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pH regulation during long-term swimming in the mullet, *Rhinomugil corsula* (Hamilton)

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Abstract. The regulation of blood pH during long-term swimming in mullet, *Rhinomugil corsula* (Hamilton) acclimated to 30° C and forced to swim in a tunnel type apparatus for five hours continuously was studied. The accumulation of lactic acid in muscle caused a reduction of muscle pH. But in blood the reduction of pH during the 1st hour of exercise was followed by the recovery of pH to norma level during the later hours of exercise. The maintenance of pH was apparently caused by the retention of CO_2 in the form of HCO_3 ions in blood.

Keywords. pH regulation; muscle lactate; *Rhinomugil corsula*; CO_2; HCO_3^-.

1. Introduction

Some information on regulation of blood pH in resting fish under changing ambient condition such as ambient CO_2 and salinity, is available (Randall 1975; Randall *et al* 1975; Janson and Randall 1975). Black *et al* (1959) have measured blood pH simultaneously with glucose, lactate and bicarbonate in blood of rainbow, trout forced to swim intensely for relatively short duration (less than 5 minutes) and also during subsequent recovery. No information is however available on the pH regulation of fish subjected to long-term exercise (5 hr) although it is known that physiological responses of fish during the initial phase and steady phase (subsequent to 1–2 hr of continuous swimming) are quite different. During the steady phase of swimming (after 2–3 hr) the initial increased metabolic rate, O_2 consumption (Brett 1964; Smith 1965) and CO_2 output (Kutty 1968) come down and so does the anaerobic component of metabolism (lactic acid accumulation and RQ; Kutty 1968; Karuppannan 1972; Sukumaran 1976), while NH_3 excretion and ammonia quotient (molar ratio of NH_3 excretion and O_2 consumption) increase (Kutty 1972; Karuppannan 1972). In view of these observations it would be worthwhile studying the regulation of blood and muscle pH during the steady phase of long-term swimming, since most of the physiological and biochemical activities can be expected to be pH-dependent. Also results of long-term swimming experiments can yield information on fish swimming during migrations. In the present investigation blood and muscle pH of the mullet, *Rhinomugil corsula*, forced to swim for 5 hr have been studied. Blood and muscle lactate measurements have also been made simultaneously.

2. Materials and methods

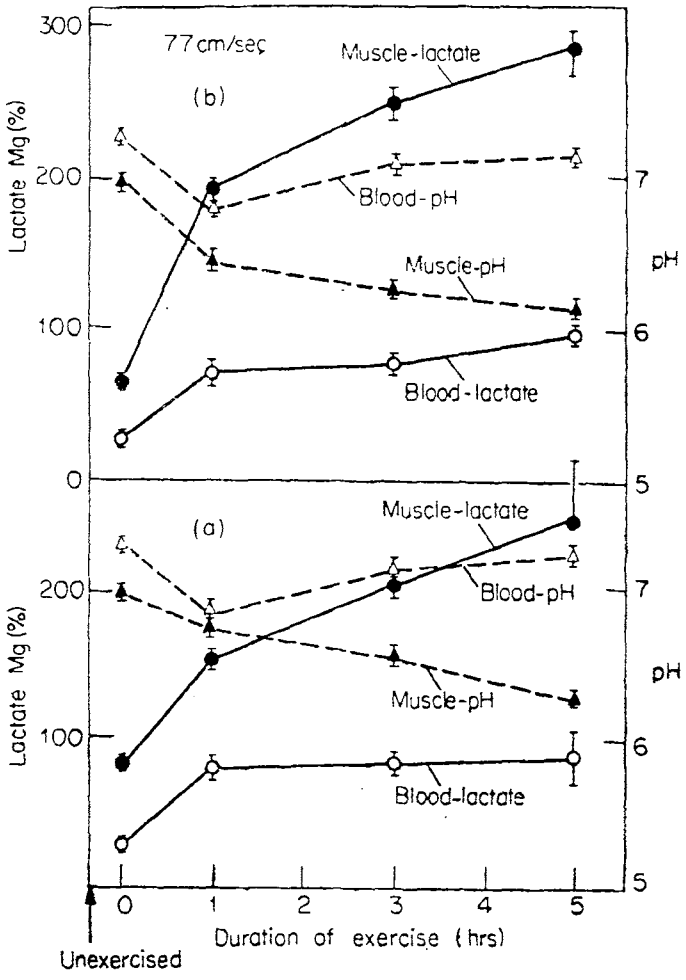
The fish used (17·1 to 18·8 cm; 38·4 to 47·2 g) were acclimated and tested at 30° C in fresh water. The fish were subjected to exercise continuously for 5 hr at two swimming speeds (20 and 77 cm/sec) in a tunnel-type respirometer (capacity 7·1 l) (Blazka *et al* 1960) described in Sukumaran and Kutty (1977). The muscle and blood pH were measured by using special pH electrodes (Toshniwal and Co–C64, made for pH measurements in fruits, cheese, meat, etc., and C14/02, specially made for pH measurements in small quantities (0·2–0·4 ml) of liquids, respectively). Blood and muscle lactate was estimated following the method of Barker and Summerson (1941), as outlined by Oser (1965).

