

STUDIES ON MUSCLE PROTEIN PROFILE OF FISH REARED IN POULTRY DROPPINGS FERTILIZED PONDS

Muhammad Hassan¹ & M. Javed²

¹Department of Zoology, Government College, Faisalabad

²Fisheries Research Farms, Department of Zoology and Fisheries, University of Agriculture, Faisalabad

The fish (*Catla catla*, *Labeo rohita* and *Cirrhina mrigala*) muscle protein profile viz. sarcoplasmic, myofibrillar and stromal proteins has been determined after one year pond fertilization trial under polyculture system. Fish were reared under five nitrogen levels (0.10, 0.13, 0.16, 0.19 and 0.22 g N) from broiler droppings and a control without additives. The present investigation reveals significant interactions between treatments and species for sarcoplasmic, myofibrillar and stromal proteins. Higher concentration of sarcoplasmic proteins in *Catla catla* corresponded with the comparative decrease of myofibrillar proteins. The correlation coefficients between sarcoplasmic and myofibrillar proteins of the three fish species were significantly negative while the same between stromal and myofibrillar proteins were negatively significant for *Cirrhina mrigala* only. This shows that stroma would increase at the expense of myofibrillar proteins, in *Cirrhina mrigala*. The correlation between sarcoplasmic and stromal proteins was positively significant in *Cirrhina mrigala* and negatively so ($P < 0.01$) in *Catla catla*.

Key words: major carps, myofibrillar, sarcoplasmic and stromal proteins, poultry droppings

INTRODUCTION

There are three types of muscle proteins viz. sarcoplasmic, myofibrillar and stromal proteins. Sarcoplasmic and myofibrillar proteins are salt-extracted proteins. Myofibrillar proteins comprise myosin, actin and regulatory proteins like tropomyosin, troponin and actinin. These proteins collectively play an important role in coagulation and gel formation during processing. Choi et al. (1986) compared the muscle protein profile of wild and cultured eels. The wild eel was composed of 30.78 % sarcoplasmic, 59.02 % myofibrillar, 9.73 % residual intracellular and 2.47 % stroma fractions and that of cultured eel consisted of 31.80, 58.37, 8.16 and 1.80 % respectively. Barlas (1986) presented the comparison of sarcoplasmic, myofibrillar and stromal proteins of flat fish, common carp and beef/rabbit and found carp to be the best as it contained less stroma (3.00 %) as compared to flat fish and beef which were 5 and 16-18 % respectively. However, the myofibrillar and sarcoplasmic protein concentrations in beef/rabbit were 39 to 68 and 16 to 18 % respectively. Javed et al. (1992) reported that *Labeo rohita*, reared under broiler manure fertilization (at the rate of 0.10 g N/100 g of fish) gave the maximum myofibrillar protein content as compared with other two fish species viz. *Catla catla* and *Cirrhina mrigala*. The meat of *Cirrhina mrigala*, cultured under layer manure fertilization of pond, had the least ($2.65 \pm 0.02\%$) stromal protein, while it ($4.88 \pm 0.03\%$) was the

maximum in *Catla catla* under control treatment (without additives). However, in all the three fish species, muscle stroma increased at the expense of myofibrillar proteins and the proportions between the major body constituents of fish varied with feeding levels.

MATERIALS AND METHODS

Factorial experiment, with two replications for each of the treatments, was conducted under ambient condition using earthen ponds. After preliminary preparations (Javed, 1988), all the ponds were initially fertilized, separately, with 40 kg broiler droppings (3333.33 kg/ha) as a starter dose to stimulate primary productivity. Fingerling major carps, 6-7 months old (induced bred, procured from Fish Seeds Hatchery, Faisalabad), average weight 21.32 ± 1.99 g, were randomly stocked, from a selected population, in each of the ponds with stocking density of 25, 60 and 15% for *Catla catla*, *Labeo rohita* and *Cirrhina mrigala* respectively (64 fish in each of the ponds). Fertilization of ponds with broiler droppings (4.37% nitrogen) was started on the basis of nitrogen contents. Five levels viz. 0.10, 0.13, 0.16, 0.19 and 0.22g nitrogen per 100 g of wet fish weight daily, were designated as T1, T2, T3, T4, and T5 respectively, while T6 served as control without additives.

At the end of one year experiment, fish muscle proteins viz. sarcoplasmic, myofibrillar and stromal proteins, in fish species (connective tissue proteins) were analysed. Fresh fish meat samples (five from each of the three fish species under all the

Table 1. Average final fish weights (g) after one year pond fertilization trial

Species	Treatments :t S.O.					
	T1 (0.10g N/level)	T2 (0.13g N/level)	T3 (0.16g N/level)	T4 (0.19g N/level)	T5 (0.22g N/level)	T6 (Control)
<i>Catla catla</i>	786.92±12.25 b	791.59±8.31 b	823.88±10.14 a	798.70±9.58 b	792.06±12.80 b	128.45±6.91 c
<i>Labeo rohita</i>	563.42±12.50 c	590.40±10.63 c	670.35±8.34 ab	686.92±12.12 a	650.02±8.53 b	142.02±3.72 d
<i>Cirrhina mrigala</i>	677.04±10.23 a	668.19±9.55 a	680.26±11.82 a	639.36±8.78 b	517.24±12.11 c	157.36±6.15 e
Mean	675.79	683.39	724.83	708.33	653.11	142.61

Means with similar letters in a row are statistically similar at $P < 0.05$; S.D. = Standard deviation.

