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Research Article

EFFECT OF PINEALECTOMY ON PLASMA $Na^{+,}$ K^{+} AND Ca^{+2} IN CATFISH *CLARIAS BATRACHUS* UNDER DIFFERENT SALINITY LEVELS

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ABSTRACT
For study Effect of pinealectomy on plasma Na^{+}, K^{+} and Ca^{+2} levels catfish, *C. batrachus* were allow to face three different salinity levels (0ppt, 5ppt and 7ppt). The observations of present study indicated that in pinealectomized catfish the plasma Na^{+} and K^{+} levels were increased with increased in salinity levels. No significance difference ($p > 0.05$) was observed in the plasma calcium of the pinealectomized as well as control fish under different salinity levels. This study indicates that pineal gland of catfish play an important role in the osmoregulation.

Keywords: *Pinealectomy, Plasma, Osmoregulation, Salinity*
Abbreviation: *ppt- parts per thousand*

INTRODUCTION
Information on the osmoregulatory physiology of the teleost fishes is based on euryhaline species which can regulate the osmoionic balance of their body fluids within narrow limits even when subjected to a sudden or the radical changes in envionmental salinity (Johnson 1973; Maetz 1974; Evans, 1980). The cat fish *C. batrachus* has a limited range of salinity tolerance, from fresh water to about 15% salt water. Much information on the influence of the pineal on plasma electrolytes levels is not available. Delahunty *et al., (*1977) studied that Pinealectomy affect the serum level of the cortisol in goldfish. According to de Vlaming *et al.,* (1979), pinealectomy affects the electrolyte balance in goldfish. The studies of Garg and Sundararaj (1986) suggest that pinealectomy altered plasma electrolyte balance in the Indian catfish, *Heteropeustes fossilis*. The present investigations were deals with the effects of pinealectomy on plasma Na^{+}, K^{+} and Ca^{+2} levels different salinity (ppt) of catfish *C. batrachus.*

MATERIALS AND METHODS
The catfish, *Clarias batrachus* (mean body weight (BW) 70-80g) were obtained from a commercial supplier. Fishes were then maintained at the laboratory in glass aquaria under 12L:12D (12 hours light and 12 hours darkness) photoperiod and 25 ± 2°C for 2 weeks.
To carry out the study three salinity levels were made i.e. 0ppt, 5ppt and 7ppt. Animals were then divided into two groups. Group-I assigned control group and group-II assigned as group of pinealectominzed fish. Catfish of both of groups were placed at different salinity levels. The fishes during the course of the experiment were fed 5% of their body weight with formulated feed (having 40% protein). The experiment lasted for 90 days.

Pinealectomy
Pinealectomy (Px) of catfish was performed following the procedure of Ghosh and Nath (2005). The individual catfish was wrapped with a napkin and a sharp incision was made on the skin covering the pineal fontanelle on the head to form a 'v' shaped flap which was then folded posteriorly to expose the pineal area. The membranous connective tissue covering the region was cut along the edge. The pineal vesicle along a major portion of the pineal stalk along with the connective tissue of *C. batrachus* was extirpated by gently pulling out by means of a pair of fine forceps. The complete process was carried out under the binocular microscope. After the surgery, the fontanelle was cleaned with 70% alcohol and the skin flap was gently pressed back to its position. The operated fish was then returned to aquarium water

which was maintained at 25 °C. The complete surgery was performed within three to five minutes. The wound healed within one week. In intact control, fishes were placed as such without performing any surgery.

Sampling Procedure: For Plasma, sodium and potassium blood was collected in heparinized 3-ml tubes and then plasma was separated by centrifuging blood at 4000 rpm/10 min. For potassium 50µl of plasma samples were taken and diluted to one hundred times while for sodium 50µl plasma samples were diluted to one thousand times. Plasma sodium and potassium level was determined by using flame photometer. Potassium chloride and sodium chloride were used as standards to obtain standard curve. Calcium was determined by atomic absorption methods. The statistical analysis of the data was performed using ANOVA followed by t-test. Significance was taken at $P<0.05$. Values are presented as means ± S.E.

