ON THE ACTIVITY OF SOME INTESTINAL ENZYMES IN *CYPRINUS CARPIO, ARISTICHTHYS NOBILIS* AND *CARASSIUS AURATUS GIBELIO* SPECIES

GABRIELA DUMITRU [1*], ELENA CIORNEA [1], SILVIA VASILE [2]

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Abstract: The present paper develops a comparative analysis on the activity of some enzymes (amylase, pepsin, trypsin) from the median part of the gastro-intestinal tractus, known as involved in the hydrolytic degradation of exogenous glucides and proteins, in three, two summer-old fish species, from a system of controlled growing. Both the amylase and the peptid-hydrolases under investigation evidenced a quite obvious enzymatic activity, variable from one species to another, the maximum values being attained, each time, in the case of *Cyprinus carpio* species.

INTRODUCTION

The metabolism of proteins and glucides plays a central part in the extremely numerous metabolic processes characteristic for the living world, once generally acknowledged that life itself is characterized by the way in which proteic substances are being organized, as well as by their permanent renewal from the chemical substances occurring in the environment.

As enzymes represent a special class of functional proteins, the metabolism of which is identical with that of structural proteins, there results that the biosynthesis of proteins in the living cell constitutes the basis of the metabolism of substances, viewed as a whole, and characteristic for each species of microorganisms, plants and animals in part (BUCKOW, 2006).

Animal organisms are dependent on the permanent contribution of exogenous glucides and proteins, their utilization beginning with their hydrolytic degradation under the action of some specific enzymes, defined as ***hydrolases*** and ***proteinases***, localized at the level of the digestive tractus. Unlike the case of glucides, in which digestion begins as early as the buccal cavity, under the action of the salivary amylase, digestion of proteins begins in the stomach, where the ***pepsin*** - resulted from the activation of pepsinogene and which, at acid pH hydrolyses the alimentary proteins up to proteoses and peptones - is active.

After its partial degradation in the stomach, under the action of gastric pepsin, the alimentary bolus reaches the duodenum where, due to acidity it gained, determines the secretion - by the duodenal mucous membrane - of a specific hormone, the secretine, which stimulates pancreatic secretion, thus inducing the release of water, minerals salts and of some proteolytic enzymes (COJOCARU and SANDU, 2004; COJOCARU *et al*., 2007).

The investigations where meant at determining the activity of some enzymes of the intestinal tractus (amylase, pepsin and trypsin) involved in the metabolism of glucides and proteins in three, two summer-old cyprinids species grown in a controlled system, namely: *Cyprinus carpio* (common carp), *Aristichthys nobilis* (bighead carp) and *Carassius auratus gibelio* (crucian carp).

MATERIALS AND METHOD

The experiments - performed on five individuals of two summer-old common carp, bighead carp and crucian carp from the Piscicultural Farm of Vlădeni, the county of Iaşi - were meant at evidencing the possible differences observed in the intestinal enzymatic activity, known as closely correlated with the trophic spectrum of each species in part. To this end, after sacrifice and after a previous removal of the intestinal rests through scraping, samples have been taken over from the median part of the digestive tractus.

Precisely-weighed amounts of intestinal tissues were homogenized through jarring and, after centrifugation, the activity of the hydrolases under study was determined from the obtained extracts. Thus, the amylase activity was evaluated by the Méthais-Bieth method, while that of pepsin and trypsin - by the color reaction induced by the hydrolysis products, soluble in trichloracetic acid, with the Folin-Ciocâlteu reactive, in an alkaline medium, color intensity being directly proportional with their concentration and, implicitly, with their proteinazic activity (COJOCARU *et al*., 2009).

Mention should be made of the fact that, for each individual in part, three parallel determinations were made, the average values thus obtained being graphically illustrated.

RESULTS AND DISSCUSION

The process of polyglucide catabolization involves a series of specific hydrolases - known as **amylases** - which catalyze the hydrolytic scission of the α-1,4-glicozidic links, with formation of some smaller fragments, named dextrins. If considering that the species under study have no individualized stomach, determination of enzymatic activity was performed in the median part of the digestive tube (between the esophagus and the duodenum), once known that, in some cyprinids, it may play the role of a so-called "prestomach" (SZLAMINSKA, 1982).

