



Research Paper

PLASMA CORTISOL LEVELS IN TWO TYPES OF FRESH WATER CARP FISHES, *Labeo rohita* AND *Cirrhanus mrigala* COLLECTED IN WILD

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Abstract

The hormone cortisol secreted from the adrenocortical cells of the head kidney is a principle corticosteroid in a teleost fish and plays pivotal role in the teleost stress response. Plasma cortisol in two types of fresh water carp fishes cultured in an aquatic body and collected in wild was measured by applying Electro-Chemiluminiscence Immunoassay Analyser (ECLIA) in a small volume of plasma to assess the stress response. The mean values obtained for plasma cortisol was found to be 4.75 µg/ml (4750 ng/ml) for *Labeo rohita* and 5.75µg/ml (5750 ng/ml) for the fish, *Cirrhanus mrigala*. The values were compared with cortisol level in other fish species. The results indicated that the higher level of cortisol in these two fishes compared with other reported fishes may be because of stress condition. The type of rough method of capturing, rough handling and transportation under hypoxic condition may be the reason for increase in cortisol level, suggesting a kind of stress response.

Key words: *Labeo rohita*, *Cirrhanus mrigala*, Cortisol hormone.

INTRODUCTION

The stress response in vertebrates involves two major classes of hormones such as corticosteroids and catecholamines. In fish, the corticosteroids have been reasonably well studied in variety of fishes and cortisol is the principle corticosteroid in teleost fishes, its plasma concentration rise dramatically during stress (Mommsen *et.al.*, 1999). The cortisol hormone released into the circulation in fish from adrenocortical cells of the head kidney. The steroid hormone cortisol plays a pivotal role in the teleost stress response but relatively few species have been studied in depth with respect to the levels of cortisol in the blood under different stages of development and environmental conditions (Pottinger *et al.*, 2000). One of the most commonly measured indicators of stress in fish is the concentration of the major circulating corticosteroid, cortisol and this hormone can be measured easily and accurately using commercially available radioimmunoassay technique (Gamperl *et al.*, 1994). In addition, the level of cortisol in the blood of both unstressed and stressed fish is subject to modulation by a number of internal and external factors (Pottinger, 1998). However, the changes in stress hormone particularly cortisol levels in response to physical and environmental stress conditions especially amongst the Indian fishes are very less studied. Hence, in the present investigation plasma cortisol level was investigated in two types of fresh water carps,

Labeo rohita and *Cirrhanus mrigala* to assess whether these fishes are under stress condition because of rough handling, capturing and their transportation from an aquatic body. The cortisol levels of these two fishes were also compared with other fishes harboured in similar conditions.

MATERIAL METHODS:

The live fresh water carp fishes of *Labeo rohita* and *Cirrhanus mrigala* which are cultured in an aquatic body called Khaja kotnoor reservoir around 25 km. away from Kalaburagi were captured by cast net by fisherman. The fishes were transported in a vehicle to the market area in live condition. The blood sample was collected immediately after the fishes landed from the vehicle. The size of the fish varied from 26.5 ± 1 cm in length and 105 ± 10 gm in weight. All sexes were used without discrimination. The fish were then placed belly upwards and blood samples obtained from the caudal circulation with the aid of a heparinized 2 cm disposable plastic syringe and a 21 gauge disposable hypodermic needle. The use of plastic syringe is a necessary precaution with fish blood because contact with glass results in decreased coagulation time. The site chosen for puncture (about 3 to 4 cm from the genital opening) was wiped dry with tissue paper to avoid contamination with mucus. The needle was inserted at right angle to the vertebral column of the fish and was gently aspirated during penetration. It was then pushed gently down until blood started to enter as the needle punctured caudal blood vessel. Blood was taken under gentle aspiration until about 2 ml has been obtained. Thereafter the needle was withdrawn and the blood gently transferred into plastic containers. Blood plasma was obtained by centrifugation and then used for the determination of cortisol. All the determinations were carried out in duplicates for each sample. The plasma was separated and further processed for the estimation of cortisol by applying Electro-Chemiluminescence Immunoassay Analyser (ECLIA) in a small volume of plasma with assistance in Am Path central Reference Laboratory, Hyderabad.

