*J. exp. Biol. 158, 463–476 (1991)*
*Printed in Great Britain © The Company of Biologists Limited 1991*



# EFFECT OF ENDURANCE SWIMMING ON THE LACTATE KINETICS OF RAINBOW TROUT

BY JEAN-MICHEL WEBER*

*Department of Zoology, University of British Columbia, Vancouver, British Columbia, Canada V6T 2A9*

*Accepted 13 March 1991*

## Summary

The lactate turnover rate of rainbow trout (*Oncorhynchus mykiss*) was measured by bolus injection of [U-$^{14}$C]lactate at rest and during prolonged swimming at 85 % $U_{crit}$ to determine the importance of this metabolic fuel for endurance locomotion in fish, to assess whether lactate exchange between white and red muscle could be a possible mechanism for supplying oxidizable fuel to their lateral red muscle, and to compare the contribution of lactate to total energy provision between teleost and mammalian species. Turnover rate only increased from 4.41±0.33 to 9.71±1.69 $\mu$mol kg$^{-1}$ min$^{-1}$ between rest and prolonged swimming, and the contribution of lactate oxidation to total metabolism declined during exercise. Lactate exchange between white and red muscle is, therefore, not a significant mechanism to fuel the active lateral red musculature during prolonged swimming. The lactate turnover rate of teleosts is one or two orders of magnitude lower than in mammals of equivalent size, but lactate has the same importance as a fuel in both vertebrate groups. However, lactate turnover rate and oxidation rate do not scale with body mass in the same fashion as does metabolic rate. The slope of the mammalian relationship for whole-body lactate turnover and oxidation is much lower (0.58) than the slope of the classic relationship for metabolic rate (0.75), indicating that lactate is a much more important oxidative substrate for small than for large animals.

## Introduction

During submaximal exercise, lactate can become an important substrate for oxidative ATP production in mammals (Eldridge, 1975; Issekutz *et al.* 1976; Jorfeldt, 1970; Weber, 1988) and lactate release by glycolytic muscle fibers has been suggested as a potential mechanism for providing carbohydrate fuel to oxidative muscles (Brooks, 1985; Molé, 1983). In teleosts, large glycogen stores in white muscle could be exploited to supply aerobic energy to the lateral red muscles

*Present address: Biology Department, University of Ottawa, 30 George Glinski, Ottawa, Ontario, Canada, K1N 6N5.

Key words: metabolism, muscle fuels, lactate turnover, scaling, biochemical adaptation, exercise, *Oncorhynchus mykiss*.

used in sustained swimming (Weber *et al.* 1986; Wittenberger *et al.* 1975; Wokoma and Johnston, 1981). These white muscle stores cannot be exported as blood glucose because glucose-6-phosphatase is virtually absent from this tissue (Walton and Cowey, 1982). Lactate exchange between white and red muscle could be a way to circumvent the problem of supplying carbohydrates to red muscles, and endurance exercise would cause a marked increase in lactate turnover rate if such a mechanism were operational in fish. Except for tuna (Weber *et al.* 1986), however, only resting values for lactate turnover are available for teleosts under a variety of conditions, including hypoxia (Dunn and Hochachka, 1987), prolonged starvation (Cornish and Moon, 1985) and recovery from intense exercise (Milligan and McDonald, 1988).

The goals of this study were, therefore, to measure the lactate turnover rate of rainbow trout during sustained exercise: (1) to determine the importance of this substrate as a metabolic fuel for endurance locomotion in fish and to compare the contribution of lactate to total energy provision between teleost and mammalian species, and (2) to assess whether lactate exchange between white and red muscle could be a possible mechanism used by teleosts for supplying oxidizable fuel to their lateral red muscles.

## Materials and methods

### *Animals and catheterizations*

Adult rainbow trout [*Oncorhynchus mykiss* (Walbaum)] of both sexes were obtained from a single stock of the Sun Valley trout farm, Mission BC, Canada. Before the experiments, they were kept for 2 months in outdoor holding tanks supplied with fresh, aerated, dechlorinated tapwater (7–9°C). During this acclimation time, they were familiarized with the exercise protocol by being placed individually in a swim tunnel for 30 min at varying water flow rates. Animals that did not swim normally at the end of this trial session were discarded.

The fish were fed commercial trout pellets *ad libitum*, but feeding was interrupted 2 days before catheterization. Animals were anaesthetized in a 1:10 000 solution of buffered MS-222 (Sigma). The dorsal aorta was then chronically cannulated with polyethylene tubing (Clay-Adams PE 50) while the gills were irrigated continuously with a dilute solution of MS-222 (1:20 000, pH 7). Catheters were filled with heparinized (10 i.u. $ml^{-1}$) Cortland saline without glucose (Wolf, 1963). The fish were placed individually in darkened Plexiglas boxes supplied with aerated fresh water at 7–9°C and were allowed to recover for at least 2 days before the measurement of lactate turnover. Animals with hematocrits lower than 25 % were not used.

