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Activity of Some Enzymes in Muscles of Some Local Carnivorous Fish Species

[*1] Duha B Diab, [2]Ruqad Faisel Ali Al-Bayati and [3]Bashar Adham Ahmed

[*1,2,3] Department of Animal Production, College of Agriculture, University of Diyala, Diyala, Iraq.

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***Corresponding Author**
Duha B Diab
Department of Animal Production, College of Agriculture, University of Diyala, Diyala, Iraq.

Abstract
This study aims at investigating the variation in protein levels in the muscles of three local carnivorous fish species across three different seasons. The species studied were *Leuciscus vorax*, *Silurus triostegus*, and *Coptodon zillii*. The results revealed differences in muscle protein levels among these species. In autumn, the average muscle protein concentration was 19.5 ± 0.5% for *L. vorax*, 22.9 ± 0.5% for *S. triostegus*, and 16.3 ± 0.5% for *C. zillii*. During winter, the protein concentration was 18.9 ± 0.4% for *L. vorax*, 20.5 ± 0.5% for *S. triostegus*, and 18.9 ± 0.5% for *C. zillii*. In spring, the protein concentration increased to 20.4 ± 0.2% for *L. vorax*, 19.9 ± 0.5% for *S. triostegus*, and 18.8 ± 0.5% for *C. zillii*. The study also examined the activity of digestive enzymes such as ALP, GPT, and GOT in the flesh of these three species across the seasonal changes. This study aims at determining the optimal muscle protein levels in these carnivorous fish species, which vary with the seasons and the ecological characteristics of their habitats. This information can help assess the nutritional value of consuming the flesh of these fish.

Keywords: Nutrition, Digestion, Natural Food, Freshwater, Season

Introduction

The concentration of protein and some digestive enzymes is affected by the amount of protein that fish obtain from their natural diet or the feed provided to them, especially when the feed contains animal-based protein sources. This relationship is directly proportional, indicating that as the protein concentration in the diet increases, the body protein also increases, accompanied by a noticeable rise in the activity of certain digestive enzymes, as illustrated by Nazir *et al.* (2023). Fish rely on high levels of protein in their bodies and the strong activity of digestive enzymes through increasing the concentration of protein in their feed, which is evident in farmed fish when the levels of protein concentrates in their diets are raised (Silva & Chamul: 2000). However, concentrations of protein in fish feed, particularly for farmed fish, are not the only means of increasing the levels of some digestive enzymes; but there are many natural protein alternatives that can enhance enzyme activity. Thus, high protein concentrations in fish bodies can be attained, especially from natural food sources (Li *et al.*, 2022; Shiyou *et al*., 2024).

The concentration of protein in the stomach content of predatory or carnivorous fish varies due to several environmental factors that affect the availability of food for these fish, as indicated by Ahmed *et al*. (1984). Among these factors are water temperature and pH. Temperature changes influence the consumption of available nutrients by fish in their environment, with feeding behavior varying by species, as noted by El-Sayed & Moharram (2007), particularly in predatory fish. These fish select their food based on its availability and type. Since temperature is influenced by the season or time of year, this in turn affects the availability of natural food for these species, leading to variations in protein concentration in their stomachs, as observed by Ahmed *et al.* (1984). Consequently, this results in differences in protein levels and the activity of certain enzymes in the flesh and bodies of these fish.

Other possible reasons include differences between sexes and age, as indicated by Hbashy (1972) and Nargis (2006). Additionally, metabolic rate and feed conversion efficiency in fish vary with temperature and other environmental changes that occur with seasonal shifts, as confirmed by Al-Shekh *et al.* (1991) and Rasul *et al.* (2021). Depending on the species

and the type of available food in the environment, we observe an increase in metabolic rate with higher temperatures. Regarding the role of different enzymes among species, enzyme concentrations vary by species and within genders of the same species, and follow seasonal patterns along with factors affecting food availability and temperature, as noted by Shafi & Jasim (1982), Al-Selah *et al.* (2010), and Rasul *et al.* (2021). It is worth mentioning that most dominant species in Iraqi inland waters belong to the Cyprinidae family, which share many external morphological traits and feeding habits, as noted by Hadi *et al*. (2023).