Table 2. Muscle protein profile of major carps

Species	T1 CO.10gN level)	T2 CO.13gN level)	T3 CO.16gN level)	T4 CO.19gN level)	T5 CO.22gN level)	T6 (Control)
	CO.10gN level)	CO.13gN level)	CO.16gN level)	CO.19gN level)	CO.22gN level)	(Control)
Sarcoplasmic proteins (%)						
<i>Catla catla</i>	30.43±0.45 d	31.23±0.41 be	31.00±0.38 c	31.38±0.47 b	32.41±0.41 a	29.96±0.18 e
<i>Labeo rohita</i>	27.66±0.46 b	28.50±0.45 a	27.08±0.42 c	26.94±0.38 c	26.82±0.36 c	28.08±0.19 b
<i>Cirrhina mrigala</i>	24.03±0.24 d	25.49±0.41 b	24.90±0.55 c	24.78±0.37 c	26.03±0.25 b	28.73±0.19 a
Mean	27.37	28.41	27.66	27.70	28.42	28.92
Myofibrillar proteins (%)						
<i>Catla catla</i>	66.00±1.06 a	65.50±0.89 b	65.20±0.58 b	65.93±0.47 a	64.70±0.51 c	66.22±0.25 a
<i>Labeo rohita</i>	69.32±0.77 b	68.13±0.42 d	69.41±1.10 b	69.80±0.46 ab	69.97±0.60 a	68.70±0.25 c
<i>Cirrhina mrigala</i>	73.59±0.49 a	71.50±0.46 c	71.71±0.58 c	72.58±0.51 b	71.25±0.59 c	68.09±0.18 d
Mean	69.64	68.38	68.77	69.44	68.64	67.67
Stromal Proteins (%)						
<i>Catla catla</i>	3.47±0.25 be	3.27±0.25 c	3.80±0.20 a	2.69±0.25 e	2.99±0.31 d	3.72±0.18 a
<i>Labeo rohita</i>	3.02±0.17 e	3.45±0.25 abc	3.51±0.30 a	3.26±0.12 cd	3.21±0.31 de	3.27±0.30 bed
<i>Cirrhina mrigala</i>	2.38±0.16 e	3.01±0.17 c	3.39±0.14 a	2.64±0.28 d	2.72±0.22 d	3.22±0.24 b
Mean	2.96	3.24	3.57	2.86	2.97	3.40

Means with similar letters in a row are statistically similar at $P < 0.05$; NeNitrogen; S.D.= Standard deviation.

treatments) were taken, separately, from nape, center and tail, mixed them to have a representative sample. Extraction of sarcoplasmic and myofibrillar proteins were performed in a temperature controlled room at 2°C (Helander, 1957). The filtrates for both sarcoplasmic and myofibrillar proteins were analysed to determine

concentrations by using biuret procedure (Gornall et al., 1949). The residue (stromal proteins) after myofibrillar protein extraction, was dissolved in 40 % NaOH solution and total proteins were estimated by using micro-Kjeldahl method of A.O.A.C. (1984), which gave the contents of stroma in fish meat. Data were analyzed using ANOVA and DMR tests.

Correlation and regression analyses were also performed to find out relationships / trends among various parameters under study.

RESULTS

Table 1 shows the final weights of three fish species under six treatments. The muscle protein profile of the finally harvested fish and relationship among various proteins have been presented in Tables 2 and 3 respectively.

Sarcoplasmic Proteins : Sarcoplasmic protein contents of *Catla catla* differed significantly under all the treatments except for T2, T3 and T4. Sarcoplasmic proteins were significantly higher due to T5 (32.41 ± 0.41 %). In case of *Labeo rohita*, T2 was the source for maximum contents of sarcoplasmic proteins (28.50 ± 0.45 %) in fish body followed by T6 (28.08 ± 0.19 %). However, the differences among T3, T4 and T5 were statistically non-significant. *Cirrhina mrigala* had the maximum content of sarcoplasmic proteins (28.73 ± 0.19 %) in control treatment and minimum (24.03 ± 0.24 %) in T1. The differences between T3 and T4, and T2 vs. T5 were statistically non-significant (Table 2). The significant interaction (treatments x species) was due to the performance of both *Labeo rohita* and *Cirrhina mrigala* under T6.