RESULTS AND DISCUSSION

No significance difference ($p> 0.05$) was observed in the electrolytes of the pinealectomized and the intact control fish. But the values for the plasma sodium and the plasma potassium were high in the pinealectomized fish than the intact control fish under different levels of salinity levels.

Plasma sodium (Na^+)

Plasma sodium in both groups (control as well as pinealectominzed) was increase as salinity level increased. But the value remained high in pinealectomized fish at different salinity levels (Table-1).

Table 1: Effect of pinealectomy on plasma Na^+ of catfish *C. batrachus* at different salinity levels

Parameter	Salinity levels (ppt)	Groups	
		Control	Pinealectomized
Plasma Na^+ (meq/l)	0	138.9±3.1	153.58±6.08
	5	150.71±2.9	168.41±3.48
	7	159.0±2.3	179.09±3.12

All the values are means ± S.E. of mean

Plasma potassium (K^+)

There were slight increased were observed in values of plasma potassium under both groups. But plasma potassium was recorded comparatively high in pinealectominzed fishes (Table-2).

Table 2: Effect of pinealectomy on plasma K^+ of catfish *C. batrachus* at different salinity levels

Parameter	Salinity levels (ppt)	Groups	
		Control	Pinealectomized
Plasma K^+ (meq/l)	0	4.82±0.51	8.01±1.12
	5	5.1±0.50	9.21±0.98
	7	5.9±0.30	10.98±1.16

All the values are means ± S.E. of mean

Plasma calcium (Ca^{+2})

The plasma calcium remained nearly the same in both group fishes. Salinity as well as pinealectomy does not affect this parameter in catfish (Table-3).

Table 3: Effect of pinealectomy on plasma Ca^{+2} level of catfish *C. batrachus* at different salinity levels

Parameter	Salinity levels (ppt)	Groups	
		Control	Pinealectomized
Plasma Ca^{+2} ($mmol^{-1}$)	0	1.95±0.02	1.99±0.03
	5	2.06±0.06	2.06±0.06
	7	2.09±0.05	2.10±0.01

All the values are means ± S.E. of mean

In present study pinealectomy in catfish elevated the plasma electrolytes Na^+, K^+ and Ca^{+2} as salinity levels were increased. The plasma electrolyte concentrations in the upper two salt concentrations (5ppt and 7ppt) were elevated constantly above the normal values in the fresh water catfish. Our results are in agreement with Ellory *et al.,* (1971) where when goldfish were exposed to the hypertonic salt solutions the concentrations of the plasma electrolytes rise and become equal to or slightly higher than the external concentration of the plasma electrolytes.

Therefore, the tolerance of the brackish water in catfish, and perhaps in the goldfish, probably reflect the ability of these fish to withstand an increased osmotic pressure of the internal fluids. Fenwick (1970) suggested that pinealectomy neither caused change in the potassium and sodium levels nor altered the osmotic concentration. The data reported here for sodium and the potassium differ from those of goldfish (de Vlaming *et al.*, 1979) where after pinealectomy the serum potassium levels were lowered while the sodium levels were not affected. How the pineal influence plasma electrolyte rhythms in the catfish do is yet not known. However, it is presumed that the pineal hormones may be acting as chronomodulators for the various circadian parameters. The effects of pineal on various physiological and the metabolic parameters in the catfish may also be mediated via the endocrine pathway or possibly the pineal mat acting as a transducer which participates in the mediation of the photoperiod effect on these functions (de Vlaming and Olcese, 1981). The data presented here show that in catfish the pineal participates in the control of the locomotory rhythms and some aspects of the osmoregulation. However, further work is needed to elucidate the role of pineal in the regulation of circadian locomotors activities and the metabolism in the different seasons of the year.

Conclusion

The findings clearly indicates that pineal of catfish is involved in the regulation some aspects of osmoregulation. The observations show that the pinealectomy increased the plasma Na^+ and K^+. However, there was no significant difference was observed in the plasma calcium of the pinealectomized as well as control group fish under different salt concentrations (ppt) (0, 5 and 7).

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