Materials and Methods

Twenty fish of the local carnivorous species, including *Leuciscus vorax, Silurus triostegus,* and *Coptodon zillii* were caught from various locations in Baghdad, Diyala, and Wasit using standard hand nets. The fish were collected in three separate batches, one in each of the three different seasons: autumn, winter, and spring, resulting in a total of 180 fish over the three seasons. The species were identified at the Fish Laboratory of the Iraqi Natural History Research Center and Museum. The stomach contents of each species were isolated individually and preserved in refrigerated bottles until analysis could be conducted. Each type of analysis was performed in replicates to ensure the accuracy of the final results. A micro-Kjeldahl apparatus was used to measure the protein concentration in the stomach contents of the three species. For enzyme activity measurement, muscle samples of the studied species were prepared as follows:

Measuring the Effectiveness of Glutamic-Oxaloacetic Transaminase (GOT)

The effectiveness of this enzyme was measured in blood serum using several kits obtained from the Institute of Serums and Vaccines. These kits relied on the colorimetric method, and the testing procedures were carried out according to the instructions provided by RANDOX, the supplier of the kits. The samples were read with a spectrophotometer at a wavelength of 546 nanometers, based on the method described by Reitman & Frankel (1957).

Measuring the Effectiveness of Glutamic-Pyruvic Transaminase (GPT)

The examination was conducted using kits obtained from the Institute of Serums and Vaccines, following the same procedures as the previous test. The process was carried out according to the instructions provided with the kits, in accordance with the method described by Reitman & Frankel (1957).

Measuring the Effectiveness of Alkaline Phosphatase (ALP)

The examination was carried out using kits obtained from the Institute of Serums and Vaccines, following the procedures outlined in the accompanying brochure provided by the French company (bioMérieux). This method, based on a colorimetric assay, was used to measure the enzyme activity. The samples were read using a spectrophotometer at a wavelength of 510 nanometers, following the method described by Kind & King (1954).

Results and Discussion

Figures 1, 2, and 3 illustrate the local carnivorous species that were captured and identified in this study.

Fig 1: *Leuciscus vorax*

Fig 2: *Silurus triostegus*

Fig 3: *Coptodon zillii*

Table 1 presents the results of protein analysis in the stomach contents of the three species across the three seasons. It shows variations in protein concentrations for each species within a single season, as well as differences in these concentrations among the different species across the three seasons. These findings indicate that seasonal changes in protein levels for these species vary with changes in the season, temperature fluctuations, and the corresponding changes in the pH levels of the water, particularly during autumn and spring (Dembergs, 1964; Marais & Erasmus, 1977).

During the analysis of the flesh from these different fish species, it was observed that there was a notable increase in moisture content compared to the protein levels in the stomachs of these species. Moisture content was high across all species, particularly in spring, which is a preparation period for reproduction and the maturation of reproductive organs, as described by Nabi & Hossain (1989). Conversely, protein levels in the diet of these species increased with a decrease in moisture content after sexual maturity and full development of reproductive organs in preparation for spawning, as noted by Epler *et al.* (2001).

Table 1: Protein concentration in the stomach content of the three species during the three different seasons

Species	Protein concentration in stomach content% of dry matter		
	Autumn	Winter	Spring
Leuciscus vorax	19.80	18.20	20.67
Silurus triostegus	23.50	20.54	19.50
Coptodon zillii	16.34	18.10	18.90

Table 2 provides details on the concentrations of certain enzymes in the flesh of the three species across the three different seasons.

Table 2: Concentrations of some enzymes in the meat of shuck, jari, and tilapia fish in three seasons of the year

Season	Enzyme concentration	Species		
		Leuciscus vorax	Silurus triostegus	Coptodon zillii
Autumn	ALP	23	23	19
	GPT	7.1	7.3	6.1
	GOT	69	71	60
Winter	ALP	21	22	21
	GPT	5.9	6.1	5.7
	GOT	60	63	57
Spring	ALP	20	20	20
	GPT	4.7	4.7	4.6
	GOT	55	56	54

It is observed that the concentration of the enzyme GOT increased by 14 units/gram in the summer. This enzyme plays a crucial role in amino acid metabolism in the body and is responsible for cell proliferation. This effect becomes evident with the rise in temperatures during the summer, as noted by Yang *et al.* (2019) and Li *et al.* (2022). On the other hand, the concentration of the enzyme GPT increased by 2.4 units/gram in summer. Both enzymes are important in the protein metabolism cycle in the body and in the deposition of proteins as fibrous tissues in fish flesh. They transport essential amino groups needed for building muscle cells, which supports the findings of Karmen *et al*. (1955), Shafi & Jasim (1982), and Giannini *et al*. (2005).

