# COMPARATIVE ANALYSIS OF THE FATTY ACID COMPOSITION OF NATURAL FOOD AND SUPPLEMENTAL FEED IN CARP FROM A FISH FARM

IVANA ŽIVIĆ<sup>1</sup>, DEJANA TRBOVIĆ<sup>2</sup>, ZORKA DULIĆ<sup>3</sup>, MARKO STANKOVIĆ<sup>3</sup>, MIROSLAV ŽIVIĆ<sup>1</sup>, ZORAN MARKOVIĆ<sup>3</sup>

<sup>1</sup>University of Belgrade, Faculty of Biology, 11000 Belgrade Serbia <sup>2</sup>Institute of Meat Hygiene and Technology, Kaćanskog 13, 11000 Belgrade, Serbia <sup>3</sup>University of Belgrade, Faculty of Agriculture, 11080 Belgrade Serbia Corresponding author:e-mail: ivanas@bio.bg.ac.rs

# KOMPARATIVNA ANALIZA SASTAVA MASNIH KISELINA U PRIRODNOJ I DODATNOJ HRANI ŠARANA SA RIBNJAKA

**Apstrakt** 

Rezultati prikazani u radu ukazuju na daleko veći značaj prirodne hrane kao izvora polinezasićenih masnih kiselina u odnosu na peletiranu hranu u ishrani šarana u poluintenzivnom sistemu gajenja. Utvrđeno je da su od organizama koji su činili prirodni obrok šaranu: predstavnici familija Chironimidae sa dominaciojm vrste *Chironomus plumosus* iz faune dna i zooplanktonski organizmi (sa dominacijom Cladocera) imali znatno veći sadržaj omega 3 i omega 6 masnih kiselina nego peletirana hrana. Istraživanja su obavljena u 4 ribnjačka jezera šaranskog ribnjaka "Despotovo" sa poluintezivnim sistemom gajenja baziranom na prihrani peletiranom hranom.

Peletiranu hranu odlikuje najmanja raznovrsnost masnih kiselina i izrazita dominacija linolne i oleinske kiseline. Larve Chironomidae i zooplankton karakteriše veća zastupljenost zasićenih masniih kiselina(SFA:  $49.36 \pm 1.92$  i  $38.70 \pm 0.67$ ) u odnosu na peletiranu hranu ( $15.47\pm0.41$ ), a zooplankton odlikuje i najveća raznovrsnost i zastupljenost n-3 masnih kiselina. Odnos n-3/n-6 masnih kiselina deset puta je veći kod zooplanktona nego kod Chironomidae, a sedam puta veći kod Chironomidae nego kod peletirane hrane.

Ključne reči: šaran, prirodna hrana, peletirana hrana, masne kiseline Keywords: carp, natural food, pelleted feed, fatty acids

#### INTRODUCTION

The analysis of fatty acid (FA) composition of carp meat showed that different rearing methods and diets provide considerable differences in the contribution of n-3 and n-6 polyunsaturated fatty acids (PUFA). Carp fed with row cereals contains less n-3 PUFA (Steffens and Wirth, 2007) compared to carp fed with extruded and pelleted feed. The n-3/n-6 ratio of PUFA in carp fed commercial pellets is very similar to those in the feed. Numerous studies demonstrated that the fatty acid composition of food has significant effects on the fatty acid composition of fish meat (Steffens and Wirth, 2007). Food richer in n-3 PUFA in the same fish production conditions, substantionaly improves the n-3/n-6 ration in fish tissues.

The natural food of carp in semi-intensive production systems mainly consists of zoo-plankton and bottom fauna (Marković, 2010). The significance of these organisms for carp culture is in their nutritive value. Except proteins, these organisms are a source of lipids, fatty acids, vitamins and enzymes for all age classes of carp (Dulić et al., 2011, Živić et al., 2012). Deficiency of natural food leads to the deficiency of vitamins, minerals, amino acids and fatty acids in carp diet. This can affect the immunity and fitness of fish, increase diseases and fish mortality (Marković, 2010). Therefore the enhancement of natural food (zooplankton and Chironomidae larvae) development in carp fish ponds is one of the feasible ways to improve the meat quality of produced carp (Steffens and Wirth, 2007; Živić et al., 2012). The aim of this investigation was to analyze the composition of PUFA in natural food and pelleted feed for fish.

