ON THE IONIC REGULATION IN SOME FISHES FROM THE COCÓ RIVER ESTUARIES IN FORTALEZA (CEARÁ STATE, BRAZIL)*

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ABSTRACT

Teleost fishes, both stenohaline and euryhaline, keep a ionic concentration of its plasma at a higher level in the sea than in freshwater. In general, regulation of hidromineral balance is not rigid, but rather it varies with changes that allow survival (HOLMES & DONALDSON⁸, JOHNSON⁹).

Little information is avaiable on the osmoregulatory response of teleost living permanently in hiposmotic brackish water, to environmental salinity stress. In the present paper 500 fish of Elops clupeoides, Bagre saurus, Anchova marinus, Oligoplites palometa, Eugerres fournieri. brasilianus, Micropogonias Mugil curema and Gobionnelus oceanicus were exposed to lower and higher salinities than usual, and measurements were then made of plasma concentration of CI^- , Na^+ , K^+ , Mg^{+2} and Ca^{+2} . Histological analysis of the skin, gills and

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kidney were made to find out a relationship between the structure of this organs and the osmoregulatory response from the fishes studied.

The values of salt content in the blood suggest a behaviour of osmoconformist for all the studied species.

The hystological analysis of the skin, gills and kidneys has not identified very sgnificant differences between the species, but the presence of a number of mucus-secretory cells in *Gobionnelus oceanicus* has been observed. It is also noteworthy the larger size of Malpighi's glomeruli in the kidneys of *Elops saurus*.

RESUMO

Peixes teleósteos, tanto estenohalinos como eurihalinos mantêm uma concentração iônica de seu plasma num nível mais elevado no mar do que na água doce. Geralmente, a regulação do balanço hidromineral não é rígida, mas varia com as mudanças que permitem sua sobrevivência (HOLMES & DONALDSON⁸; JOHNSON⁹).

Existem poucas informações sobre a resposta osmorregulatória de teleósteos que vivem permanentemente em águas estuarinas hiposmótica em condições de "stress". No presente trabalho 500 peixes pertencentes as espécies *Elops saurus, Anchova clupeoides,*

brasilianus, Micropogonias fournieri, Mugil curema e Gobionnelus oceanicus foram expostos a altas e baixas salinidades do que a usual e medidos a concentração de CI^- , Na^+ , K^+ , Mg^{+2} e Ca^{+2} do seu plasma. Análise histológica da pele, brânquias e rim também foi feita na tentativa de correlacionar estas estruturas com a resposta osmorregulatória dos peixes estudados.

Os valroes do conteúdo de sal do sangue sugere um comportamento osmoconformista para todas as espécies estudadas.

A análise histológica efetuada não apresentou diferença significativa entre as espécies, mas foi observada a presença de numerosas células secretoras de muco em *Gobionnelus oceanicus*. Também é digno de nota o maior tamanho dos glomérulos de Malpighi no rim de *Elps saurus*. *Key-Words:* lonic regulation, teleost fishes, osmoregulatory response.

INTRODUCTION

Estuarine waters, as a rule, do not have a typical fauna, but rather a group of marine, estuarine and freswater species (OLIVEIRA¹⁰).

CAMERON & PRITCHARD⁴ and PRITCHARD¹³ define an estuary as a body of coastal, semienclosed water, that connect with the sea and within which the sea is gradually diluted by the freshwater derived from earth drainage.

The number of marine species that enter inland water outweighs that of freshwater species (GUNTHER^{6,7}). The main factor to control entry of brackish water fish is the revel of water salinity. On the other hand, many eurihaline fishes keep the same salt salinity levels (POTTS & PARRY¹²). However, a certain degree of tolerance to tolerance to internal changes is a requirement of some anadromous and catadromous fishes when they migrate between the sea and the freshwater. Those species are capable of osmoregulation, to some extent in both environments and they do not stay isosmotic for a great length of time.

A few species require a gradual adaptation so as to change their osmotic condition, but others are able to undergo very rapid metabolic adjustment.

Teleost fishes, both stonohaline and euryhaline, keep a ionic concentration of its plasma at a higher level in the sea than is freshwater. In general, regulation of the hidromineral balance is not rigid, but rather it varies with changes that allow survival (HOLMES & DONALD-SON⁸; JOHNSON⁹).

The present paper aims at determining, by means of laboratory studies, the tolerance shown by estuarine fishes at different levels of salinity, as a way to arriving at their mechanisms of ionic regulation.