### *Experimental protocol*

All experiments were performed between 07:00 and 11:00 h to minimize the variability caused by circadian fluctuations in metabolism. Fish were randomly assigned to a resting or to an exercise group. The resting measurements wer

performed in the dark Plexiglas boxes where the cannulated animals were kept. Only the end of the catheter was held out of the box to prevent the fish from seeing the experimenter. Exercise experiments were carried out in a Brett-type swim tunnel (Brett, 1964) supplied with aerated, dechlorinated fresh water at 8±1 °C. On the evening preceding the turnover measurement during exercise, the animal was quietly transferred by net to the swim tunnel to avoid struggling. It was left there overnight without light or water current. Oxygen tension and temperature were held constant by continual replacement of the swim tunnel water with fresh dechlorinated water. On the following morning the animal had to swim for 3 h at $1.5\,L\,s^{-1}$ (where $L$ is body length) while lactate kinetics were quantified. Preliminary measurements on five animals showed that $1.5\,L\,s^{-1}$ represents 85±3 % $U_{crit}$ for cannulated animals ($U_{crit}$ is maximal sustained swimming speed or critical velocity, see Kiceniuk and Jones, 1977). This swimming velocity was selected because not only red muscle but also white muscle fibers are recruited (Bone, 1978; Hudson, 1973). In addition, this speed is ecologically relevant because ultrasonic tracking studies have shown that salmon maintain it for prolonged periods during migration (Quinn, 1988).

### *Measurement of lactate turnover*

The bolus injection technique was used to determine turnover rate (Hetenyi *et al.* 1983). The advantage of this method is that it requires only one catheter. For this reason, bolus injection has been selected instead of continuous infusion here, as in other fish studies, where double catheterization would be technically difficult or sometimes impossible (Bever *et al.* 1977; Machado *et al.* 1989; Milligan and McDonald, 1988; Weber *et al.* 1986). Because the bolus injection technique assumes a steady plasma lactate concentration, the radioactive bolus was injected either at rest or after the animal had already been swimming for 60 min and was therefore in a steady state for lactate. About 10 $\mu$Ci of [U-$^{14}$C]lactate (specific activity: 150 mCi mmol$^{-1}$; Amersham, Oakville, Ontario, Canada) was mixed with 0.5 ml of saline without heparin and injected *via* the catheter at time zero. The exact amount administered was measured separately for each experiment by weighing the syringe before and after injection, and by counting a sample of the bolus solution. The catheter was flushed with 1 ml of saline immediately after injection and blood samples (150 $\mu$l each) were drawn after 2, 3, 5, 8, 12, 20, 30, 60 and 120 min. The catheter was flushed with 150 $\mu$l of saline between samples.

### *Sample analyses*

Blood samples were deproteinized immediately by putting them in preweighed Eppendorf tubes containing 0.3 ml of cold perchloric acid (8 %). They were mixed carefully, weighed again to determine the exact dilution, and centrifuged for 2 min at 10 000 *g*. The supernatant was stored at −4 °C until further analysis.

Metabolite concentrations were determined enzymatically in the neutralized perchloric acid extracts. Lactate concentration was measured at 340 nm with lactate dehydrogenase and NADH (Bergmeyer, 1974). Glucose concentration was

also determined to confirm that carbohydrate metabolism was in steady state throughout the experiments and to make sure that the animals were not becoming hypoglycemic after several hours of swimming. Glucose concentration was measured on a Beckman glucose analyzer with the glucose oxidase method. All metabolite assays were performed in duplicate and they were repeated when the two original measurements differed by more than 10 %.

Lactate activity was determined by subtracting glucose activity from total activity, as described previously (Weber *et al.* 1986). Preliminary experiments on trout plasma comparing the glucose separation method used here with lactate separation by column chromatography (Katz *et al.* 1981) showed no difference in measured lactate activity. Furthermore, it has been shown in another ectotherm that, after bolus injection of [$^{14}$C]lactate, all measurable activity in plasma is restricted to lactate and glucose (Gleeson and Dalessio, 1989). Scintillation counting was performed on a Beckman LS6300 with external quench correction after the samples had been mixed with Hydrofluor (National Diagnostics, Manville, NJ) and left at 4 °C in the dark for 12 h. Counting efficiency ranged from 93 to 95 %.

### *Calculations*

Mass-specific turnover rate (in $\mu$mol kg$^{-1}$ min$^{-1}$) was calculated by dividing the dose injected (in disints min$^{-1}$) by the surface area under the specific activity decay curve (in disints $\mu$mol$^{-1}$) and by the mass of the animal (in kg) (Katz *et al.* 1981). Decay curves were fitted with triple exponential functions (Enzfitter, Cambridge, UK) and integrated as described previously (Weber *et al.* 1986). The maximum activity was calculated as the dose injected divided by the volume of the rapidly mixing pool, estimated at 6 % of body volume. Lactate clearance (MCR) was calculated as turnover rate divided by mean lactate concentration. Relationships between concentration and turnover, concentration and clearance, and log (body mass) *vs* log (whole-body turnover) were analyzed by linear regression and analysis of variance (ANOVA). Values given are means±S.E.M..

## Results

### *Blood metabolite concentrations*

All the animals were in lactate steady state during the turnover measurements at rest and during exercise (Figs 1 and 2, Table 1). The mean resting blood lactate concentration was 0.84±0.09 mmol l$^{-1}$ ($N$=6). While the fish were swimming, lactate concentration remained steady within each individual, but mean values ranged from 0.74 to 3.53 mmol l$^{-1}$ in the different animals (Table 1). Blood glucose concentration stayed constant in all fish at rest as well as during exercise. However, mean values ranged from 6.15 to 14.35 mmol l$^{-1}$ between individuals (Table 1).

