

## **INFLUENCE OF FATTY ACID COMPOSITION OF FISH FEED ON FATTY ACID COMPOSITION OF CARP MEAT**

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### **UTICAJ MASNOKISELINSKOG SASTAVA HRANE ZA RIBE NA SASTAV MASNIH KISELINA MESA ŠARANA**

#### *Apstrakt*

Riblje meso već dugi niz godina privlači pažnju nutricionista i preporučuje se u ishrani ljudi pre svega zbog visokog sadržaja nezasićenih masnih kiselina, od kojih su naročito značajne polinezasićene omega-3 masne kiseline. Ova jedinjenja smatraju se esencijalnim, jer ih sisari, a samim tim i ljudi, ne mogu sintetisati u organizmu, već ih moraju uneti putem hrane. Dobro je poznato da ove masne kiseline blagotvorno deluju na zdravlje ljudi, smanjuju rizik od pojave oboljenja srčanog mišića i krvnih sudova.

U našoj zemlji, u ishrani se u značajnoj meri koristi šaransko meso. Od ukupnog uzgoja rečnih riba u svetu, šaran zauzima prvo mesto sa 70 % celokupne proizvodnje. Uglavnom se uzgaja u ribnjacima, gde se prirodna ishrana planktonom dopunjava pretežno cerealijama. Kako cerealije sadrže vrlo malo esencijalnih masnih kiselina, i masnokiselinski sastav šaranskog mesa se menja, tako da u mesu tovljenih šarana prevladuje oleinska kiselina, dok su omega-3 masne kiseline manje zastupljene. Najjednostavniji način da se poboljša sastav masnih kiselina ribljeg mesa jeste da se u hranu za ribe dodaju sirovine sa poželjnim masnokiselinskim sastavom, kao što su laneno, ili riblje ulje.

Cilj ovog rada bio je upravo da se utvrdi kako dodatak lanenog ili ribljeg ulja u eksperimentalnu hranu utiče na promenu masnokiselinskog sastava hrane za ribe, a potom posredno i na njihov sastav u mesu šarana. U eksperimentu je upotrebljavana ekstrudirana hrana za šarane sa naknadno dodatim uljem u vakuum zamašćivaču. Ribe su gajene u plastičnim tankovima sa protokom vode, kako bi se obezbedilo snabdevanje svežim kiseonikom.

Rezultati ovog istraživanja su pokazali da je dodatak obe vrste ulja u značajnoj meri unapredio masnokiselinski sastav hrane za ribe. Dodatak lanenog ulja povećao je relativni udeo  $\alpha$ -linolenske kiseline u odnosu na kontrolnu hranu (7,08 % u kontrolnoj hrani u odnosu na 27,57 % u eksperimentalnoj hrani 1), dok je dodatak ribljev ulja uticao na povećanje sadržaja dokosaheksaenoične (DHA) i eikosapentaenoične (EPA) masne kiseline, koji su u hrani 2 iznosili 4,14 % i 7,32 % (u kontroli su relativni udeli bili 0,36 % i 0,80 %). Kada se govori o ribljem mesu, dodatak obe vrste ulja pozitivno je uticao na masnokiselinski sastav mesa. Tako je odnos n-6/n-3 masnih kiselina u grupi hranjenoj obrokom 1 iznosio 4,64, a u grupi hranjene obrokom 2 bio je 1,87. U oba slučaja odnos je snižen u poređenju sa kontrolnom grupom gde je n-6/n-3 odnos bio 8,97. Prilikom poređenja dve eksperimentalne grupe međusobno, kod mesa šarana hranjenih ekstrudiranom hranom sa dodatkom ribljev ulja uočava se nešto povoljniji sastav masnih kiselina u odnosu na meso šarana hranjenih obrokom sa dodatkom lanenog ulja, pre svega jer im je značajnije povećan udeo EPA i DHA masnih kiselina. Tako je relativni udeo EPA u ovim uzorcima iznosio 1,0 %, dok je u kontrolnoj grupi bio 0,21 %. U eksperimentalnom uzorku sadržaj DHA je bio 2,47%, a u kontrolnom uzorku 0,67 %. Dakle, sardžaj EPA se povećao skoro 5 puta, a sadržaj DHA oko 3,5 puta u poređenju sa kontrolnim uzorkom.

