ISSN 1313 - 8820
Volume 8, Number 1
March 2016

AGRICULTURAL SCIENCE AND TECHNOLOGY

2016

An International Journal Published by Faculty of Agriculture,
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The journal is accepted to be indexed with the support of a project № BG051PO001-3.3.05-0001 "Science and business" financed by Operational Programme "Human Resources Development" of EU.

The title has been suggested to be included in SCOPUS (Elsevier) and Electronic Journals Submission Form (Thomson Reuters).

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ISSN 1313 - 8820

Volume 8, Number 1
March 2016

AGRICULTURAL SCIENCE AND TECHNOLOGY

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An International Journal Published by Faculty of Agriculture,
Trakia University, Stara Zagora, Bulgaria

AGRICULTURAL SCIENCE AND TECHNOLOGY, VOL. 8, No 1, pp 90 - 93, 2016
DOI: 10.15547/ast.2016.01.016

Heavy metal content in the meat of common carp (*Cyprinus carpio* L.) and rainbow trout (*Oncorhynchus mykiss* W.), cultivated under different technologies

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(Manuscript received 15 October 2015; accepted for publication 19 February 2016)

Abstract. *Water pollution from industrial production and developing agriculture is a serious problem in aquaculture. The aim of this study was to determine the content of heavy metals Zn (zink), Pb(lead), Ni (nickel) and Cd (cadmium) in the muscles of common carp (Cyprinus carpio L.) and rainbow trout (Oncorhynchus mykiss W.), grown under different technologies. In the current study were investigated common carp (Cyprinus carpio) and rainbow trout (Oncorhynchus mykiss), cultivated in net cages, earthen ponds and raceways. The concentration of heavy metals in the muscles of fish was determined by the methods of AAS in the Scientific laboratory of the Faculty of Agriculture. The influence of different production technologies on the bioaccumulation of Zn, Pb, Ni and Cd (in the flesh of common carp and rainbow trout) was found. The Ni content in muscles was 31.25% higher in common carp, cultured at earthen ponds, compared with its content in the flesh of the fish raised in net cages. The concentration of Pd and Ni in rainbow trout, raised in raceways was higher than that determinated for rainbow trout cultivated in net cages, by 25.0% and 7.14%, respectively. The concentration of Cd and Zn of these species, grown in raceways were lower by 33.33% and 2.14%, respectively, compared with their concentration in rainbow trout, cultivated in net cages.*

Keywords: aquaculture, *Cyprinus carpio* L., *Oncorhynchus mykiss* W., heavy metals, meat, production technologies

Introduction

The most widely cultivated fish species in Bulgaria are common carp (*Cyprinus carpio* L.) and rainbow trout *(Oncorhynchus mykiss* W.*)*.The first one is the most commonly consumed species by households in the country.The consumption of common carp is 35.7% of the total fish production in Bulgarian. It is followed by rainbow trout (17.5%), crucian carp (11.0%), bighead carp (9.9%) and pike perch (8.3%). The cultivation of the most consumed fish species in the country (common carp and rainbow trout) uses different production technologies. In Bulgaria common carp is grown mainly in earthen ponds, and rainbow trout is raised in raceways (Zaykov and Staykov, 2013). There is a tendency to increase the number of farms used for cultivation of fish species in net cages in the last years (Zaykov and Staykov, 2014). It is important to know that aquatic organisms are characterized by the accumulation of many harmful elements in the body. The water, used for aquaculture, which contains very high concentrations of various toxic substances can affect adversely the quality of the final product cultivated fish and consumer health, too (Staykov, 2001). Increased concentrations of heavy metals in the environment is mainly due to erosion and anthropogenic factors. Since these elements are very persistent pollutants, they could accumulate in soil, water and last, but not at least in the different trophic food chains (Szyczewski et al., 2009). Heavy metals have low threshold toxicity and a wide range of disabilities (Atanasov et al., 2013). The accumulation of heavy metals in the body of fish mainly depends on their concentration in food and water, in this aspect the technology of cultivation has an essential role. Being in the body of the hydrobionts, heavy metals accumulate mainly in liver and gill than in muscle. Heavy metal levels in tissues of common carp were increased in summer and winter and decreased in autumn and spring (Selda and Ismail, 2008). Along the food chain eating these fish is dangerous to human health, according to authors working in this field (Farkas et al., 2001; Chen et al., 2000).The attention of researchers in this area is focused on toxic metals, such as iron, arsenic, selenium, chromium, manganese, mercury, lead, cadmium, zinc and copper, which accumulate in the body of fish species (Maffuci et al., 2005; Čelechovská et al., 2007, Selda and Ismail, 2008). The metals classified as heavy metals include: cobalt, selenium, cadmium, chromium, zinc, iron, lead, tin, manganese, mercury, nickel, molybdenum, vanadium, tungsten. In the group of heavy metals one can distinguish both the elements essential for the living organisms (microelements) and the elements whose physiological role is not known, and thus they are "inactive" towards animals, plants and people. Their deficit and surplus badly affects the living organism (Szyczewski et al., 2009). Heavy metals have low threshold of toxicity and high degree of damage in cultured fish. According to many authors, the content of heavy metals in water may have devastating effects on the ecological balance in the environment and in the diversity of aquatic organisms (Ashraj, 2005; Vosyliene and Jankaite, 2006; Farombi et al., 2007). The result of this is large losses of fish production in the aquafarms. The studies are mainly related to tracking quantitative relationship between the concentration of heavy metals in the water and their accumulation in the body of free-living hydrobionts, while the investigations retraced the impact of growing technology on bioaccumulation of these harmful elements in aquatic organisms are strongly limited.

