Ciência Rural, Santa Maria, v.30, n.6, p.1041-1045, 2000 
ISSN 0103-8478

# $Na^+$ and $K^+$ BODY LEVELS AND SURVIVAL OF FINGERLINGS OF *Rhamdia quelen* (SILURIFORMES, PIMELODIDAE) EXPOSED TO ACUTE CHANGES OF WATER pH.

## NÍVEIS CORPORAIS DE $Na^+$ E $K^+$ E SOBREVIVÊNCIA DE ALEVINOS DE *Rhamdia quelen* (SILURIFORMES, PIMELODIDAE) SUBMETIDOS A VARIAÇÕES AGUDAS DO pH DA ÁGUA.

**Maria Ignez Zaions[1] Bernardo Baldisserotto[2]**

**SUMMARY**

*The objective of this study was to investigate the effect of water pH on survival and $Na^+$ and $K^+$ body levels of fingerlings of* ***Rhamdia quelen****, a freshwater catfish. Survival was 100% at pH 4.00 to 9.0 throughout the experiment (96h), and death of all fingerlings was observed only at pH 3.75 or lower and at pH 10.50. There was a significant correlation between water pH and body $Na^+$ levels (but not $K^+$ levels) 72h after transfer. There was a clear reduction in $Na^+$ levels when the transfer was to pH outside the 5.0 - 9.0 range, which is in agreement with the fact that fishes exposed to such pH's present loss of ions by diffusion and/or a decrease in absorption. Growth experiments with this species probably would present best results within the 5.0 – 9.0 pH range, in which the decrease of body $Na^+$ levels is reduced.*

**Key words:** ***Rhamdia quelen****, fingerlings, pH, body ion levels, survival*

**RESUMO**

*O objetivo deste estudo foi analisar o efeito do pH da água na sobrevivência e níveis corporais de $Na^+$ e $K^+$ de alevinos de* ***Rhamdia quelen****, um peixe de água doce. A sobrevivência foi de 100% ao longo do experimento (96h) na faixa de pH de 4,0 a 9,0, e foi observado mortalidade total dos alevinos em pH abaixo de 3,75 (e inclusive) e em pH 10,5. Houve uma correlação significativa entre o pH da água e os níveis corporais de $Na^+$ (mas não de $K^+$) 72h após a transferência. Quando os alevinos foram transferidos para um pH fora da faixa de 5,0 – 9,0, houve uma nítida redução dos níveis corporais de $Na^+$, concordando com o fato que peixes expostos a esses níveis de pH apresentam perda de íons por difusão e/ou uma redução na sua absorção. O crescimento dessa espécie, provavelmente, é estimulado dentro da faixa de pH 5,0 – 9,0, na qual a diminuição dos níveis corporais de $Na^+$ foi menor.*

**Palavras-chave:** ***Rhamdia quelen****, alevinos, pH, níveis iônicos corporais, sobrevivência*

## INTRODUCTION

Water pH is usually regulated by the carbonic gas-bicarbonate-carbonate system, and remains within the 6.0-8.0 range. Oscillations in water pH over this range may be due to the abundance of phytoplankton or the presence of high concentrations of dissolved $HCO_3^-$ and $CO_3^{2-}$ salts (alkaline pH) or of mineral and organic acids (acidic pH) (WILKIE & WOOD, 1996; ARANA, 1997). Most fish species survive acute exposure (a few days) to pH 4.5 to 9.0 (FREDA & McDONALD, 1988; VAN DIJK ***et al.***, 1993) but some species can resist even pH 3.5-4.4 (GONZALEZ & DUNSON, 1987) or a pH near 10.0 (WILKIE ***et al.***, 1993, 1994). Survival in acidic waters seems to be mainly related to the ability to prevent loss of $Na^+$ (FREDA & McDONALD, 1988) while adaptation to alkaline waters implies adjustments to nitrogen waste metabolism and excretion patterns (WILKIE & WOOD, 1996).