#### MATERIALS AND METHODS

### Site location

The fish farm "Despotovo" is located in the village of Despotovo, in Vojvodina Province. The carp farm consists of six earthen fishponds that cover a surface area of about 200 ha. For the study, four fish ponds (J1, J3, J4 and J6) were used with surface area of: 100ha, 25ha, 25ha and 16ha, respectively. Fish were fed daily with commercial pelleted feed "BAFI" (Futog, Serbia) with 25% proteins (plant origin) and 7% fat.

Sample collection and preparation

Samples of Chironomidae larvae (*Chironomus plumosus*) and zooplankton were taken monthly from June to October 2012 from three points in every pond (inflow, middle, and outflow). Chironomidae larvae were sampled with Van Veen grab, grasping area of 260 cm². In order to analyze fatty acid composition of *Chironomus plumosus* larvae, 279 individuals were collected from the pond J1, J3, J4 and J6 (fourth and fifth larval instar). Zooplankton samples were collected with 250 µm mesh plankton net. Samples were kept at the temperature of -18 °C until analysis. Fatty acid content and total lipid analysis of of Chironomidae, zooplankton and pelleted feed were completed at the Institute of Meat Hygiene and Technology in Belgrade (Serbia) using standard methods.

Statistical Methods

Correspondence analysis (CA) was used to analyze relationship between fatty acids in Chironomidae, zooplankton and pelleted feed. CA was performed using the Brodgar program (Highland Statistics Ltd, UK). To obtain differences between mean values of the two samples, t-test with statistical importance of P<0.05 was used.

## RESULTS AND DISCUSSION

We analyzed 20 fatty acids from three main sources: carp feed, zooplankton, Chironomidae and pelleted feed. Data were collected monthly during four months from four neighboring fish ponds. ANOVA showed that there was no significant difference between lakes in analyzed zooplankton, Chironomidae and pelleted feed fatty acids composition, thus the results were pooled, and average values used in further analysis. Average values of all investigated fatty acids are presented in Table 1.

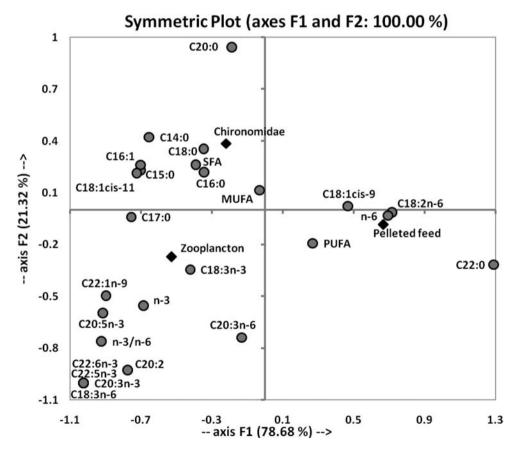
Correspondence analysis of studied fatty acids from all samples is presented on Fig 1. The two axes describe 100% of variance. F1 axis explains the difference between the fatty acid concentrations in supplemental and natural food. F2 axis explains the difference between two types of natural food. Since F1 axis explains most of the variability (78.68%) it is clear that there is a bigger difference between supplemental and natural food than between zooplankton and bottom fauna. The main cause for these differences is the domination of linoleic and to a lesser extent oleic acid. Pelleted feed contain up to 80.48% of these acids, while Chironomidae and zooplankton contained only 27.78% and 14.53% respectively (Tab. 1). In pelleted feed only palmitic (10.96%), stearic (3.73%) and linolenic (3.74%) acids were represented in larger amounts, while the rest of the fatty acids were either absent (six of them) or accounted for less than 0.32% of total amount of fatty acids (Tab. 1). Particularly evident is the lack of long chain PUFA n-3 fatty acids in pellet feed.

Contrary to this, the fatty acid content of natural food, especially zooplankton, was considerably more balanced compared to pelleted feed. This is especially evident in the case of monounsaturated fatty acids (MUFA) with four of them (C16:1, C18:1cis-9, C18:1cis-11 and C22:1n-9, Tab. 1) significantly represented in zooplankton and Chironomidae.

