

# Composition and the Contents of Fatty Acids of Some Organs of Gastropod Mollusk *Volutopsius Castaneus*

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## Abstract

The composition and the contents of Fatty Acids (FAs) some organs (a foot, gonads, a liver) edible gastropod mollusk *Volutopsius castaneus* are investigated. Dependence of FA content on time of gathering of a mollusk is shown. Sixty three FAs are identified. The contribution of saturated FAs in lipids of the investigated organs includes 14:0, 16:0, 17:0, 18:0. The contribution of polyunsaturated FAs (40-60 %) is especially considerable. That fact is remarkable, that total content polyunsaturated FAs (PUFAs) during all season changed slightly. The highest level PUFA was observed in a foot (55-60 %). The main C20 PUFAs were arachidonic (20:4n6) and eicosapentaenoic (20:5n3) acids. At tissues investigated were present characteristic for mollusks non-methylene interrupted FAs: 20:2Δ5, 11 and 20:2Δ5, 13 (to 4.5 %).

**Keywords:** essential fatty acids; gastropod mollusk; influence time catch;

## Introduction

Research objective - search of the food objects which are a source of essential Fatty Acids (FAs) and also to track dynamics of changes in composition and contents FAs of a mollusk depending on its time catch. For research three organs of a mollusk are taken: a foot, a liver, gonads. There is no winter period because of catch problems. The data obtained are presented in the table 1.

*Volutopsius castaneus* is a gastropod mollusk, widely distributed in the waters of the Japan and the Okhotsk Seas. Despite the fact that the gastropods are the most numerous class of mollusks, most of the works relate to the study of their biology. There is a well-known reference book on the ecology of marine gastropods. It is the single in the world literature generalizing work on the ecology of marine gastropods [1]. At the same time, data on their chemical composition, in particular on the composition of FAs, are limited. This class of mollusks refers to predators, they hunt for other mollusks, mostly bivalves, polychaetes, echinoderms and feed on even carrion. The saliva of the *V. castaneus* has a paralyzing effect on the victim. Inhabit the Gastropod Mollusks (GM) on the bottom.

The chemical composition of GM includes a sufficiently large number of indispensable useful compounds of natural origin. Mollusks play an important role not only in the food industry, but also in the pharmacological, medical, manufacturing and cosmetic industries [2]. Besides positive properties, there is a danger to humans from mollusks. Some mollusks can cause fatal harm to the human body due to the special chemical composition of toxic poisons, which are equipped with the animal's body for self-defense or attack. In some carnivorous GM secretions of salivary glands contain sulfuric acid (2-4%) or some organic acids. Such mollusks feed on other mollusks and echinoderms. It is shown that in the Far Eastern GM to the group of Biologically Active Substances (BAS) are terpenoids that affect the functions of various parts of the central nervous system. Also GM contains homarin, serotonin, histamine and other biogenic amines of the heterocyclic series, capable of altering the activity of smooth muscles, the tone of blood vessels [3]. The most valuable edible part of the trumpeter is the leg meat. Water content in the blower is 71.8%, protein - 18.5%, lipids - 1.2%, mineral substances - 6.3%, carbohydrates - 4.1%. Eleven fatty acids were identified. Among them there are saturated, monoenoic and Polyunsaturated Fatty Acids (PUFA). Among the lasts there are arachidonic (20: 4) to 2.5% in the sum of lipids and docosahexaenoic PUFA (22: 6w3) also with a content of 2.5% (in the sum of lipids).

In the literature there is information about the GM of other regions. Thus, the biogeochemical characteristics of the dominant GM species of the stony littoral of southern Baikal are given [4]. Two types of GM have been studied for the content of microelements in them. It is shown that in the tissues of the leg the content of microelements is lower than in the body. The prospects of industrial use of the Barents Sea GM (*Buccinum undatum* and *Neptunea despecta*) [5] are investigated. Data on exchange of substances in the foot of the GM family Muricidae of South Vietnam are given in the Miroshnichenko's work [6].

## Experimental Section

**Animal material:** *Volutopsius castaneus* (family Buccinidae) collected at coast of Tatar strait (an island Sakhalin) on depth of 0.5-1.0 m in May, June, July and September, 2015.

**Lipid extraction:** It is performed with a method Bligh – Dyer [7].

**Methyl esters FAs** received on method J.P. Carreau and J.P. Dubacq [8]. Clearing of methyl esters FAs spent with the help microcolumn chromatography in system of solvents hexane: diethyl ether (98:2 v/v) [9].