The objective of this study was to investigate the effect of water pH on survival and body $Na^+$ and $K^+$ levels in fingerlings of the silver catfish ***Rhamdia quelen***, a freshwater fish living in Central and South America (SILFVERGRIP, 1996). The use of this species for fish culture is increasing in South Brazil because this is a native species

[1] Biologist, Master in Animal Husbandry.
[2] Oceanologist, Doctor, UFSM, Departamento de Fisiologia, 97105-900, Santa Maria, RS. E.mail: bernardo@ccs.ufsm.br. Author for correspondence.
**Received for publication 26.11.99. Aproved 12.04.00**

which can survive at the low water temperature that occurs in winter (CHIPPARI GOMES ***et al.***, in press) and can feed on artificial food (PIAIA ***et al.***, 1997), and whose spawning is easily induced with hormones (RADÜNZ NETO, 1981). However, nothing is known about the survival and physiological responses of ***R. quelen*** to changes in water pH. In this region surface waters present pH from 3.0 to 8.4 (CORSAN, 1996), but several fish farmers use underground water whose pH can vary from 4.2 to 9.4 (CRPM, 1994). Consequently, it is important to determine the pH range tolerated by this species to reduce mortality in fish culture.

## MATERIAL AND METHODS

Fingerlings of ***Rhamdia quelen*** (Pimelodidae) (weight - 1.68 ± 0.87g and length - 5.96 ± 0.70cm) were bought from a local supplier and kept in a 1400ℓ running freshwater tank at 23-25°C, pH 6,8, with hardness of 30mg/ℓ $CaCO_3$, for 3 to 7 days. Fish were fed commercial dry pellets (Tembiú) containing 30% crude protein.

To determine the effect of pH on the survival and general feeding behavior of the ***R. quelen*** fingerlings, groups of 50 fishes were transferred to continuously aerated 250ℓ freshwater tanks (hardness of 30mg/l $CaCO_3$) with water pH's of 3.50, 3.75, 3.87, 4.00, 5.00, 7.00, 8.50, 9.00, 9.50, 9.75, 10.00, or 10.50 (three replicates per treatment). Water pH was previously adjusted to the desired experimental pH by the addition of sulfuric acid or sodium hydroxide. These tanks contained a water re-use system (24±1°C), and water pH was measured with a DMPH-2 pHmeter (Digimed, São Paulo, Brazil) and adjusted daily. Mortality was determined at 12, 24, 48, 72, and 96h after transfer. Fishes were fed ad libitum once a day with the food described above. All feces and pellet residues were removed daily by suction, and consequently 10% of the water of the tank was replaced.

In another series of experiments, the experimental pH's were those at which mortality was not 100% within 24h, i.e., 3.87, 4.00, 5.00, 7.00, 8.50, 9.00, 9.50, 9.75, and 10.00 (one replicate per treatment). In these experiments, other groups of 50 fingerlings were placed in the tanks. The procedures of maintenance and measurement of tank water were the same as in the first series of experiments. To determine the influence of pH on body $Na^+$ and $K^+$ levels, samples of 10 fingerlings were collected from water at each pH at 12, 24, 48, 72, and 96h after transfer. These fishes were killed with a blow to the head, weighed, immersed individually in hydrochloric acid, and left to dissolve in an oven (80°C) for 24h according to the method of OGNER (1983). The $Na^+$ and $K^+$ levels of the solutions originated by the acidic dissolution of the fingerlings and also the levels of these ions in the tank water were measured with a B262 flame spectrophotometer (Micronal, São Paulo, SP, Brazil, precision 1mg/ℓ).

Survival was analized by the Chi-square test, using the GraphPad Instat statistical sofware (version 2.05a.). The correlation between water pH and $Na^+$ and $K^+$ levels was calculated with the aid of the Slide Write Plus program (Advanced Graphics Software, Inc.). Data were expressed as mean ± SEM, and the minimum significance level was $P < 0.05$.