Myofibrillar Proteins: Myofibrillar proteins are salt-extracted proteins comprising myosin, actin and regulating proteins such as tropomyosin, troponins and actinins. These collectively play an important role in coagulation and gel formation during processing. In *Catla catla*, T1, T4 and T6 showed significant differences with other treatments for the contents of myofibrillar proteins. *Catla catla* had the maximum myofibrillar protein content of 66.22 ± 0.25 % under T6 closely followed by the content of 66.00 ± 1.06 % under T1 (Table 2). In case of *Labeo rohita*, the maximum value for these proteins being 69.97 ± 0.60 % for T5, while minimum value of 68.13 ± 0.42 % was found under T2. As regards *Cirrhina mrigala*, the differences were non-significant among T2, T3 and T5. This species of fish showed significantly high myofibrillar protein content of 73.59 ± 0.49 % under T1, while the same was minimum in the fish given T6 (68.09 ± 0.18 %). On overall basis, *Cirrhina mrigala* appeared to be the species which had better ability to synthesize myofibrillar proteins than the other two species. Significant interaction (treatments x species) for myofibrillar protein was due to the performance of

Labeo rohita and *Cirrhina mrigala* under T6 (Table 2).

Stromal Proteins : *Catla catla* reared under T3 and T6 contained the maximum stroma of 3.80 ± 0.20 and 3.72 ± 0.18 % respectively while the same was significantly minimum under T4 (2.69 ± 0.25 %). As regards *Labeo rohita*, the maximum accumulation of stroma was observed under T3 and T2 with non-significant differences, T3 also caused significantly higher accumulation of stromal proteins in *Cirrhina mrigala* than with rest of the treatments (Table 2).

As regards the overall picture of observations on the stromal contents of three fish species, *Cirrhina mrigala* appeared as the best species, since it contained less stroma, followed by *Labeo rohita* and *Catla catla*. Significant differences among the treatments for fish stroma contents reflected the differential response of three fish species for acquiring stromal proteins under pond fertilization conditions because of highly significant interaction between treatments and species (Table 2).

DISCUSSION

Significantly higher percentages of stromal proteins in all the three fish species under 0.16 g nitrogen level (T3) were due to higher weight attainment of fish in this treatment (Tables 1 and 2). The interactions (treatments x species) for sarcoplasmic, myofibrillar and stromal protein contents of fish meat were highly significant indicating that the three fish species responded differently under the six treatments for these variables. Sarcoplasmic proteins of *Labeo rohita* differed significantly from those of *Catla catla* and *Cirrhina mrigala* (Table 2). The latter two species showed similar response in all the treatments except for T4, T5 and T6. Higher concentration of sarcoplasmic proteins of *Catla catla* in all the six treatments corresponded with the comparative decrease in myofibrillar proteins in fish and correlation coefficients between sarcoplasmic and myofibrillar protein contents of all the three fish species were significantly negative (Tables 2 and 3). The correlation coefficients between stromal and myofibrillar proteins in all the three fish species were negative but significant for *Cirrhina mrigala* only. The correlation between sarcoplasmic and stromal proteins of *Catla catla* was negatively significant while that of *Cirrhina mrigala* was positive and significant (Table 3).

From these findings it is concluded that stroma increased at the expense of myofibrillar proteins, but this relationship was significant for *Cirrhina mrigala* only, whereas myofibrillar proteins also showed

significantly ($P < 0.01$) negative correlation with sarcoplasmic proteins for all the three fish species (Table 3). Javed et al. (1992) reported highly significant but negative correlation between sarcoplasmic and myofibrillar proteins of *Catla catla*, *Labeo rohita* and *Cirrhina mrigala* reared under inorganic and organic fertilization treatments. Barlas (1986) compared sarcoplasmic, myofibrillar and stromal proteins of flat fish, *Cyprinus carpio* and beef/rabbit and found carp to be the best as it contained less stroma (3.00%) than flat fish or beef which had 5 and 16-18% stroma respectively. Choi et al. (1986) reported 31.80 % sarcoplasmic and 58.37 % myofibrillar proteins in the muscle of cultured eel, whereas stroma accounted for 1.80 % of the total proteins. During present study, significantly higher percentages of stroma in all the three fish species under T3 was the result of significantly higher weight gains of fish under this treatment (Tables 1 and 2).

Table 3. Relationships among sarcoplasmic, myofibrillar and stromal proteins in three fish species

	Myofibrillar protein	Stromal protein
<i>Catla catla</i>	**	**
Sarcoplasmic proteins	-0.502	-0.603 NS
Myofibrillar proteins		-0.170
<i>Labeo rohita</i>	**	NS
Sarcoplasmic proteins	-0.486	0.091 NS
Myofibrillar proteins		-0.084
<i>Cirrhina mrigala</i>	**	**
Sarcoplasmic proteins	-0.919	0.428 **
Myofibrillar proteins		-0.574

** = Significant at $P < 0.01$; NS = Non-significant.

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