## ACID-BASE TRANSFERS IN THE LONG-HORNED SCULPIN (MYOXOCEPHALUS OCTODECIMSPINOSUS) FOLLOWING EXPOSURE TO 20% SEAWATER AND LOW EXTERNAL CHLORIDE

James B. Claiborne<sup>1</sup> and Erin Perry<sup>2</sup>

<sup>1</sup>Department of Biology, Georgia Southern University, Statesboro, GA 30460

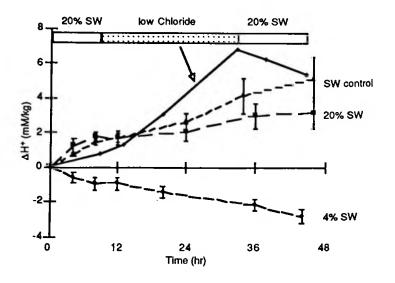
<sup>2</sup>The Mount Dessert Island Biological Laboratory, Salsbury Cove ME 04672

We have shown previously that sculpin lose large amounts of HCO3<sup>-</sup> to the water when exposed to decreased salinities (Walton & Claiborne, Bull MDIBL 27:4-5, 1988). Plasma pH remains relatively stable even though plasma TCO2 increases by ~50% (Claiborne et al., Bull MDIBL 29:60-61, 1990). While bone demineralization (Cameron, J. Exp. Biol. 117:307-18, 1985) may be the source of the observed HCO3<sup>-</sup> loss (Claiborne et al., ibid.), the transbranchial mechanism responsible for the net base loss remains unclear. The low salinities utilized in the majority of our previous experiments were only tolerated by the fish for several days (usually 4% seawater; ~ 20 mM NaCl). Therefore, we know little about the acid-base responses of these animals when exposed to dilutions within the adaptive salinity range of these somewhat "euryhaline" fish. It was the purpose of this experiment to observe acid-base transfers between the animals and the water during exposure to 20% seawater. By partial Cl<sup>-</sup> substitution of the external water, we have also attempted to characterize the mechanism by which these animals lose HCO3<sup>-</sup> during the low salinity exposure (LSE).

Long-horned sculpin (Myoxocephalus octodecimspinosus) were cannulated and placed in experimental chambers according to the methods described by Walton & Claiborne (ibid.). Following an overnight seawater recovery period the animals were subjected to a LSE (to water which had been prediluted to ~100 mM NaCl, measured as [Cl-]; approximately 20% seawater) of two or more days. During the LSE period, water NH<sub>4</sub><sup>+</sup> and HCO<sub>3</sub><sup>-</sup> were measured so that cumulative transfers of H<sup>+</sup> between the fish and the water could be calculated (Claiborne & Evans, Bull. MDIBL 25:32-34, 1985 and J. Exp. Biol. 140:89-105, 1988). In two animals (after a LSE of ~6 days), the 20% seawater bath was changed to one containing 100 mM sodium gluconate + 4% seawater for 24 hours. This "low chloride" (~20 mM) exposure was followed by a return to standard 20% seawater once again.

46 hours of LSE to 20% seawater (Fig. 1) induced an insignificant decrease in  $\Delta H^+$  transfer when compared to control animals  $(3.1\pm0.9~{\rm vs}~5.1\pm1.3~{\rm mmol~kg^{-1}}$  respectively;  $\mu\pm S.E.$ , n=4, p>0.1). This was in contrast to a significant negative  $\Delta H^+$  (a net base loss) observed during LSE in 4% seawater (-2.9  $\pm$  0.4 mmol kg<sup>-1</sup>; control and 4% data calculated from Walton & Claiborne, ibid.). Previous studies had shown that long-horned sculpin were able adapt to relatively low salinities in the range of those tested in this study (Oikari, Bull MDIBL 20:57-60, 1980). Clearly, animals in 20% seawater were capable of maintaining acid-base transfers (this study) and near normal plasma NaCl levels (Claiborne & Perry; unpublished).

Figure 1. Cumulative fish to water ΔH<sup>+</sup> transfers in sculpin exposed to various salinities. Solid line represents 2 animals preadapted to 20% seawater for 6 days then placed in a low Cl<sup>-</sup> bath for 24 hours.



When 20% adapted fish were placed in low chloride media, cumulative H<sup>+</sup> efflux increased rapidly and after 24 hours, ΔH<sup>+</sup> had risen above both 20% and control seawater groups (Fig. 1; solid line). This effect was due to a reversal in ΔHCO<sub>3</sub><sup>-</sup> excretion during the period (100 mM pre-exposure: 0.10 mmol kg<sup>-1</sup> hr<sup>-1</sup>, low chloride: -0.14 mmol kg<sup>-1</sup> hr<sup>-1</sup>, 100 mM post-exposure: 0.22 mmol kg<sup>-1</sup> hr<sup>-1</sup>; n=2). These data imply that external [Cl<sup>-</sup>] is linked to the transfer of HCO<sub>3</sub><sup>-</sup> to the water (as has been postulated in both marine and freshwater species; see Evans, in "Fish Physiology", eds. W. S. Hoar & D. J. Randall, Vol Xb, pp. 239-283, 1984). Indeed, after the ambient [Cl<sup>-</sup>] is decreased, the observed negative HCO<sub>3</sub><sup>-</sup> transfer may be due to a continuing H<sup>+</sup> efflux in exchange for Na<sup>+</sup> uptake across the gills (Evans, ibid.). That ΔH<sup>+</sup> efflux is elevated during low chloride exposure but decreases when both external Na<sup>+</sup> and Cl<sup>-</sup> are low (Fig. 1; low chloride verses 4% group) may indicate that the change in external [Na<sup>+</sup>] is the major cause of the acid-base transfer imbalance encountered at the lowest salinities. Substitution of the Na<sup>+</sup> in the media will allow a more direct test of this hypothesis. (Funded by NSF DCM 86-02905 to JBC and a Hearst Foundation Stipend to JBC and Scholarship to EP).