The aim of this study was to determine the content of heavy metals zinc (Zn), lead (Pb), nickel (Ni) and cadmium (Cd) in the muscles of common carp (*Cyprinus carpio* L.*)* and rainbow trout (*Oncorhynchus mykiss* W.), which were grown under different production technologies.

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Material and methods

The six pieces of fish were collected from each fish farm for the current study. The common carp and rainbow trout were selected to have similar size and body weight. The fish were taken from the following fish farms:

- Tunja 73, Nikolaevo town – carp earthen pond farm;
- Forest group, Jrebchevo Dam – net cage farm for common carp growing;
- Reya Fish, Dospat Dam – net cage farm for rainbow trout growing;
- Bukovets, Tvarditsa town – raceways fish farm for growing rainbow trout.

The fish were transported in a cooler box with ice to the laboratory of the Department of Biology and Aquaculture, Trakia University and were segregated, according to their species and production technologies. The total length (L, cm) and body weight (BW, g) were measured in the studied fish. The fish were rinsed with deionized water, placed in polythene bags and stored at -20°C, until performance of the respective tests.

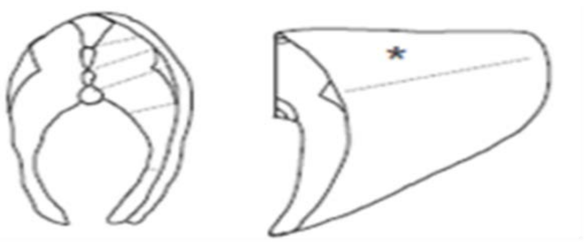

Figure 1. Place for sampling of the muscles of the studied fish (Periago et al., 2005)

Samples for testing the concentration of heavy metals were taken from the dorsal muscle by methodology, described in Periago et al. (2005) (Figure 1).

The fish samples for chemical analyzes were prepared by their filleting and homogenization. The analysis was carried out in the Scientific Laboratory of the Faculty of Agriculture, Trakia University, Stara Zagora. Pretreatment of the samples of the muscles of common carp (*Cyprinus carpio* L.*)* and rainbow trout (*Oncorhynchus mykiss* W.*)* were ashed in a dry manner and dissolved in 6n HCl in order to obtain a solution, with an optimum concentration of the elements Zn, Pb, Cd and Ni.These heavy metals were measured by the atomic absorption spectrophotometry method (Perkin Elmer, AAS) (AOAC, 2007). The concentration of heavy metals was calculated with the help of a standard curve. For each element there is a standard solution with specific concentration to which the content of each tested muscle sample is determined. The content of Zn, Pb, Ni and Cd in the meat of the tested fish species were measured in $\mu g.g^{-1}$. The received concentrations of the studied heavy metals were compared with their reference quantities, specified for the fish intended for human consumption.The statistical processing of data was performed by ANOVA T-test (MS Office, 2010).

Results and discussion

The weight and total length of the studied fish species are given in Table 1. The differences of total length (L,cm) and body weight (BW,g) in common carp, raised in earthen ponds and common carp, grown in net cages were not statistically proven (p>0.05) (Table 1). There were also no statistically significant differences found in the same parameters between rainbow trout, cultured in both production technologies (p>0.05).

According to the authors, who conducted research in this area, the accumulation of heavy metals in the different tissues of the carp is in the following order: intestine > skin > liver > spleen > gills > muscle (Yousafzai et al., 2012). Our research on heavy metal content in the meat of two of the most consumed fish – common carp and rainbow trout raised at earthen ponds and net cages technology, showed that they are lower than the limit values: Pb (0.2 – 0.4 $\mu g.g^{-1}$), Cd (0.05 – 0.1 $\mu g.g^{-1}$), Zn (up to 50 $\mu g.g^{-1}$ and Ni (0.5 $\mu g.g^{-1}$) (Table 2) (2004/22/EC, No: 466).

The presence of heavy metals in the tested muscles of fish is a good indicator for pollution of water bodies with these metals (Carla et al., 2004; Al-Khatani, 2009). The results of the current study showed that in the flesh of common carp cultured under earthen pond technology, the content of Ni was by 31.25% higher than its content in the muscles of fish, grown in net cages ($p \leq 0.01$). In the earthen pond production system Zn concentration in the muscles was 15.39% lower compared to that in fish grown in net cages ($p \leq 0.01$) (Table 2), but the quantity of Ni and Zn in fish muscles was within the permissible limit value. Cultivated in the raceway rainbow trout showed higher Zn content in the muscles (37.28 ± 0.05 g), than that found for the rainbow trout, raised at net cages and the difference was statistically significant ($p \leq 0{,}01$). Similar results are obtained by other researchers, who worked in this area (Fallah et al., 2011).