In general, fish typically release their reproductive products during the spring, from mid-February to mid-March. Following this period, the fish enter full sexual maturity, accompanied by seasonal sexual morphological changes, especially in males. This was observed by Ibrahim and Ghazwan (2016) in their study of *L. vorax* during the mating season, as illustrated in Figure 4. The changes in these enzymes are accompanied by alterations in the size and shape of the reproductive organs in both sexes. This is clearly evident from the analysis of amino acid transaminases during the spring season. This pattern generally applies to most Iraqi fish species during this spring period, as shown in Figures 5 and 6. Amino acids are lost due to reproductive stress and pressure on fish during spawning, impacting their vitality as they expend energy to maintain the species and search for food to enhance reproductive capacity in spring, especially in Iraq (Reitman & Frankel, 1957; Hbashy, 1972).

Conversely, it is observed that the levels of amino acid transaminases increase during the summer, as fish compensate for the weight lost during spawning and reproduction in the spring, particularly (Reitman & Frankel, 1957; Wootton, 1974; Wootton, 1998; Satyanarayana, 2018; Jeyasanta & Patterson, 2023).

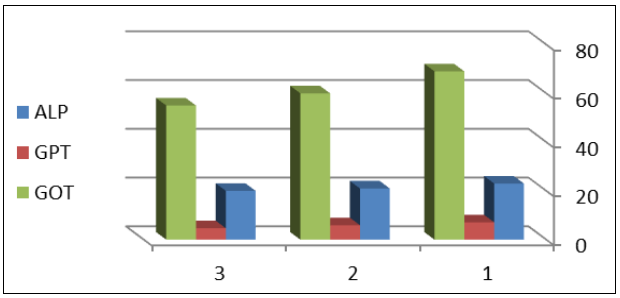

Fig 4: Enzyme concentration of *Leuciscus vorax*

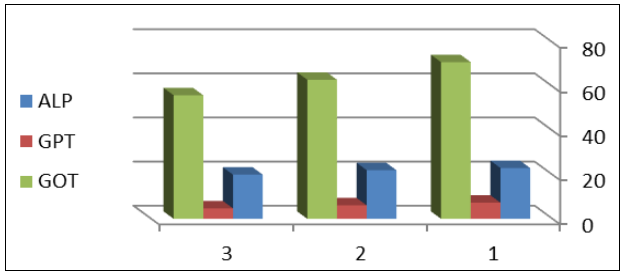

Fig 5: Enzyme concentration of *Silurus triostegus*

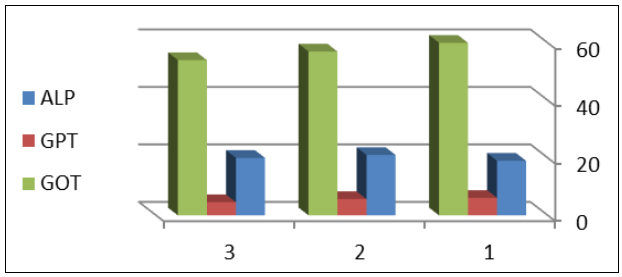

Fig 6: Enzyme concentration of *Coptodon zillii*

There was no significant change observed in the concentration of the enzyme Alkaline Phosphatase (ALP) across the three studied species. Additionally, the values of this enzyme showed only slight variations throughout the three seasons of this study, as illustrated in Figures 4, 5, and 6. ALP is considered a good indicator of overall bodily condition and vitality, particularly in assessing malnutrition, damage to internal organs, and liver activity across vertebrates, including fish (Shipman *et al.*, 2013; Lowe *et al.*, 2023).

The seasonal variation in food availability significantly impacts the protein content in the diet of fish, which in turn affects the protein content in their stomachs (Ahmed *et al*., 1984). During the breeding season, which generally occurs in spring for local species, changes in fish bodies have a significant impact on the levels of body components, particularly proteins and fats. Additionally, variations in different enzymes correspond to the season, leading to fluctuations in protein levels (Phillips, 1966; Wootton, 1974; Wootton, 1998; Satyanarayana, 2018; Jeyasanta & Patterson, 2023).

