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Tissue esterase patterns of muscle and brain of channiformes and perciformes fishes

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Tissue and species specific distribution of esterases were studied in eight fishes of two tissue were *viz.*, brain and muscle of channiformes and perciformes. Variation was noticed in muscle and brain. Muscle exhibited fast moving zones in both channiformes and perciformes while in brain exhibited slow moving zones and ArE esterase were dominant in perciformes fishes. ArE esterases were absent in channiformes fishes. Esdp esterases were noticed in muscle of channiformes fishes.

Key words : Electrophoresis, Esterases, Brain, Muscle, Perciformes, Channiformes fishes

INTRODUCTION

Heterogencity and multiplicity of esterases are probably more extensive and diverse than that observed for any other enzymes. A single tissue in some cases exhibits the presence of 30 or more heteromorphy. One important characteristic of these enzymes is that the homologous tissue of different species exhibit wide divergence in the extent of their heterogenecity and the characteristics of multiple enzyme forms even in closely related species. (Master and Holmes, 1975). Electrophoretic patterns of esterase's are increasingly used in identifying the species from microbial fauna to plant and animal species. The tissue specific proteins and enzymes in recognizing species and establishing their taxonomic relationship in number of animal group (Whitt, 1987). Inter specific patterns in several organisms within these compare studies on tissue esterase's patterns were confined only to compare the tissue specific and species specific differences existing in different animal group. (Master and Holmes, 1974). Besides, the enzymes are used in recognizing the stages of differentiation of specific tissue during the development of the organism (Holmes and Whitt, 1970; Lakshmipathi and Sujatha, 1991). In this report eight fishes belonging to two different orders (Jayram, 1981) are observed for their esterase patterns and comparison were made between channiformes and perciformes fishes.

MATERIALS AND METHODS

Adult fishes were caught from fresh water tanks (ponds) located with in a radius of 25 km. from the Laboratory. They were immediately brought to the laboratory contains water in plastic buckets and acclimatized to laboratory condition for about a week in aquaria. They were fed in natural habitats. Fishes were immobilized by hitting them on the head and the tissue were dissected out from the animals, four tissues were selected for the study. They were muscle and brain. The tissue extracts were centrifuged at 2000 rpm for 10 minutes. The supernatants were mixed with equal volumes of 20% sucrose solutions containing 0.05% bromophenol blue as the tracking dye in aliquot of 0.1 ml of this mixture was used for loading the sample for separation of esterase patterns. Esterase patterns were separated on thin layer (1.5 mm thick) polyacrylamide gel. The gel mixture was prepared according to the procedure of clarke (1974). Gelling was allowed for 45 minutes and sample was loaded directly on to the gel, constant current of 20 mA for the first 15 minutes followed by 40 mA for the test of run. After terminating the current supply, the plate were removed from the chamber. The gel was removed from the glass moulds and the stain solution was poured directly on the gel so as to immerse it completely, the patterns of esterases appeared within 10 minutes. Type of esterase's were identified by using the inhibitors like paraxon esterine, and physostgmine. Esterases were classified in according with the procedure of (Holmes and Masters 1976). Hart and Cook (1976) on the basis of their sensitivity of specific inhibitors, phyostigimine (corbomate) PcMB (thiol active compound) and paraxon (op compounds) were used for

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classification of esterases.

RESULTS AND DISCUSSION

The results obtained from the present investigation are summarized below:

Muscle:

Muscle consisted of seven zones with Rm value .92, .89, .80, .75, .73, .70 and .65. Among these, the zone with Rm value .92 was present in *Chana punctatus, Oreochromis mossambicus, Glossogobius giuris* and *Colisa fasciata* with CHsP, ER, CE and ER esterases, respectively. The zone with Rm value .89 was present only in perciformes fishes of *Oreochromis mossambicus* and *Colisa fasciata* with ER esterases. The zone with Rm value .80 was present in only *Glossogobius giuris* with ER esterases. The zone with Rm value .75 exhibited in *Etroplus maculates* and *colisa fasciata* with CE and ChE esterases, respectively. The zone with Rm value .73 was present in *Channa punctatus* and *Glossogobius giuris* with Esdp and CE esterases, respectively. In *Glossogobius giuris* this zone exhibited higher activity (+++) but in *Channa punctatus* exhibited low activity

Table 1: Esterase patterns of muscle of channiformes 1-4 and Perciformes fishes (5-8)

Sr. No.	Fish species \ Rm value	1 .92	2 .89	3 .80	4 .75	5 .73	6 .70	7 .65
1.	*Channa marulius*							+ Esdp
2.	*Channa orientalis*							+ CE
3.	*Channa punctatus*	+ CHsp				+ Esdp		
4.	*Channa striatus*							++ CE
5.	*Etroplus maculates*				++ CE		++ ER	
6.	*Oreochromis mossambicus*	+ ER	+ ER					
7.	*Glossogobius giuris*	++ CE		+ ER		+++ CE		
8.	*Colisa fasciata*	++ ER	++ ER		++ ChE			

Table 2: Esterase patterns of brain of channiformes 1-4 and perciformes fishes (5-8)