Table 1. *Lactate concentration, turnover rate, and clearance rate in resting and swimming rainbow trout*

| Experiment | Body mass (g) | [Glucose] ($mmol\,l^{-1}$) | [Lactate] ($mmol\,l^{-1}$) | Lactate turnover ($R_T$) ($\mu mol\,kg^{-1}\,min^{-1}$) | Lactate clearance (MCR) ($ml\,kg^{-1}\,min^{-1}$) |
|---|---|---|---|---|---|
| Rest | | | | | |
| 1 | 428 | 8.56±0.35 | 1.12±0.16 | 4.73 | 4.22 |
| 2 | 341 | 14.23±0.32 | 1.08±0.07 | 3.88 | 3.59 |
| 3 | 299 | 9.25±0.29 | 0.64±0.05 | 3.05 | 4.77 |
| 4 | 311 | 14.35±0.34 | 0.74±0.06 | 5.15 | 6.96 |
| 5 | 253 | 7.23±0.10 | 0.75±0.05 | 4.55 | 6.07 |
| 6 | 299 | 7.30±0.09 | 0.69±0.05 | 5.09 | 7.38 |
| Exercise | | | | | |
| 7 | 260 | 8.13±0.21 | 0.74±0.03 | 3.96 | 5.35 |
| 8 | 260 | 6.96±0.18 | 0.75±0.06 | 5.38 | 7.17 |
| 9 | 290 | 6.16±0.15 | 0.91±0.02 | 4.28 | 4.70 |
| 10 | 294 | 7.59±0.21 | 1.10±0.06 | 5.38 | 4.89 |
| 11 | 278 | 8.47±0.27 | 1.72±0.15 | 9.36 | 5.44 |
| 12 | 274 | 10.24±0.25 | 2.00±0.32 | 7.02 | 3.51 |
| 13 | 289 | 11.08±0.25 | 2.40±0.12 | 15.91 | 6.63 |
| 14 | 283 | 9.60±0.24 | 2.51±0.05 | 12.54 | 5.00 |
| 15 | 257 | 7.83±0.19 | 2.91±0.19 | 14.70 | 5.05 |
| 16 | 249 | 6.15±0.11 | 3.53±0.16 | 18.55 | 5.25 |

Values are concentration means±S.E.M. ($N$=9).

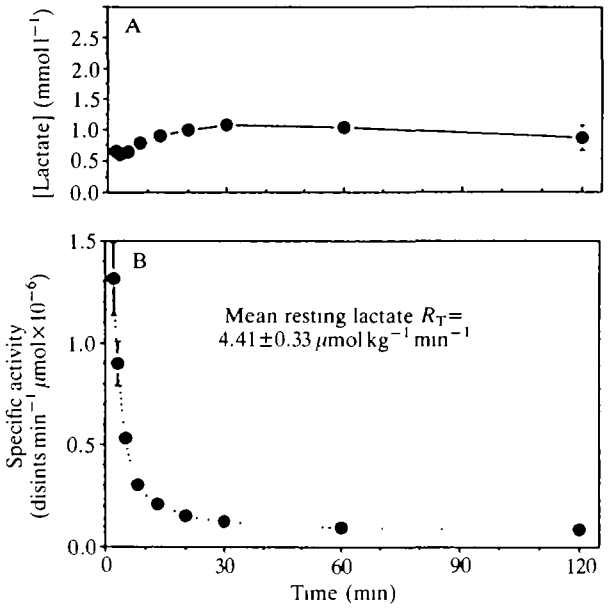


Fig. 1. Mean lactate concentration (A) and decay curve for mean lactate specific activity (B) in resting rainbow trout after injection of $23.5\times10^6$ disints min$^{-1}$ (range $22.02\times10^6$ to $24.73\times10^6$ disints min$^{-1}$) [U-$^{14}$C]lactate at time zero. The decay curve was fitted with a triple exponential function. Standard errors are shown only when larger than the symbols ($N$=6). $R_T$, rate of turnover.

*Lactate turnover and clearance rates*

Mean activity of the [U-$^{14}$C]lactate boluses injected into resting and exercising animals was 22 640 585±508 disints min$^{-1}$ ($N$=16). In all cases, triple exponential functions fitted the specific activity decay curves very well. The lactate turnover rates calculated from these curves are presented in Table 1. Fig. 1B shows the mean decay curve for resting individuals, which had an average lactate turnover rate of 4.41±0.33 μmol kg$^{-1}$ min$^{-1}$ ($N$=6). During exercise, mean turnover rate only increased to 9.71±1.69 μmol kg$^{-1}$ min$^{-1}$ ($N$=10), but the effect of exercise varied between individuals. This result was first observed in a group of five fish and subsequently confirmed after increasing the sample size of the exercise group to 10 individuals (Table 1). Particular care was taken to acclimate the animals and to keep them undisturbed while they were swimming. All exercise results were pooled because no significant difference was found between the two subsequent groups of five swimming fish. Results are summarized in Table 1, where the exercise experiments were ranked according to mean lactate concentration. The specific activity decay curves from the two animals that showed the weakest and the strongest response to exercise are shown in Fig. 2B.

The relationship between blood lactate concentration and turnover rate is presented in Fig. 3. The two variables were related linearly (for swimming fish: $r$=0.95; slope=5.17, significantly different from 0 at $P$<0.0001).

