

pH PROFILE ALONG THE GUT OF THE SILVER EEL AND ITS MODIFICATION DURING SEAWATER ACCLIMATION

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It is generally accepted that the gut plays an essential role in the water economy of marine and seawater-acclimated teleosts, water losses being balanced by continuous ingestion of drinking water (e.g. Maetz, 1970). Desalting of the ingested sea water (SW) takes place in the oesophagus, which is very permeable to Na and Cl but relatively impermeable to water, and Na, Cl and water resorption continues in the intestine (Hirano, Morisawa, Ando & Utida, 1976; Kirsch & Meister, 1982). Since 1930, it has been believed that the intestinal fluids of marine teleosts are alkaline (Smith, 1930). This has since been confirmed by Hickman (1968) for the rectal fluid of SW-adapted southern flounders and by Oide (1973) for SW-adapted Japanese eels, whose intestinal fluid was shown to have an average pH of 8.7. Oide has also shown that an increase in the pH of the bathing medium results in greater water movement across everted eel intestine. Hence the alkalinity of intestinal fluid has been invoked as a possible factor involved in water and salt resorption in marine or in SW-adapted teleosts. However, the precise role, or the origin, of this alkalinity has not been elucidated. Moreover, pH measurements in samples collected from dead animals and exposed to the air should always be considered with extreme care, especially in this case, since alkaline pH values are somewhat uncommon in biological fluids.

Our aim here is to report on the pH profile in the gut of silver eels (*Anguilla anguilla* L.) acclimated either to fresh water (FW) or to SW, using a flexible glass pH probe (Pharmacia Electronics microelectrode), in order to re-evaluate the importance of the alkaline nature of the intestinal fluid in ion and water absorption.

Silver eels caught in Meuse river tributaries were kept in FW or in SW tanks at 20 °C for 3 weeks. The pH of the tanks was checked once a week and kept between 7.5 and 7.8. After decapitation, each animal was pithed and the microelectrode was immediately introduced into the oesophagus. The advancement of the pH probe along

Table 1. *pH values at different levels of the gut of FW and SW silver eels*

	FW (<i>N</i> = 8)	SW (<i>N</i> = 7)
Anterior oesophagus	6.60 + 0.72	5.73 + 0.34*
Posterior oesophagus	6.66 + 0.58	5.99 + 0.38*
Stomach	5.88 + 1.88	4.64 + 1.50
Prepylorus	6.81 + 0.83	6.27 + 0.23
Postpylorus	7.33 + 0.29	7.07 + 0.24
Anterior intestine	7.18 + 0.45	7.14 + 0.23
Posterior intestine	7.33 + 0.42	7.14 + 0.47
Rectum	7.55 + 0.49	7.36 + 0.47

pH values are means \pm s.d.

* *t*-test: the difference is significant at the 5% level.

N, number of individuals.

the digestive tract was facilitated by progressive dissection behind the electrode tip. The electrode was calibrated with two buffer solutions, before and after each experiment, but was very stable. Measurements made using this procedure may not exactly reveal the *in vivo* situation; stress effects due to handling and decapitation are factors which may modify the pH profile of the gut. This method, however, has the advantage of measuring pH in fluids which have not been exposed to the air, within 10 min of death.

Table 1 presents the average pH values recorded at the different levels of the gut. They are very similar for FW and SW animals for all parts of the digestive tract, except for the oesophagus. In this region, for SW eels, pH is quite acid (5.73) while the pH of the tank is slightly basic; for FW eels, oesophageal pH is less acid (6.60) than for SW animals but still more acid than the environment. It could be argued that in SW, since they are drinking large amounts of water, the acidity in the oesophagus could be due to a leakage of stomach fluid. If such was the case, the posterior oesophagus would be expected to be more acid than the anterior oesophagus but for each animal the reverse situation is observed.

Increases in Na and Cl net fluxes have been shown to take place in the oesophagus of eels adapted from FW to SW, but permeability to water remains low (Hirano & Mayer-Gostan, 1976; Kirsch, 1978). On the other hand, the SW ingested by SW animals is very rapidly diluted at this level (Kirsch & Laurent, 1975; Kirsch & Meister, 1982). Furthermore, important modifications in structure and ultrastructure of the oesophagus have been shown in several teleosts adapted from FW to SW (Yamamoto & Hirano, 1978; Ezeasor & Stokoe, 1980; Meister, Humbert, Kirsch & Vivien-Roels, 1983). The possible role and the origin of acidification, occurring at the same time as these changes in structure and function of the oesophagus, remain to be investigated. However it could be speculated that an increase in Na and Cl permeabilities without change of the water permeability involves a very specific carrier-mediated process. The high efficiency of the dilution mechanism also favours this view. According to Kirsch & Meister (1982) seropositive electrical potentials are always recorded in the oesophagus of SW eels and Cl exchanges at this level could be mainly passive. But is Na resorption an active process?