MATERIAL AND METHODS

The study material is comprised of 500 specimens belonging to the species Elops saurus, Anchovia clupeoides, Bagre marinus, Oligoplites palometa, Eugerres brasilianus, Micropogonias fournieri, Mugil curema, Gobionnelus oceanicus, for which data on length are set out in Table 1.

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Statistics of length (mm) of the specimens used in the study of ionic regulation of estuarine fishes, through salinity variations, in normal temperature conditions (28°C)

Species	X (mm)	S (mm)	CV (%)		
Elops saurus	164.4	5.56	3.40		
Anchovia clupeoides	105.2	5.47	5.20		
Bagre marinus	98.0	6.87	7.02		
Oligoplites palometa	84.8	6.57	7.65		
Eugerres brasilianus	88.2	3.19	3.60		
Micropogonias fournieri	116.0	2.50	2.18		
Mugil curema	106.5	4.60	4.32		
Gobionnelus oceanicus	183.7	2.66	1.45		

At sampling time the O_2 content and temperature were estimated by means of an Oxygen Meter, Model 51 – AYSI, while the pH value was estimated by a meter pH FANEM-ORION, Model 301 (Table 2).

TABLE 2

Characteristics of the water in the estuarine region of Cocó River, in the period July/1980 - June/1981

Statistical parameters	02	pН	Temp °C
x	3.93	5.63	27.8
S	0.55	1.98	1.95
CV	0.44	0.17	0.16

The specimens caught with a trawlnet were placed in containers with water from the sampling site, previously filtered, and left for acclimation in laboratory, for a while. As food supply oats was used in a 4 pen cent proportion of the live weight.

In testing tolerance to salinity, sea and freshwater, were mixed at the following proportions: 0 - 20 - 30 - 40- 50 - 60 - 70 - 80 - 90 - 100%. Salinity was measured by the Knudsen method with the modifications introduced by SWINGLE¹⁴. Water temperature in all experimental phases varied from 27 to 29°C.

The salt content in the blood for species submitted to the tolerance test was estimated from a blood sample taken from the cardiac region just before the line which links the insertion of the pectoral fins, as recommended by AMLACHER¹.

As an average, 0.2 ml of blood have been collected, submitted to centrifugation, the plasma being used for the estimates according to the method of spectrophotometry. The values of Na⁺, K^+ , Cl⁻, Mg⁺² and Ca⁺² were calculated after dilution of the plasma in destilled water, in a proportion of 1: 500, in mEg/1 units.

For histological studies, randomly chosen specimens of the different species

had fragments of the skin, gills and kidneys taken, and fixed at 10% formalin for microtomic cuts of 5 micra of the pieces embedded in paraphin, by the usual method via xylol. For colorations, hematoxilin of Delafield and eosine at 1% have been used.

RESULTS AND DISCUSSION

The variability of environmental conditions ensures the appearance of species highly sensitive to this factor as well as others which adapt themselves easily to a certain range of values.

One of the most important factors brought to bear on fish behaviour is resistence to the water salinity in their respective habitats.

Testing of tolerance to salinity in saurus. Anchova clupeoides, Elops Bagre marinus, Oligoplites palometa, brasilianus, **Micropogonias** Eugerres fournieri, Mugil curema and Gobionnelus oceanicus, at different levels, show that among the studied species *Elops saurus*. Eugerres brasilianus and Micropogonias fournieri are less able to support changes salinity than the other species in (Table 3).

After the experimental tests, the quantities of salt underwent changes in all species, as shown by the t – tested statistical differences between means (Table 4).

Some species require a gradual adptation in order to change their osmotic condition, but other are capable of a very quick metabolic adjustment. The present data seem to characterize perfectly the condition of osmoconfor - mist in the studies species.

According to $BLACK^2$, it seems that the likelihood of stenohaline fish surviving in water higher salt concentrations may depend on the histology and surface areas of the gills, O_2 intake, tolerance of the tissues to salts and control of permeability. This control, on its turn, may be the result of the neurosecretory action or the hormonal reaction

he study of ionic regulation of estuarine	
tests of saline shock (30 days period)	

	Seawater proportion (%)										
Species	0	10	20	30	40	50	60	70	80	90	100
Elops saurus	_	_	_								100
Anchovia clupeoides	80	100	100								100
Bagre marinus	_	20	20								100
Olígoplites palometa		_	10								100
Eugerres brasilianus	100	100	100								_
Micropogonias fournieri	-	_	40								100
Mugil curema	100	100	100								100
Gobionnelus oceanicus	50	100	100								50

TABLE 4

Data of t - tested statistical differences between means, after the experimental tests.