Lactate clearance was maintained between rest and exercise ($P>0.05$). Mean values were 5.50±0.63 at rest ($N=6$) and 5.30±0.32 ml $kg^{-1}$ $min^{-1}$ during exercise ($N=10$). Fig. 4 shows the relationship between blood lactate concentration and

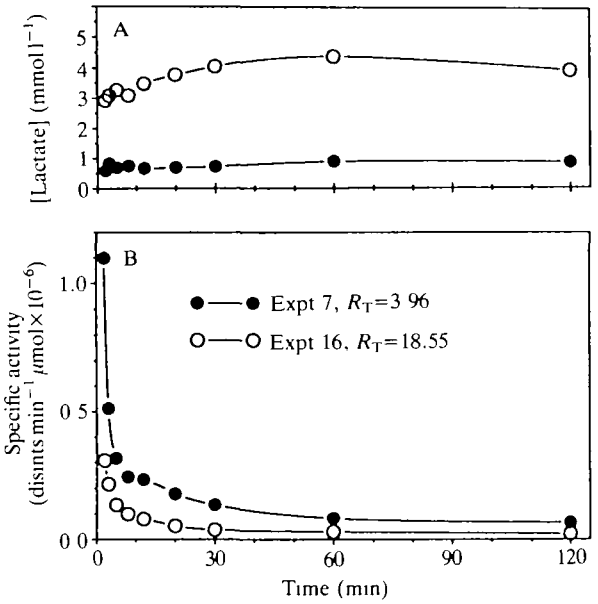


Fig. 2. Lactate concentration (A) and lactate specific activity decay curves (B) for two rainbow trout swimming at 1.5 body lengths $s^{-1}$, after injection of $23.18\times10^6$ disints $min^{-1}$ (○) and $18.47\times10^6$ disints $min^{-1}$ [U-$^{14}$C]lactate (●). The two individuals shown here had the weakest and the strongest responses to exercise (expts 7 and 16, respectively).

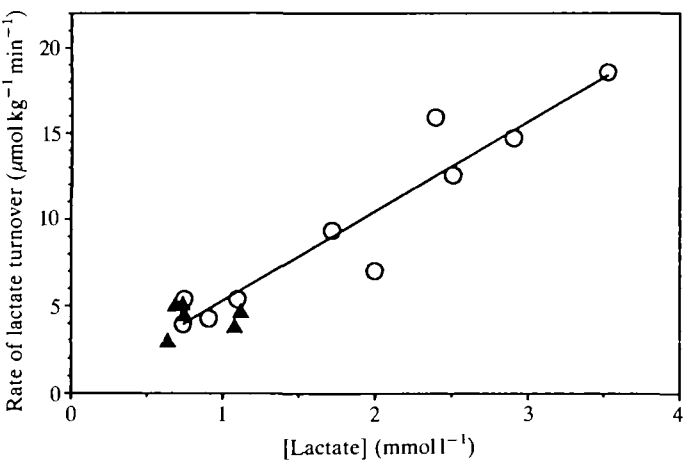


Fig. 3. Relationship between blood lactate concentration and lactate turnover rate in resting (▲) and swimming (○) rainbow trout. The line was fitted through values for swimming fish only.

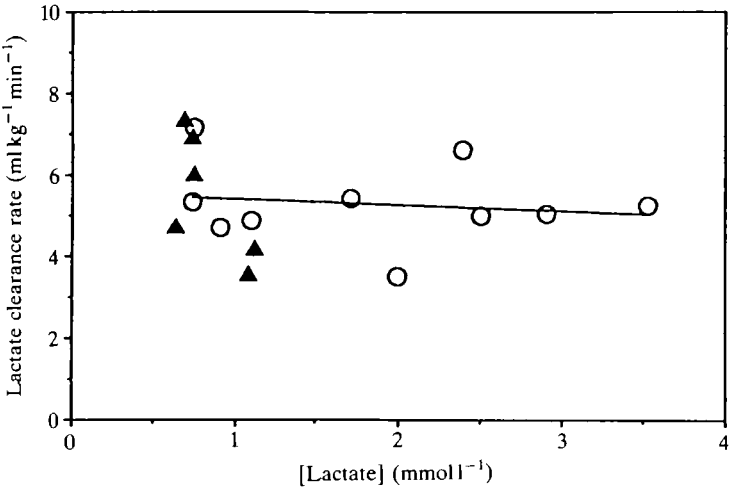


Fig. 4. Relationship between blood lactate concentration and lactate clearance rate in resting (▲) and swimming (○) rainbow trout. The line was fitted through values for swimming fish only.

clearance. The two variables were not significantly correlated (for swimming fish: $r$=0.15; slope=−0.16, not significantly different from 0, $P$>0.5).

