A Study of General Proteins in the Freshwater Fishes, Labeo Rohita and Cirrhina Mrigala

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***Abstract*—In the present investigation, an attempt has been made to study the relative mobility, intensity and thickness of the electrophoretic bands of general proteins of *Labeo rohita* and *Cirrhina mrigala.* The general proteins of these fishes have been separated with the help of SDS-PAGE. Molecular weights have been estimated with the help of standard proteins. All the bands have been observed between molecular weights 14,400 to 98,000 in both the species. The relative concentration of various polypeptides has been estimated by gel densitometric scanning.**

***Index Terms*—Protein, Indian major carp, densitometry, polymorphism, etc.**

I. INTRODUCTION

Biochemical analyses by various types of electrophoresis are very effective in detecting species-specific proteins and to demonstrate hybrid and polyploid specimens which otherwise are hardly distinguishable from their parental species at the level of morphological or meristic criteria (Campton, 1987). Densitometric studies employed in the present study established that general proteins of *L. rohita* have more mobility as compared to that of *C. mrigala*.

II. MATERIALS AND METHODS

Two Indian major carps *Labeo rohita* and *Cirrhina mrigala*, belonging to family Cyprinidae and order Cypriniformes, have been analyzed for general proteins (total muscle proteins). The specimens of these fishes were collected from 'National Fish Seed Farm', Jyotisar near Kurukshetra University, Kurukshetra. The fishes were brought alive to the laboratory and acclimatized in well-aerated aquaria. The extract of muscle tissue of each fish was prepared in 0.125 M Tris-HCl buffer (pH 6.8) containing 2% sodium dodecyl sulphate (SDS) and stored in eppendorf tubes at –10°C.

14 % SDS-PAGE (sodium dodecylsulphate-polyacrylamide gel electrophoresis), using discontinuous system of Davies (1964) and Ornstein (1964), was used for the analysis of tissue extract. Electrophoresis was carried out under non-dissociating conditions using formulations of Laemmlie (1970). The gels were stained in Commassie Brilliant Blue (0.05 %), dissolved in a mixture of solvents containing methanol, acetic acid and distilled water in the ratio of 50: 7: 48 (v/v). The gels were destained (in case of over staining) in the above solvents lacking in Commassie Brilliant Blue. Molecular weights of polypeptides separated on SDS gels were determined following the method given by Laemmlie (1970). Polypeptides constituting various proteins and separated as band by SDS-PAGE were scanned at 580nm using Pharmacia-1 KB ultra scan spectrophotometer.

III. RESULTS

The polypeptide patterns of protein fraction of *L. rohita* and *C. mrigala* on SDS-PAGE can be seen in Fig 1. The molecular weights have been estimated from the calibration curve. The following are the results:

A large number of polypeptides in the range of 14,400 to 84,000 daltons could be seen in the protein fraction. In case of *L. rohita*, the polypeptides of molecular weights 14,400, 16,900, 20,000 and 23,000 are relatively darkly stained than the other polypeptides while in *C. mrigala*, the polypeptides of molecular weights 14,400, 20,000 and 23,000 are

relatively darkly stained than the other polypeptides The relative mobility and densitometric measurements showing relative concentration of polypeptides and molecular weights have been presented in table 1 for *L. rohita* and in table 2 for *C. mrigala*. Gel densitometric scanning profile of *L. rohita* can be seen as dark line while that of *C. mrigala* as light line in the Fig. 2.

IV. DISSCUSSION

In the present analysis, rohu (*L. rohita*) and mrigal (*C. mrigala*) exhibited species-specific differences in relative mobility, intensity and thickness of bands relating to general proteins. All the protein bands of rohu and mrigal were observed within the range of molecular weight of 14,400 to 98,000 daltons. Similar results were earlier obtained by Ponniah and Sahu (1990). Ponniah and Sahu (1990) have also reported lack of variations among the carp samples. Salmonids have also been reported to be monomorphic (Altukhov et al., 1969). According to Rab et al. (1994), the monomorphic nature of fish proteins may be used successfully for studies on fish taxonomy. In many other species, muscle protein, haemoglobins and transferrins are reported to be monomorphic and species-specific and, therefore, can be used for evolution studies (Huntsman, 1970).

Proteins of skeletal muscle are polymorphic in a number of other fish species eg. trouts, common sucker, hake, whiting, cod, trout, tunas and hagfishes etc. (Sick, 1965; Tsuyuki, 1966; Grag & Mckenzie, 1970; Utter & Hodgins, 1971; Allendorf et al., 1977; Huriaux et al., 1992; Lee et al., 1993; Sezaki et al., 1994).