OBSERVATION:

Plasma cortisol level was measured in two types of fresh water carp fishes cultured in an aquatic body, captured and collected in wild. The fishes were captured by cast net by local fisherman and transported in a vehicle to the market area in live condition with minimum care. The steroid hormone cortisol plays a pivotal role in the teleost stress response but relatively few species have been studied in depth with respect to the levels of cortisol in the blood under different natural and environmental stress conditions. The plasma cortisol was measured by applying Electro-Chemiluminescence Immunoassay Analyser (ECLIA) in a small volume of plasma to assess the stress condition and response. The mean values obtained for plasma cortisol is presented in the table-1 and fig.1 and the level was found to be 4.75 µg/ml (4750 ng/ml) for *Labeo rohita* and 5.75 µg/ml (5750 ng/ml) for the fish, *Cirrhanus mrigala*. The fish, *Cirrhanus mrigala* has higher level of plasma cortisol than the fish, *Labeo rohita*. However, in both the fishes the values were found to be higher as compared with cortisol levels in other fish species. The results indicated that the two fresh water fishes found to be under stress condition as the levels were higher comparing with other reported fishes and this may be attributed to the rough method of capturing, handling stress and their improper transportation under hypoxic condition.

Table-1: Showing plasma cortisol in two fresh water carps collected in wild from a aquatic body.

Sl.No	Name of the fish species	Plasma cortisol
1	<i>Labeo rohita</i>	4.75 µg/ml/ 4750 ng/ml
2	<i>Cirrhonus mrigala</i>	5.75 µg/ml/ 5750ng/ ml

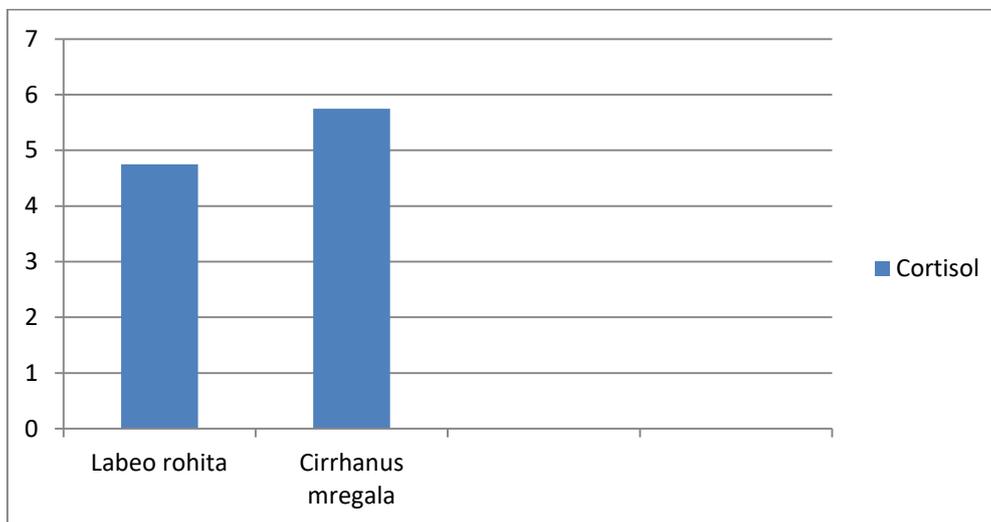


Fig.1: Showing plasma cortisol in two fresh water carps collected in wild from a aquatic body.

