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THE LIPID COMPONENT OF CHITIN AND CHITOSAN STRUCTURES

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The qualitative and quantitative compositions of the lipids isolated from the chitin, chitosan, and shell of the crab *Paralitodes camtshatica* have been investigated for the first time. The chitin was isolated from the shell of the crab *Paralitodes camtshatica* by two demineralizations with 1 N hydrochloric acid for 1 h at room temperature, alternating with two deproteinizations with 1 N caustic soda for 1 h at 90°C. Chitosan was obtained by deacetylating the chitin with 50% aqueous caustic soda at 100°C for 0.5 h.

The lipids were extracted by the usual Folch [1] and Bligh-Dyer [2] procedures. A weighed sample was covered with a 1:2 mixture of chloroform and methanol, and 20% of water was added to the combined extract. After phase separation, the chloroform layer was taken off, and the solvent was eliminated in a rotary evaporator. The yields of lipids from the crab shell and from the chitin and chitosan were 0.50, 0.20, and 0.18%, respectively.

The qualitative compositions of the lipids were studied by microthin-layer chromatography [3, 4]. Phospholipids were found in all the materials. In samples of the crab shell we found a set of three phospholipids: phosphatidylcholine (PC), phosphatidylethanolamine (PE), and phosphatidylserine (PS). In the sample of chitin we found PC and in the chitosan diphosphatidylglycerol (DPG).

The quantitative determination of the phospholipids was carried out by a known spectrophotometric method at 825 nm [4] (SF-26 spectrophotometer). The amounts of phosphorus in the samples studied are given in Table 1.

The qualitative and quantitative compositions of the fatty acids from the lipids were determined (Table 2).

The methyl esters of the fatty acids were obtained by the method of Hartman et al. [5]. Chromatograms were obtained on a Tsvet-100 GLC with a flame-ionization detector. Glass column (3 mm × 3 m); phase — 6.5% of diethyleneglycol adipate on Chromaton NAW, 0.160—200 μm. Evaporator temperature 250°C, column temperature 197°C. Rate of flow of carrier gas (argon) 35 ml/min; internal standard — palmitic acid. The sample of shell contained a broad spectrum of fatty acids. Among them the monoenic acids 14:1, 16:1, and 18:1 predominated. The contribution of the $18{:}1^{9}$ acid was particularly large (22.56%). The rarely encountered 20:3 and 22:5 fatty acids were detected. According to our findings, the lipids of the chitin contained 12 fatty acids. Among them the 16:0 neo-acid predominated (22.66%). In the lipids of the chitosan, as compared with the chitin, the acids of the 16:0-neo, 16:0-iso, $16{:}1^{6}$, 18:-iso, $18{:}2^{6,9}$, $18{:}3^{9,12,15}$, and 18:4 families disappeared, while a series of new acids — 18:0, $18{:}2^{9,12}$, and others — appeared.

TABLE 1. Amounts and Compositions of the Phospholipids in the Shell, Chitin, and Chitosan

Sample	Phospholipids	Amount of phosphorus	
		in the lipid extract, %	in the sample, $\%\cdot10^{-3}$
Shell	PC	1.46	9.5
	PE	0.88	5.7
	PS	1.04	6.8
Chitin	PC	0.81	1.6
Chitosan	DPG	1.41	2.4

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TABLE 2. Qualitative and Quantitative Compositions of the Fatty Acids from the Lipids

Acid	Amount of fatty acids, % by weight		
	shell	chitin	chitosan
12:0	1.2	—	5.9
13:0			4.2
14:0	6.3	12.4	7.6
14:1	3.8		
15:0	3.2		4.2
15:0 anteiso			5.4
16:0	1.1	2.7	16.5
16:0 neo		22.6	
16:0 iso	10.7		
$16:1^{6}$	8.8	12.1	
16:2	3.5		6.7
18:0	2.0		3.3
18:0 iso	4.6	2.6	
$18:1^{9}$	22.5	19.1	6.2
$18:2^{9,\ 12}$	10.1		18.3
$18:2^{6,\ 9}$		5.4	
$18:3^{9,\ 12,\ 15}$		3.4	
18:4		5.8	
19:0			9.0
19:yn	4.1		2.7
20:0	2.2		1.9
20:yn	5.1	3.0	6.1
$20:3^{8,\ 11,\ 14}$	3.0		1.6
$20:4^{5,\ 8,11,\ 14}$		1.5	Сл.
21:0	1.8		0.3
Unidentified	4.4		
$22:5^{7,\ 10,\ 13,\ 16,\ 19}$	1.6		
Unidentified		9.3	

Thus, the first information has been obtained on the presence of concrete phospholipids and fatty acids in the rigid structures of chitin and chitosan. The presence of phospholipids in these polymers may make a definite contribution to their pharmacological activity since phospholipids themselves are pharmaceutical preparations. Furthermore, with the presence of lipid components in the chitin and chitosan it is possible to speak of a partial similarity of their structure to lipid A [6]. There are, in fact, results showing a similarity of some pharmacological properties of chitin and of lipid A, especially immunomodulatory properties.

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