## RESULTS

***R. quelen*** fingerlings presented 100% survival and normal feeding behavior within the 4.00 to 9.00 pH range (mortality value at pH 9.00 is not significantly different from 0%) throughout the experiment (96 h). Death of all fingerlings was observed only at pH 3.87 (mortality value at pH 3.87 is not significantly different from 100%) or lower and at pH 10.50 (table 1).

There was a significant correlation (calculated by a Gaussian equation) between water pH and body $Na^+$ levels (but not between body $K^+$ levels and experimental pH) 72h after transfer to the

Table 1 - Accumulated mortality of ***Rhamdia quelen*** fingerlings as a function of time after transfer to the experimental pH.

| | Mortality (%) | | | | |
|---|---|---|---|---|---|
| Time (h) | 12 | 24 | 48 | 72 | 96[1] |
| pH | | | | | |
| 3.5 | 96 | 100 | 100 | 100 | 100[a] |
| 3.75 | 68 | 100 | 100 | 100 | 100[a] |
| 3.87 | 10 | 46 | 70 | 86 | 98[a] |
| 4.0 | 0 | 0 | 0 | 0 | 0[b] |
| 5.0 | 0 | 0 | 0 | 0 | 0[b] |
| 7.0 | 0 | 0 | 0 | 0 | 0[b] |
| 8.5 | 0 | 0 | 0 | 0 | 0[b] |
| 9.0 | 0 | 0 | 0 | 2 | 1[b] |
| 9.5 | 0 | 2 | 6 | 10 | 50[c] |
| 9.75 | 6 | 10 | 16 | 50 | 87[d] |
| 10.0 | 12 | 14 | 40 | 70 | 87[d] |
| 10.5 | 60 | 100 | 100 | 100 | 100[a] |

[1] Means identified by different letters in the column were significantly different ($P < 0.05$) as determined by Chi-square test comparison of mean values.

experimental pH. Exposure of fingerlings to acidic or alkaline pH reduced body $Na^+$ levels (figure 1). The correlation was calculated up to 72h because at 96h there were no data for these parameters at pH 10.0 (all the fingerlings died before 96h). The adjustment of tank water to alkaline pH with NaOH increased $Na^+$ levels from 0.086 to 0.551mEq/ℓ. Tank water $K^+$ levels were the same at all experimental pH values (0.008mEq/ℓ).

## DISCUSSION

Several studies have dealt with the survival of fishes in relation to pH changes, but all experiments analyzed the survival of a given species at acidic (GONZALEZ & DUNSON, 1987; FREDA & McDONALD, 1988; VAN DIJK ***et al.***, 1993) or alkaline pH (WILKIE ***et al.***, 1993, 1994), and not at both extremes of pH. Several teleost species tolerate acute exposure up to pH 4.5 in soft water, but at pH 4.0 some species, like ***Catostomus commersoni***, die within 48h (FRASER & HARVEY, 1984). Survival at lower pH in soft water is obtained only for species that usually live in acidic habitats like the Rio Negro in the Amazon region (GONZALEZ, 1996). Comparisons of survival at alkaline pH are complicated by the fact that some experiments were carried out in hard water (YESAKI & IWAMA, 1992) or alkaline-saline water (WILKIE ***et al.***, 1993, 1994). Salmonids in general can survive some days in soft water at pH 9.5, but a pH of 9.7 is lethal within one day (ALABASTER & LLOYD, 1982). Consequently, the pH range at which ***R. quelen*** can survive (4.00 to 9.00) is similar to that for most teleosts that live in circumneutral waters.