Može se zaključiti da oba dodata ulja imaju pozitivno dejstvo na masnokiselinski sastav mesa šarana i da ne pokazuju negativan uticaj na rezultate odgoja.

*Ključne reči: masnokiselinski sastav, riblja hrana, meso šaran, esencijalne masne kiseline*

*Keywords: fatty acid composition, fish feed, carp meat, essential fatty acids*

## INTRODUCTION

For many years, much attention is given to fish meat and its presence in human nutrition. The reason for that lies in significant amount of polyunsaturated fatty acids (PUFA), particularly omega-3 fatty acids (n-3 FAs) which are present in all types of fishes. Long chain n-3 PUFAs are essential because mammals, and therefore humans, cannot synthesize them and must adopt them exogenously from dietary sources (Beare-Rogers et al, 2001; Ivanov et al, 2012). They are derived from  $\alpha$ -linolenic acid (ALA), which is the precursor of docosahexaenoic (DHA) and eicosapentaenoic (EPA) acids (Gorjão et al, 2009). These two FAs are known to decrease risk of cardiovascular diseases, especially coronary heart disease (WHO/FAO, 2003).

Carp represents one of the largest groups of cultured fish with around 70% of freshwater aquaculture production. The majority of European carp production is placed in central Europe where it is produced in ponds using traditional semi-intensive techniques.

Fish reflect the lipid pattern of its diet to a high extent. Carp is traditionally reared in earthen ponds and its nutrition is based on natural food with cereal supplementation (Buchtova et al, 2007). Since cereals are rich in carbohydrates and have very low level of n-3 fatty acids, the flesh of the farmed carps generally contains a high level of oleic acid and low level of favourable n-3 PUFA (Chengeri, 1996). An alternative approach to influence muscle lipid composition might be inclusion of rich sources of omega FA in fish feed.

Raw materials which are known as a good source of essential FA are linseed oil and fish oil. Linseed oil is rich in linoleic (LA, 18:2, n-6) and especially  $\alpha$ -linolenic acid

(ALA, 18:3, n-3) (Łukaszewicz et al., 2004), while fish oil contains significant amount of DHA and EPA (Jonzo et al, 2000). Fish oil is the main lipid source in feeds of carnivorous farmed fish and aquafeeds use 87% of the global supply of fish oil, of which over 66% is used for salmonids (Tacon et al, 2008).

The aim of this study was to examine how addition of linseed oil and fish oil influences FA composition of fish feed, and to register if there are any changes in fatty acid composition of meat of carps fed with experimental diets.

## MATERIALS AND METHODS

In the experiment, carp feeds were formulated according to commercial and physiological consideration. Following ingredients were used for production of basic diet: corn (30 %), wheat (10 %), corn gluten (10 %), fish meal with 70% of protein (10 %), soybean meal (36 %), yeast (3%), and premix (1%). Experimental diets were prepared by addition of 6% of linseed oil (Diet 1) and 6% of fish oil (Diet 2).

Fish feed was produced on twin-screw extruder (Mu Yang MY 90, China) with a screw diameter of 85 mm, length-to-diameter ratio of 20:1, and maximum temperature of 135 °C. Extruder was equipped with differential diameter conditioner (DDC). Addition of oil was done on laboratory vacuum coater (model F-6-RVC, Forberg International AS, Norway) with capacity of 6 L per batch.

Experimental carps were bred in 2.0 m<sup>3</sup> plastic square-shaped tanks divided into two compartments (about 1.0 m<sup>3</sup> each) by a screen wall positioned along one diagonal of each tank, each with two compartments and with 12 fishes per compartment. Water inflow into the tanks was regulated to ensure oxygen concentration of the outflow water at least 80 % of saturation. During the experimental period natural day-light regime was ensured. Feeding was performed by automatic belt feeders during 10 hours per day.