The analysis of methyl esters FAs spent on gas chromatograph Shimadzu GC-2010 plus with flame - ionization detector and

capillary column Supelcowax-10 (length - 30 m, internal diameter - 0,25 mm, a thickness of a phase - 0,25 microns; injector temperature - 250°C, column - 200°C, the detector - 270°C). Fatty acids were identified by means of standards and on values of equivalent length of a chain [10]. For more strict identification from methyl esters FAs synthesized 4'4'- dimethyl-2-oxozoline derivatives by Svetashev's method [11] which then were analyzed by gas- liquid chromatography – mass spectrometry on device Shimadzu GCMS-QP5050A with column MDN-5S (the temperature program - 160°C, then 2°C/minutes to 250°C).

## Results

The data received are presented in the table.

**Table 1:** Seasonal change fatty acid composition of some organs of gastropod mollusk *Volutopsius castaneus*

FAs	Gonads				Liver				Foot			
	May	June	July	September	May	June	July	September	May	June	July	September
14:00	1,10	2,40	1,17	1,03	4,47	3,99	4,69	5,37	1,21	1,16	1,26	1,68
16:00	10,53	9,74	11,50	11,19	10,66	10,78	13,32	13,08	5,93	5,88	6,30	6,08
16:1n7	0,51	0,85	1,15	1,19	2,21	2,09	4,20	4,10	0,00	1,08	1,00	1,54
18:00	5,09	4,93	5,00	5,72	4,45	4,27	4,71	4,17	9,05	8,55	10,57	8,20
18:1n11	0,00	0,00	0,33	0,44	1,17	1,78	1,31	1,02	0,00	0,00	0,30	0,21
18:1n7	1,18	1,36	1,63	1,90	2,55	2,49	3,71	4,03	0,98	1,09	1,35	1,35
18:1n9	1,71	1,95	2,71	3,93	2,94	2,24	3,87	3,04	3,89	4,21	4,47	4,69
18:2n6	0,56	1,37	1,27	2,12	0,95	0,89	0,96	1,62	1,74	1,96	2,08	2,35
18:3n3	0,74	0,72	0,98	1,07	0,98	0,89	0,95	1,96	1,04	1,12	1,06	1,60
18:4n1	1,31	0,96	0,00	0,00	0,55	0,58	0,47	0,41	5,26	5,46	5,67	5,53
18:4n3	0,46	0,46	0,00	0,00	0,82	0,80	1,12	1,33	0,00	0,25	0,00	0,00
20:1n11	9,04	10,08	10,69	10,18	10,02	11,14	8,90	6,74	3,94	3,46	5,35	6,24
20:1n7	4,50	3,45	5,36	4,25	4,21	4,10	4,38	4,00	0,76	0,78	0,88	0,75
20:1n9	5,77	3,99	4,42	3,67	3,55	3,04	3,18	2,12	1,89	1,77	1,90	1,39
20:2n6	1,83	3,30	2,32	3,58	1,78	1,58	1,66	1,67	2,87	2,84	2,75	2,37
20:2Δ5,11	1,22	1,51	0,39	0,32	2,19	2,64	1,86	0,53	0,18	0,00	0,00	0,07
20:2Δ5,13	0,96	1,11	0,79	0,52	1,56	1,86	1,11	0,74	0,16	0,00	0,00	0,10
20:3n3	1,24	1,01	0,62	0,66	1,15	0,99	0,71	0,66	0,23	0,23	0,14	0,19
20:4n6	11,14	9,68	5,98	5,07	7,58	7,85	6,07	3,94	14,32	12,87	12,29	11,59
20:5n3	8,91	9,89	13,53	11,47	10,36	9,54	12,61	14,76	11,39	12,02	9,59	11,10
22:1n11	0,00	1,51	0,00	0,21	1,42	1,69	0,86	0,24	0,00	0,00	0,00	0,00
22:1n9	0,56	0,77	0,00	0,00	1,07	1,33	0,89	0,00	0,00	0,00	0,00	0,00
22:2Δ7,13	5,89	5,73	3,86	3,90	4,14	4,28	2,81	1,94	4,50	4,05	4,78	3,35
22:2Δ7,15	4,34	4,84	6,35	4,49	4,91	4,89	3,50	2,92	3,38	3,36	4,26	3,67
22:4n6	1,47	1,20	1,07	1,16	0,82	0,82	0,62	0,87	1,59	1,44	1,30	1,34
22:5n3	5,69	5,06	7,41	6,45	3,72	3,30	2,85	4,33	11,19	11,30	9,18	10,35
22:6n3	0,71	1,03	2,48	4,18	2,95	2,56	2,25	5,15	1,44	1,80	1,20	1,92
DMA-16: 0	1,63	1,12	0,00	0,42	0,37	0,43	0,26	0,29	1,53	1,42	1,58	2,01

