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## NUTRITIONAL VALUE OF PLANKTONIC CLADOCERAN *DAPHNIA MAGNA* FOR COMMON CARP (*Cyprinus carpio*) FRY FEEDING

I.Bogut<sup>1\*</sup>, Z. Adámek<sup>2</sup>, Z. Puškadija<sup>1</sup>, D. Galović<sup>1</sup>, D. Bodakoš<sup>3</sup>

#### Summary

Chemical composition and contents of amino acids and fatty acids in the planktonic cladoceran Daphnia magna were investigated, aiming to evaluate its value for feeding of young carp. Crude protein, crude fat, crude fibre, ash and moisture contents were analyzed according to standard laboratory procedures. Amino acids content was determined by LKB 4101 automatic analyzer and that of fatty acids by Chrompack CP 9000 chromatograph, using a flame ionizing detector. Protein contents amounted to 1.18 and 39.24% of fresh and dry mass, respectively. These amounts of proteins completely meet nutritional requirements both of carp fry and its older categories and other omnivorous fishes. Raw fat and fibre contents in dry weight were 4.98 and 4.32%, respectively, which is suitable for the commercial carp breeding. Methionine and phenylalanine are partially in deficit, whereas other essential amino acids identified in dry mass of Daphnia magna were present in amounts adequate for all carp categories. The proportions of saturated and unsaturated fatty acids in lipids of Daphnia magna were 18.70 and 66.20%, respectively. Among the unsaturated fatty acids, the omega-3 group was present with 27.30%. The omega-3: omega-6 fatty acids ratio was 5.68:1, which fully meets the carp nutrition requirements.

**Key words:** Daphnia magna, Cyprinus carpio, proteins, unsaturated fatty acids

Prof. Ivan Bogut, Ph. D., Prof. Zlatko Puškadija, Ph. D., MSc. Dalida Galović, University of J. J. Strossmayer, Faculty of Agriculture, Osijek, Croatia; Assoc. Prof. Zdenek Adamek, Ph. D., University of South Bohemia in Ceske Budejovice, Faculty of Fisheries and Protection of Waters, Research Institute of Fish Culture and Hydrobiology, Vodnany, Czech Republic; Prof. Drago Bodakoš, Ph. D., University of J. J. Strossmayer, Faculty of food technology, Osijek, Croatia

<sup>\*</sup>Contact address: Prof. Ivan Bogut, Ph. D., Faculty of Agriculture, Trg Svetog Trojstva 3, 31000 Osijek, Croatia, Tel. ++385 31 224 258, Fax ++385 31 207 017, e-mail: ihtiologija@gmail.com

#### INTRODUCTION

Natural food is an optimal diet for carp juveniles. An appropriate usage of aquacultural measures on the fishpond prior to filling and subsequent manuring after fry stocking result in rich and rapid development of zooplanktonic organisms (Adámek et al., 2008). At higher fish stocking densities, the natural food is consumed faster than it is restored. Therefore, the efforts in preparing suitable commercial feeds, that will meet nutritional and physiological requirements of the fish and replace the natural food, are still in progress. Such feed should contain (depending on fish species and size) around 25-50% protein, 10-15% carbohydrates and 12-15% of fat, as claimed by Berka (1982) and Jirásek and Mareš (2001a; 2001b). However, the authors stress the importance of natural food, explaining some basic differences being important in ingestion and digesting the natural food, compared to the commercially prepared feeds. Live organisms as natural food are moving actively and can be adapted to mouth size and shape immediately after capture as they contain 85-95% of water. Live food mobility is an important factor since most fish respond to prey movements. While the commercial food is characterized by the stable shape, natural food is easily shapeable. Main disadvantage of the industrially prepared feeds is dry matter content which is 10 times higher compared to the natural food. So, if it is not completely consumed, the food residues contribute substantially to water pollution in farming facilities.

Among freshwater zooplankton used in feeding of juvenile carp, planktonic rotifers, cladocerans and copepods are most important. The first report on chemical composition of the planktonic cladoceran *Daphnia magna* was brought by Mann (1935). Chemical analyses showed that fresh matter of *Daphnia magna* contained 91.6, 2.98, 0.78, 0.62, 2.62 and 1.62% of water, protein, chitin, lipid, N-free extract and ash, respectively. Similar results for the composition of *Daphnia magna* body were reported by Wenig (1949), Farkas (1958) and Farkas and Herodek (1960); Schäperclaus (1966) and Albrecht and Breitschprecher (1969). After the one-year observation period, Bogatova et al. (1971) concluded that fresh mass of *Daphnia magna* contains 86.4–97.6, 1.3–5.4, 0.1–0.8 and 0.6–4.0% of water, protein, crude lipid and carbohydrates, respectively. The same authors reported that younger individuals of *Daphnia magna* contain considerably higher amounts of protein in comparison to older ones and that lipid content increased with aging.