Fig. 5 shows the relationship between body mass (in g) and whole-body lactate

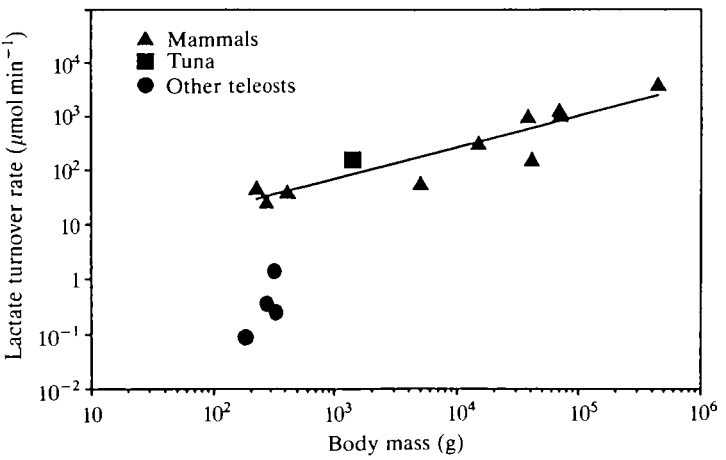


Fig. 5. Relationship between body mass and whole-body lactate turnover rate plotted on a double logarithmic scale for teleosts and mammals at rest. The line was fitted through mammalian values only. References for teleosts: American eel, Cornish and Moon (1985); salmon and flounder, Milligan and McDonald (1988); skipjack tuna, Weber *et al.* (1986); rainbow trout, this study. References for mammals: rat, Donovan and Brooks (1983), Donovan and Pagliassotti (1989) and Okajima *et al.* (1981); guinea pig, Freminet and Leclerc (1980); wallaby, Hochachka *et al.* (1985); dog, Issekutz *et al.* (1976); seal, Davis (1983); sheep, Reilly and Chandrasena (1977); human, Brooks *et al.* (1984) and Stanley *et al.* (1985); Thoroughbred racehorse, Weber *et al.* (1987*b*).

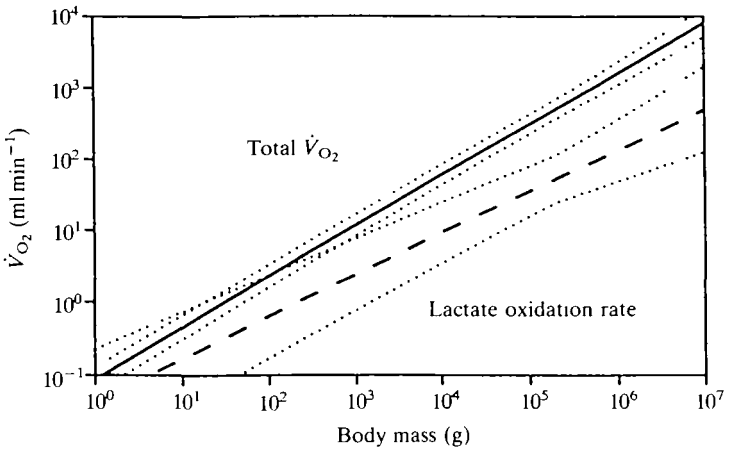


Fig. 6. Relationships between whole-body $\dot{V}_{O_2}$ (solid line; adapted from Schmidt-Nielsen, 1990), whole-body lactate oxidation (dashed line; same references as Fig. 5) and body mass plotted on a double logarithmic scale for mammals at rest. Fifty per cent of the lactate turned over was assumed to be oxidized in species where oxidation was not measured directly. Dotted lines represent 95 % confidence limits.

turnover rate (in $\mu$mol min$^{-1}$) for resting teleosts and mammals on double logarithmic coordinates. For resting mammalian species, the relationship was linear with a slope of 0.58 and an intercept of 0.11 ($r$=0.93; slope significantly different from 0 at $P$<0.0001).

Linear regressions for whole-organism $\dot{V}_{O_2}$ (in ml $O_2$ min$^{-1}$; from Schmidt-Nielsen, 1990) and for $\dot{V}_{O_2}$ accounted for by lactate oxidation (ml $O_2$ min$^{-1}$) *versus* body mass in resting mammalian species are presented in Fig. 6 on logarithmic coordinates. The slopes of these two regression lines are significantly different (0.75 *vs* 0.58, analysis of covariance, $P$<0.05).

## Discussion

### *Effect of prolonged exercise on lactate flux*

This study shows that the mean lactate turnover rate of trout only doubles between rest and prolonged swimming. Such a weak response is not due to their inability to augment rates of lactate production and utilization beyond a low maximal level. Indeed, much larger changes have been observed for salmonids and other teleosts in response to a variety of stresses. For example, prolonged starvation causes a 2.5-fold increase in the lactate turnover rate of American eel (Cornish and Moon, 1985), and resting trout subjected to environmental hypoxia show a sevenfold increase (Dunn and Hochachka, 1987). In addition, a ninefold change between rest and recovery from maximal exercise was measured in coho salmon (Milligan and McDonald, 1988). Stronger responses have also been observed in mammalian species at the same or lower exercise intensities [rats, threefold increase (Donovan and Brooks, 1983); humans and dogs, fourfold

increase (Stanley *et al.* 1985; Issekutz *et al.* 1976), Thoroughbred horses, sixfold increase (Weber *et al.* 1987*b*)].

The data also reveal that 4 out of 10 fish did not alter lactate flux between rest and prolonged exercise, even though they were perfectly capable of swimming at 85 % $U_{crit}$ for 3 h (Table 1, Fig. 3). These results demonstrate that the ability of rainbow trout to exercise strenuously for long periods does not depend on their capacity to metabolize lactate rapidly.

Undoubtedly, much bigger changes in lactate flux will occur at higher work rates than those used in the present study, when the animal reaches maximal oxygen consumption. At such exercise intensities, unfortunately, lactate metabolism gets out of steady state and the non-steady-state methods presently available (i.e. continuous tracer infusion and Steele equations; Steele, 1959) become unreliable for lactate and would be technically very difficult to use in a swimming fish. With the bolus injection technique performed here, turnover rate can only be measured accurately when the animal is in dynamic steady state, and when enough and timely blood samples are drawn to establish a reliable specific activity decay curve (Hetenyi *et al.* 1983). All these conditions were met in the present experiments, where lactate concentration was clearly in steady state for both resting and exercising animals (Figs 1 and 2, Table 1). Furthermore, concentration was maintained at a low level throughout the measurements, indicating that the fish were not particularly stressed by the experimental procedure.

