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OXYGEN CONSUMPTION AND SWIMMING PERFORMANCE IN HYPOXIA-ACCLIMATED RAINBOW TROUT *SALMO GAIRDNERI*

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SUMMARY

1. Swimming performance and oxygen consumption of normoxic (control) and hypoxia-acclimated (P_{O_2} = 40 mmHg) rainbow trout, *Salmo gairdneri* Richardson, were monitored at >145, 60 and 40 mmHg.

2. Maximum swimming velocity at 40 mmHg was reduced from >54·8 cm s^{-1} to 41·4 cm s^{-1} in controls and to 40·6 cm s^{-1} in hypoxia-acclimated fish.

3. Normoxic oxygen consumption of control fish ranged from 97·5 mg O_2 kg^{-1} h^{-1} (5·5 cm s^{-1}) to 318·5 mg O_2 kg^{-1} h^{-1} (54·8 cm s^{-1}) and did not differ significantly from that of hypoxia-acclimated fish in normoxia.

4. Reduction of ambient P_{O_2} from normoxia to 60 mmHg or 40 mmHg did not significantly change oxygen consumption in control animals, although no fish (control or hypoxia acclimated) completed swimming trials at 54·8 cm s^{-1} in 40 mmHg.

5. Oxygen consumption of hypoxia-acclimated fish at 5·5 cm s^{-1} and 40 mmHg was significantly higher than oxygen uptake in normoxia at the same speed. This relative increase was not maintained, however, as oxygen consumption at higher swimming speeds was similar to that in normoxia.

6. Blood studies showed that hypoxia-acclimated fish had lower ATP concentrations and P_{50} values. While these factors may increase the blood oxygen loading capacity, the change is apparently not enough markedly to improve swimming performance or oxygen consumption in hypoxia and/or exercise.

INTRODUCTION

Fish swimming at high sustained speeds may experience an oxygen limiting situation similar to that incurred during ambient hypoxia. At swimming velocities approaching the critical swimming speed, the metabolic demand for oxygen may be greater than can be provided by ventilatory and circulatory systems (Jones & Randall, 1978). When hypoxia and exercise are combined, this oxygen limitation must be enhanced. Jones (1971) demonstrated that either moderate hypoxia or anaemia resulted

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in a reduction of critical swimming speed in trout. Presumably, hypoxia and exercise together act to reduce the ability of the gills to take up oxygen, while exercise and anaemia will affect the oxygen exchange as well as the oxygen transport system. Hypoxia-acclimated fish have been demonstrated to have blood respiratory properties resulting in improved oxygen loading capabilities (Wood & Johansen, 1972). It is not known whether this property is advantageous during exercise in normoxic or hypoxic water. The aim of this study was to determine whether hypoxia acclimation of trout results in an improved ability to maintain oxygen consumption and swimming performance, compared to normoxic fish, when both groups are tested at variable ambient P_{O_2} and swimming speed in a specially designed swimming respirometer. A similar study on goldfish (Kutty, 1968) demonstrated that hypoxia acclimation had no significant influence on swimming performance.

MATERIALS AND METHODS

Animals

Rainbow trout (*Salmo gairdneri* Richardson) of 250–350 g weight and 29–30 cm total length, were used. Fish were held in several well-aerated tanks at 15 °C and fed trout pellets regularly. Three days before the first swimming trial, 10 fish were selected and moved to a smaller aquarium under similar conditions, but not fed. A separate group of 10 fish was acclimated to hypoxia in an aquarium in which P_{O_2} of the water was controlled (±3 mmHg) by an oxygen regulating system (Wood, Johansen & Weber, 1975). Oxygen tension was lowered over a 4-day period to the final oxygen tension of 40 mmHg. Fish were acclimated to stable conditions for at least 2 weeks before the first experimental run was commenced. Feeding was also ended 3 days prior to the first run for the hypoxia-acclimated fish.