Carp requirements for essential amino acids depend on the ratio between energy and protein in food, fish age and size category, partial deficiency of any of essential amino acids, content of non-essential amino acids in food and on the biological availability of essential amino acids. The protein and amino acids utilization depends on ecological factors, mainly water temperature and

dissolved oxygen concentration; food particle size (milling fineness), food ration amount and number of daily rations (Tacon, 1990). All growth categories of carp are sensitive to the lack of lysine, which is manifested in slower growth and disease outbreak. Lysine can be successfully obtained from soybean (Degani et al., 1997; Noble et al., 1998; Yamamoto et al., 1998) or by adding synthetic lysine (Bogut et al., 2000).

Natural food for the carp fry, containing *Daphnia magna* from a small fishpond, was collected at the end of June for the nutritional analyses. The purpose of the study was to determine the chemical composition, essential amino acids and fatty acids content of the freshwater crustacean, *Daphnia magna*, and to compare the results with carp feeding requirements.

#### MATERIAL AND METHODS

The samples of *Daphnia magna* were collected from the concrete pool (5x9 m), which is in use exclusively for cladoceran culture. Samples were collected with 5–liter sampler, then filtered through a 25  $\mu$ m mesh net and preserved in 4% formaldehyde (HCHO). Identification of *Daphnia magna* individuals was done according to Koste (1978) using the Jenaval microscope at various magnifications. Filtrate containing *Daphnia magna* was frozen at temperature of –18°C prior to chemical analyses.

Water content in *Daphnia magna* samples was determined by oven drying at  $105^{\circ}$ C up to constant mass. Ash was determined by burning the samples in a muffol furnace at  $550^{\circ}$ C. Total protein content was determined with Kjel–Foss 16200 type nitrogen analyzer (protein content = N% x 6.25), and fat content was assessed according to Soxhlet. Amino acid content in the hydrolysates was determined by the LKB 4101 (LKB Biochrom, England) automatic analyzer using the Merck (Merck, Germany) standards (Csapo et al., 1986a). Lipids were extracted according to the method of Folch et al., (1975) and fatty acid composition was determined in Chrompack CP 9000 chromatograph using a flame ionizing detector. The calculation of specific fatty acids concentration was performed by the method of Csapo et al. (1986b).

### RESULTS AND DISCUSSION

The results of chemical analyses, presented in Table 1, showed that water content of fresh cladoceran  $Daphnia\ magna$  was 97.28%. The results achieved in our analyses do not correspond with those of Bogatova et al. (1971) who reported the values from 86.40% to 95.60%. They explained the difference in water content by the age of individuals in samples, as being reported by Mann (1935) as well.

Table 1. Chemical composition of live forms and dry matter Daphnia magna (%)\* Tablica 1. Kemijski sastav živih oblika i suhe tvari Daphnia magna (%)\*

Component Sastojak	Daphnia magna		
	Live forms svježi oblici	Dry matter suha tvar	
Dry matter — suha tvar	$2.72 \pm 0.06$	$90.46 \pm 2.76$	
Crude proteins — sirove bjelančevine	$1.18 \pm 0.03$	$39.24 \pm 1.65$	
Crude fats — sirova mast	$0.15 \pm 0.01$	$4.98 \pm 0.33$	
Crude fibres — sirova vlakna	$0.13 \pm 0.02$	$4.32 \pm 0.21$	
N-free extract — NET	$0.82 ~\pm~ 0.05$	$27.29 \pm 2.32$	
Ash — pepeo	$0.44 \pm 0.06$	$14.63 \pm 1.41$	