Species	Na ⁺	<u></u> κ ⁺	cı [–]	Mg ²⁺	Ca ²⁺
Elops saurus					
Anchova clupeoides					
Bagre marinus					
Oligoplites palometa					
Eugerres brasilianus					
Micropogonias fournieri					
Mugil curema					
Gobionnelus oceanicus	1799 B. S. 199 JT. JANS STR. 1995			ar an had shares as an a subscription over the solution	

to the new environment, as well as of a direct effect on cellular surfaces.

From the behaviour of the fish species and through histological analyses signs were sought of changes in the skin, gills and kidneys which could be related to this condition of osmoconformist of the species. A description of the analysed structures is given below, separately, pointing out that it applies to all species, but with emphasis on observed differences in some cases.

1 – Skin

The skin, with its auxiliary structures, stands for an external wrapper of the fish's body, through which most of the contacts with the external medium are made, It carries out many important functions in fish that are, essentially, of a protective nature (OOSTEN¹¹).

As in the other fishes, the skin of the species here studied is made up of two

basic layers: on other one, the epidermis and an inner one, the dermis, also known as "corium". These layers differ from one another according to position, origin, structure, character and function.

In the considered species, the epidermis at the intermediate region of the body, by dorsal fin, is comprised of eight to ten layers. In *Gobionnelus oceanicus*, at the insertion point of the fin, it is comprised of five layers.

The epitelial cells are bound by a viscous intercellular element. The germinative stratum is composed of colunar cells and there from new cells are derived. Reproduction in this layer occurs by mitotic division. The new cell substitutes the one from which it originated, while pushing it upward as far as the surface, where it takes the place of another cell that become useless from wearing or damage.

cell undergoes a chemical change and gradually takes on a scal or flat aspect. On the surface it finally dies and is eliminated. This scaling off is а continous process and thus the whole of the epidermis stays alive, save for the superficial layer of the cells that have already run their courses and are replaced continuosly.

Nutrition for growth and reproduction of the basal cells is obtained by the vascular underlying system of the dermis.

The dermis originates from the embrionic mesenquima of mesodermal origin (OOSTEN¹¹). In this studied species, no noteworthy detailed specific structures have been put to evidence, but rather a fiberelastic conective tissue showing in all of then with relatively few cells. The upper layer, very fine, comprises the vascular or spongy stratum, the lower layer is trick and dense standing for the compact stratum.

In the skin of Gobionnelus oceanicus there are several mucus-secretive cells, which carry out a lubrication function. They derive from the basal layer of the epidermis. in great numbers. and secrete a large quantity of mucus, of which it was not possible to determine the specific composition. However. OOSTEN¹¹ reports that in species that produce mucus this would interfere in the process of osmosis. This same author also emphasizes the role of the mucus in the protection against infestation by fungi, and coagulation power of blood, in addition to the precipitation action as a result of neutralization of negative electric changes in coloidal suspension by means of ionization of an acid another agent which liberates positive ions.

The other estuarine species hold few mucus-secretive cells, scattered in the epidermis.

The skin of teleost fishes is, as rule, permeable to water, but to varying degrees it has little or no permeability to organic substances or ions. PermeabiAccording to OOSTEN¹¹, experiments have shown direct losses to occur through damaged skin of freshwater fish, a large loss of water and a salts diffusion in damaged marine species.

Among the studied species, the ones with higher resistance to salinity fluctuations are exactly those which hold a skin with thick epidermis, and mucus cells in larger quantities.

2 – Gills

The gills consist of a row of folds, the branchial filaments, placed at a dorso-lateral position, from radial to ventral-lateral of the tips of the pharingean clefts. Those filaments are settled in two vertical semi-gills anterior and posterior to the opening and each semi-gill is supported in the foregoing and hinder sept, respectively.

The branchial filaments support on each side a series of transversal structures, the secondary lamellae, responsible for the respiration, and consist of central tissue with a single layer of auxiliary cells (FIG. 1).

The lamellae on the adjacent sides of two adjacent filaments hold a serie of minute grooves, through which water flows in a perpendicular direction to the three ends of the filament. This way, water flows straight from the pharingean cavity into the epibranchial space through several minute holes introduced by the intermediate lamellae.

Another anatomic feature that contributes largely to the gill's efficiency is the behaviour of blood supply. The disposition of the efferent and afferent blood vessels is such that blood flows through the branchial lamellae in the opposite direction to that of water's flow. This counter-current system entrances a wider exchange of respiratory gases and other substances.