In general, the composition of fish bodies, the contents of their digestive tracts, and the nature of their flesh are directly influenced by the species, age, and sex of the fish, as well as the type and nature of their diet. Moreover, the feeding behavior of these species when accessing different food sources varies according to the type of fish (Phillips, 1966; Gras *et al.*, 1967; Nasef, 2021; Krishna *et al*., 2023).

Conclusion

Based on this study conducted on three local species of carnivorous fish, we can infer that protein concentration levels increase directly with the increase in the proportion of protein components in the fish's diet. Additionally, an increase in the activity of certain digestive enzymes, which play a crucial role in the synthesis and formation of essential amino acids used in muscle cell construction, was observed. Consequently, the relationship between growth and changes in environmental factors such as temperature and the availability of natural food for each species is positively correlated. In other words, growth increases with the rise of these factors, particularly as warmer seasons like spring approach.

References

1. Ahmed AT, Mustafa G, Alam MZ, Rubbi SF, M Moslemuddin M. Biochemical composition of seven species of Gobi fish. *J. Asiatic Soc. Bangladesh (Sc.),* 1984, 10107-111.
2. Al-Selah F, Hammoud V, Al-Hussein A. Reproduction – Biology of Aspius vorax (Heckle,1843) Within the Middle Euphrates River. 2010; (26).
3. Al-Shekh MA, Mansor Q, Al-luss SB. Fish breeding and production, P2, Dar Al- Hekma for Printing and Publishing, Baghdad, 1991, 59-62.
4. Dembergs N. Extractions of fish muscles, 4.Seasonal variation of fat, water soluble protein and water in Cod (*Gadno morlia*) fillets. *J. Fish Res. Canada.* 1964; (21):705-709.
5. El-sayed HK, Moharram SG. Reproductive Biology of Tilapia zilli (Gerv. 1848) from Abu Qirbay, Egypt, of Aqua res. 2007; 33(1):379-394.
6. Epler P, Sokotowska M, Popek W, Bieniarz K, Bartel R, Szczerboski J. reproductive biology of selected fish species from lakes Tharthar and habbaniya in Iraq, *Arch. Pol. Fissh.* 2001; 9(1):199-209.
7. Giannini EG, Testa R, Savarino V. Liver enzyme alteration: a guide for clinicians. CMAJ. 2005; 172(3):367-79. https://doi.org/10.1503/cmaj.1040752
8. Gras J, Reynand R, Gamoty L, Frey J, Henry JC. Biochemical study of Fishes. 1. Six monthly variations in the water and protein content of the muscle tissue of rain bow trout (Salmo gairderi Rich). Experiments. 1967; (23):430-431.
9. Habashy AP. Seasonal variation of moisture, protein, fat and ash in the mirror carp (Cyprinus carpio Linnaeus, 1758. A R E Zool. Listy. 1972; 22:85-89.
10. Hadi HD, Baraaj AH, Ali AH. Morphological and molecular studies of kais kingfish cyprinion kais heckel, 1843 (piscies, cypriniformes, cyprinidae) from the middle of Iraq, Bulletin of the iraq natural history museum. 2023; 17(4):611-628. https://doi.org/10.26842/binhm.7.2023.17.4.0611
11. Ibrahim AS, Ghazwan MI. Study of sexual dimorphism in *Aspius vorax* fish (Heckel, 1843) (Cypriniformes: Cyprinidae), The 4th International Scientific Conference on Genetics and Environment, Cairo, Egypt from 23-30. 2016, 256-268.
12. Jeyasanta KI, Patterson J. Seasonal Variation in the Nutritional Composition of Fishes in Tuticorin, Southeast Coast of India. *Journal of Advances in Food Science & Technology*. 2023; 10(1):29-47. https://doi.org/10.56557/jafsat/2023/v10i18067
13. Karmen A, Wroblewski F, Ladue JS. Transaminase activity in human blood. *J Clin Invest.* 1955; 34(1):126-31. https://doi.org/10.1172/JCI103055