As illustrated by the comparative graphical representation, the *Cyprinus carpio* species shows the highest amylasic activity, with an average value of 0.357 mg hydrolyzed starch/30 min. while, in the case of the bighead carp and crucian carp, the enzyme records somewhat lower values, representing 97, respectively 80% of the value recorded in the common carp, which has to be closely correlated with the greater appetite and higher degree of assimilation of the nutritive substances characteristic to this species (Fig. 1).

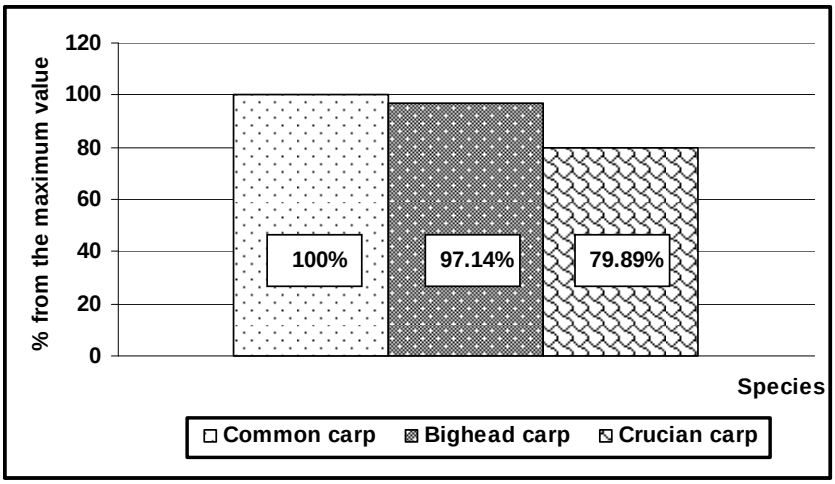

Fig.1. Comparative graphical representation of intestinal amylase activity in common carp, bighead carp and crucian carp

The hydrolytic degradation of proteins occurs under the action of some specific enzymes, defined as proteases or peptid-hydrolases, and it may be either *partial*, with formation of peptides, when only some peptidic links, from the vicinity of some aminoacid rests, towards which the proteolytic enzyme evidences specificity, are split, and *total*, when aminoacids - the final products of proteins enzymatic hydrolysis - are formed. Peptid-hydrolases are to be met in absolutely all living organisms (microorganisms, plants and animals), where they perform some specific functions, of vital importance for the metabolism (VOET and VOET, 1992; PELMONT, 1995).

According to current opinions, proteins degradation and re-synthesis is a complex, permanently-developing process, assuming the utilization of both endogenous and exogenous precursors. As a function of the environmental conditions, physiopathological peculiarities etc., the two resources are utilized at unequal speeds.

The activity of pepsin was determined at intestinal level, the literature of the field suggesting that, in cyprinids, species possessing no-stomach, a weak pepsinic activity, or no activity, may be registered (GUILLAUME *et al*., 1999).

Analysis of the obtained results permits the observation that the intestinal pepsin evidenced some enzymatic activity, in this case, too, variable from one species to another, the maximum value being once again registered in the common carp, followed by the crucian carp while, in the bighead carp, the peptic activity represents less than 50% of the one evidenced in *Cyprinus carpio* (Fig. 2).