Sr. No.	Fish species \ Rm value	1 .58	2 .51	3 .45	4 .40	5 .35	6 .33	7 .27	8 .8
1.	*Channa marulius*	± CHsp	± CE			+++ CE		+ CHsp	+ CHsp
2.	*Channa orientalis*					+++ CE		+ ER	+ CHsp
3.	*Channa punctatus*					++ CHsp	+ ER		
4.	*Channa striatus*					+ ChE		+ CE	+ CHsp
5.	*Etroplus maculates*			+++ ER		+ ChE		++ ChE	
6.	*Oreochromis mossambicus*			+++ ER				++ ChE	
7.	*Glossogobius giuris*				+ Esdp	+ ArE			
8.	*Colisa fasciata*			++ ER				+++ ER	

Rm = Relative mobility; CE = Carboxyl Esterase; ER = Esterase Resistant to inhibitors; ChE = Choline Esterase; CHsp = Choline esterases like enzymes; ArE = Aryl esterases; Esdp = Esterase sensitive to organo phosphates and pCMB. +++ = High activity; ++ = Moderate activity; + = Low activity; ± = Very low activity

Table 3: Inhibitor sensitivity of individual esterase zones in muscle of eight fishes (1-4 Channiformes and 5-8 Perciformes)

Name of species	*Channa marulius*	*Channa orientalis*	*Channa punctatus*		*Channa striatus*	*Etroplus maculatus*		*O. mossambicus*		*Glossogobius giuris*			*Colisa fasciata*		
Rm values	.65	.65	.92	.73	.65	.75	.70	.92	.89	.92	.80	.73	.92	.89	.75
Activity	+	+	+	+	++	++	++	+	+	++	+	+++	++	++	++
pCMB	-	+	+	-	+	+	+	+	+	+	+	+	+	+	-
Eserine	+	+	-	+	+	+	+	+	+	+	+	+	+	+	-
Paraoxon	-	-	-	-	-	-	+	+	+	-	+	-	+	+	-
Classification	Esdp	CE	CHsp	Esdp	CE	CE	ER	ER	ER	CE	ER	CE	ER	ER	ChE

(+). The zone with Rm value .70 exhibited in only *Etroplus maculates*. It was classified as ER esterases. The zone with Rm value .65 exhibited Esdp esterase in *Channa punctatus* while in *Channa orientalis* and *Channa striatus* exhibited CE esterases.

Brain:

Brain exhibited eight zones on zymogram in 8 fishes, with Rm value .58, .51, .45, .40, .35, .33, .27 and .8. Among these, the zone with Rm value .58 and .51 were present in only *Channa marulius*. It was classified as CHsp and CE esterases, respectively. The zone with Rm value .45 was present in only perciformes fishes in *Etroplus maculates, Oreochromis mossambicus* and *Colisa fasciata*. It was classified as ER esterases in all three perciformes fishes with higher (+++) activity. The zone with Rm value .40 was present in *Glossogobius giuris* with Esdp esterase.

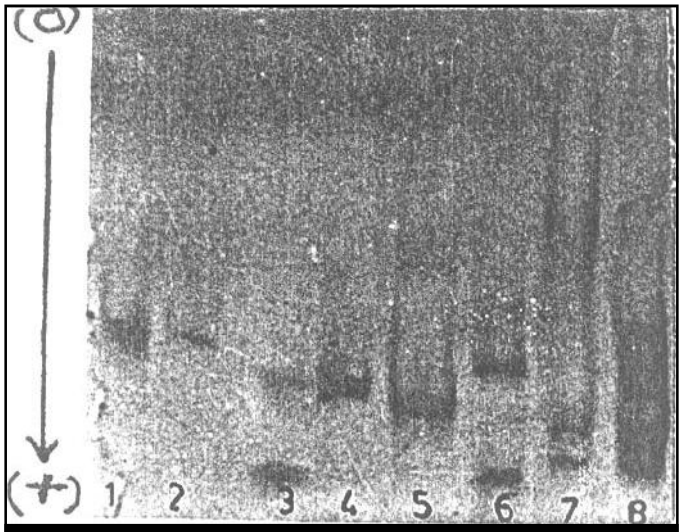

Fig. 1: Esterase patterns of muscles of Channiformes 1-4 and Perciformes 5-8
Channa marulius, Channa orientalis, Channa punctatus, Channa striatus (Channiformes); Etroplus maculatus, Oreochromis mossambicus, Glossogobius giuris, Colisa fasciata (Perciformes)

The zone with Rm value .35 was present in all the 6 fishes except *Colisa fasciata* and *Oreochromis mossambicus*. In *Channa marulius* and *Channa orientalis* this zone exhibited CE esterases. In *Channa striatus* and *Etroplus maculates* exhibited ChE esterases. While *Glossogobius giuris* exhibited ArE esterases and in *Channa punctatus* CHsp esterase was noticed. The zone with Rm value .33 was present only *Channa punctatus* with ER esterases. In remaining fishes this zone was not noticed. The zone with Rm value .27 was noticed in *Channa marulius* as CHsp, in *Channa orientalis* and *Colisa fasciata* as ER esterases. While in *Etroplus maculates* and *Oreochromis mossambicus* with ChE esterases, but in *Channa striatus* CE esterases were noticed. The zone with Rm value .8 was present in *Channa orientalis*, *Channa striatus* and *Channa orientalis* with CHsp esterases.