Two polymorphic protein system, serum transferring and serum esterase, were studied in population of *Ictiobus cyprinellus* by Koehn & Johnson (1967). There were no genetic differences between population samples, which may result from selective pressures that maintain homogeneity at the transferring and esterase loci. Proteins of crystalline lens, much like serum proteins and muscle proteins proved to be subjected to considerable individual variability (Eckroat et al., 1969). On the basis of comparative analysis of protein profiles, hybrids of interspecific and intergeneric nature as well as polyploid forms can be easily identified. Densitometric studies employed in the present study established that general protein of *Labeo rohita* have more mobility as compared to that of *Cirrhina mrigala*. Polyploid individuals can be easily distinguished from diploids on the basis of comparative densitometeric analysis.

V. ACKNOWLEDGEMENTS

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LEGENDS

Fig. 1 General proteins of *C. mrigala* and *L. rohita.*

Fig. 2 Comparative densitometeric profile of *C. mrigala* and *L. rohita.*

Table 1 showing relative mobilities, relative concentrations and approximate molecular weights of various polypeptides from muscle extracts of *L. rohita*.

Table 2 showing relative mobilities, relative concentrations and approximate molecular weights of various polypeptides from muscle extracts of *C. mrigala*.

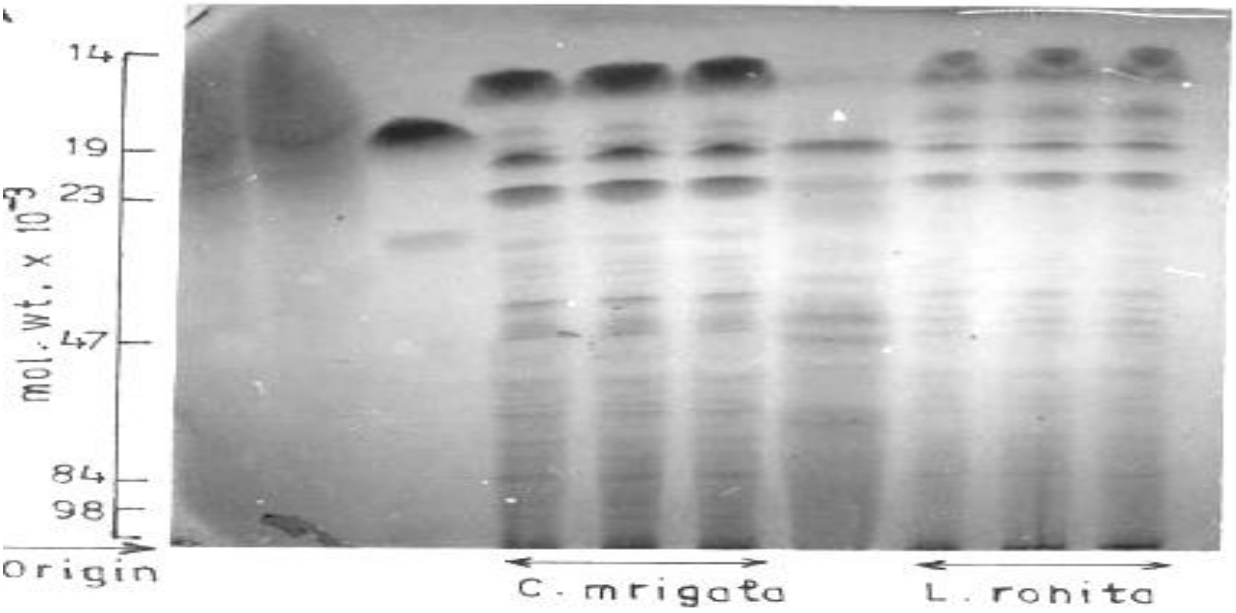