<sup>\*</sup> mean value of 10 analyses — srednja vrijednost od 10 uzoraka

Protein and dry matter content in live Daphnia magna individuals corresponded to 1.18 and 39.24%, respectively. Compared to our assessments, higher protein amount was reported by Steffens (1986), who found out 4.10 and 46.60% in fresh and dry Daphnia magna mass, respectively. Results reported by Bogatova et al. (1971) indicate large differences in protein content of Daphnia magna in dry matter, ranging from 30.80 to 61.00%. Protein content depends on the age of individuals and on the physiological state of analyzed specimens. This can be influenced e.g. by food composition, egg numbers etc. Hhan and Siddaiqui (1971) recorded in their investigations lower protein fluctuations depending on the age of individuals and confirmed the nitrogen content. The highest nitrogen and protein contents in Daphnia magna were recorded in spring, while the lowest ones in autumn. From the comparison of protein values in Daphnia magna, determined in our study and reference data with requirements of carp growth fish categories, as reported by Tacon (1990), it can be concluded that in terms of protein content, Daphnia magna is an optimal food for all growth categories of carp, except larvae.

Lipid content (Table 1) was 0.15 and 4.98% in fresh and dry *Daphnia magna* mass, respectively. Almost identical values were reported by Bogatova et al. (1971) and Ghioni et al. (1996). The requirements of carp for fat depend on the growth category (Tacon, 1990). From the results presented it can be concluded that *Daphnia magna* meets the requirements for carp nutrition in terms of total fat content. The increase of fat content in fish food results in fat accumulation inside muscles and liver (Hilge, 1998). Optimal ratio between protein and fat percentage in intensive carp fry farming

Table 2. Amino acid composition of live forms and dry matter Daphnia magna and carp amino acids requirements

Tablica 2. Aminokiselinski sastav živih oblika i suhe tvari Daphnia magna i potrebe šarana za aminokiselinama

Amino acid aminokiselina	Daphnia Live forms% svježi oblici %	magna  Dry matter % suha tvar %	Carp requirements in dry matter % potrebe šarana u % suhe tvari
Arginine — arginin	$0.059 \pm 0.002$	$1.60 \pm 0.1$	1.96
Histidine — histidin	$0.026 \pm 0.002$	$0.80 \pm 0.02$	0.86
Isoleucine — izoleucin	$0.053 \pm 0.003$	$0.90 \pm 0.04$	1.76
Leucine — leucin	$0.080 \pm 0.003$	$1.30 \pm 0.06$	2.66
Valine — valin	$0.048 \pm 0.002$	$1.40 \pm 0.03$	1.59
Lysine — lizin	$0.071 \pm 0.003$	$2.20 \pm 0.63$	2.36
Phenylalanine — fenilalanin	$0.059 \pm 0.003$	$2.50 \pm 0.02$	1.96
Methionine — metionin	$0.037 \pm 0.002$	$1.20 \pm 0.03$	1.23
Threonine — treonin	$0.047 \pm 0.002$	$1.50 \pm 0.02$	1.56
Tryptophan — triptofan	$0.028 \pm 0.002$	$0.30 \pm 0.02$	0.93

<sup>\*</sup> mean value of 10 analyses — srednja vrijednost od 10 uzoraka

is 40%: 7% which is slightly lower in fat compared to *Daphnia magna* dry mass (39.24%: 4.98% — see Table 1). N-free extract content in *Daphnia magna* dry mass in our investigations was 27.29%, which is by 15% higher compared to the figures presented by Steffens (1986).

Comparing carp requirements for amino acids (Nose, 1977) with our results, it can be concluded that *Daphnia magna* meet all amino acid requirements except partial phenylalanine shortage. Values of amino acids, higher than in *Daphnia magna*, were determined in rotifers (Dabrowski and Rusecki, 1983; Guisande and Serrano, 1989).

The importance of fatty acids in animal nutrition was first noticed by Burr and Burr (1930). When salmonid culture under intensive conditions was iniated at the beginning of 1960s, it was observed that they were unable to synthesize \*\*de novo\*\* highly unsaturated fatty acids in their organisms, un-

less they received them in food. Watanabe et al. (1975) and Csengeri (1993) pointed out that carp had pronounced requirements for fatty acids of the omega–3 group but they were less sensitive to their shortage than salmonids. It was determined that 0.5% of highly unsaturated fatty acids (20:5 and 22:6 omega–3) were more efficient than 1% linoleic fatty acid. Excessive content of essential fatty acids in carp diet leads to their degradation and conversion into oleic acid. Reduction in the content of essential fatty acids below carp requirements induces slowing down the growth and increasing of 20:3 omega–9 acids in carp flesh.