A large majority of metals have great affinity to bind with amino acids and SH groups of proteins, and therefore act as enzyme inhibitors. An important negative characteristic of metals is their ability to accumulate in organs, especially in the liver, spleen, kidneys and gonads (Spurný et al., 2002; Yilmaz, 2006; Andreji et al., 2006b). To better understand not only the physiological effects but also the toxicological and hygienic effects of metals, it is necessary to know the distribution patterns of individual metals in the organism. The distribution of metals in tissues of different species of fish has

Table 1. Measurements of the studied fish species

Fish species	Fish farm	n	Total body length, cm	Body weight, g
			Mean ± SD	Mean ± SD
Common carp	Tunja 73	6	31.81 ±1.86	1950 ± 0.20
Common carp	Forest Group	6	32.72 ± 1.51	2100 ± 0.14
Rainbow trout	Bukovets	6	23.9 ± 1.19	241.33 ± 5.54
Rainbow trout	Reya Fish	6	21.07 ± 0.47	229.33 ± 16.72

Table 2. Content of heavy metals in the muscles of common carp and rainbow trout (µg.g^{-1})

Fish species	Fish farm	n	Pb	Cd	Ni	Zn
			Mean ± SD	Mean ± SD	Mean ± SD	Mean ± SD
Common carp	Tunja 73	6	0.20 ± 0.002	0.05 ± 0.002	0.16 ± 0.005^{a}	15.66 ±0.296^{a}
Common carp	Forest Group	6	0.20 ± 0.003	0.05 ± 0.001	0.11 ± 0.002^{a}	18.07 ± 0.144^{a}
Rainbow trout	Bukovets	6	0.20 ± 0.002^{c}	0.06 ± 0.004^{b}	0.28 ± 0.001^{c}	37.28 ± 0.048^{b}
Rainbow trout	Reya Fish	6	0.25 ± 0.016^{c}	0.04 ± 0.002^{b}	0.30 ± 0.002^{c}	36.48 ± 0.067^{b}

$^{a, b, c}$ – Values with thw same superscripts in a vertical row differ significantly (p ≤ 0.001).

been described by, e.g., Stancikiene et al. (2006) (perch, roach, silver bream, chub, smelt, tench and pike) and Spurný et al. (2002) (chub).

The Pb content in the muscles of common carp showed the same concentration in both fish farms (Table 2). The concentration of Cd in the muscles of common carp, cultivated in earthen ponds and net cages was the same. The order of accumulation of heavy metals in muscles of the tested common carp in the current study, raised in both production technologies was the following: Zn> Pb> Ni> Cd. Yousafzai et al. (2012) reported, that the order of accumulation of the heavy metals in the muscles of carp raised in earthen pond was the same as the one obtained by us: Zn > Pb > Ni > Cd.

The concentration of Pb and Ni in rainbow trout, raised in raceways was lower than that determined for the same species cultivated in net cages by 25.0% and 7.14%, respectively. The concentration of Cd and Zn in rainbow trout raised in raceways was higher respectively by 33.33% and 2.14%, compared to their concentration in rainbow trout cultivated in net cages. (Table 2). In studies by Cretì et al. (2010) for other fish species it was found that regardless of the production technology, the accumulation of Pb in the internal organs is in the following order: kidney>liver>gills>gut> muscle. Cd in the tissues of some hydrobionts shows sequence similarities of accumulation of the highest levels in the kidney > liver > gills > gut (Ishii et al., 1985). According to Usero et al. (2004) the heavy metal content in fish liver was considerably higher than in the muscle. The heavy metal content in fish may serve as an indicator for the quality of the aquatic environment (Henry et al., 2004; Farkas et al., 2001).

Conclusion

The Ni content in muscles was 31.25% higher in common carp, cultured in earthen ponds, compared with its content in the flesh of carp, raised in net cages. But Zn concentration in the muscles was 15.39% lower, compared to that in fish, grown in net cages. The concentration of Pb and Ni in rainbow trout raised in raceways was lower compared to that determined for rainbow trout cultivated in net cages, by 25.0% and 7.14%, respectively. The concentrations of Cd and Zn in rainbow trout raised in raceways were higher by 33.33% and 2.14%, respectively, compared to their concentration in rainbow trout cultivated in net cages. In the analyzed samples of muscles from common carp and rainbow trout, cultivated in different technologies, the content of Zn, Pb, Ni and Cd was below the limit values for these heavy metals (2001/22/EC, No: 466).

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Todorov N and Mitev J, 1995. Effect of level of feeding during dry period, and body condition score on reproductive performance in dairy cows, IX^{th} International Conference on Production Diseases in Farm Animals, September 11–14, Berlin, Germany.

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AGRICULTURAL SCIENCE AND TECHNOLOGY

Volume 8, Number 1
March 2016

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