EFFECT OF CAPTURE STRESS ON PLASMA AND RED CELL PARAMETERS IN ATLANTIC MACKEREL (SCOMBER SCOMBRUS) EVIDENCE FOR IN VIVO RED CELL VOLUME REGULATION

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Capture stress in marine teleosts has been the subject of a number of investigations in recent years. Characteristic physiological changes during capture include increases in plasma osmolality, electrolyte concentrations, metabolites, proteins and hormones (e.g. Wells and Davie, Comp. Biochem. Physiol. 81A (3): 643-646, 1985; Ling and Wells, Comp. Biochem. Physiol. 82A (3): 609-612, 1985 and 82C(1): 231-234, 1985; Wells et al., Comp. Biochem. Physiol. 84A(3): 565-571, 1986). The observation of an increase in plasma osmolality raises the question of how cells exposed to hypertonic extracellular fluids regulate their volume <u>in vivo</u> during the stress response.

Simultaneous <u>in vivo</u> measurements of stress-induced changes in plasma and red cell ionic contents have been reported only for fresh-water adapted salmonids (Thomas et al., Am. J. Physiol. 250: R319-R327, 1986; Fievet et al., Am. J. Physiol. 252: R269-R275, 1987). Marine teleosts, however, face a entirely different set of problems in regulating the volume and composition of body fluids. In this report, we present <u>in vivo</u> measurements of changes in plasma osmolality, cell water content and ion concentrations of both plasma and red cells during capture stress in a stenohaline marine teleost, the Atlantic mackerel (<u>Scomber scombrus</u>).

Adult mackerel were caught by hook and line in Frenchman Bay, Salsbury Cove, Maine. Immediately after landing (30-60 seconds fight time) 2 mls of blood were drawn from the caudal vein with a heparinized syringe. After tagging, the animals were placed in a live car immersed in sea water, and then bled again after 10 minutes, 2 hours and 4 hours. A second set of fish were placed directly into a live car without being bled, and held for 8 days to assess recovery from capture.

After centrifugation of the whole blood, plasma osmolalities were measure on 10 microliter aliquots with a Wescor vapor-pressure osmometer (5100B). Perchloric acid (3.6% PCA/1.5 mM CsNO₃) extracts were made from aliquots of plasma and packed red cells. Cell and plasma Na and K were determined on an IL flame photometer and Cl on a Radiometer CMT-10 chloridometer. Water content of the cells was assayed gravimetrically as previously described (Schmidt and McManus, J. Gen. Physiol. 70: 59-79, 1977).

Plasma osmolality, Na, K and Cl changes after capture are presented in Figure 1. Within 10 minutes, there was a marked rise in osmolality, Na and Cl that continued until at 4 hours the percent increase was 45%, 38% and 29%, respectively. During this period, plasma K remained relatively constant. The increase in Na and Cl accounted for most of the change in osmolality. After 8 days in captivity, all values had returned to normal. The response of the red cells to these changes is shown in Figure 2. Despite the drastic increase in plasma osmolality during the first 2 hours, red cell water and K concentration was unaffected. By 4 hours, however, cell K was significantly increased (19%). Cell Na and Cl also rose sharply during the first 2 hours, leveling off between 2 and 4 hours at 189% and 50% over initial values. In animals kept in captivity for 8 days, all parameters returned to normal. Cell water and Cl are actually somewhat lower compared to those bled immediately after capture, suggesting perhaps that even the earliest data taken after capture were already showing an increase over normal. The animals bled after 8 days were collected promptly from the live car by net without the struggle associated with the initial hook and line technique, therefore they may be closer to a physiological steady state.

These in vivo changes in ionic and osmotic parameters during capture stress in mackerel are decidedly different than those reported for fresh-water adapted trout (<u>Salmo gairdneri</u>) stressed by hypoxia (Thomas et al., op. cit.). In that study, 20 minutes exposure to hypoxia caused only a minor elevation in trout plasma Na and K—changes too small to have a significant effect on osmolality. On the other hand, cell Na went up 3 fold with smaller increases in cell K and Cl. Since plasma osmolality remained constant, the red cells swelled due to a net gain of salt and water. It is evident that a marine teleost, such as the mackerel, under stress can suffer a rapid uptake of solute—mainly NaCl—from the environment. It is not clear whether this influx occurs across the gill or from the gastrointestinal tract, since as is well known most marine teleosts swallow sea water (Smith, Quart. Rev. Biol. 7: 1-26, 1932), although the amount varies between species.

In summary, we have shown that plasma osmolality of Atlantic mackerel increases sharply under stress, due mainly to an increase in Na and Cl. The red cells, however, maintain a constant volume by taking up an osmotic equivalent of these ions, thus demonstrating hypertonic volume regulation <u>in vivo</u>. These acute changes are reversible as the animals adjust to captivity. The adaptive significance of this hyperosmotic response is unclear. The freezing point of the blood must certainly decrease, which may be significant for active cold water species. The gain of Na and Cl by the red cells is probably due to activation of Na/H exchange by catecholamines, which we have been able to demonstrate *in vitro* (unpublished experiments). In this respect the response is similar to that seen in fresh-water adapted trout, except that mackerel cells do not swell because of the concurrent increase in plasma osmolality.

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Plasma Values



