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Fish physiology

**IMPACT OF HIGH LEVEL OF CORTISOL, OBSERVED IN THE BLOOD
OF INTENSIVELY REARED TWO-YEAR-OLD CARP (*CYPRINUS
CARPIO* L.), ON THE LEVELS OF ELECTROLYTES AND BLOOD
OSMOLARITY**

**WPŁYW WYSOKIEGO STĘŻENIA KORTYZOLU, OBSERWOWANEGO
WE KRWI INTENSYWNIE HODOWANEGO KROCZKA KARPIA
(*CYPRINUS CARPIO* L.), NA POZIOMY ELEKTROLITÓW
I OSMOLARNOŚĆ KRWI**

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The present study revealed that intensive culture of two-year-old carp, using a high-fat diet (group 1) or a high-carbohydrate diet (group 2), causing a statistically significant rise of cortisol level, resulted in statistically significant rise in potassium ion concentrations as well as a statistically significant drop in chloride ion concentrations (group 1 only).

INTRODUCTION

Ionic composition of the organismic fluids of fishes depends on a number of environmental factors. Among the most important are: water salinity (Eddy 1981), its temperature (Kłyszajko 1986), and contamination level (Andrew 1989).

The ionic-osmotic regulation in fishes proceeds under the control of many hormones. Among the latter the most important role is played by prolactin, glucocorticoids, catecholamines. In view of the above it seems possible, that also other factors of environmental aggression influence the electrolyte balance of fishes, when their action causes a release of those hormones (Thomas 1990).

Taking into account glucocorticoids, it is necessary to mention, that the data concerning their impact on the water-mineral balance of organism are still insufficient, in-

complete, and often contradictory. Some sources report that glucocorticoids play a significant role in water and electrolyte transport through cell membranes, particularly during stress reactions (Swingle and Swingle 1966; Kolpakov et al. 1969) and cause water diuresis (Bailis and Brenner 1978), although the mechanism of the latter is not completely known. It was demonstrated that glucocorticoids cause retention of sodium and chlorides in an organism (Swingle et al. 1967) as well as exhibit natriuretic activity (Krecek et al. 1969). It is also suggested that cortisol participates in regulating the sodium-potassium ratio in extra- and intracellular spaces of the skeletal muscles (Emeljanov 1967) and also takes part in regulating the volume of water spaces in the organism in an extrarenal way on the level of cell-intercellular fluid (Kolpakov et al. 1969).

The aim of this work was to determine the influence of a very high concentration of cortisol, observed in the blood of two-year-old carp subjected to an intensive feeding with feeds of a variable content of fats and carbohydrates, on the levels of basic electrolytes and blood osmolarity.

MATERIAL AND METHODS

The experiment was conducted in July and August on clinically healthy carp in their second year of life. The fish weighed 237 ± 21 g and were supplied by a fish farm in Nowe Czarnowo. The carp were randomly divided into two groups, 90 specimens in each. They were cultured in cages measuring $2.0 \times 0.75 \times 0.8$ m. The cages, stocked with 30 fish each, were kept in the post-cooling discharge canal of the Dolna Odra power plant.

The fish were fed accordingly to the following formula:

Composition of the standard diet before the experiment started:

protein—56 g = 224 kcal (60% of energy),

fat—11 g = 99 kcal (27% of energy),

carbohydrates—12 g = 48 kcal (13% of energy),

additives—1 g.

Total calorific value of 100 g of feed—371 kcal.

Group 1—fed with feed of elevated content of fats (addition of 10 g of poultry fat to 100 g of feed):

protein—35 g = 140 kcal (29% of energy),

fat—17.5 g = 157.5 kcal (33% of energy),

carbohydrates—45.5 g = 182 kcal (38 % of energy),

additives—2 g.

Total calorific value of 100 g of feed—479.5 kcal.

Group 2—fed with feed of elevated content of carbohydrates (addition of 10 g of wheat flour to 100 g of feed):

protein—35 g = 140 kcal (32% of energy),

fat—7.5 g = 67.5 kcal (16% of energy),

carbohydrates—55.5 g = 222 kcal (52% energy),

additives—2 g.

Total calorific value of 100 g of feed—429.5 kcal.

The fish were fed daily from 8:00 to 17:00 every 60 minutes. The feed was served manually on the water surface in the amount of 2% of the metabolic dose. In order to correct the dosage of feeds, the fish were weighed, every 7 days.

The feeds used in the experiment were made in the Experimental Station of the Chair of Inland Fisheries, University of Agriculture in Poznań.

Blood for the studies was sampled before the commencement of the experiment and after its completion. A total of 20 carp was sampled for each group between 7:30 and 8:00. Blood was taken from the caudal vein. The following parameters were determined:

1. Cortisol concentration—employing radioimmunological method, using labeled ^{125}J Cortisol of the Orion Diagnostica Finland reagent set.
2. Concentration of Na^+ and K^+ —using photometric method, on flame photometer Flapho-4.
3. Concentration of chloride ions—employing the impulse coulometric titration, using Spexton 100 Cl chlorimeter.
4. Osmolarity—on automatic Knauer microosmometer.

The results acquired were statistically processed using Statgraphics computer program version 6.0.

Significance of the differences was determined using the t-Student test and the Duncan test.

RESULTS AND DISCUSSION

The present study revealed that intensive rearing of the two-year-old carp, using high-fat or high-carbohydrates diets, causing statistically significant ($P < 0.01$) cortisol rise—resulted, in the aspect of ionic-osmotic balance, in the rise of K^+ and Cl^- ion concentrations in group 1 which was fed using the high-fat diet (Tab. 1).

Similar tendencies of the changes in the electrolyte concentrations were observed also in group 2, fed with the high-carbohydrates diet. The changes, however, were small and statistically insignificant.

