±0.02 ^{AB}
16:2(n-7)	0.17±0.02 ^{BC}	0.09±0.01 ^{A}	0.10±0.02 ^{A}
18:2(n-7)	0.05±0.01 ^{C}	0.07±0.01 ^{C}	0.31±0.07 ^{AB}
Sum (n-7) PUFA	0.22±0.02	0.16±0.02 ^{C}	0.41±0.09 ^{B}
16:2(n-6)	0.41±0.05 ^{BC}	0.20±0.03 ^{A}	0.20±0.02 ^{A}
16:3(n-6)	0.35±0.03 ^{BC}	0.21±0.03 ^{AC}	0.01±0.00 ^{AB}
18:2(n-6)	1.31±0.05 ^{C}	1.21±0.02 ^{C}	1.06±0.05 ^{AB}
18:3(n-6)	0.15±0.01 ^{B}	0.11±0.01 ^{AC}	0.15±0.01 ^{B}
20:2(n-6)	0.22±0.01 ^{B}	0.20±0.01 ^{AC}	0.24±0.01 ^{B}
20:4(n-6)	0.28±0.02	0.30±0.01	0.29±0.02
Sum (n-6) PUFA	3.52±0.16 ^{BC}	2.70±0.07 ^{A}	2.58±0.09 ^{A}
18:3(n-3)	0.76±0.06 ^{B}	0.92±0.03 ^{A}	0.93±0.07
18:4(n-3)	1.33±0.10 ^{BC}	1.75±0.07 ^{A}	2.12±0.16 ^{A}
20:4(n-3)	0.42±0.04 ^{BC}	0.66±0.03 ^{AC}	0.53±0.03 ^{AB}
20:5(n-3)	6.51±0.52	6.61±0.31	7.38±0.47
22:6(n-3)	5.25±0.45 ^{C}	5.11±0.27 ^{C}	7.01±0.65 ^{AB}
Sum (n-3) PUFA	15.12±1.15 ^{C}	15.84±0.68 ^{C}	19.19±1.35 ^{AB}
Sum PUFA	21.48±1.38	20.71±0.74	23.92±1.48
(n-6)/(n-3)	0.26±0.02 ^{BC}	0.18±0.01 ^{AC}	0.15±0.01 ^{AB}

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18:3(n-3)/18:2(n-6)	0.58±0.04 ^{BC}	0.76±0.02 ^{AC}	0.89±0.05 ^{AB}
16:0/18:1(n-9)	1.10±0.31 ^{C}	1.07±0.27 ^{C}	1.24±0.05 ^{AB}

The fatty acid spectrum of total lipids of the White Sea herring from all investigated bays were performed by monounsaturated fatty acids (MUFA) (from 38.81 to 42.06% of total FA), among which 18:1(n-9) and 16:1(n-7) FA dominated (within the limits 18.40 – 20.14% and 7.73 – 11.45% of total FA, respectively) (Table 1). The herring from Kandalaksha Bay had higher ($p \leq 0.05$) level of 18:1(n-9) FA than the fish from Onega Bay, whereas the last one was characterized by low ($p \leq 0.05$) level of 16:1(n-7) FA. It was determined that the amount of other MUFAs – 18:1(n-7), 20:1(n-9), 22:1(n-11), 22:1(n-7), 24:1(n-9) – did not exceed 5.0 % of total FA but their levels were significantly different ($p \leq 0.05$) among fishes from different bays of the sea.

It was shown that the level of saturated FA (SFA) was high. Among SFA – 16:0 and 14:0 FA dominated and ranged from 21.40 to 22.26% and from 8.80 to 12.40% of total FA, respectively in all investigated groups of herring. Low concentration of 18:0 FA (1.66 – 3.21% of total FA) was detected in relation to fish from all sea regions.

Polyunsaturated FA (PUFA) amount was within the limits from 20.71 to 23.92% of total FA, mainly because of (n-3) polyenic FA (15.12 – 19.19% of total FA) in the White Sea herring from all investigated areas. Among (n-3) PUFA – 20:5(n-3) and 22:6(n-3) were dominated. The White Sea herring from Onega Bay was slightly distinguished by high level ($p \leq 0.05$) of these acids (mainly 22:6(n-3)) and (n-3)/(n-6), 18:3(n-3)/18:2(n-6) and 16:0/18:1(n-9) ratios.

