

SHORT COMMUNICATION
**RECOVERY OF CHLORIDE UPTAKE IN SEAWATER-
ADAPTED RAINBOW TROUT (*SALMO GAIRDNERI*) AFTER
TRANSFER TO FRESH WATER**

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The ability of rainbow trout (*Salmo gairdneri* Richardson) to acclimate to sea water or partially diluted sea water has been studied many times (Houston, 1959; Parry, 1960; Eddy and Bath, 1979; Prunet and Boeuf, 1985). However, the reverse transition from sea water (SW) to fresh water (FW) has been less well studied. The transition from a hyperosmotic medium to a hypo-osmotic medium reverses the burden on the osmoregulatory organs, i.e. the gills and the kidney. SW teleost fish normally show net salt (NaCl) efflux, whereas FW fish need to acquire salt from the medium. Recent work on salmon (*Salmo salar*) transferred from SW to FW (Talbot and Potts, 1989; Talbot *et al.* 1989; Potts *et al.* 1989) has described the changes in sodium balance, osmoregulation and renal function in adult and juvenile fish. Potts *et al.* (1985) showed that salmon are capable of taking up sodium immediately on transfer to FW. Chloride uptake *via* the gills of FW fish is of equal importance to sodium uptake, and the two ions are thought to be transported independently (Maetz and Garcia-Romeu, 1964). This paper examines the changes in chloride transport that occur during the transition from SW to FW.

Rainbow trout (approx. 125 g) were obtained from College Mill Trout Farm, Almondbank, Perthshire, and maintained in Dundee tap water (Na^+ 0.2 mequiv l^{-1} ; Cl^- 0.25 mequiv l^{-1} ; Ca^{2+} 0.25 mequiv l^{-1} ; pH 7.2–7.4) at 10°C. Some fish were transferred to SW (29‰, North Sea) at 10°C and acclimated for at least 3 weeks without feeding. The transfer of fish to the loading tank was performed rapidly in a similar manner to that of Cameron (1976) and Eddy *et al.* (1977), with minimal handling, and in less than 3 s; the fish were lifted by hand from the holding tank into the experimental tank which contained 4 l of aerated Dundee tapwater at 10°C containing 10 μCi of Na^{36}Cl (Amersham International). After 15 min the fish were removed in a net, rinsed in tap water to remove any external activity and killed with a blow to the head. Blood samples (1 ml) were

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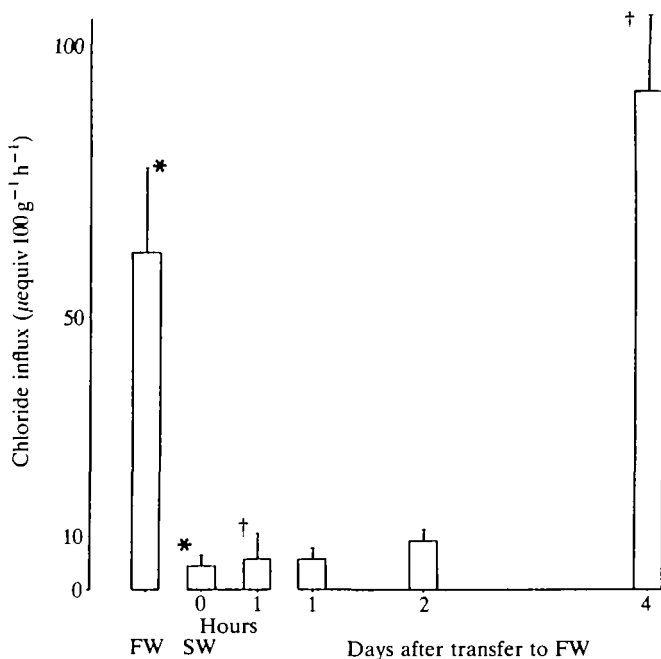


Fig. 1. Chloride influx ($\mu\text{equiv } 100 \text{ g}^{-1} \text{ h}^{-1} + \text{s.e.}$, $N=6$) at 10°C into rainbow trout in fresh water. FW, freshwater-acclimated control fish; SW, seawater-acclimated fish after 15 min in FW loading solution containing $10 \mu\text{Ci}$ of ^{36}Cl (0 h), 1 h 15 min (1 h) in FW, and 1, 2 and 4 days in FW. Two further SW fish left in FW for 8 days showed influxes of 13.5 and $56.8 \mu\text{equiv } 100 \text{ g}^{-1} \text{ h}^{-1}$. *, values for FW-adapted fish compared with values for SW fish; $P < 0.01$; †, values for SW fish at 0 h compared with fish after 4 days in FW; $P < 0.001$ (paired t -test).

withdrawn immediately into a heparinized syringe by cardiac puncture. Plasma chloride was measured using a Radiometer CMT10 chloride titrator and plasma radioactivity was counted in a Packard scintillation counter. Chloride uptake was measured in seawater-adapted fish transferred directly from SW to the FW loading solution, and after 1, 24, 48 and 96 h in FW. Two fish which were left after the 96 h experiment were allowed to remain in FW for a further 4 days and chloride uptake was subsequently measured at 8 days post-transfer. Chloride uptake and plasma chloride concentration of starved freshwater-adapted fish were also measured to provide control values. Flux rates were calculated by the methods presented in McWilliams and Potts (1978). Significance of results was tested by means of a paired t -test.