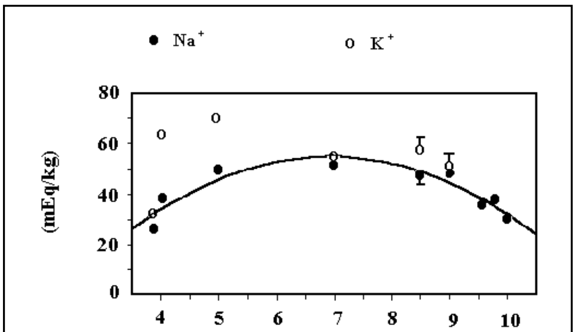


Figure 1 - Effect of pH on body $Na^+$ and $K^+$ levels 72 h after transfer. At some points error bars are not shown because they are inside the marker. The following equation was fitted to the $Na^+$ data:

$$y = -2387.15 + 2442.13\, e^{-0.5\left(\frac{x-6.94}{22.78}\right)^2}$$

where y = body level of $Na^+$ (mEq/kg) and x = pH of the water

$r^2 = 0.833$ ($P<0.05$)

According to the equation for the correlation between water pH and body $Na^+$ levels 72h after transfer to an experimental pH, higher levels of this ion could be observed in specimens transferred to pH 7.0, which was the pH nearest to that of the water where fingerlings were maintained before transfer. There was a clear reduction in $Na^+$ levels when the transfer was to pH outside the 5.0-9.0 range. The high $Na^+$ losses of ***R. quelen*** fingerlings after transfer to acidic or alkaline water certainly contributed to the mortality observed in this experiment.

Reduction of $Na^+$ and $K^+$ levels was also observed in specimens of ***C. commersoni*** exposed to pH 4.2 - 4.3 (HÕBE 1987; HÕBE ***et al.***, 1983). After transfer to water at pH 4.5, $Na^+$ levels remained lower than control up to 120h in this species (FRASER & HARVEY, 1984). A decrease in $Na^+$ levels was also determined in ***Perca flavescens***, ***Notropis cornutus***, and ***Salmo gairdneri*** exposed to pH 4.00 (FREDA & McDONALD, 1988) and in ***Salmo salar*** and ***Alosa pseudoharengus*** living in acidic rivers (LACROIX, 1985). Transfer of ***Oncorhynchus clarki henshawi*** specimens from water at pH 9.4 to pH 10.0 reduced $Na^+$ and $K^+$ levels of the muscle (WILKIE ***et al.***, 1994). Plasma $Na^+$ and $Cl^-$ levels also decreased in ***Oncorhynchus mykiss*** transferred to soft water at pH 10.0 (YESAKI & IWAMA, 1992) or buffered water at pH 10.5 (McGEER & EDDY, 1998).

In acidic water, excess $H^+$ can inhibit the $Na^+/H^+$ exchanger (POTTS, 1994) and create a gradient too steep for further extrusion of protons (LIN & RANDALL, 1991), reducing $Na^+$ uptake by the gills. Moreover, high $H^+$ concentrations disrupt the tight junctions of gill epithelia, increasing ion loss by a paracellular route. Fishes which survive in acidic water show a high affinity of the branchial tight junctions for $Ca^{++}$. This ion is important in maintaining the integrity of the tight junctions and in decreasing ion loss (GONZALEZ, 1996). Exposure to alkaline water enhances ion loss probably due to an inhibition of branchial $Na^+/H^+$, $Na^+/NH_4^+$ and $Cl^-/HCO_3^-$ exchangers (WILKIE & WOOD, 1996).

The decrease of body $Na^+$ and $K^+$ levels in ***R. quelen*** even after transfer to a pH close to the pH of acclimatization is surprising, but could be explained by the stress of handling. Specimens of ***Tilapia mossambica*** (DHARMAMBA ***et al.***, 1975), ***Fundulus heteroclitus*** (MAETZ ***et al.***, 1967) and salmonids in general (McDONALD & MILLIGAN, 1997) also presented loss of ions when stressed. The decline in body ions in stressed fish could arise mainly from increased branchial efflux (McDONALD & MILLIGAN, 1997). Electrolyte disturbances due to stress vary among species. For example, ***O. mykiss*** fingerlings exposed to severe confinement stress for 4h presented $Na^+$ loss, and recovery to pre-stress levels of this ion took 40h (McDONALD & MILLIGAN, 1997). On the other hand, ***Morone saxatilis*** submitted to transport stress (5h at 180g/ℓ load density) showed high $Na^+$ and $Cl^-$ losses and mortality reached 100% by 4 weeks post-transport (MAZIK ***et al.***, 1991).