DMA-18: 0	2,66	1,79	0,00	0,55	0,71	0,80	0,76	0,32	5,26	4,43	6,01	4,72
i-17: 0	0,23	0,34	1,18	1,12	0,55	0,53	0,64	1,26	0,20	0,21	0,28	0,37
S NIK	18,11	18,46	18,79	19,38	20,76	20,09	23,82	24,04	17,25	16,61	18,93	16,86
MNJK	24,66	25,23	26,85	26,37	30,41	31,22	32,46	26,23	12,36	12,57	15,34	16,39
PJJK	48,01	49,36	47,56	46,35	45,25	44,52	40,65	44,16	60,33	59,63	54,74	56,55
DMA	4,63	3,04	0,00	0,97	1,08	1,30	1,02	0,61	7,52	6,18	8,39	7,05
$\Sigma$ n-3	18,86	19,24	25,53	24,37	20,54	18,83	21,03	28,84	25,75	27,19	21,61	25,70
$\Sigma$ n-6	15,43	15,97	10,64	12,06	11,36	11,44	9,31	8,25	21,10	19,57	18,42	18,13

Comment: in table the data of three measurements are given  
 $\Sigma$  SFAs – sum of saturated FAs  
 $\Sigma$  MUFAs – sum of monounsaturated FAs  
 $\Sigma$  PUFAs – sum of polyunsaturated FAs  
 $\Sigma$  DMA - sum of dimethyl acetals

## Discussion

It is appropriate to note here why FAs have interested us. The fact is that FAs are represented not only in practically every class of lipids, but also in many other structures of natural compounds of non-lipid nature.

By present time from natural sources it is isolated more than 1200 FAs [Internet]. However about concrete functions in an organism it is known only for limited number FAs. It should be noted that gastropod mollusks, though the enough studied class of organisms, but data about their chemical composition are limited [12,13,14,15]. In the organs of a mollusk investigated by us have been identified 63 FAs from C12 to C24, however are included in the table only what contents at least in one of samples has exceeded 1% from sum FAs. It is necessary to notice, that on a share resulted FAs it are more than 90% from the sum of all found out FAs.

Making comments on the table, it is possible to tell, that the contents Saturated FAs (SFAs) smoothly increased from May till September in gonads and a mollusk liver (with 18.1 to 19.4 % and with 20.8 to 24.0 %, accordingly) while in a foot their level fluctuated in a range 16.6 - 18.9 %. The main FA in a liver and gonads was palmitic acid (16:0). Its contribution in gonads and a liver reached 11-13 % whereas in a foot it did not exceed 6.3 %. Myristic acid (14:0) was present at all samples. Its greatest content has been noted in a liver (to 5.4 %). Its isomer iso-14:0 was absent, and anteiso-14:0 have found out only in a foot. Margaric acid (17:0) in small quantities (less than 1%) has been identified in all investigated organs. The content stearic acid (18:0) changed in a range 5.0 - 10.6 %. Most of all it was in a foot during the summer period (8.2 - 10.6 %), in gonads and a liver its share has made about 5%.

Level Monounsaturated FAs (MUFAs) in lipids of gonads and foot increased during all investigated season while in a liver during the period from August till September their contents decreased sharply from 32.5 to 26.2%. More than half of all MUFAs it was consisted from family C20, namely: 20:1n11, 20:1n9 and 20:1n7.

The contribution 20:1n11 in gonads has made 9.0 - 10.7%, in a liver 6.7 - 11.1% with gradual fall by September. In a foot the content of it an isomer varied within 3.9 - 6.2%. Level 20:1n9 in gonads reached 3.7-5.8%, in a liver of 2.1-3.6%, in a foot of 1.9 %. The contents oleic (18:1n9) and vakcenic (18:1n7) acids reached to 7% in a liver in September and July. Share C22:1 FA was rather insignificant and has exceeded 3% from the sum of all FAs only in June samples of a liver. This FA is absent in a foot.

The highest content palmitooleinic acid (16:1n7) has been noted in a liver (from 2.1 to 4.2%) while in other organs it did not exceed 1.5%.