In the analyses of *Daphnia magna* samples, 18.70 and 66.20% of saturated and unsaturated fatty acids, respectively, were determined. The ratio between unsaturated and saturated fatty acids was 3.54. Among unsaturated fatty acids, monounsaturated and polyunsaturated fatty acids were present with 34.10 and 32.10%, respectively. Total amount of omega–3 and omega–6 acids was 27.3 and 4.80, respectively, with the ratio between them corresponding to 5.68:1 which confirms that omega–3 fatty acids, essential for the fish, are present in very favourable form and amount in *Daphnia magna* bodies.

#### **CONCLUSION**

Concerning the protein content, *Daphnia magna* is an optimal food for carp and other omnivorous fishes. In fresh and dry *Daphnia magna* mass, the protein content corresponded to 1.18 and 39.24%, respectively. Crude fat content in *Daphnia magna* meets the requirements of carp adults but carp fry and juveniles need higher proportion (around 8%). All essential amino acids, except partial deficit of phenylalanine, are present in appropriate quantities for the nutrition of all growth categories of carp and other omnivorous fishes.

The concentration of omega–3 fatty acids, being essential for the carp, the ratio between omega–3 and omega–6 fatty acids, as well as the ratio between saturated and unsaturated fatty acids in the lipids of *Daphnia magna* body completely meet the feeding requirements of all growth categories of carp.

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#### Sažetak

## HRANIDBENA VRIJEDNOST PLANKTONSKOG RAČIĆA DAPHNIA MAGNA ZA HRANIDBU ŠARANSKE MLAĐI (Cyprinus carpio)

I. Bogut<sup>1\*</sup>, Z. Adámek<sup>2</sup>, Z. Puškadija<sup>1</sup>, D. Galović<sup>1</sup>, D. Bodakoš<sup>3</sup>

Istraživanja kemijskog, aminokiselinskog i masnokiselinskog sastava planktonskog račića Daphnia magna obavljena su radi utvrđivanja pogodnosti za hranidbu šaranske mlađi. Sadržaj sirovih bjelančevina, masti, vlaknine, pepela i vlage utvrđeni su uobičajenim metodama. Sadržaj aminokiselina određen je LKB 4101 automatskim analizatorom, a sadržaj masnih kiselina Chropack CP 9000 kromatografom s pomoću plamenoga ionizirajućeg detektora. Sadržaj bjelančevina u svježoj tvari iznosio je 1,18 %, a u suhoj tvari 39,24 %. Navedene vrijednosti bjelančevina u potpunosti odgovaraju hranidbenim potrebama mlađi i starijim kategorijama šarana i drugih omnivornih riba. Količina sirove masti u suhoj tvari iznosila je 4,98 %, a vlaknine 4,32 %, što odgovara potrebama konzumnog šarana. Od esencijalnih aminokiselina u suhoj tvari Daphnia magna, fenilalanin je djelomično nedostatan dok su ostale esencijalne aminokiseline zastupljene u pogodnoj količini za sve kategorije šarana. Količina zasićenih masnih kiselina u masti Daphnia magnae iznosila je 18,70 %, a nezasićenih 66,20 %. Od nezasićenih masnih kiselina, omega-3 masne kiseline zastupljene su s 27,30 %. Odnos omega-3 i omega-6 masnih kiselina iznosio je 5,68:1, što u potpunosti odgovara hranidbenim potrebama šarana.

Ključne riječi: Daphnia magna, Cyprinus carpio, bjelančevine, nezasićene masne kiseline

Prof. dr. sc. Ivan Bogut, prof. dr. sc. Zlatko Puškadija, mr. sc. Dalida Galović, Sveučilište J. J. Strossmayera, Poljoprivredni fakultet, Osijek, Hrvatska; prof. dr. sc. Zdenek Adamek, Sveučilište Južna Bohemija, Češke Budejovice, Fakultet za ribarstvo i zaštitu voda, Istraživački institut za uzgoj ribe i hidrobiologiju, Vodnany, Republika Češka; prof. dr. sc. Dragutin Bodakoš, Sveučilište J. J. Strossmayera, Prehrambeno–tehnološki fakultet, Osijek, Hrvatska

<sup>\*</sup>Kontakt adresa: prof. dr. sc. Ivan Bogut, Poljoprivredni fakultet, Trg Svetog Trojstva 3, 31000 Osijek, Hrvatska, tel. ++385 31 224 258, fax ++385 31 207 017, e-mail: ihtiologija@gmail.com

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