Oxygen consumption measurement

Oxygen consumption ($\dot{V}_{O_2}$) was measured in a Brett-type tunnel respirometer, previously described by Christiansen, Lomholt & Johansen (1982). Oxygen tension, temperature and water velocity in the respirometer were monitored and recorded on magnetic tape by a Hewlett-Packard 9825 A calculator. Temperature and P_{O_2} were also recorded on a Hewlett-Packard 7132 A two-channel chart recorder. The HP-computer was programmed automatically to sample P_{O_2}-data, and control and shift between preset water P_{O_2} values.

Oxygen tension of the water was measured continuously by a Radiometer oxygen electrode (E-5046) mounted in a cuvette (D616) and thermostatted at the experimental temperature of 15 °C. A constant flow past the electrode was maintained by an Ismatec Mini-s roller pump mounted downstream of the cuvette. The oxygen electrode was repeatedly calibrated with zero P_{O_2} solution and air-saturated water.

Oxygen tension in the water feeding the respirometer section of the swimtunnel was maintained and controlled by shunting a fraction of the water leaving this section through a Sci-Med Kolobow membrane gas exchanger (lung) with a 3·5 m^2 surface area.

During measurement periods, water and air flow through the external gas exchanger were halted, and oxygen tension recorded every 30 s for 40 min. A linear regression

was performed by the HP-computer of the time course of the P_{O_2} change, and the slope of the line ($\Delta P_{O_2}/\Delta t$) determined in this manner was used to calculate oxygen consumption according to the equation:

$$\dot{V}_{O_2} = V \cdot \Delta P_{O_2}/\Delta t \cdot \alpha \cdot bw^{-1}.$$

Oxygen tension in the water passing the respirometer section of the swimtunnel was not allowed to fall more than 20 mmHg during swimming runs in normoxia and below 30 mmHg during the hypoxic tests.

Experimental protocol

$\dot{V}_{O_2}$ of control or normoxia-adapted fish was determined at four water speeds (5, 20, 35 and 50 cm s^{-1}) and three oxygen tensions, normoxia (145 mmHg) and hypoxia (40 and 60 mmHg).

Swimming velocities of fish in the respirometer were corrected for solid blocking effects of fish body cross-sectional area (Webb, 1975). Actual swimming velocities at the four pre-selected water speeds were therefore 5·5, 21·9, 38·3 and 54·8 cm s^{-1}. These corrected swimming velocities have been used in all further discussions.

The fish to be tested was acclimated overnight to the swimming chamber at the first trial swimming speed of 5·5 cm s^{-1}. Oxygen tension was kept higher than 145 mmHg throughout this period. $\dot{V}_{O_2}$ was measured by starting at the lowest swimming speed (5·5 cm s^{-1}) and increasing the speed in a stepwise manner to 54·8 cm s^{-1}. In order to ensure that oxygen consumption was at a steady state, the fish were made to swim for 30 min at the chosen swimming speed before $\dot{V}_{O_2}$ measurements began. At this time, air and water flow through the gas exchanger was stopped and the fish allowed to swim for an additional 40 min to measure $\dot{V}_{O_2}$. Water speed was then increased, and P_{O_2} was brought back to the level of 145 mmHg. At the conclusion of measurements at 54·8 cm s^{-1} in normoxia, swimming speed was reduced to 5·5 cm s^{-1} whereafter normoxic conditions were maintained for 30 min. P_{O_2} was then lowered over the next 30 min by flushing the membrane gas exchanger with nitrogen. When P_{O_2} had stabilized at the pre-determined oxygen tension (40 or 60 mmHg) for 5 min, $\dot{V}_{O_2}$ measurements were again started. Oxygen tension during the 30 min steady state swimming period was kept at the hypoxic test condition. In the case of animals swimming at 40 mmHg, P_{O_2} was raised to 45 mmHg 3–4 min before $\dot{V}_{O_2}$ measurements began, and was never allowed to fall below 30 mmHg during this phase. An animal was judged to have collapsed when it could no longer keep itself off the electrified grid at the rear of the swimming chamber.