**Table 1.** Average values (±1SE) of fatty acids representations in zooplankton, Chironomidae and pelleted feed.

	7 1 1	CI: :I	D II + 1 C I		7 1 1.	Cl. :1	_
	Zooplankton	Chironomidae	Pelleted feed		Zooplankton	Chironomidae	)
C14:0	$3.07\pm0.23$	$4.39 \pm 0.05$	$0.070\pm0.004$	C20:3n-6	$0.21 \pm 0.02$	n.d	
C15:0	$1.61\pm0.11$	$1.66\pm0.06$	$0.030\pm0.004$	C20:3n-3	$0.46\pm0.08$	n.d	
C16:0	$23.03\pm0.53$	$27.91 \pm 1.68$	10.96±0.41	C22:1n-9	$3.61 \pm 0.25$	$0.94 \pm 0.26$	
C16:1	$8.94 \pm 0.92$	$9.71 \pm 1.08$	$0.100\pm0.009$	C20:5n-3	$7.86 \pm 0.42$	$1.56\pm0.18$	
C17:0	$3.31\pm0.18$	$2.18\pm0.30$	$0.080\pm0.004$	C22:0	n.d	n.d	
C18:0	$7.67 \pm 0.37$	$11.95\pm0.24$	$3.73\pm0.01$	C22:5n-3	$0.66 \pm 0.09$	n.d	
C18:1cis-9	$7.21 \pm 0.48$	$11.41\pm0.29$	$25.52\pm0.46$	C22:6n-3	$6.70\pm1.08$	n.d	
C18:1cis-11	$6.79 \pm 0.45$	$6.75\pm0.71$	0.00	SFA	$38.70\pm0.67$	$49.36 \pm 1.92$	
C18:2n-6	$7.33 \pm 0.40$	$16.36\pm0.92$	54.96±0.47	MUFA	$22.93 \pm 1.20$	$27.87 \pm 1.50$	
C20:0	$0.14 \pm 0.02$	$1.26\pm0.17$	$0.28 \pm 0.01$	PUFA	$34.76 \pm 1.30$	$21.82 \pm 0.16$	
C18:3n-6	$0.51 \pm 0.04$	n.d	0.00	n-3	$26.30 \pm 1.22$	$5.46\pm0.92$	
C18:3n-3	$10.68\pm0.67$	$3.90\pm0.94$	3.74±0.05	n-6	$8.46 \pm 0.40$	$16.36\pm0.76$	
C20:2	$0.42\pm0.05$	n.d	$0.050\pm0.009$	n-3/n-6	$3.19 \pm 0.20$	$0.34 \pm 0.06$	

Fatty acid composition of zooplankton and Chironomids are clearly separated along the F2 axis. This is mainly due to the higher level of SFA in Chironomids that are, with the exception of the C22:0, grouped near Chironomids (Fig. 1). High level of SFA in Chirnomids (*Chironomus plumosus*) from fish pond has been observed in earlier studies (Bogut *et al.*, 2007, Živić, et al., 2013) therefore it can be considered as their important characteristic. Major difference found between Chironomids and zooplankton is due to a much higher diversity and content of n-3 FA in zooplankton (Tab. 1, Fig. 1). Moreover, three most important FA: α linolenic (ALA), eicosapentaenoic (EPA) and docosahexaenoic (DHA) are equally distributed in samples of zooplankton. Numerous studies observed the high content of EPA and DHA in freshwater mesozooplankton (>250 μm), with the prevailing content of eicosapentaenoic acid in Cladocerans and docosahexaenoic in Copepods (Kainz et al., 2004). In our study the Cladocerans were dominant in zooplankton. Additionally, ALA is a precursor of EPA (Von Elert, 2002), thus was also prominent in zooplankton samples. The FA composition found in samples show that, compared to pelleted feed, zooplankton has a 10 fold higher n-3/n-6 ratio, while Chirnomids have a 7 fold higher n-3/n-6 ratio (Tab.1).