The last fish starved for 24 hr was left in respirometer overnight in running water and was exercised for 1 to 5 hr at swimming speeds of 20 and 77 cm/sec. After an exercise of 1, 3, or 5 hr, the fish was removed and stunned by a hit on the head and immediately thereafter the muscle pH was measured by introducing the pH electrode in an incision made on the lateral muscle. Blood sample was collected in a heparinized syringe from direct heart puncture and transferred to a vial under liquid paraffin. The blood pH was measured immediately after collection. The blood and muscle samples were used subsequently for lactic acid analysis. Four fish from the same acclimation tank as that of the test fish were used as control.

3. Results and discussion

The data obtained for the two swimming speeds tested are summarised graphically in figures 1a and b. The muscle and blood pH obtained for 1st, 3rd and 5th hour of five hour continuous swimming are plotted along with the concentration of lactic acid in muscle and blood values for unexercised fish (control). These are taken as initial values and plotted at zero time on the *X* axis in the figures.

The muscle pH in both the exercises shows a steady decline, whereas the muscle lactic acid shows the opposite trend of a steady increase which has also been observed in other studies on fish (Suyama *et al* 1960; Black *et al* 1962), but under short-term exercise. But the blood pH of exercised mullet, after a decrease during the initial phase (1st hour) of swimming increased subsequently to reach the same level as that of unexercised fish. The accumulation of lactic acid during the later phase of exercise is negligible as also the changes in other parameters. Janssen and Randall (1975) injected 0·025 ml HCl to the dorsal aorta of *Salmo qairdneri* (unexercised-resting) and observed that the arterial pH fell drastically, but returned to pre-injection levels within 90 min. Randall *et al* (1975) again demonstrated that the arterial pH is restored within 3–5 hr in dog fish during chronic hypercapnia and this process is associated with a net uptake of $-HCO_3$ from sea water across the gills. So it is likely that in mullet as well the $-HCO_3$ ions are used to restore the blood pH and also that in exercised mullet the blood condition as that of one created by acid injection in salmon exist. The increase in the level of lactic acid in muscle and blood during the initial phase of exercise would certainly cause the observed reduction in muscle and blood pH. The regaining of resting pH towards the end of a 5 hr exercise (the fish could swim at



Figures 1a and b. Concentration of lactate and pH in muscle and blood in relation to duration of exercise in *Rhinomugil corsula* exercised continuously upto 5 hr at 20 cm/sec (lower panel—A) and 77 cm/sec (upper panel—B). Values from unexercised fish are plotted at zero time. Each value is a mean of four individual values and one standard error is represented by vertical bars.

this speed much longer) might be due to retention of respiratory carbon-dioxide inside the animal as $^{-}HCO_3$. The RQ which was 1·3 in the first hour of exercise decreased to 0·7 to the 5th hour and the relative carbon dioxide excretion was also less (Sukumaran 1976). The 5th hour RQ of *Tilapia mossambica* exercised continuously at a swimming velocity of 93 cm/sec was 0·5 (Karuppannan 1972). The total CO_2 in blood was found to be higher during long-term exercise of *Tilapia mossambica* (unpublished data) again suggesting the retention of the respiratory CO_2 for the restoration of $^{-}HCO_3$ ions to bring back the blood pH. While the regulation of blood pH might be largely by the retention of CO_2 and adjustment of bicarbonate: acid ratio the role of other blood buffers like phosphates and proteins cannot be ruled out (Albers 1970). A remarkable feature is the

maintenance of the difference in blood and muscle pH even at the end of the 5th hour exercise. This suggests a blood-muscle barrier and also active regulation of blood pH in swimming mullet.

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