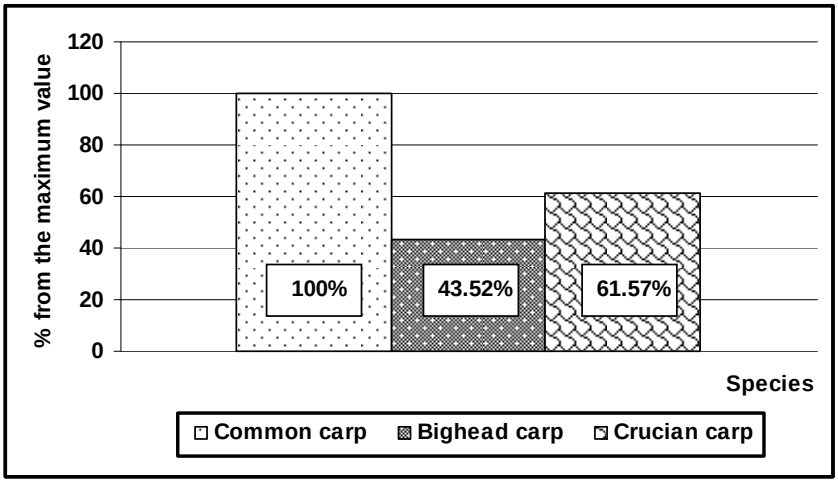

Fig.2. Comparative graphical representation of intestinal pepsin activity in common carp, bighead carp and crucian carp

Another objective of the researches was to determine the activity of trypsin, an enzyme synthesized and secreted by the exocrine pancreas in the form of its inactive zymogen - namely trypsinogen. Its conversion into an active enzyme is initiated by enterokinase (enteropeptidase), an enzyme secreted in the intestinal juice, after which the process continues in an autocatalytic manner - *i.e*., the first trypsin molecules thus formed catalyze the transformation of the new trypsinogen molecule (BRANDEN and TOOZE, 1991).

The maximum activity of trypsin occurs at pH values between 7 and 9, the enzyme hydrolyzing preponderantly the peptidic links, to the formation of which the -COOH groups of arginine and lysine take part. At the level of the thin intestine, where the pancreatic juice is released, trypsin hydrolyses these links in the polypeptides resulted from the action of pepsin.

Figure 3 shows that proteins are mobilized and hydrolytically degraded at higher speed in the same species, *Cyprinus carpio* while, in the *Aristichthys nobilis* representatives, the tryptic activity represents 72.5% and, in *Carassius auratus gibelio* - approximately 65%.

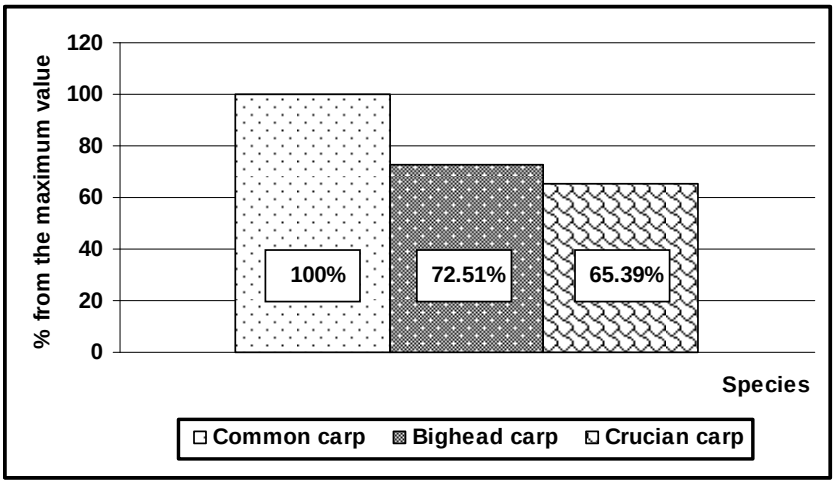

Fig.3. Comparative graphical representation of intestinal trypsin activity in common carp, bighead carp and crucian carp

CONCLUSIONS

A comparative analysis on the activity of some intestinal enzymes involved in the metabolism of glucides and proteins in three cyprinids species occurring in the same developmental stage evidences significant differences from one species to another, as to the rate of enzymatic degradation of alimentary polyglucides and proteins, all enzymes under investigation attaining maximum values in the case of common carp, a species characterized by a more active feeding regime than that of the bighead carp and crucian carp.

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1 - "Alexandru Ioan Cuza" University of Iaşi, Faculty of Biology, Bd. Carol I, Nr. 20 A, 700506, Iaşi, Romania
2 - Ştiubei Comprehensive School, Buzău County, Romania
* - gabriela.dumitru@uaic.ro