Fig. 1 General proteins of *C. mrigala* and *L. rohita.*

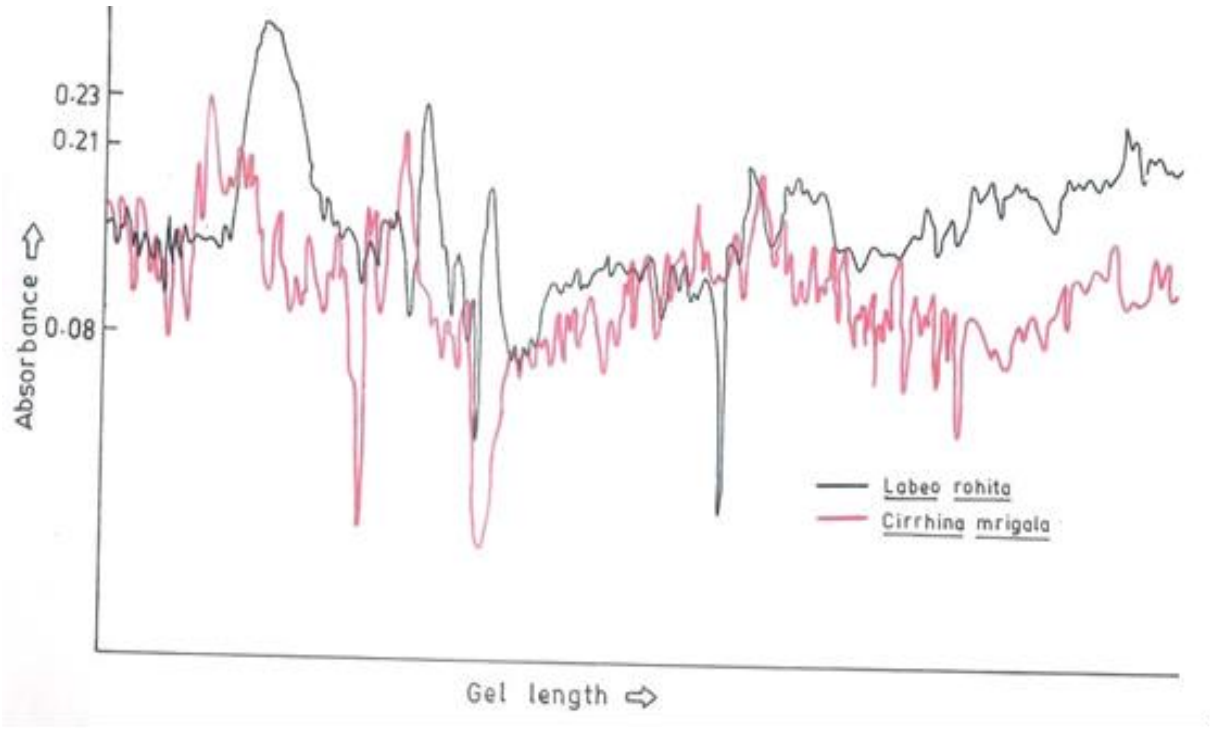

Fig. 2 Comparative densitometeric profile of *C. mrigala* and *L. rohita.*

Table No. 1

Relative mobilities, relative concentrations and approximate molecular weights of various polypeptides from muscle extracts of *Labeo rohita.*		
Relative mobility	Relative concentration	Molecular weight
.86	2.0	14,400
.81	7.8	16,900
.76	10.3	18,500
.73	8.7	20,000
.67	5.2	23,000
.61	2.5	27,000
.53	2.3	32,000
.48	7.7	36,000
.45	3.0	39,000
.42	3.3	42,000
.41	3.8	45,000
.31	3.0	54,000
.27	2.4	60,000
.24	2.6	64,000
.14	3.2	70,000
.13	2.3	74,000
	3.5	80,000
	3.0	84,000

Table 1 showing relative mobilities, relative concentrations and approximate molecular weights of various polypeptides from muscle extracts of *L. rohita*.

Table No. 2

Relative mobilities, relative concentrations and approximate molecular weights of various polypeptides from muscle extracts of *Cirrhina mrigala.*		
Relative mobility	Relative concentration	Molecular weight
.86	7.6	14,400
.81	3.9	16,900
.76	3.9	18,500
.73	3.2	20,000
.67	6.7	23,000
.62	3.2	26,500
.57	4.8	29,000
.56	3.0	30,000
.54	2.7	31,000
.52	2.1	33,000
.51	3.0	34,000
.44	2.0	40,000
.42	6.6	42,000
.37	1.9	47,000
.32	1.9	53,000
.31	2.0	54,000
.28	2.5	59,000
.26	2.7	61,500
.20	2.1	70,000
.18	3.5	74,000
.15	5.3	78,000
.14	4.2	80,000
.07	3.9	94,000
.05	3.0	98,000

Table 2 showing relative mobilities, relative concentrations and approximate molecular weights of various polypeptides from muscle extracts of *C. mrigala*.