In a separate experiment to measure possible oxygen debt, fish from the control group were put through the same experimental protocol used to measure $\dot{V}_{O_2}$ at the four swimming speeds in normoxia. At the conclusion of the swimming trial at 54·8 cm s^{-1}, water velocity was reduced to 5·5 cm s^{-1} and $\dot{V}_{O_2}$ monitored for the next hour.

Hypoxia-acclimated fish were tested in a manner similar to control animals (normoxia-acclimated). During overnight acclimation to the chamber, P_{O_2} was kept at 40 mmHg by the oxygen regulating system that controlled water and air flow through the membrane gas exchanger. At the end of this period, oxygen consumption was measured at the acclimation P_{O_2} (40 mmHg) and swimming speed (5·5 cm s^{-1}).

Oxygen tension was then raised to air saturation and $\dot{V}_{O_2}$ measurements in normoxia were begun at 5·5 cm s^{-1} and proceeded through the four speeds in normoxia and hypoxia. Therefore, unlike the control group, two measurements of $\dot{V}_{O_2}$ at 5·5 cm s^{-1} in hypoxia were made for the hypoxia-acclimated group. The first, taken at the conclusion of the overnight chamber acclimation and before P_{O_2} was raised for normoxia swimming trials, will be referred to in the text and table as 'prenormoxia' $\dot{V}_{O_2}$. The second was made when P_{O_2} was reduced to hypoxic levels at the beginning of hypoxia trials and is similar in its temporal relationship to the measurement made for control animals.

Maximum swimming velocity (V_{max}) was calculated for all fish failing to swim at the highest velocity. An interpolation was performed to determine swimming velocity if the fish did not collapse exactly at the beginning or end of the swimming period by adjusting the last velocity increment in proportion to the amount of time spent at that velocity (Brett, 1964; Webb, 1971).

V_{max} for fish completing trials at 54·8 cm s^{-1} could not be calculated since maximum swim-velocity was never attained due to water speed limitations of the respirometer. For the purpose of calculation and discussion, however, V_{max} for these animals was taken to be 54·8 cm s^{-1}. V_{max} reported for conditions at which some fish completed all trials (normoxia and 60 mmHg) will therefore probably be underestimates of the true maximum swimming velocity (critical swimming speed).

Blood respiratory properties

For blood sampling, fish were netted and held between two wet towels to facilitate rapid (30 s) sampling. A heparinized plastic syringe was used to draw approximately 2 ml blood, percutaneously from a caudal vessel. Blood Hct, pH, oxygen affinity (P_{50}), ATP and GTP concentrations were determined as described by Tetens & Lykkeboe (1981).

Statistics

Statistical analysis of results was based on the Student's *t*-test. All values in the text, table, and figures are given as mean ± standard deviation.

RESULTS

Results of oxygen consumption determinations in normoxia (control) and hypoxia-acclimated fish in normoxia and hypoxia are summarized in Table 1 and Figs 1 and 2. Neither group of fish had any apparent difficulty completing swimming trials through all four velocities in normoxia. Differences in swimming performance began to emerge in hypoxia trials. At water O_2 tensions of 60 mmHg, three out of eight control fish could not complete the test at 54·8 cm s^{-1}. Swimming trials at a water P_{O_2} of 40 mmHg resulted in two out of eight fish failing at 38·3 cm s^{-1}, while all failed at 54·8 cm s^{-1}. All eight hypoxia-acclimated fish successfully completed the 38·3 cm s^{-1} trial in hypoxia (40 mmHg), but none were successful at 54·8 cm s^{-1}.