Table 1

Impact of intensive rearing and feeding of two-year-old carp with high-fat diet (group 1) or high-carbohydrate diet (group 2) on concentration of cortisol, concentration of Na^+ , K^+ , Cl^- ions, and blood osmolarity

Component	Before experiment (a)	After experiment		
		Group 1 (b)	Group 2 (c)	Significant differences
Cortisol (mmol/dm^3)	188.2 ± 31.2	737.8 ± 372.5	1079.2 ± 534.6	a-b**, a-c**, b-c**
Na^+ (mEq/dm^3)	136.2 ± 8.6	128.8 ± 3.9	128.1 ± 3.2	
K^+ (mEq/dm^3)	3.10 ± 0.26	3.75 ± 0.48	3.21 ± 0.26	a-b*, b-c*
Cl^- (mEq/dm^3)	123.4 ± 2.6	112.3 ± 1.9	116.2 ± 3.5	a-b*
Osmolarity ($\text{mosmol/kg H}_2\text{O}$)	253.1 ± 6.2	246.2 ± 8.6	252.9 ± 6.7	

* $p < 0.05$.

** $p < 0.01$.

Change in the diet composition or its partial modification, through forcing into action other metabolic and enzymatic activities, is perceived by the organism as a factor of environmental aggression, which manifests itself, among others, by changes in glucocorticoid levels (Schüz-Ahrens et al. 1991; Friedrich 1995). Such effect was observed also in the course of intensive rearing and feeding of two-year-old carp with diets of higher values of fats or carbohydrates.

It is a common belief nowadays, that the mechanism of the cortisol action is based on stimulating rise of activities of Na^+ - and K^+ -ATPases. The latter two are responsible for the acceleration of ionic transport in the organs of fishes, which in turn are responsible for maintaining water-mineral balance (Hirano and Mayer-Gostan 1978). It was also demonstrated that in freshwater fishes, cortisol prevents salt losses and reduces Na^+ deficits in the blood serum (Fletcher 1975). In the presently conducted experiment, it was revealed, however, that despite the very high concentration of cortisol observed in the blood of both diet-groups, the effect of its action manifested itself only in the carp of group 1—fed with the high-fat diet. It seems to be linked to a specific impact of this hormone on changes of the volume of the water spaces in the external way, at the cell-intercellular-liquid level (Kolpakov et al. 1969), and also to cortisol-induced changes in the volume of blood serum (Swingle and Swingle 1966). Such increase in the volume of serum may, to some degree, mask the concurrent increase in the content of the electrolytes. Only after exceeding the defined level of their concentrations, the effect of the action of glucocorticoids in this aspect can be visible. It would be confirmed by the lack of statistically significant changes in group 2, in which the concentration of the cortisol was higher (statistically significant) than in group 1. It must be kept in mind, however, that the volume of the blood serum circulating in a young, intensively-developing organism is, according to the majority of re-

searchers, substantially higher, than in the adult individuals (Swingle and Swingle 1966; Kolpakov et al. 1969; Janus et al. 1988).

In view of the above context, the statistically significant ($P < 0.05$) increase in the concentration of K^+ ions in the group 1, seems to be interesting and noticeable. The literature concerning the impact of glucocorticoids on the K^+ level in the blood of fishes reports diversified impacts of them on the concentrations of this ion. Kłyszajko (1986) stated that lowering the water temperature caused the rise of the K^+ level in the blood of the fish kept in that water. Similarly Fletcher (1975) using handling stress, observed, induced by it, a rise in the level of potassium. The different effect of cortisol action was observed by Friedrich (1996) who stated a statistically significant drop in K^+ ion concentration in the course of transport-induced stress. Also Proser et al. (1970) observed a statistically significant decrease in K^+ ions in the blood of goldfish subjected to thermal stress, with the temperature rise from 15 to 25°C.

Summing up, it cannot be ruled out, that the lack of statistically significant changes of the analyzed electrolytes, which under such high concentration of cortisol, particularly in group 2, should be visible—was linked to:

1. General adaptation reactions of the organism which occurred in the course of the experiment (Fitko 1983; de Kloet 1989).
2. Sensitivity decrease of the steroid receptors under prolonged action of high concentrations of cortisol (de Kloet 1989) and resulting decrease of its action symptoms.
3. Protective action of carbohydrates in the water-mineral balance—in group 2, fed with the high-carbohydrate diet, in which cortisol concentration was higher (statistically significant) compared to that of group 1

CONCLUSION

A statistically significant increase in the cortisol level, observed throughout the intensive rearing of two-year-old carp, fed with the diets of high-fat, high-carbohydrates level—caused a statistically significant increase in K^+ ion concentration and a statistically significant decrease in Cl^- ion concentration, but only in the blood of the fish fed with the high-fat diet.

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STRESZCZENIE

Doświadczenie przeprowadzono na kroczeniu karpia, w drugim roku życia, hodowanym w kanale wody pochlódniczej Elektrowni Dolna Odra. Celem pracy było określenie wpływu wysokiego stężenia kortyzolu, obserwowanego we krwi ryb żywionych dietą z podwyższonym udziałem tłuszczu lub węglowodanów, na stężenie podstawowych elektrolitów i osmolarność krwi. Stwierdzono statystycznie istotny wzrost stężenia jonów potasu ($P < 0.05$) i statystycznie istotny spadek stężenia jonów chlorkowych ($P < 0.05$) tylko we krwi ryb żywionych dietą bogatą w tłuszcz. Podobne tendencje zmian stężeń obserwowano w grupie ryb żywionych dietą bogatą w węglowodany, jednak zmiany te były niewielkie i statystycznie nieistotne.

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