Glucose homeostasis was also achieved in both resting and exercising animals, but the set point differed between individuals, as was observed in skipjack tuna (Weber *et al.* 1986). In trout, however, nutritional differences cannot explain such large interindividual differences in plasma glucose concentration and their cause remains unclear (Table 1).

The linear relationship between lactate concentration and turnover rate (Fig. 3) indicates that trout, like mammals (Weber *et al.* 1987*a*,*b*), use changes in plasma metabolite concentration to modulate turnover rate during exercise. In addition, rainbow trout were capable of maintaining a constant lactate clearance over a wide range of metabolic rates (Fig. 4).

### *Lactate as a metabolic fuel*

The simultaneous measurement of lactate turnover and lactate oxidation rates has only been performed in mammals where 50 % of the lactate turned over is oxidized at rest and 75 % during sustainable exercise (Donovan and Brooks, 1983; Issekutz *et al.* 1976; Mazzeo *et al.* 1986; Stanley *et al.* 1985). If one assumes that the same proportions of total lactate flux are oxidized in fish and mammals, lactate oxidation accounts for 25 % of the metabolic rate of rainbow trout at rest ($\dot{M}_{O_2}$=26 $\mu$mol $O_2$ $kg^{-1}$ $min^{-1}$ at 8°C, see Kiceniuk and Jones, 1977), but for only 15 % during exercise ($\dot{M}_{O_2}$=140 $\mu$mol $O_2$ $kg^{-1}$ $min^{-1}$, Kiceniuk and Jones, 1977). These calculated values are not unrealistic for fish because (1) another ectotherm, the lizard, also appears to oxidize 50 % of its lactate flux at rest (Gleeson and Dalessio, 1989), and (2) all animals measured show a large increase in percenta

Table 2. *Fraction of metabolic rate accounted for by lactate oxidation*

| | Assumed percentage of lactate turned over oxidized | | |
|---|---|---|---|
| | 50 | 75 | 100 |
| Rest ($N$=6) | 25.4±1.9 | 38.1±2.9 | 50.9±3.8 |
| Exercise ($N$=10) | 10.4±1.8 | 15.6±2.7 | 20.8±3.6 |

Values are means±S.E.M.

oxidation with exercise (Donovan and Brooks, 1983; Issekutz *et al.* 1976; Mazzeo *et al.* 1986; Stanley *et al.* 1985). Until lactate oxidation is measured directly in fish, however, the values calculated above must remain tentative. Table 2 shows the importance of lactate as an oxidative fuel in trout for different assumed values of percentage lactate flux oxidized. These calculations reveal that the fraction of the metabolic rate accounted for by lactate oxidation decreases between rest and exercise under all possible circumstances (i.e. even if 100 % of the lactate turned over is oxidized during exercise). In addition, the maximum possible contribution of lactate oxidation to metabolic rate during prolonged swimming is only 20 %.

### *Lactate exchange between white and red muscle*

The very small increase in turnover rate and the reduction in the contribution of lactate oxidation to $\dot{M}_{O_2}$ observed between rest and exercise are not compatible with the notion that lactate exchange between fiber types is an important mechanism for supplying oxidative fuel to the red lateral muscle. Indeed, a much greater increase in turnover would have occurred if such a shuttle mechanism for carbon fuel was used by trout during prolonged swimming. Metabolite movements between white and red fibers may be very difficult to achieve in fish, where the two muscle types are spatially separated and where diffusion distances for lactate would be very large, particularly through the relatively poorly perfused white muscle mass. In mammalian mixed muscle, in contrast, lactate exchange may be possible between adjacent glycolytic and oxidative fibers by simple diffusion without a need for circulatory transport (Brooks, 1985; Molé, 1983). This issue has been addressed very recently for resting mammalian muscle (Pagliassotti and Donovan, 1990). In these experiments, no evidence could be found to support the hypothesis that lactate exchange between fiber types of a mixed muscle bed actually takes place. Similar measurements for exercising muscles of mammals are not yet available, however, and the situation may be quite different during work. Further research is necessary to determine whether direct exchange without transit *via* the blood really occurs in contracting muscles.

### *Teleosts* versus *mammals: lactate metabolism and body size*

To compare turnover rates of fish and mammalian species, the effect of body

mass must be taken into account. Indeed, fluxes of metabolic substrates such as glucose vary with body size as does oxygen consumption (Weber *et al.* 1986). The known resting lactate turnover rates of teleosts and mammals were plotted on logarithmic coordinates as a function of body mass to provide a size-independent comparison (Fig. 5). The values reported here for trout were similar to those measured previously in other fish species, except tuna, but the teleost rates were 1–2 orders of magnitude lower than those of mammals of equivalent size. At present, it is not possible to determine whether the teleost and mammal lines are parallel, because the range of fish sizes for which data are available is too restricted. However, the pattern of differences in lactate turnover between mammals and fish reflects the well-known pattern of differences in metabolic rate (Schmidt-Nielsen, 1990). The ratios between lactate turnover and metabolic rate of teleosts and mammals of the same size are therefore very similar, implying that the metabolic role of lactate is equivalent in the two vertebrate groups.

