

**SODIUM UPTAKE KINETICS AND RESPONSES TO HIGH pH IN
FISH INHABITING A SEASONALLY ALKALINE LAKE**

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EXTENDED ABSTRACT ONLY - DO NOT CITE

Introduction

The fish used in this study inhabit a shallow freshwater lake (Slapton Ley, Devon, UK), which has suffered on-going cultural eutrophication since the 1940s. As a consequence blooms of cyanobacteria occur in most summers, causing deterioration of water quality, namely a rise in water pH associated with accelerated algal photosynthesis (Halstead and Tash, 1982). The average pH of Slapton Ley ranged between 9.2 and 9.5 (max pH 10.1) in the summers of 2000 and 2001. Little is known of the physiology of the fish species native to Slapton Ley (perch, pike, roach and rudd), and the pHs (> 9.0) recorded in Slapton are in the range seen to have severe effects on rainbow trout physiology. These physiological disturbances include a drop in plasma electrolytes (Na⁺ and Cl⁻), associated with a reduction in ion uptake rates (Laurent et al., 2000), and inhibition of ammonia excretion attributed to reversal of the NH₃ gradient across the gill (Wilkie, 1997). In order to understand the function of ionic transport systems it is important to analyse their transport kinetics (Shaw, 1959). This can be done by measuring the transport rate at a variety of substrate (external ion) concentrations, and can reveal possible differences in ionoregulatory strategies that may be correlated to an animal's environment and lifestyle (Gonzalez and Wilson, 2001). The aim of this study was to investigate the relative tolerances of the Slapton species to alkaline water, with respect to ion regulation and ammonia excretion disruption, as well as characterising their ion uptake mechanisms as a potential means to explain any tolerance.

Materials and Methods

Perch (*Perca fluviatilis*), pike (*Esox lucius*), roach (*Rutilus rutilus*) and rudd (*Scardinius erythrophthalmus*) were obtained from Slapton Ley by electrofishing, and rainbow trout (*Oncorhynchus mykiss*) from a local trout farm. Animals were held in dechlorinated Exeter tap water ([Ca²⁺] ~ 600.5

$\mu\text{mol l}^{-1}$; $[\text{Na}^+] \sim 437.3 \mu\text{mol l}^{-1}$; TAlk $\sim 0.90 \text{ mmol l}^{-1}$; 10.7°C ; pH 7.4) and withheld food in the week prior to an experiment. Experiments were carried out with individual fish held in flux chambers of varying volumes of aerated water. Fish were exposed to high pH (9.5 ± 0.1 ; using KOH) for 1 h, and increasing external $[\text{Na}^+]$ (using NaCl) for 0.75 h in order to analyse Na^+ kinetics. In both experiments sodium influxes ($J_{\text{Na}}^{\text{in}}$) were measured by monitoring the disappearance of ^{22}Na radioisotope from the external medium. Net sodium fluxes ($J_{\text{Na}}^{\text{net}}$) were calculated from changes in total $[\text{Na}^+]$ measured by flame photometry, and effluxes ($J_{\text{Na}}^{\text{out}}$) were calculated as the difference between net flux and influx. Ammonia excretion was determined by its increase in the external medium, measured by colourimetric assay. Kinetic parameters (K_m and V_{max}) were determined for individual fish using enzyme kinetics software (GraFit). All values are expressed as means \pm standard error. Negative and positive flux values indicate a loss from or gain to the fish, respectively, and were compared using repeated measures ANOVA ($P \leq 0.05$) with each fish as its own control.

Results

Acute exposure to pH 9.5 water caused significant changes to the Na^+ fluxes of all species (Figure 1). Perch and roach showed significantly more negative $J_{\text{Na}}^{\text{net}}$ during high pH exposure associated with an increased $J_{\text{Na}}^{\text{out}}$, only perch showed recovery of $J_{\text{Na}}^{\text{net}}$. Rudd and trout displayed no change in $J_{\text{Na}}^{\text{net}}$ throughout the experiment, and both species had reduced $J_{\text{Na}}^{\text{in}}$ and $J_{\text{Na}}^{\text{out}}$ in the recovery period. Trout showed elevated $J_{\text{Na}}^{\text{in}}$ and $J_{\text{Na}}^{\text{out}}$ during high pH exposure. Sodium uptake in all four species displayed typical saturation kinetics, and K_m and V_{max} varied between the different species (Table 1). All species (including pike) showed similar inhibition of ammonia excretion during alkaline exposure (40-60%).