Since fingerlings of ***R. quelen*** showed significant losses of body $Na^+$ and $K^+$ when transferred to a new medium (even at neutral pH), it would be interesting to conduct further studies with the addition of $Ca^{++}$ to the water to reduce osmoregulatory stress, since survival in acidic and alkaline environments is improved in hard water (FRASER & HARVEY, 1984; YESAKI & IWAMA, 1992).

Our results allow us to conclude that this species can survive at least 96h in the 4.0-9.0 pH range without any significant mortality. Any variation outside this pH range must be quickly corrected because high fingerling mortality can occur in less than 24h, mainly at acidic pH. Growth experiments probably would present best results within the 5.0-9.0 pH range, in which the decrease of body $Na^+$ levels is reduced. The maintenance of the fingerlings within this pH range would reduce the energy requirement for osmoregulatory work, improving growth.

## REFERENCES


ALABASTER, J.S., LLOYD, R. **Water quality criteria for freshwater fish**. 2. ed. Cambridge : FAO, 1982. 361p.

ARANA, L.V. **Princípios químicos da qualidade da água em aqüicultura**. Florianópolis : UFSC, 1997. 166p.

CHIPPARI GOMES, A.R., GOMES, L.C., BALDISSEROTTO, B. Lethal temperature for ***Rhamdia quelen*** fingerlings. **Journal of Applied Aquaculture**, v.9, n.4, p.11-21, 1999.

CRPM (Companhia de Pesquisa de Recursos Minerais) **Programa levantamentos geológicos básicos do Brasil**. Hydrological map, Santa Maria : Rio Grande do Sul, Brazil, 1994.

CORSAN (Companhia Regional de Saneamento) **Controle da qualidade da água** – regiões Centro Sul do Rio Grande do Sul. Porto Alegre, 1996. 14p. (Technical report).

DHARMAMBA, M., BORNANCIN, M., MAETZ, J. Environmental salinity and sodium chloride exchanges across the gill of ***Tilapia mossambica***. **Journal of Physiology**, Paris, v.70, p.627-636, 1975.

FRASER, A.G., HARVEY, H.H. Effects of environmental pH on the ionic composition of the white sucker (***Catostomus commersoni***) and pumpkinseed (***Lepomis gibbosus***). **Canadian Journal of Zoology**, v.62, n.2, p.249-259, 1984.

FREDA, J., McDONALD, D.G. Physiological correlates of interspecific variation in acid tolerance in fish. **Journal of Experimental Biology**, v.136, p.243-258, 1988.

GONZALEZ, R.J. Ion regulation in ion poor waters of low pH. In: VAL, A.L., ALMEIDA-VAL, V.M.F., RANDALL, D.J. **Physiology and biochemistry of the fishes of the Amazon**. Manaus : INPA, 1996. p.111-121.

GONZALEZ, R.J., DUNSON, W.A. Adaptations of sodium balance to low pH in a sunfish (***Enneacanthus obesus***) from naturally acidic waters. **Journal of Comparative Physiology B**, v.157, p.555-566, 1987.

HÕBE, H. Sulphate entry into soft-water fish (***Salmo gairdneri***, ***Catostomus commersoni***) during low ambient pH exposure. **Journal of Experimental Biology**, v.133, p.87-109, 1987.

HÕBE, H., WILKES, P.R.H., WALKER, R.L., ***et al.*** Acid-base balance, ionic status, and renal function in resting and acid-exposed white suckers *(**Catostomus commersoni***). **Canadian Journal of Zoology**, v.61, p.2660-2668, 1983.

LACROIX, G.L. Plasma ionic concentration of the atlantic salmon (***Salmo salar***), white sucker (***Catostomus commersoni***), and alewife (***Alosa pseudoharengus***) in some acidic rivers of Nova Scotia. **Canadian Journal of Zoology**, v.63, p.2254-2261, 1985.