The branchial filaments are dressed up by a single cavernous epitelium, containing a layer of conective tissue into which the afferent an efferent

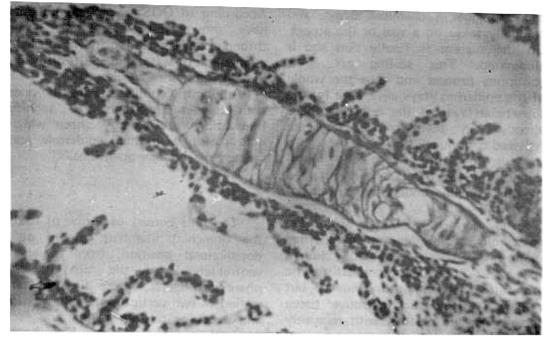


Figure 1 — Branchial filaments and secondary lamellae of *Mugil curema*. They are dressed up by a single cavernous epitelium.

arteriolae enter, the some holding for nervous sheaves.

The filaments are supported by a cavernous tissue wich stretches in the base and penetrate in each one by means of rather broad extension. This tissue is made out functionally of branchial cartilagenous columns which harden the filaments.

Keys & Wilmar (1932, 41), Liu (1942) and Copeland (1948), reported by BERTIN² have identified in the base of the branchial lamellae and in its two faces, bulky epitelial cells acidophilous and a large nucleus, comparable to those of HCl of mammal's stomach. The existence of those cells would probably be related to a great osmoregulatory power and would have as function to secrete sodium and potassium chlorides and to oppose to the animal's dismineralization on its way through water of different salinities.

No cells with such characteristics have been found in the branchial filaments of any of studied species.

3 – Kidney

Nash (1931), Romer & Groove (1935) and Smith (1930), according to report by WORSMANN et alii¹⁵, have pointed out the presence or absence of glomeruli in the habitat, discussing the various factors involved in the absorption, excretion and osmoregulation in marine and freshwater fishes.

GERARD⁵ reports a renal differentiation of some teleost species, especially in the breeding season. Such difference has not been observed in the studied species, but it must be said that the sampled individuals were not in reproductive activity.

With reference to the glomeruli of Malpighi the mesonephros of teleosts may be glomerular, glomeruless or functionally glomeruless. In freshwater fishes those structures are well developed, to a variable extent, and fractional or full counts of a number of glomeruli have been used as a parameters for fish grouping (WORSMANN et alii ¹⁵).

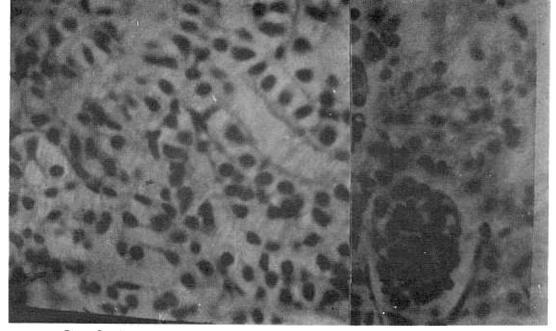


Figure 2 – A transversal cut of the kidney of *Elops seurus*, emphasizing a glomerulus of Malpighi and proximal tubules.

All species here considered present a small glomerulus of Malpighi, with renal parenquima comprised of tubules, most of them proximal with have an ample light with a spheric and central nucleus.

A transversal cut of the kidney of *Elops saurus* is show in Figure 2 enphasizing a glomerulus of Malpighi and proximal tubules tranversally observed. This species was the one with the most developed glomeruli.

The existence of well depeloped Malpighi's glomeruli is probably related to the tolerance shown by the species to variation in the environment's salinities.

GENERAL CONCLUSION

From the study of the species's behaviour to the saline shock, it has been shown that Anchova clupeoides. marinus, Mugil Bagre curema and Gobionnelus oceanicus tolerate large variations of salinity, ranging from (0,4°/₀₀) freshwater to sea water $35,7^{\circ}/_{\circ\circ}$). Elops saurus, Eugerres *brasilianus, Oligoplites palometa* and *Micropogonias fournieri* are less able to suport changes in salinity than the other species.

The values of salt content in the blood suggest a behaviour of osmoconformist for all the studied species.

The hystological analysis of the skin, gills and kidney has not identified very significant difference between the species, but the presence of a number of mucus-secretory cells in *Gobionnelus oceanicus* has been observed. It is also noteworthy the larger size of Malpighi's glomeruli in the kidney of *Elops saurus*.

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