DISCUSSION:

In teleost fishes, the adrenal consisting of steroidogenic and chromaffin cells called inter-renal cells are distributed in the head kidney, mostly along the posterior cardinal veins and their branches, the steroidogenic cells lie in close proximity to the chromaffin cells (Kulkarni and Sathyanesan, 1982). The inter-renal (adrenocortical) cells are the source for cortisol hormone in fish and is known to be involved in the general homeostasis of energy as well as acute and chronic response to stress (Mommsen *et al.*,1999). Plasma levels of steroids reflect the serum of synthesis, secretion, metabolism and clearance (Pankhurst and Van Der Kraak,2000). If severe enough, periods of acute stress in animals may be associated with the release of catecholamine hormones (noradrenaline and adrenaline) into the circulation; a response termed the acute humoral adrenergic stress response and release of catecholamines from chromaffin cells is under neuroendocrine control (Perry and Capaldo, 2011). In teleost fishes plasma and head kidney has been shown to contain with considerable variation in the concentration of hormones, the concentration is very low in fish compared to with other vertebrates (Matty, 1985). Plasma concentration of cortisol /or cortisone have been reported in numerous teleosts and although show considerable variation, their general range of 100 – 400 ng/ml is comparable to that found in mammals. The plasma cortisol level has been estimated in variety of fishes such as rainbow trout, *Oncorhynchus mykiss*, pacific salmon, *oncorhynchus spp.*, brown *Salmo trutta* and Atlantic salmon, *Slmo solar* (Gamperl *et al.*,1994, Mommsen *et al.*,1999). The cortisol level estimated in cyprinids have been compared with the level found in salmonid species and found that the level is higher in cyprinids. In the present study as the two fishes belong to the cyprinids the level found to be at higher level. The cortisol levels in unstressed common carp (*Cyprinus carpio*) has been reported to be -50 – 150

mg ml⁻¹ rising to 300- 450mg ml⁻¹ during exposure to stressors (Dabrowska *et al.*, 1991, Pottinger, 1998). In the gold fish, *Carassius auratus* the cortisol level was found to be between 5 and 300mg ml⁻¹ (Spielar and Noeske, 1984) between 7 and 110mg ml⁻¹ (Paxtan *et al.*, 1984) and between 25 and 50 mg ml⁻¹ (Singley and Chavin, 1975). In the fresh water fish, *Notopterus notopterus* in our earlier studies (Shankar,2001), the cortisol level found to be ranged between 15µg/dL for male fish and 5µg/dL for female fish in the blood collected during morning hours. In the present study the cortisol level in two fresh water carps found to be 4.75µg/ml (4750 ng/ml) for the fish, *Labeo rohita* and 5.75µg/ml (575 ng/ml) for the fish, *Cirrhinus mrigala* such variation was found with different species of fish. Intra and inter species differences in blood cortisol levels have been reported for salmonid species. Pottinger *et al.*(2000) have reported that in contrast to the relating minor intra and inter species differences in blood cortisol levels reported for salmonid species, those in a more pronounced distinction between cortisol levels among the salmonidae and the cyprinidae, with basal and stress induced cortisol levels markedly higher in the latter. In the present study also it was found that in both *Labeo rohita* and *Cirrhinus mrigala* cortisol found to be higher in these two types of carp fishes. The plasma cortisol in *Notopterus notopterus* was measured by RIA detection and it was found that the values for plasma cortisol were 5-15µg/ml.(Shankar, 2001). Seema Langer (2014) studied the effect of starvation on the cortisol level of fresh water fish, *Garra gotyla gotyla* and found that after 5th week, the cortisol level increased from 115.0 ng/ml to 210.5 ng/ml and remained higher than control even after 9 weeks. The author has suggested that food deprivation influence the cortisol level. The cortisol measured in the present study is by applying ECLIA method and the values were higher possibly, these fishes were under stress condition may be because of rough capture, handling and transportation method. There are some reports on the increased level of catecholamines subjected to hypoxia (low level oxygen availability) and also under experimental conditions of inducing fish to some exercises (Steve *et.al.*, 1991). It is possible, that in the present study increase in the level of plasma cortisol in both the fishes may be because of problem with capture method, handling stress and also transportation of the fish under hypoxic condition.

CONCLUSIONS:

The plasma cortisol level was measured in two species of carp fish, *Labeo rohita* and *Cirrhinus mrigala* collected in wild. The results indicated that the cortisol was at higher level in comparison to other fishes reported. Thus, indicating that both these fishes are under stressful conditions and this could be because of the method of capture, handling stress and transportation of fish under hypoxic condition.

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