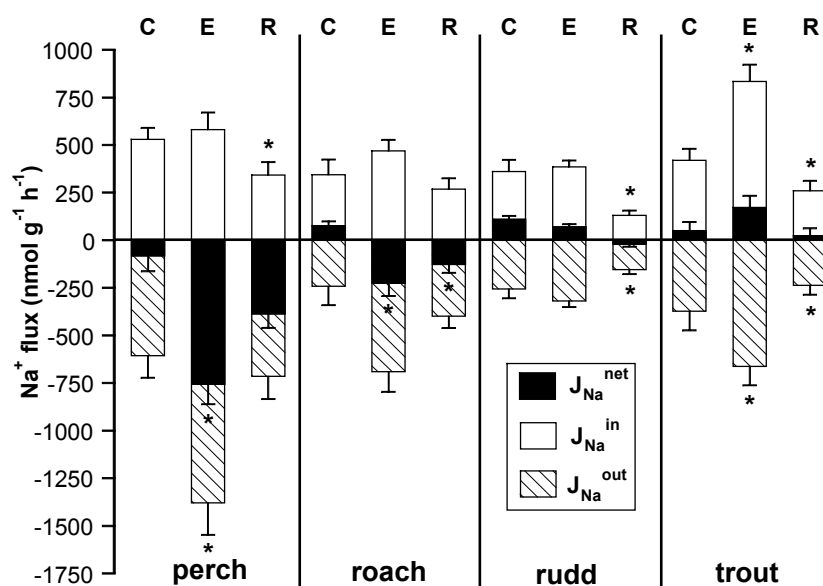


Figure 1. Effect of pH 9.5 water on the Na⁺ fluxes of 3 Slapton fish species and rainbow trout. C = control flux (pH 7.4), E = experimental flux (pH 9.5) and R = recovery flux (pH 7.4)

Table 1. Mean K_m and V_{max} values for each species.

	K _m (μmol l ⁻¹)	V _{max} (nmol g ⁻¹ h ⁻¹)
perch	69.2 ± 13.4	645.3 ± 83.9
roach	190.2 ± 56.9	713.7 ± 124.7
rudd	465.5 ± 73.8	832.4 ± 83.5
rainbow trout	163.7 ± 37.6	618.6 ± 174.7

Conclusions

Short term exposure to pH 9.5 had no effect on the net Na⁺ balance of rainbow trout, and caused increased Na⁺ uptake. There may be a varying degree of tolerance to high pH water with regard to Na⁺ balance in the Slapton species, with perch being the least tolerant, followed by roach and rudd. Varying Na⁺ uptake characteristics were also recorded, with the fish most susceptible to Na⁺ loss (perch) having the highest affinity transporter

and vice versa. The effect that high pH exposure had on ammonia excretion was markedly similar in all species.

Acknowledgements

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References

- Gonzalez, R.J. and Wilson, R.W. 2001. Patterns of ion regulation in acidophilic fish native to the ion-poor, acidic Rio Negro. *J. Fish Biol.* 58:1680-1690
- Halstead, B.G. and Tash, J.C. 1982. Unusual diel pHs in water as related to aquatic vegetation. *Hydrobiologia.* 96:217-224
- Laurent, P., Wilkie, M.P., Chevalier, C. and Wood, C.M. 2000. The effect of highly alkaline water (pH 9.5) on the morphology and morphometry of chloride cells and pavement cells in the gills of the freshwater rainbow trout: relationship to ionic transport and ammonia excretion. *Can. J. Zool.* 78:307-319
- Shaw, J. 1959. The absorption of sodium ions by the crayfish, *Astacus pallipes* Lereboullet I. The effect of external and internal sodium concentrations. *J. Expt. Biol.* 36:126-144
- Wilkie, M.P. 1997. Mechanisms of ammonia excretion across fish gills. *Comp. Biochem. Physiol.* 118A:39-50