**Figure 1.** The CA biplot showing relationships between zooplankton, Chironomidae and pelleted feed based on their fatty acids concentrations.

Feeding carp mainly with supplemental feed would probably lead to the domination of oleic acid, already dominant in carp meat regardless of diet type, and linoleic acid and unfavorable n-3/n-6 ratio. Conversely, enhancement of natural food in fish ponds would provide a more diverse and balanced fatty acid composition with a highly favorable n-3/n-6 ratio, and especially increased level of long chain n-3 fatty acids (Bogut et al., 2007). It should be taken into account that supplemental feed has a dominant effect on the FA composition in carp compared to natural food (Živić et al., 2013), since in the presence of supplemental feed carp switches to easily accessible artificial food, while depending on the natural food availability, carp will preferably feed on bottom fauna or zooplankton (Rahman, 2010).

#### CONCLUSIONS

Comparative analysis of FA content of natural food and pelleted feed from the fish farm "Despotovo" showed that the dominant natural food for carp were Chironomids and Cladocerans with a significantly higher level of n-3 and n-6 fatty acids than in pelleted feed. Zooplankton is characterized by a higher diversity and content of n-3 FA compared to bottom fauna, with the domination of the most valuable essential fatty acids, EPA, DHA and ALA.

#### ACKNOWLEDGEMENT

The present study was supported by the Serbian Ministry of Education, Science and Technological Development (project No. TR 31075)

#### REFERENCE

Bogut, I., Has-Schön, E., Adámek, Z., Rajković. V., Galović, D. (2007). *Chironomus plumosus* larvae - a suitable nutrient for freshwater farmed fish. Agriculture, Vol. 13, No. 1, 1-5.

Dulić, Z., Stanković, M., Rašković, B., Spasić, M., Ćirić, M., Grubišić, M., Marković, Z. (2011): Role and significance of zooplankton in semi-intensive carp production. Fifth International Conference "Aquaculture & Fishery" Conference Proceedings. Faculty of Agriculture, Belgrade-Zemun, Serbia, p. 66 – 71.

Elerte v. E, . 2002. Determination of limiting polyunsaturated fatty acids in Daphnia galeata using a new method to enrich food algae with single fatty acids. Limnology and Oceanography, 47: 1764-1773.

Kainz, M., Arts, M.T. and Mazumder, A. (2004): Essential Fatty Acids in the Planktonic Food Web and Their Ecological Role for Higher Trophic Levels. Limnology and Oceanography, 49: 1784-1793

Marković, Z. (2010). Šaran, Gajenje u ribnjacima i kaveznim sistemima. Prof. dr Zoran Marković, Beograd, 1-152.

Rahman M., Kadowaki S., Balcombe S. and Wahab M. (2010): Common carp (Cyprinus carpio L.) alters its feeding niche in response to changing food resources: direct observations in simulated ponds. Ecol. Res., 25, 303–309.

Steffens, W., Wirth, M. (2007). Influence of nutrition on the lipid quality of pond fish: common carp (*Cyprinus carpio*) and tench (*Tinca tinca*). Aquacult. Int. 15: 313-319

Živić, I., Bjelanović, K., Dulić, Z., Stanković, M., Rašković, B., Poleksić, V., Marković, Z. (2012). Significance of carp (*Cyprinus carpio*) feeding with Chironomidae larvae for meat quality improvement in the semiintensive production system. 6th Central European Congress on Food, CEFood2012. Novi Sad. Conference proceedings, 1570-1575.

Živić, I., Trbović, D., Živić, M., Bjelanović, K., Marković, Z.S., Stanković, M., Marković, Z. (2013). The influence of supplement feed preparation on the fatty acids composition of carp and Chironomidae larvae in a semi-intensive production system. Arch. Biol. Sci., Belgrade, 65 (4): 1387-1396.

Živić, I., Živić, M., Bjelanović, K., Spasić, M., Rašković, B., Stanković, M., Marković, Z. (2014). Fatty acid profile in muscles of carp (*Cyprinus carpio* L.) raised in a semi-intensive production system fed with grains, pelleted and extruded feed. Arch. Biol. Sci., Belgrade, 66 (2): 877-887.