Fat for fatty acid analyses was extracted from the samples of feed and fish meat using Folch method (Folch, 1957). The preparation of fatty acid methyl esters was done by transesterification that use 14% wt. boron trifluoride/methanol solution (Sigma Aldrich, MO, USA), as recommended method for this type of substrates. Fatty acid composition analyses were done on a gas chromatograph Agilent 7890A system (Agilent Technologies, Santa Clara, CA, USA) with flame ionization detector (GC-FID), equipped with fused silica capillary column (SP-2560, 100 m x 0.25 mm, I.D., 0.20 µm). Split ratio was 30:1. Carrier gas was helium (purity > 99.9997 vol %, flow rate = 1.5 ml/min). All determinations were done in triplicates.

## RESULTS AND DISCUSSION

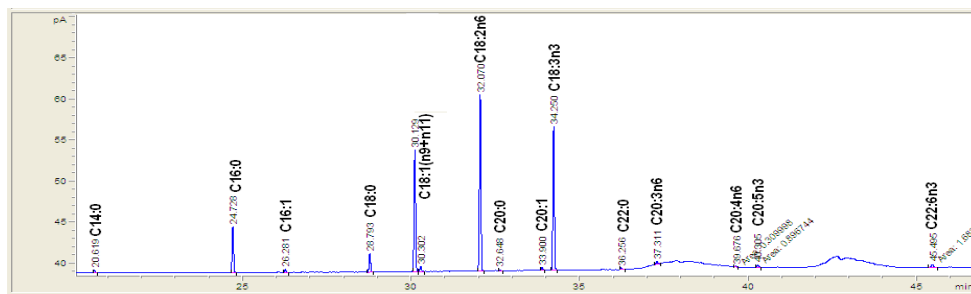
Results of FA composition of fish feed and carp meat from the control and experimental groups are presented in Table 1. As it can be seen, both experimental diets had significantly ( $p \leq 0.05$ ) higher relative content of n-3 FA, while content of n-6 FA decreased. These changes resulted in more favourable n-6/n-3 ratio in comparison with control diet (n-6/n-3 ratio of 5.85), which was 1.14 in Diet 1 and 2.25 in Diet 2. According to WHO and FAO, this ratio should be less than 4 (FAO/WHO, 2003). It can be concluded that only control diet did not fulfil this recommendation.

PUFA/SFA ratio was the highest in Diet 1 (5.02), mostly due to the very high content of ALA, which was 27.57 % in this sample. The lowest level of PUFA/SFA ratio recommended by WHO and FAO is 0.4 (FAO/WHO, 2003). Diet 2 showed significant

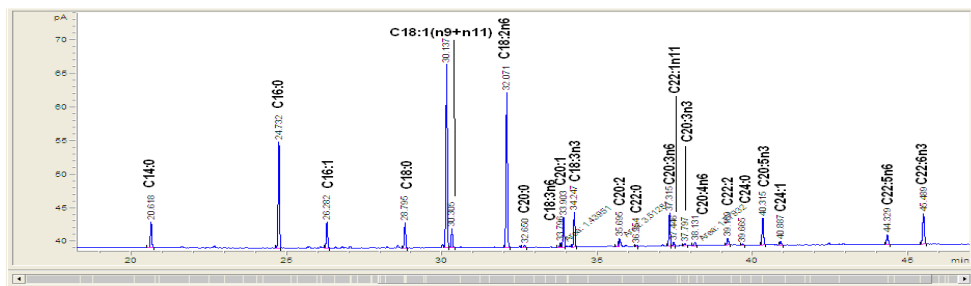
increase in EPA (%) and DHA (%) in comparison with control diet (4.14 % and 7.32 %, respectively). Figure 1 presents GC-chromatograms of Diet 1 (a) and Diet 2 (b). Mentioned differences are even more obvious in fatty acid composition of carp meat. Both groups fed with experimental diets had better n-6/n-3 ratio than control group, and carps fed with Diet 2 showed the best results with n-6/n-3 ratio of 1.87.

In this sample, EPA content increased almost five times in comparison with control group (1.01 % vs. 0.21 %), while DHA content increased more than 3.5 times (2.47 % vs. 0.67 %). EPA content in meat of carp fed with Diet 1 increased to 0.52 % and DHA content increased to 1.2 %. It can be concluded that addition of fish oil in fish feed gave more favourable fatty acid composition of carp meat than addition of linseed oil.