Thus, high level of total MUFAs ($p \geq 0.05$) was typical for the White Sea herring from all the studied inhabitations of the White Sea. The variations of some FA: with high level 17:0, 17:1(n-7), 18:0, 18:2(n-7), 22:1(n-7), 24:1(n-9), 22:6(n-3) and low level - 14:0, 16:1(n-7), 16:3(n-6), 18:1(n-5), 18:2(n-6), - differed the herring from Onega bay from others. Besides that, this fishes were characterized by high level of (n-3)/(n-6)PUFA, 18:3(n-3)/18:3(n-6) and 16:0/18:1(n-9) ratios.

IV. DISCUSSION

Fatty acid status of the White Sea herring from different bays of the White Sea (Dvina, Kandalaksha and Onega Bays) in autumn season was investigated. High level of MUFAs is one of the typical features of aquatic marine organisms in northern latitudes. MUFA were the dominant FA group in all the herring groups in studied sites of the White Sea. Among MUFA – 18:1(n-9) and 16:1(n-7) were the most abundant. It is known that some MUFAs are dietary derived and they are trophic biomarkers for marine organisms: 18:1(n-9) FA is the main biomarker of dinoflagellates and bacterial plankton while 16:1(n-7) FA is the biomarker of diatoms microalgae [12-16]. Dinoflagellates and diatoms microalgae make the main contribution to biomass of the White Sea phytoplankton [17]. In the end of October (time of material collection) the phytoplankton bloom period finishes. In water there are just mass species of flagellates and diatoms algae disappears [18]. This fact explains lower level of 16:1(n-7) FA in herring from Onega Bay in comparison with fish from other places of the White Sea.

It was outlined that herrings of the studied bays had low level of 20:1(n-9) and 22:1(n-11) FA (1.46 – 2.10 % and 0.88 – 1.44 % of total FA, respectively) that are synthesized *de novo* only by planktivorous species *Calanus* spp. [15] [19]. Therefore, in this period these zooplankton species made up small amount of herring diet that was evidently reflected on FA spectrum of the White Sea herring.

Respectively high level of SFA, the domination of 16:0 and 14:0 FA, was connected not only with *de novo* synthesis but also with their accumulation related to feeding on phyto- and zooplankton, presence and availability of which differed in food web and food habits of herring from different areas of the White Sea. Low level of 14:0 as well as 16:1(n-7) FA, biomarkers of diatoms, in herrings from Onega Bay in comparison with herrings from other studied area might be connected with decrease of producing capacity of diatoms algae in this period. The 16:0/18:1(n-9) ratio was higher in herring from Onega Bay. It indicated higher degree of intensity of lipid metabolism [20].

Low level of PUFA, especially 22:6(n-3) FA among the White Sea herring in comparison with other arctic and subarctic fish species shows that these FA played secondary role in terms of biomembrane functioning in these conditions. The correlation between (n-3) and (n-6) PUFA plays a specific role in organism due to competitive relations in the process of their metabolism [21]. It is also shown that cold water pelagic fishes have higher (n-3) PUFA level that satisfy their need in essential fatty acids more than (n-6) and (n-9) PUFA [4] [22] [23]. The (n-3)/(n-6) PUFA ratio was higher in herring from Onega Bay than among fish groups in other bays of the sea. The change of lipid unsaturation degree is one of the basic mechanisms of regulation of activity of membrane connected enzymes under environment fluctuations [1] [24].

V. CONCLUSION

The difference in terms total lipids and fatty acids among the White Sea herring from the studied bays of the White Sea was presented. The results of the paper present information about some features of mechanisms of adaptions on the level of lipid spectra and the role of fatty acid constituents in ecological and biochemical adaptations of fishes under study. It showed various influence of the complex of ecological factors, in this research mainly biotic (trophic). The determined spectra of fatty acids among the White Sea herring from its different bays confirm that in the period of fattening the main source of their nutrition were the diatoms algae and dinoflagellates and the income of copepods was small. Such lipid status reflects ecological and biochemical features of fish adaptation in relation to specific hydrobiological and trophoecological factors of the inhabitation areas of the White Sea and choice of optimal and adequate survival strategy in future. Detailed analysis of

separate fatty acids, their ratios of herrings from different sites of the White Sea in respect to abiotic factors (temperature, depth, water currents) will be given in future. The results of this work will contribute to the current knowledge about the role and functions of lipids and lipid components in biochemical adaptations of fishes from high latitudes.

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