Rainbow trout are not very efficient at absorbing chloride after abrupt transfer to FW and the recovery of chloride uptake to FW levels takes several days to develop. After 4 days in FW the rate of chloride uptake increased to levels higher than in the FW control (Fig. 1), which shows compensation for the chloride loss from the plasma that occurs in the first 2 days after transfer (Fig. 2). After 8 days in FW, compensation of plasma chloride is probably complete, and chloride uptake

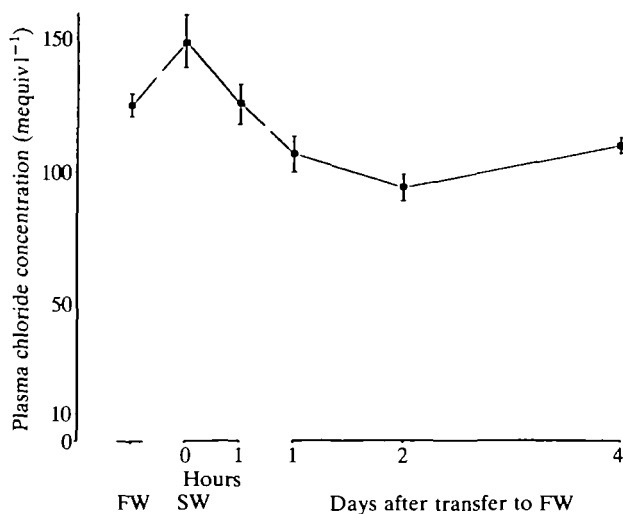


Fig. 2. Plasma chloride concentration (mequiv l⁻¹, \pm s.e., $N=6$) of rainbow trout. FW, freshwater-acclimated control fish; SW, seawater-acclimated fish after 15 min in FW (loading solution), 1 h 15 min in FW, and 1, 2 and 4 days in FW. Two further SW fish left in FW for 8 days had plasma chloride concentrations of 125 and 125.5 mequiv l⁻¹.

rates are likely to be near the FW level, assuming that a larger sample of fish would show values similar to those for the two fish measured (Fig. 1). Talbot and Potts (1989) demonstrated a net loss of chloride from the plasma of salmon post-smolts transferred directly to FW, with a lowest value of about 85 mequiv after 3 days in FW, and recovery after about 10 days.

Normal values for chloride uptake in FW-adapted trout depend on the composition of the medium, and values of $48 \mu\text{equiv } 100 \text{ g}^{-1} \text{ h}^{-1}$ (Williams and Eddy, 1986) for rainbow trout and $15\text{--}25 \mu\text{equiv } 100 \text{ g}^{-1} \text{ h}^{-1}$ for brown trout *Salmo trutta* (Battam, 1988) have been recorded. The value for FW-adapted rainbow trout of $60 \mu\text{equiv } 100 \text{ g}^{-1} \text{ h}^{-1}$ recorded here is closest to the value of Williams and Eddy (1986) measured in the same aquarium in similar water.

Chloride efflux was not measured in the experiments above but it is assumed that a net loss of chloride occurs and, from the data in Fig. 2, which shows that on transfer to FW between 0 and 1 h the fish lose approximately 16.5% of their plasma chloride, and using the value of $50 \mu\text{equiv Cl}^{-} \text{ g}^{-1}$ (Battam, 1988) for total body chloride, the loss of chloride would be $825 \mu\text{equiv } 100 \text{ g}^{-1} \text{ h}^{-1}$ during the first hour in FW, for a 100-g fish. Against this the average uptake of chloride after 1 h in FW is only $5 \mu\text{equiv } 100 \text{ g}^{-1} \text{ h}^{-1}$. Were losses of this magnitude to be maintained, the consequences would be obvious. However, the rate of chloride loss is rapidly reduced (Fig. 2).

The ability of rainbow trout to take up chloride immediately upon transfer from SW to FW is very interesting and of major significance to the fish. Sodium uptake rates in adult SW-adapted salmon measured less than 5 min after transfer to FW

were similar to those of FW-adapted salmon, but it could be argued that 'the pump is already triggered before the fish enters fresh water' (Potts *et al.* 1985). However, Talbot and Potts (1989) showed that SW-adapted salmon post-smolts were also capable of sodium uptake from FW within the first hour of transfer to FW, but at much lower rates than the adults. Evans (1973) found that the SW-adapted sailfin molly (*Poecilia latipinna*) maintained sodium influx in sea water, while the hagfish (*Myxine glutinosa*), a fish without freshwater relatives, also had a component of active sodium and chloride influx (Evans, 1984). It is therefore possible to argue that the rate of chloride uptake by trout in the first hour of transfer from SW to FW is the same as that which occurred in SW; this uptake would normally be masked by a massive efflux of chloride ($1406 \mu\text{equiv } 100 \text{ g}^{-1} \text{ h}^{-1}$; Eddy and Bath, 1979) and would feature as a small proportion of the total chloride influx, of which $1106 \mu\text{equiv } 100 \text{ g}^{-1} \text{ h}^{-1}$ is the calculated passive influx (Eddy and Bath, 1979). These observations also provide further evidence that the chloride cell is not responsible for salt uptake in fresh water, because the chloride cell is generally accepted as the site of active chloride efflux in SW teleosts (Foskett *et al.* 1983) and is unlikely to be the site of simultaneous chloride influx and efflux. It is more likely that salt uptake occurs *via* different cells, presumably the respiratory (pavement epithelial) cells of the branchial epithelium (Payan *et al.* 1984) which, in sea water, maintain a small influx component to assist acid-base regulation (Evans, 1984). The role of the chloride cells of the branchial epithelium of FW fish may be that of calcium uptake, for which there is both physiological (Payan *et al.* 1981) and ultrastructural evidence (Ishihara and Mugiya, 1987).

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