Maximum swimming velocity, shown in Table 1, in control fish decreased from a value of $\geq$54·8 cm s^{-1} in normoxia to 51·5 ± 6·0 cm s^{-1} at 60 mmHg. A further reduction of P_{O_2} to 40 mmHg resulted in a decrease of critical swimming speed to

Table 1. *Oxygen consumption and maximum swimming velocity of normoxic (control) and hypoxia-acclimated rainbow trout in normoxia and at two levels of hypoxia*

Swimming velocity ($cm\,s^{-1}$)	Oxygen consumption ($mg\,O_2\,kg^{-1}\,h^{-1}$)				
	Normoxia		60 mmHg	40 mmHg	
	Hypoxia-acclimated	Control	Control	Control	Hypoxia-acclimated
0·0*	66·75 ± 16·2 (8)	82·48 ± 27·4 (15)	96·69 ± 25·1 (8)	97·14 ± 20·9 (8)	105·04 ± 24·0† (8)
					124·87 ± 30·5 (8)
5·5	75·91 ± 14·6 (8)	97·51 ± 36·7 (15)	115·11 ± 21·7 (8)	111·54 ± 21·8 (8)	110·07 ± 20·9‡ (8)
					142·71 ± 35·9 (8)
21·9	131·78 ± 19·4 (8)	129·76 ± 32·7 (15)	132·30 ± 39·3 (8)	135·31 ± 22·6 (8)	150·66 ± 26·4 (8)
38·3	217·49 ± 23·8 (8)	220·10 ± 45·5 (15)	200·64 ± 46·9 (8)	213·92 ± 36·5 (6)	227·01 ± 37·2 (8)
54·8	345·75 ± 73·2 (7)	318·50 ± 71·0 (15)	278·44 ± 44·7 (5)	—	—
	Maximum swimming velocity ($cm\,s^{-1}$)				
	≥ 54·8 (8)	≥ 54·8 (15)	51·5 ± 6·0 (8)	41·4 ± 5·7 (8)	40·6 ± 2·2 (8)

* Extrapolated from linear regression.
† Calculated using 'prenormoxia' value.
‡ 'Prenormoxia' value.
Values given are mean ± S.D. Number of fish in parentheses.

41·4 ± 5·7 $cm\,s^{-1}$, which was significantly lower ($P < 0·001$) than V_{max} recorded at 60 mmHg. Results were similar for hypoxia-acclimated animals. V_{max} fell from ≥54·8 $cm\,s^{-1}$ in normoxia to 40·6 ± 2·2 $cm\,s^{-1}$ at 40 mmHg.

Standard metabolic rates (SMR) listed in Table 1 were calculated from linear regressions of data from individual fish. SMR of control fish was 82·5 ± 27·4 $mg\,O_2\,kg^{-1}\,h^{-1}$ and did not change significantly when P_{O_2} was lowered to 40 mmHg. Hypoxia-acclimated animals in normoxia had lower, but insignificantly different SMRs. Reduction of P_{O_2} during swimming trials in this group increased SMR by 87% ($P < 0·001$) to 124·87 ± 30·5 $mg\,O_2\,kg^{-1}\,h^{-1}$, which is significantly higher ($P < 0·05$) than control fish at 40 mmHg. Recalculation of SMR in hypoxia-acclimated fish in hypoxia, utilizing the 'prenormoxia' $\dot{V}_{O_2}$ values measured at 5·5 $cm\,s^{-1}$, considerably alters the slope of the regressions and lowers the SMR to 105·04 ± 24·0 $mg\,O_2\,kg^{-1}\,h^{-1}$. This is not significantly different from controls in hypoxia, but is still significantly higher than SMR of hypoxia-acclimated fish in normoxia ($P < 0·001$).

Oxygen consumption of control fish in normoxia averaged 97·5 $mg\,O_2\,kg^{-1}\,h^{-1}$ at 5·5 $cm\,s^{-1}$ and increased to 318·5 $mg\,O_2\,kg^{-1}\,h^{-1}$ at the highest velocity (Fig. 1).