Such an analysis also shows that mammalian lactate turnover, unlike glucose turnover (Weber *et al.* 1986), does not scale with body mass, as does metabolic rate. At the whole-organism level, the slope of the relationship for lactate turnover is much lower (0.58) than the slope of the classic relationship for metabolic rate (0.75). This discrepancy is shown in Fig. 6, where the regression lines for metabolic rate and lactate oxidation rate are plotted together. The two lines clearly diverge as body mass increases, indicating that lactate is a much more important oxidative substrate for small than for large species. A preliminary look at the limited data available for active animals suggests that the divergence between these two lines is even more accentuated than at rest (references are given in the legend to Fig. 5).

The relationship between body mass and mass-specific lactate oxidation has a negative exponent of −0.42. This is in sharp contrast with the positive values found for mass-specific muscle LDH activity in mammals (0.15; Emmett and Hochachka, 1981) and in pelagic fish, including rainbow trout (0.40; Somero and Childress, 1990). These positive values show that, per kilogram of muscle, large animals have a higher potential for maximal anaerobic work. Here, mass-specific lactate oxidation scales with a negative exponent like all other indices of aerobic potential (Emmett and Hochachka, 1981; Somero and Childress, 1990). In view of the above results, one can predict that LDH activity for the forward (pyruvate to lactate) and for the reverse (lactate to pyruvate) reactions may scale very differently with body mass. In contrast to pyruvate reduction, the scaling of LDH activity for lactate oxidation should have a negative exponent, particularly in oxidative muscle and in liver.

During this study, I was supported by a post-doctoral Fellowship from the Medical Research Council of Canada. I thank Peter Hochachka for financing these experiments through an NSERC grant, Dick Taylor for very helpful criticism of an early draft of the manuscript, and David Randall for lending the fish respirometer.

## References


BERGMEYER, H. U. (1974). *Methods of Enzymatic Analysis*, vol. 3. New York: Academic Press.

BEVER, K., CHENOWETH, M. AND DUNN, A. (1977). Glucose turnover in kelp bass (*Paralabrax* sp.): *in vivo* studies with [6-$^{3}$H, 6-$^{14}$C]glucose. *Am. J. Physiol.* **232**, R66-R72.

BONE, Q. (1978). Locomotor muscle. In *Fish Physiology*, vol. VII (ed. W. S. Hoar and D. J. Randall), pp. 407–408.

BRETT, J. R. (1964). The respiratory metabolism and swimming performance of young sockeye salmon. *J. Fish. Res. Bd Can.* **21**, 1183–1226.

BROOKS, G. A. (1985). Lactate: Glycolytic end product and oxidative substrate during sustained exercise in mammals – The 'lactate shuttle'. In *Comparative Physiology and Biochemistry – Current Topics and Trends*, vol. A, *Circulation, Respiration, and Metabolism* (ed. R. Gilles), pp. 208–218. Berlin: Springer-Verlag.

BROOKS, G. A., MAZZEO, R. S., SCHOELLER, D. A. AND BUDINGER, T. F. (1984). Lactate turnover in man during rest and graded exercise. *Med. Sci. Sports Exercise* **16**, 121.

CORNISH, I. AND MOON, T. W. (1985). Glucose and lactate kinetics in American eel *Anguilla rostrata*. *Am. J. Physiol.* **249**, R67–R72.

DAVIS, R. W. (1983). Lactate and glucose metabolism in the resting and diving harbor seal (*Phoca vitulina*). *J. comp. Physiol.* **153**, 275–288.

DONOVAN, C. M. AND BROOKS, G. A. (1983). Endurance training affects lactate clearance, not lactate production. *Am. J. Physiol.* **244**, E83–E92.

DONOVAN, C. M. AND PAGLIASSOTTI, M. J. (1989). Endurance training enhances lactate clearance during hyperlactatemia. *Am. J. Physiol.* **257**, E782–E789.

DUNN, J. F. AND HOCHACHKA, P. W. (1987). Turnover rates of glucose and lactate in rainbow trout during acute hypoxia. *Can. J. Zool.* **65**, 1144–1148.

ELDRIDGE, F. L. (1975). Relationship between turnover and blood concentration in exercising dogs. *J. appl. Physiol.* **39**, 231–234.

EMMETT, B. AND HOCHACHKA, P. W. (1981). Scaling of oxidative and glycolytic enzymes in mammals. *Respir. Physiol.* **45**, 261–272.

FREMINET, A. AND LECLERC, L. (1980). Effect of fasting on glucose, lactate, and alanine turnover in rats and guinea-pigs. *Comp. Biochem. Physiol.* B **65**, 363–367.

GLEESON, T. T. AND DALESSIO, P. M. (1989). Lactate and glycogen metabolism in the lizard *Dipsosaurus dorsalis* following exhaustive exercise. *J. exp. Biol.* **144**, 377–393.

HETENYI, G. JR, PEREZ, G. AND VRANIC, M. (1983). Turnover and precursor–product relationships of nonlipid metabolites. *Physiol. Rev.* **63**, 606–667.

HOCHACHKA, P. W., RUNCIMAN, W. B. AND BAUDINETTE, R. V. (1985). Why exercising tammar wallabies turn over lactate rapidly: implications for models of mammalian exercise metabolism. *Molec. Physiol.* **7**, 17–28.

HUDSON, R. C. L. (1973). On the function of the white muscles in teleosts at intermediate swimming speeds. *J. exp. Biol.* **58**, 509–522.