LIN, H., RANDALL, D.J. Evidence for the presence of an electrogenic proton pump on the trout gill epithelium. **Journal of Experimental Biology**, v.161, p.119-134, 1991.

MAETZ, J., SAWYER, W.H., PICKFORD, G.E., ***et al.*** Évolution de la balanc minérale chez ***Fundulus heteroclitus*** au cors du transfert d'eau de mer en eau douce: effects de l'hypophysectomie et de la prolactine. **General and Comparative Endocrinology**, v.8, p.163-176, 1967.

MAZIK, P.M., SIMCO, B.A., PARKER, N.C. Influence of water hardness and salts on survival and physiological characteristics of stripped bass during and after transport. **Transactions of the American Fisheries Society**, v.120, p.121-126, 1991.

McDONALD, G., MILLIGAN, L. Ionic, osmotic and acid-base regulation in stress. In: IWAMA, G.K., PICKERING, A.D., SUMPTER, J.P., ***et al.*** **Fish stress and health in aquaculture**. Cambridge : Cambridge University, 1997. p.119-144.

McGEER, J.C., EDDY, F.B. Ionic regulation and nitrogenous excretion in rainbow trout exposed to buffered and unbuffered freshwater of pH 10.5. **Physiological Zoology**, v.71, n.2, p.179-190, 1998.

OGNER, G. Digestion of plants and organic soils using nitric acid, hidrogen peroxide and UV irradiation. **Communications in Soil Science and Plant Analyses**, v.14, n.10, p.937-943, 1983.

PIAIA, R., ULIANA, O., FILIPETTO, J., ***et al.*** Alimentação de larvas de jundiá (***Rhamdia quelen***), com dietas artificiais. **Ciência e Natura**, Santa Maria, v.19, p.119-131, 1997.

POTTS, W.T.W. Kinetics of sodium uptake in freshwater animals: a comparison of ion exchange and proton pump hypothesis. **American Journal of Physiology**, v.226, p.R315-R320, 1994.

RADÜNZ NETO, J. **Desenvolvimento de técnicas de reprodução e manejo de larvas e alevinos de jundiá *(Rhamdia quelen)***. Santa Maria, 1981. 77p. Dissertação (Mestrado em Zootecnia) - Curso de Pós-graduação em Zootecnia. Universidade Federal de Santa Maria, 1981.

SILFVERGRIP, A.M.C. **A systematic revision of the neotropical catfish genus *Rhamdia* (Teleostei, Pimelodidae)**. Stockholm, 1996. 156p. Dissertation (Master in Zoology) - Post-graduation course in Zoology. Stockholm University, 1996.

Van DIJK, P.L.M., Van DEN THILLART, G.E.E.J.M., BALM, P., ***et al.*** The influence of gradual acid/base status and plasma hormone levels in carp (***Cyprinus carpio***). **Journal of Fish Biology**, v.42, p.661-671, 1993.

WILKIE, M.P., WOOD, C.M. The adaptations of fish to extremely alkaline environments. **Comparative Biochemistry and Physiology**, v.113B, p.665-673, 1996.

WILKIE, M.P., WRIGHT, P.A., IWAMA, G.K., ***et al.*** The physiological responses of the Lahontan cutthroat trout (***Oncorhynchus clarki henshawi***), a resident of highly alkaline Pyramid Lake (pH 9.4), to challenge at pH 10. **Journal of Experimental Biology**, v.175, p.173-194, 1993.

WILKIE, M.P., WRIGHT, P.A., IWAMA, G.K., ***et al.*** The physiological responses of the Lahontan cutthroat trout (***Oncorhynchus clarki henshawi***) following transfer from well water to the highly alkaline waters of Pyramid Lake, Nevada (pH 9.4). **Physiological Zoology**, v.67, n.2, p.355-380, 1994.

YESAKI, T.Y., IWAMA, G.K. Survival, acid-base regulation, and ammonia excretion in rainbow trout in highly alkaline hard water. **Physiological Zoology**, v.65, n.4, p.763-787, 1992.