Diene isomer of family 18:2n6 arrives 2% and more in summer and autumn periods in gonads and leg. Maximal content of isomer 18:4n1 in a leg more than 5% for all investigational period, while isomer 18:4n3 is absent in July and September in gonads, and in a leg in May, July and September. Diene isomer of family 20:2n6 has two peaks in gonads in June and September (3.3 - 3.6%). Its content small changes in a liver for all investigational period (1.8 - 1.7%) and in a leg (2.9 - 2.4%). Diene isomers of non-methylene interrupted FAs 20:2Δ5, 11 and 20:2Δ5, 13 are in negligible quantities in a leg. In other samples their contents hesitate from 0.5 to 2.6% depending on an organ and season. While isomers 22:2Δ7, 13 and 22:2Δ7, 15 contained in all organs in great quantities (1.9-6.4%).

Considerable content is marked for arachidonic acid (20:4n6) in all organs investigated. So, in a gonads reduction of its content (11.7 - 7.6%) goes from May to September, it is marked and for a liver (7.6 - 3.9%). The most content of 20:4 (14.3 - 11.6%) is in a leg with gradual reduction to the autumn.

*V. castaneus* tissues are rich Polyunsaturated FAs (PUFAs) - from 40 to 60% depending on organ and time of gathering of a mollusk. That fact is essential, that total content PUFAs during all season changed slightly. The highest level PUFAs was observed in a foot (54.7 - 60.3%), the lowest - in a liver (40.7 - 45.3%). Group C18 PUFAs has been presented linoleic (18:2n6),  $\alpha$ -linolenic (18:3n3), stearidonic (18:4n3) and 18:4n1 by acids. The contents

linoleic and  $\alpha$ -linolenic acids smoothly increased from May till September. Stearidonic acid was present at the majority of samples in trace quantities (less than 1%) or has not been found out at all. So, 18:4n1 its big part has concentrated in lipids of mollusk foot (5.3 - 5.7%) while in other organs its share was minimum. The main C20 PUFAs were arachidonic (20:4n6) and eicosapentaenoic (20:5n3) acids. Concentration of arachidonic acid steady decreased (to 2 times) from May until September in all organs. In a liver and gonads this process was accompanied by level growth eicosapentaenoic acid. Besides 20:4n6 and 20:5n3 in mollusk tissues in significant quantities presented 20:2n6 (1.6 - 3.6%), 20:3n3 (0.1 - 1.2%), and also characteristic for mollusks [16] non-methylene-interrupted FAs 20:2 $\Delta$ 5, 11 and 20:2 $\Delta$ 5, 13 (to 4.5% at the sum).

*V. castaneus* organs strongly differed on C22 PUFAs composition. So, the greatest content docosapentaenoic acid has been noted in a foot (9.2 - 11.3%), minimum - in a liver (2.9 - 4.3%). Docosahexaenoic (22:6n3) acid was present at smaller quantities, thus its level increased by the autumn. Most considerably it proceeded in lipid of gonads. The powerful contributions have brought 22:2 $\Delta$ 7, 13 and 22:2 $\Delta$ 7, 15 (6.3 - 10.6%), synthesized by a mollusk by means of elongation chains 20:2 $\Delta$ 5, 11 and 20:2 $\Delta$ 5, 13 FAs [14].

Also during the analysis of composition of FA methyl esters have been identified Dimethyl Acetals (DMA) 16:0 and 18:0, that testifies to presence glycerophospholipidalkenyl forms in mollusk organs. Their maximum quantity was found out in a foot - from 6.2 to 8.4 % from sum FAs. Earlier Isay et al have shown that some gastropod mollusks of Japan Sea contain Glyceryl Ethers (GE). So, the content  $\alpha$  - GE in some of them made 1% (in lipid extract), and in gastropod mollusks of tropical areas of Pacific Ocean  $\alpha$  - GE reached the contents of 1.5 - 16.2% [15].

## Conclusion

Summing up to research, it is possible to tell, that the foot considerably differs from other organs investigated. By the way, the foot is an edible part of a mollusk. In it is found out not only all spectrum PUFAs, but also their highest level (55-60%). Thus on  $\omega$ -3 and  $\omega$ -6 PUFAs it is necessary to 30%. Besides, isomer anteizo-14:0 FA it is identified only in a foot. Also in it the big contribution 18:4n1 FA whereas in other organs its contribution is minimal is noted. The foot differs the high contribution 22:5 FA, at the same time in it were absent 22:1n11 and 22:1n9 FAs whereas in gonads and a liver these acids are though and in small amounts.