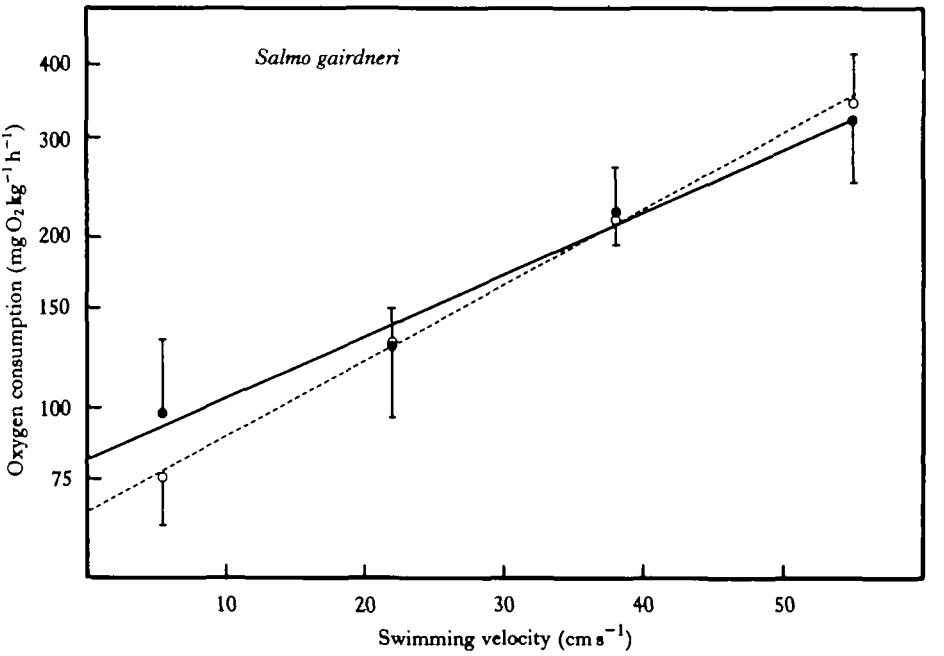

Fig. 1. Oxygen consumption *versus* swimming velocity of normoxic (control ●) and hypoxia-acclimated (40 mmHg ○) fish in normoxia (150 mmHg).