14. Kind PRN, King EJ. Estimation of plasma phosphatase by determination of hydrolysed phenol with amino-antipyrine. *J. Clin. Pathol*. 1954; 7:322.
15. Krishna PV, M Naveen Kumar, BVL Aradhya Sarma, P Dedeepya. Proximate composition and its seasonal variations of the muscle tissue of *Channa striata* from Krishna river, Andhra Pradesh. *Int J Fish Aquat Stud.* 2023; 11(1):109-115. DOI: https://doi.org/10.22271/fish.2023.v11.i1b.2769
16. Li Y, Li B, Xu Y, Qian L, Xu T, Meng G, Li H. GOT2 Silencing Promotes Reprogramming of Glutamine Metabolism and Sensitizes Hepatocellular Carcinoma to Glutaminase Inhibitors. Cancer Res. 2022; 16;82(18):3223-3235. https://doi.org/10.1158/0008-5472.CAN-22-0042
17. Lowe D, Sanvictores T, Zubair M, John S. Alkaline Phosphatase. Oct 29. In: Stat Pearls [Internet]. Treasure Island (FL): Stat Pearls, 2023-2024. PMID: 29083622. https://pubmed.ncbi.nlm.nih.gov/29083622/
18. Marais JF, Erasmus T. Body composition of Mugil cephalus, Liza domerle, L. richardsoni and L. tricaspidus (Teleostei: Mugilidae) caught in the Swartkops estuary. Aqua. 1977; (10)75-86.
19. Nabi MR, Hossain MA. Seasonal variation in the chemical composition and caloric content of Macrognathus aculeatus (Bloch) from the Chalon Beel waters. *J. Asiatic Soc. Bangladesh (Sci.).* 1989; (25):103-110.
20. Nargis A. Seasonal Variation in the Chemical Composition of Body Flesh of Koi Fish *Anabas testudineus* (Bloch) (Anabantidae: Perciformes). *Bangladesh J. Sci. Ind. Res.* 2006; 41(3-4):219-226.
21. Nasef A. Effect of Variation in Aquatic Environment Type on Biochemical Composition and Protein Quality in Some Fishes. *Egyptian Academic Journal of Biological Sciences, B. Zoology*. 2021; 13(2):207-223. https://doi.org/10.21608/eajbsz.2021.207515
22. Nazir S, Khan N, Fatima M, Azmat H, Naveed S, Ramzan MM. The influence of dietary protein concentration on digestive enzyme activities, growth, and body composition in juvenile bullseye snakehead (Channa marulius). PLoS One. 2023; 14;18(2):e0281274. https://doi:10.1371/journal.pone.0281274
23. Phillips AM, Livingsten DL, Poston HA. Use of caloric sources by brook trout. *Proc. Fish Cult.* 1966; (28):67-72.
24. Rasul MG, Jahan I, Yuan C, Sarkar MSI, Bapary MAJ, Baten MA. Seasonal variation of nutritional constituents in fish of South Asian Countries: A review. Fundamental and Applied Agriculture. 2021; 6(2):193-209. https://doi.org/10.5455/faa.65131
25. Reitman S, Frankel S. A calorimetric method for the determination of serum glutamic oxaloacetic and glutamic pyrovic transaminases. *Am. J. Clin. Path.* 1957; (28):56-63.
26. Satyanarayana MCHV. Seasonal variations in proximate composition of freshwater catfish Mystusspecies (Family: Bagaridae) from two different habitats, *International Journal of Creative Research Thoughts (IJCRT)*. 2018; *6*(1): 1585- 1606.
27. Shafi M, Jasim BM. Some aspects of the biology of a cyprinid, Asius vorax Heckle, *J. Fish Biol*. 1982; 20:271-278.
28. Shipman KE, Holt AD, Gama R. Interpreting an isolated raised serum alkaline phosphatase level in an asymptomatic patient. *British Medical Journal*. 2013; 346:976.
29. Shiyou Chen, Sahya Maulu, Jie Wang, Xiaoze Xie, Xiaofang Liang, Hao Wang. The application of protease in aquaculture: Prospects for enhancing the aquafeed industry, Animal Nutrition. 2024; 16:105-121, ISSN 2405-6545, https://doi.org/10.1016/j.aninu.2023.11.001
30. Silva JL, Chamul RS. Composition of marine and freshwater finfish and shellfish species. Marine and freshwater products handbook. Technomic Publishing, Lancaster. 2000; 4:31-45.