ISSEKUTZ, B., JR, SHAW, W. A. S. AND ISSEKUTZ, A. C. (1976). Lactate metabolism in resting and exercising dogs. *J. appl. Physiol.* **40**, 312–319.

JORFELDT, L. (1970). Metabolism of L(+)-lactate in human skeletal muscle during exercise. *Acta physiol. scand. (Suppl.)* **338**, 1–67.

KATZ, J., OKAJIMA, F., CHENOWETH, M. AND DUNN, A. (1981). The determination of lactate turnover *in vivo* with $^{3}$H- and $^{14}$C-labelled lactate. *Biochem. J.* **194**, 513–524.

KICENIUK, J. W. AND JONES, D. R. (1977). The oxygen transport system in trout (*Salmo gairdneri*) during sustained exercise. *J. exp. Biol.* **69**, 247–260.

MACHADO, C. R., GAROFALO, M. A. R., ROSELINO, J. E. S., KETTLEHUT, I. C. AND MIGLIORINI, R. H. (1989). Effect of fasting on glucose turnover in a carnivorous fish (*Hoplias* sp.). *Am. J. Physiol.* **256**, R612–R615.

MAZZEO, R. S., BROOKS, G. A., SCHOELLER, D. A. AND BUDINGER, T. F. (1986). Disposal of blood [1-$^{13}$C]lactate in humans during rest and exercise. *J. appl. Physiol.* **60**, 232–241.

MILLIGAN, C. L. AND MCDONALD, D. G. (1988). *In vivo* lactate kinetics at rest and during recovery from exhaustive exercise in Coho salmon (*Oncorhynchus kisutch*) and starry flounder (*Platichthys stellatus*). *J. exp. Biol.* **135**, 119–131.

MOLÉ, P. A. (1983). Exercise metabolism. In *Exercise Medicine: Physiological Principles and*

*Clinical Applications* (ed. A. A. Bove and D. T. Lowenthal), chapter 4, pp. 43–88. New York: Academic Press.

OKAJIMA, F., CHENOWETH, M., ROGNSTAD, R., DUNN, A. AND KATZ, J. (1981). Metabolism of $^{3}H$- and $^{14}C$-labelled lactate in starved rats. *Biochem. J.* **194**, 525–540.

PAGLIASSOTTI, M. J. AND DONOVAN, C. M. (1990). Influence of cell heterogeneity on skeletal muscle lactate kinetics. *Am. J. Physiol.* **258**, E625–E634.

QUINN, T. P. (1988). Estimated swimming speeds of migrating adult sockeye salmon. *Can. J. Zool.* **66**, 2160–2163.

REILLY, P. E. B. AND CHANDRASENA, L. G. (1977). Sheep lactate entry-rate measurements: error due to sampling jugular blood. *Am. J. Physiol.* **233**, E138–E140.

SCHMIDT-NIELSEN, K. (1990). *Animal Physiology*, fourth edition, pp. 192–198. Cambridge, New York, Melbourne: Cambridge University Press.

SOMERO, G. N. AND CHILDRESS, J. J. (1990). Scaling of ATP-supplying enzymes, myofibrillar proteins and buffering capacity in fish muscle: relationship to locomotory habit. *J. exp. Biol.* **149**, 319–333.

STANLEY, W. C., GERTZ, E. W., WISNESKI, J. A., MORRIS, L. D., NEESE, R. A. AND BROOKS, G. A. (1985). Systemic lactate kinetics during graded exercise in man. *Am. J. Physiol.* **249**, E595–E602.

STEELE, R. (1959). Influences of glucose loading and of injected insulin on hepatic glucose output. *Ann. N.Y. Acad. med. Sci.* **82**, 420–430.

WALTON, M. J. AND COWEY, C. B. (1982). Aspects of intermediary metabolism in salmonid fish. *Comp. Biochem. Physiol.* **73B**, 59–79.

WEBER, J.-M. (1988). Design of exogenous fuel supply systems: adaptive strategies for endurance locomotion. *Can. J. Zool.* **66**, 1116–1121.

WEBER, J.-M., BRILL, R. W. AND HOCHACHKA, P. W. (1986). Mammalian metabolite flux rates in a teleost: lactate and glucose turnover in tuna. *Am. J. Physiol.* **250**, R452–R458.

WEBER, J.-M., DOBSON, G. P., PARKHOUSE, W. S., WHEELDON, D., HARMAN, J. C., SNOW, D. H. AND HOCHACHKA, P. W. (1987*a*). Cardiac output and oxygen consumption in exercising Thoroughbred horses. *Am. J. Physiol.* **253**, R890–R895.

WEBER, J.-M., PARKHOUSE, W. S., DOBSON, G. P., HARMAN, J. C., SNOW, D. H. AND HOCHACHKA, P. W. (1987*b*). Lactate kinetics in exercising Thoroughbred horses: regulation of turnover rate in plasma. *Am. J. Physiol.* **253**, R896–R903.

WITTENBERGER, C., COPREAN, D. AND MORAR, L. (1975). Studies on the carbohydrate metabolism of the lateral muscles in carp (influence of Phloridzin, insulin and adrenaline). *J. comp. Physiol.* **101**, 161–172.

WOKOMA, A. AND JOHNSTON, I. A. (1981). Lactate production at high sustainable cruising speeds in rainbow trout (*Salmo gairdneri* Richardson). *J. exp. Biol.* **90**, 361–364.

WOLF, K. (1963). Physiological salines for fresh-water teleosts. *Progve Fish Cult.* **25**, 135–140.