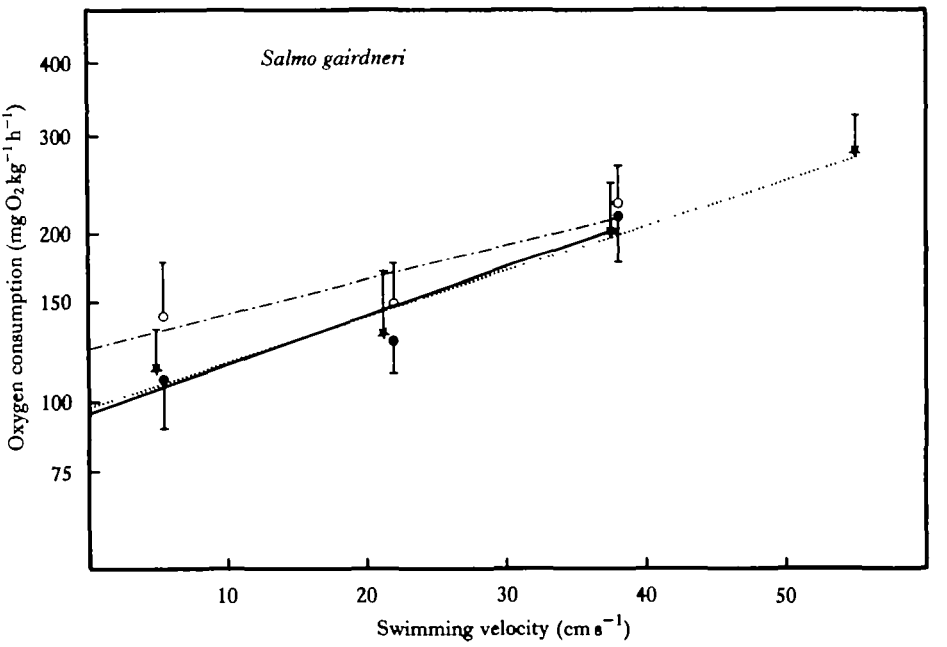

Fig. 2. Oxygen consumption *versus* swimming velocity of normoxic-acclimated (control) fish in water P_{O_2} of 60 (*) and 40 mmHg (●), and of hypoxia-acclimated fish in water P_{O_2} of 40 mmHg (○).

Swimming trials at 60 and 40 mmHg resulted in $\dot{V}_{O_2}$ values that were not significantly different from those recorded in normoxia (Fig. 2).

The 'prenormoxia' oxygen consumption of hypoxia-acclimated fish at 40 mmHg ($5{\cdot}5\,cm\,s^{-1}$) was $110{\cdot}1\,mg\,O_2\,kg^{-1}\,h^{-1}$ and fell significantly ($P<0{\cdot}005$) to $75{\cdot}9\,mg\,O_2\,kg^{-1}\,h^{-1}$ when P_{O_2} was raised to normoxia. Increasing the swimming velocity in normoxia to $54{\cdot}8\,cm\,s^{-1}$ increased $\dot{V}_{O_2}$ to an average of $345{\cdot}7\,mg\,O_2\,kg^{-1}\,h^{-1}$ (Fig. 2). There were no significant differences in normoxic $\dot{V}_{O_2}$ values between hypoxia-acclimated fish and controls swimming at any swimming speed.

Reduction of P_{O_2} from normoxia to 40 mmHg following normoxic swimming trials, resulted in an 88% increase in $\dot{V}_{O_2}$ in the hypoxia-acclimated animals at $5{\cdot}5\,cm\,s^{-1}$. This differed significantly ($P<0{\cdot}001$) from $\dot{V}_{O_2}$ measured in normoxia, as well as $\dot{V}_{O_2}$ measured under similar hypoxic conditions before normoxia swimming trials during the 'prenormoxia' phase ($P<0{\cdot}025$). Oxygen consumption increased through the remaining two swimming speeds, but did not differ significantly from $\dot{V}_{O_2}$ in normoxia or $\dot{V}_{O_2}$ of control animals in normoxia or hypoxia (Fig. 2).

No oxygen debt resulting from the swimming protocol in normoxia was noted in the $\dot{V}_{O_2}$ values. Oxygen consumption prior to swimming trials was $99{\cdot}2 \pm 24{\cdot}4\,mg\,O_2\,kg^{-1}\,h^{-1}$ ($5{\cdot}5\,cm\,s^{-1}$). Following the conclusion of swimming at the highest water velocity, $\dot{V}_{O_2}$ at $5{\cdot}5\,cm\,s^{-1}$ had returned to $99{\cdot}3 \pm 23{\cdot}6\,mg\,O_2\,kg^{-1}\,h^{-1}$ within 30 min and was $94{\cdot}7\,mg\,O_2\,kg^{-1}\,h^{-1}$ after 60 min.

Blood respiratory properties

Haematocrit and blood O_2 capacity in normoxia and hypoxia-acclimated trout, were not significantly different: average heamatocrits were $26{\cdot}3 \pm 6{\cdot}5$% and $25{\cdot}2 \pm 6{\cdot}0$% and O_2 capacities $9{\cdot}25 \pm 2{\cdot}28$ vol% and $8{\cdot}69 \pm 3{\cdot}37$ vol%, respectively, for normoxia- and hypoxia-acclimated fish. Red cell ATP concentration was, however, markedly higher in the normoxic group, averaging $4{\cdot}86 \pm 0{\cdot}54\,mmol\,l^{-1}$ packed red blood cells against $2{\cdot}51 \pm 0{\cdot}56\,mmol\,l^{-1}$ packed red blood cells ($P<0{\cdot}001$). Blood O_2 affinity was elevated in hypoxic fish expressed by a P_{50} value of 19·5 mmHg compared to 22·9 mmHg in normoxia-acclimated fish. The Bohr factor and n-value did not vary between the groups: $\phi = -0{\cdot}49$ and $n = 1{\cdot}92 \pm 0{\cdot}25$ in normoxic fish, and $\phi = -0{\cdot}45$, $n = 2{\cdot}34 \pm 0{\cdot}45$ in hypoxia-acclimated fish.