Looking at PUFA/SFA ratio of carp meat samples, it can be seen that all samples had ratio higher than 0.4. Although addition of linseed oil and fish oil decreased, this level in comparison with control carp meat samples, it is still above recommended, so it cannot be considered as undesirable effect.



(a)



(b)

GC chromatograms of Diet 1 (a) and Diet 2 (b)

**Table 1:** Fatty acid compositions of control diet, Diet 1, Diet 2 and meat of carp fed with the different diets

Fatty acid	% of fatty acid in total fatty acid content					
	C diet	D 1	D 2	C Carp	D1 carp	D2 carp
C12:0	ND	ND	ND	0.02	0.02	0.03
C14:0	0.43	0.40	2.96	0.77	0.03	1.40
C14:1	ND	ND	ND	0.44	0.05	0.06
C16:0	11.46	8.06	12.72	16.04	16.38	16.85

C16:1	0.59	0.62	3.32	0.35	0.1	6.47
C17:0	ND	ND	ND	0.21	0.21	5.94
C17:1	ND	ND	ND	4.77	0.15	0.05
C18:0	4.70	3.43	3.08	5.56	5.53	4.71
C18:1 <i>n-9c + n-11c</i>	24.85	23.78	26.42	42.79	45.11	40.21
C18:2 <i>n-6c</i>	47.42	32.83	19.85	21.32	16.48	11.43
C20:0	0.43	0.27	0.33	0.14	0.12	0.05
C18:3 <i>n-6</i>	ND	ND	0.56	0.49	0.27	0.18
C20:1	0.71	0.63	4.19	2.05	0.46	0.8
C18:3 <i>n-3</i> (ALA)	7.08	27.57	4.52	1.87	1.98	2.87
C21:0	ND	ND	ND	0.02	8.26	1.38
C20:2	ND	ND	1.57	0.12	0.26	ND
C22:0	0.41	0.30	0.16	ND	0.34	0.23
C20:3 <i>n-6</i>	0.66	0.56	4.41	0.57	0.47	0.35
C22:1 <i>n-9</i>	ND	ND	0.54	0.34	0.37	1.04
C20:3 <i>n-3</i>	ND	ND	0.24	0.06	0.07	0.11
C20:4 <i>n-6</i>	0.11	0.16	0.47	0.04	0.26	0.11
C22:2	ND	ND	0.96	0.11	0.22	1.12
C24:0	ND	ND	0.12	0.88	0.87	0.71
C20:5 <i>n-3</i> (EPA)	0.36	0.43	4.14	0.21	0.52	1.01
C24:1	ND	ND	0.49	ND	ND	0.3
C22:5 <i>n-6</i>	ND	ND	1.62	ND	ND	ND
C22:6 <i>n-3</i> (DHA)	0.80	0.97	7.32	0.67	1.2	2.47
<i>n-3</i>	8.24	28.97	16.22	2.81	3.77	6.46
<i>n-6</i>	48.19	32.99	36.52	25.23	17.48	12.07
<i>n-6/n-3</i>	5.85	1.14	2.25	8.97	4.64	1.87
SFA	17.43	12.46	19.37	7.98	46.3	54.82
MUFA	26.15	25.02	34.96	46.18	20.27	19.65
PUFA	56.42	62.52	45.67	24.67	33.43	25.53
PUFA/SFA	3.24	5.02	2.36	3.09	0.61	0.77

*ND* – not detected. results in the table are presented as mean, *n* =3; *SFA* - saturated fatty acids, *MUFA* - monounsaturated fatty acids, *PUFA* - polyunsaturated fatty acids; *C diet* – control diet, *D1* – diet 1, *D2* – diet 2

## CONCLUSIONS

Based on experimental result, it can be concluded that fish feed significantly influenced fatty acid composition of carp meat. By addition of linseed oil and fish oil in the

diet, n-3 fatty acid content of carp meat increased for 0.96 % and 3.65 %, respectively. Ratio n-6/n-3 was the most favourable in meat of carp fed with Diet 2, and it was 1.87. It is obvious that both linseed oil and fish oil were proven as a good source of omega fatty acids in carp feed, but addition of fish oil gave slightly better results.

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