Measurements of Na fluxes in short-circuit preparations have to be carried out

before a conclusion can be reached. A Na/H exchange mechanism across the epithelium could explain not only the Na resorption but also the acidification of the luminal fluid. Most probably several transport mechanisms exist at the two borders of the epithelium, the final balance being a NaCl resorption possibly accompanied by a secretion of protons. A study of the various ion-sensitive ATPases could also help to produce a clearer picture of the SW dilution mechanism in the oesophagus.

In the intestine of Japanese eels adapted to SW, a large increase in salt and water absorption has been related to higher specific activities of three enzymes: Na,K-ATPase, HCO₃-ATPase and alkaline phosphatase (Utida & Isono, 1967; Oide, 1967, 1970, 1973; Morisawa & Utida, 1976; Morisawa & Hirano, 1978). Since the latter two enzymes have an optimum pH close to 9.0, it has been hypothesized that the alkalinity of the intestinal fluid favours maximum efficiency.

Since our results indicate that the intestinal pH is very similar and close to neutrality in FW and SW silver eels, the role of alkaline phosphatase and HCO₃-ATPase in salt and water resorption should be evaluated in the same species at pH values close to 7.0. First of all, *in situ* pH measurements in the gut fluids of other species need to be made before a definite conclusion can be reached as to the involvement of pH in the process of SW adaptation in teleosts, at the level of the oesophagus as well as in the intestine.

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REFERENCES

- EZEASOR, D. N. & STOKOE, W. M. (1980). Scanning electron microscopic study of the gut mucosa of the rainbow trout, *Salmo gairdneri* R. *J. Fish Biol.* **17**, 529–539.
- HICKMAN, C. P., JR. (1968). Ingestion, intestinal absorption and elimination of sea water and salts in the Southern flounder, *Paralichthys lethostigma*. *Can. J. Zool.* **46**, 457–466.
- HIRANO, T. & MAYER-GOSTAN, N. (1976). Eel oesophagus as an osmoregulatory organ. *Proc. natn. Acad. Sci. U.S.A.* **73**, 1348–1350.
- HIRANO, T., MORISAWA, M., ANDO, M. & UTIDA, S. (1976). Adaptive changes in ion and water transport mechanism in the eel intestine. In *Intestinal Ion Transport*, (ed. J. W. L. Robinson), pp. 301–328. Lancaster: MTP Press.
- KIRSCH, R. (1978). Role of oesophagus in osmoregulation in teleost fishes. In *Alfred Benzon Symposium XI. Osmotic and Volume Regulation*, pp. 138–150. Copenhagen: Munksgaard.
- KIRSCH, R. & LAURENT, P. (1975). L'oesophage, organe effecteur de l'osmorégulation chez un téléostéen euryhalin, l'anguille (*Anguilla anguilla* L.). *C. r. hebdom. Séanc. Acad. Sci., Paris* **280D**, 2013–2015.
- KIRSCH, R. & MEISTER, M. F. (1982). Progressive processing of ingested water in the gut of sea-water teleosts. *J. exp. Biol.* **98**, 67–81.
- MAETZ, J. (1970). Mechanism of salt and water transfer across membranes in teleosts in relation to the aquatic environment. *Mem. Soc. Endocr.* **18**, 3–28.
- MEISTER, M. F., HUMBERT, W., KIRSCH, R. & VIVIEN-ROELS, B. (1983). Structure and ultrastructure of the oesophagus in sea-water and fresh-water teleosts (Pisces). *Zoomorph.* **102**, 33–51.
- MORISAWA, M. & HIRANO, T. (1978). Effects of L-phenylalanine on alkaline phosphatase activity and ion and water absorption in the eel intestine. *Comp. Biochem. Physiol.* **59C**, 111–115.
- MORISAWA, M. & UTIDA, S. (1976). HCO₃⁻-activated adenosine triphosphatase in intestinal mucosa of the eel. *Biochim. biophys. Acta* **445**, 458–463.
- OIDE, M. (1967). Effects of inhibitors on transport of water and ion in isolated intestine and Na⁺-K⁺ ATPase in intestinal mucosa of the eel. *Annotnes zool. jap.* **40**, 130–135.
- OIDE, M. (1970). Purification and some properties of alkaline phosphatase from intestinal mucosa of the eel adapted to fresh water or sea water. *Comp. Biochem. Physiol.* **36**, 241–252.

- OIDE, M. (1973). Role of alkaline phosphatase in intestinal water absorption by eels adapted to sea water. *Comp. Biochem. Physiol.* **46A**, 639-645.
- SMITH, H. W. (1930). The absorption and excretion of water and salts by marine teleosts. *Am. J. Physiol.* **93**, 480-505.
- UTIDA, S. & ISONO, N. (1967). Alkaline phosphatase activity in intestinal mucosa of the eel adapted to fresh water or sea water. *Proc. Japan Acad.* **43**, 789-792.
- YAMAMOTO, M. & HIRANO, T. (1978). Morphological changes in the oesophageal epithelium of the eel, *Anguilla japonica*, during adaptation to sea water. *Cell Tissue Res.* **192**, 25-38.