The tissue and species specific distribution of esterases were earlier reported from two cat fishes and the toad (Venkaiah and Lakshmipathi, 2006). The surface mucous of fishes were used as tools in establishing the genetic relatedness among the closely related species (Wu and Wu, 1983). Esterase of skin secretion of cat fishes was shown to be higher in their activity when compared to the extract of poison glands (Al., Hussain *et al.*, 1987), retinal specific esterases in adults (Ahuja *et al.*, 1977) as well as in embryos of fishes (Hart and Cook, 1977), retinals specific esterases in thirteen fresh water fish species (Rajaiah and Lakshmipathi, 1997). Effect of parathion in esterase patterns of channa punctatus was studied. (Rajaiah and Venkaiah, 2007). In our studies, among the eight fishes in two tissues, the patterns of esterases observed in the eight species exhibited highly tissue specific and species specific patterns. The tissue muscle exhibited seven zones in all the fishes with fast moving zones in both channiformes and perciformes fishes. *Glossogobius giuries* and *Colisa fasciata* exhibited maximum number of zones

Table 4: Inhibitor sensitivity of individual esterase zones in brain of eight fishes (1-4 Channiformes and 5-8 Perciformes)

Name of species	*Channa marulius*				*Channa orientalis*				*Channa punctatus*				*Channa striatus*				*Etroplus maculatus*				*O. mossambicus*		*Glossogobius giuris*		*Colisa fasciata*	
Rm values	.58	.51	.35	.27	.8	.35	.27	.8	.35	.33	.35	.27	.8	.45	.35	.27	.45	.27	.40	.35	.45	.27				
Activity	+	+	+++	+	+	+++	+	+	++	+	+	+	+	+++	+	++	++–	++	+	+	+++	+++				
pCMB	-	+	+	-	-	+	+	-	-	+	+	+	-	+	+	+	+	+	-	+	+	+				
Eserine	-	+	+	-	-	+	+	-	-	+	-	-	-	+	-	-	+	-	+	-	+	+				
Paraoxon	-	-	-	-	-	-	+	-	-	+	-	-	-	+	-	-	+	-	-	+	+	+				
Classification	CHsp	CE	CE	CHsp	CHsp	CE	ER	CHsp	CHsp	ER	ChE	CE	CHsp	ER	ChE	ChE	ER	ChE	Esdp	ArE	ER	ER				

Rm = Relative mobility; CE = Carboxyl Estcrasc; ER = Estcrasc Resistant to inhibitors; ChE = Choline Estcrasc;
CHsp = Choline esterases like enzymes; ArE = Aryl esterases; Esdp = Esterase sensitive to organo phosphates and pCMB.
+++ = High activity; ++ = Moderate activity; + = Low activity; ± = Very low activity

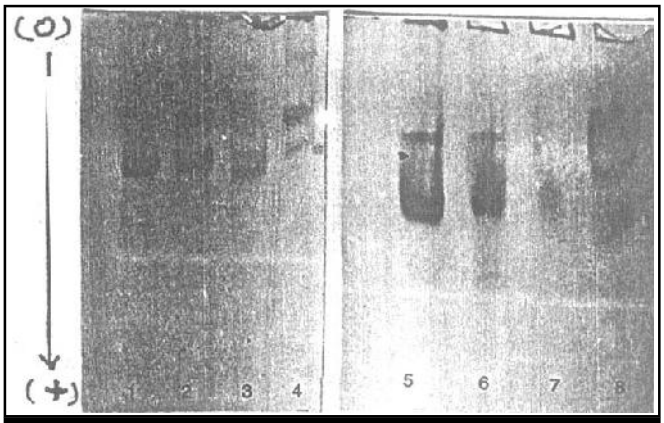

Fig. 2: Esterase patterns of brain of Channiformes 1-4 and Perciformes 5-8
Channa marulius, Channa orientalis, Channa punctatus, Channa striatus (Channiformes); Etroplus maculatus, Oreochromis mossambicus, Glossogobius giuris, Colisa fasciata (Perciformes)

(3). While remaining fishes exhibited two and single zones. Among the type of esterases observed, ER esterases were the principal contributors to esterase activity in different tissues of perciformes fishes. Total absence of ER esterase in channiformes fishes keeps these species distinct from other groups. In addition to this, Esdp esterases were noticed in channiformes fishes but not noticed in perciformes fishes indicated the channiformes fishes distinct from perciformes fishes. Brain contain eight slow moving zones in all the eight fishes. Among eight fishes, *Channa marulius* exhibited maximum number of zones in the Zymogram. Remaining all the fishes contained three and two zones. Distribution of different kinds of esterases indicates that CE esterases were present in all the channiformes fishes. But CE esterase were not noticed in perciformes fishes in brain tissue. ArE esterases were found in all the perciformes fishes but ArE esterases were not noticed in channiformes fishes indicated that channiformes order is distinct from perciformes order fishes.

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