By present time from natural sources it is isolated more than 1200 FAs [16]. However, about concrete functions in an organism it is known only for limited number FAs.

In the literature there is information on the importance concrete FAs in medical practice. So, influence oleic acid on different systems of an organism is shown [17,18]; linoleic FA possesses antioxidant properties [18,19,20]; stearidonic acid

shows cardio protection activity [20]; arachidonic acid, along with linoleic, are main FAs of phospholipids membranes of the person [13]. Linoleat also inhibits growth of a cancer of the person in vitro, reduces size of a tumour and metastasis [14,15].

## Declarations

- conflict of interests - NA
- ethical approval - NA

## References

1. Tatishvili KG, Bagdasaryan KG, Kazahashvili JR. Reference book on ecology of marine gastropod mollusks (eds. Davitashvili L. Sh., Merklin R. L.) "Science", Moskow. 1968.
2. Besednova NN. Marine hydrobionts - potential sources of medicaments. Health. Medical Ecology. Science. 2014;57(3):4-10.
3. Kozlova EY. Gastropod mollusks of Far Eastern region as source of biology active substances. International conference. Vladivostok. 2015.
4. Kulikova NN, Maksimova NV, Sutorin AN, et al. Biogeochemical characteristic of dominating species of gastropod mollusks of stone littoral of the south Baikal. Geochemistry. 2007;5:35-46.
5. Tolkacheva VF. Prospects for industrial use of the Barents sea mollusks. Fish Economy. 2016;2:103-107.
6. Miroshnichenko DA, Flerova YA. Particularities of a chemical composition of the muscle foot of gastropod mollusks Muricidae (Asteraceae) of village of South Viet-Nam. Agro-Industry Complex News. "Yaroslavskaya agricultural Academy". 2016;3:65-70.
7. Bligh EG, Dyer WJ. A rapid method of total lipid extraction and purification. Canadian Journal of Biochemistry and Physiology. 1959;37(8):911-917.
8. Carreau JP, Dubacq JP. Adaptation of a macroscale method to the microscale for fatty acid methyl transesterification of biological lipid extracts. Journal of Chromatography A. 1978;151(3):384-390.
9. Kates M. Techniques of lipidology; Isolation, analysis and identification of lipids. New York: Elsevier. 1972:341.
10. Stransky K, Jursik T, Vitek A. Standard equivalent chain length values of monoenic and polyenic (methylene interrupted) fatty acids. Journal of High Resolution Chromatography. 1997;20(3):143-158.
11. Svetashev VI. Mild method for preparation of 4,4-dimethylloxoline derivatives of polyunsaturated fatty acids for GC-MS. Lipids. 2011;46(5):463-467. doi: 10.1007/s11745-011-3550-4
12. Johns RB, Nichols PD, Perry GJ. Fatty acid components of nine species of molluscs of the littoral zone from Australian waters. Comparative Biochemistry and Physiology Part B: Comparative Biochemistry. 1980;65(2):207-214.
13. Joseph JD. Lipid composition of marine and estuarine invertebrates. Part II: Mollusca. Progress in Lipid Research. 1982;21(2):109-153.
14. Gunstone FD. Fatty acids and lipid chemistry. Springer. 1996:252.
15. Isay SV, Makarchenko MA, Vaskovsky VE. A study of glyceryl ethers - I. Content of  $\alpha$ -glyceryl ethers in marine invertebrates from the sea of Japan and tropical regions of the Pacific Ocean. Comp Biochem Physiol B. 1976;55(2):301-305.
16. Barnathan G. Non-methylene-interrupted fatty acids from marine invertebrates: occurrence, characterization and biological properties. Biochimie. 2009;91(6):671-678. doi: 10.1016/j.biochi.2009.03.020

17. JJM van den Berg, Cook NE, Tribble DL. Dietary reference intakes: The essential guide to nutrient requirement. *Lipids*. 1995;30(6):599-605.
18. Sales-Campos H, Souza PR, Peghini BC, da Silva JS, Cardoso CR. An overview of the modulatory effects of oleic acid in health and disease. *Mini Rev Med Chem*. 2013;13(2):201-210.
19. Chen ZY, Chan PT, Kwan KY, Zhang A. Reassessment of the antioxidant activity of conjugated linoleic acids. *JAOCS*. 1997;74(6):749-753.
20. Hagen RM, Rhodes A, Ladomery MR. Conjugated linoleate reduces prostate cancer viability whereas the effects of oleate and stearate are cell line dependent. *Anticancer Res*. 2013;33(10):4395-4400.