OSMOREGULATION IN ADULT SEA TROUT FOLLOWING TRANSFER FROM SEA WATER TO FRESHWATER

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Introduction

In sea trout, as in other anadromous salmonids, a well-known series of endocrine-controlled events ("smoltification") occurs prior to downstream migration to the sea. These prepare the juvenile fish for marine osmoregulation before departure from freshwater. Research has concentrated particularly on biochemical events in the gills, such as induction of the enzyme Na^+, K^+-activated ATPase, which plays a key role in the extrusion of chloride and sodium ions.

In view of the inverse osmo- and volume-regulatory problems faced by freshwater and marine teleosts, different mechanisms are required for maintenance of homeostasis in the two media. In addition to the running costs of these mechanisms, a not insignificant fraction of the fishes basal metabolism, there must be a certain cost involved in maintaining both sets of mechanism in readiness in those species which are frequently exposed to changes in environmental salinity (e.g. in estuaries). Teleosts which only migrate at certain fixed times in their life cycle may benefit energetically by going through a process of pre-adaptation (acquisition of the appropriate machinery for the environment to be entered) prior to migration. Another option, a period of gradual acclimation in the river mouth, exposes the congregated fish to predation risks.

The aim of the present study was to look for indications of changes in the freshwater osmoregulatory capacities of adult sea trout during and outside the periods when they would normally enter freshwater. The hypothesis being tested was that there was some pre-adaptation to hyperosmotic regulation (i.e. some kind of “reverse smoltification”) whilst the fish were still at sea.

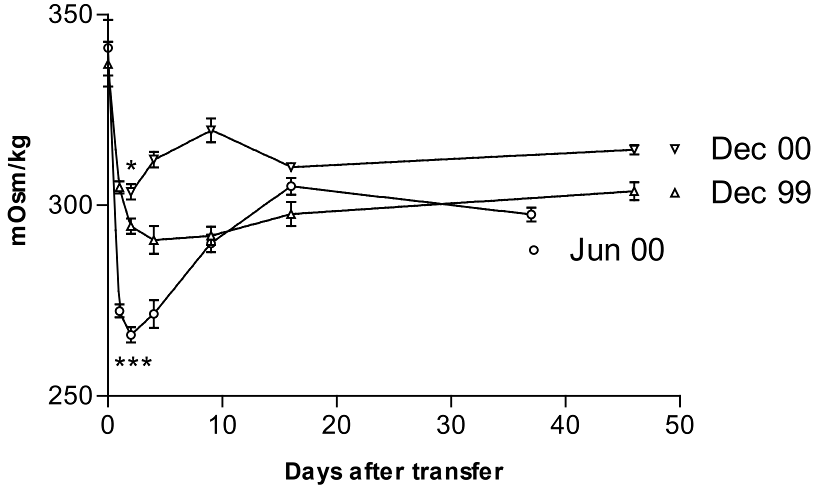
Materials and Methods

Hatchery-reared sea trout (*Salmo trutta* L) were kept at a constant 10^{o}C in a recirculating 24-28 ppt sea water system from smoltification in 1998 onwards. Groups were transferred directly to freshwater (at 10^{o}C) in December 1999 and June 2000. Blood and gill biopsy samples were taken before and at intervals after transfer, under MS 222 anaesthesia. As this was obviously a highly stressful procedure a third group was transferred in December 2000 with fewer sampling points. No sampling was carried out until 2 days after transfer to freshwater (different individuals were sampled as the pre-transfer sea water point). Plasma osmolality was measured (Wescor Vapro 5520 vapor pressure osmometer) and diluted plasma analysed for inorganic cations and anions (Dionex 4500i ion chromatograph). Gill Na^{+}, K^{+}-ATPase enzymatic activity was assayed at 25 °C using a coupled assay method. Messenger RNA was analysed with standard formaldehyde gel electrophoresis and Northern blotting. Species-specific [illegible]abelled cDNA probes were used for the quantification of Na^{+}, K^{+}-ATPase alpha-subunit and the secretory-type (NKCC1) Na^{+}, K^{+}, $2Cl^{-}$ co-transporter mRNA abundance, using a β-actin probe to correct for loading differences. Band intensities were quantified using phospor imaging (Molecular Dynamics, Sunnyvale, CA).

Results and Discussion

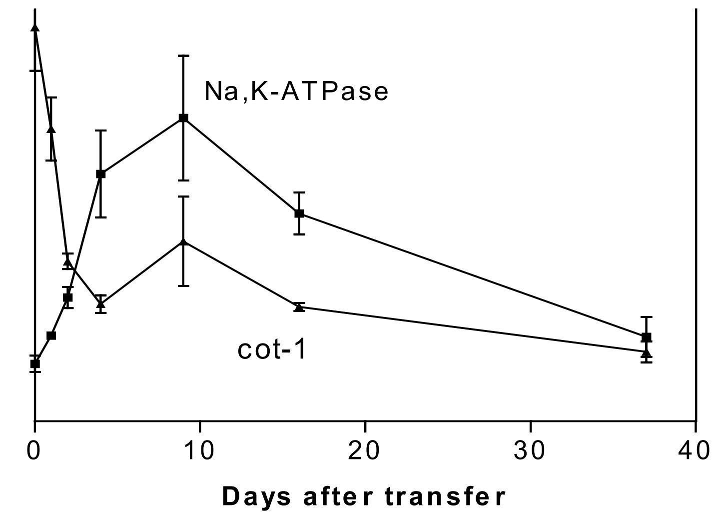
Plasma osmolality was around 340 mOsm kg^{-1} in all groups before transfer. It fell following transfer (Fig. 1), reaching a minimum of 266 mOsm kg^{-1} after 2 days in the June group ($p < 0.001$ cf. Dec. 1999 group). In the Dec.2000 group the 2 day (minimum) value was slightly higher ($p<0.05$) than the corresponding point in the Dec. 99 group and subsequent recovery was more rapid. By day 16 all groups had reached similar values.

Fig. 1. Changes in Plasma Osmolality



Na^+, K^+-ATPase activity varied a little during the the first 4 days after transfer and thereafter declined steadily to 0.92 ± 0.11 µmol ADP mg protein^{-1} h^{-1} (mean ± SEM) by day 37 in the June group and to 1.19 ± 0.24 µmol ADP mg protein^{-1} h^{-1} by day 46 in the December 1999 group from initial sea-water values of 3.27± 0.26 and 4.06 ± 0.29 µmol ADP mg protein^{-1} h^{-1} respectively. In contrast, expression of the Na^+, K^+-ATPase α-subunit mRNA increased dramatically over the first 4 days after transfer in both June and December 1999 (Fig. 2, June group).

Fig. 2 m-rna abundance (relative changes)



Expression of the Na^+, K^+, $2Cl^-$co-transporter cot-1 fell rapidly over the first 4 days in June and December (fig. 2, June group: Dec 1999 results were very similar). The contrast between the greatly increased expression of Na^+, K^+-ATPase α-subunit during the period in which the enzyme activity at first changed little and then started to fall was striking. Histological examination of the gill biopsy samples showed that this was during a period when typical freshwater chloride cells on the lamellae were replacing the seawater-type chloride cells in the filaments.

Sea trout transferred in December (when they would normally have migrated to freshwater) appeared to acclimate better to freshwater than those transferred in June (before the migration period). This raises the possibility that some pre-adaptation had already occurred.