DISCUSSION

The maximum velocity a fish can maintain for a precise period of time is termed the critical swimming speed (Brett, 1964), and will vary with the magnitude of the speed increment and the amount of time spent at each speed. Usually, a decrease in test period duration will result in an increase in critical swimming speed (Webb, 1971).

These factors, in combination with differing acclimation temperatures and size of fish will result in a wide range of values. Critical swimming speeds are, however, a useful way of quantifying swimming performance in the same species of fish. It should be pointed out that since all of the fish in this study completed trials at the highest swimming speeds in normoxia, V_{max} for these fish will not be as high as their critical swimming speeds. However, in cases where fish were unable to complete the swimming trials (40 mmHg), V_{max}, by definition, will be the same as the critical swimming

speed. Critical swimming speeds reported for rainbow trout at 15 °C (1-h swimming periods) range between 39·2 cm s^{-1} (4·3 L s^{-1}) (Beamish, 1978) and 58·1 cm s^{-1} (2·0 L s^{-1}) (Webb, 1971). Our study showed that V_{max} for rainbow trout in normoxia is at least 54·8 cm s^{-1} (1·8 L s^{-1}), since no fish failed to complete the swimming trial at this speed. This is in agreement with values of 58·1 cm s^{-1} (2 L s^{-1}) published by Webb (1971) using fish of similar size.

Reduction of ambient P_{O_2} decreased V_{max} to 51·5 cm s^{-1} (1·7 L s^{-1}) at 60 mmHg and 41·4 cm s^{-1} (1·4 L s^{-1}) at 40 mmHg in control fish. A decrease in critical swimming speeds with severity of hypoxia has earlier been noted in bass (*Micropterus salmoides*), coho salmon (*Oncorhyncus nerka*), goldfish (*Carassius auratus*) and rainbow trout (*Salmo gairdneri*) (Kutty, 1968; Dahlberg, Shumway & Doudoroff, 1968; Jones, 1971). Hypoxia acclimation in the present study had no effect on V_{max}. Kutty (1968) found the same to be true of hypoxia-acclimated goldfish.

The failure of fish to swim at reduced P_{O_2} was attributed by Kutty (1968) to be due to a peripheral or central oxygen sensing system, not exhaustion, since fish resumed steady state swimming when P_{O_2} was raised. This contention is not supported by our experiments on rainbow trout. Following collapse in hypoxia, several fish had difficulty orientating themselves in a normal swimming posture even though water velocity had been reduced and ambient P_{O_2} increased above 100 mmHg. On two occasions, $\dot{V}_{O_2}$ was measured continuously following collapse in hypoxia. Oxygen uptake of these fish in normoxia was elevated above pre-exercise values, in one case by 85 % and by 200 % in another. These observations suggest that fish were making maximal effort under the experimental conditions and ceased swimming due to exhaustion.

Standard metabolic rate (SMR) extrapolated for control fish in normoxia (82·5 mg O_2 kg^{-1} h^{-1}) falls within the range of 72·5 mg O_2 kg^{-1} h^{-1} (Webb, 1971) to 112 mg O_2 kg^{-1} h^{-1} (Rao, 1968) reported for rainbow trout at the same temperature. SMR of hypoxia-acclimated fish was somewhat lower. As severity of hypoxia increased, SMR of control animals remained stable. This was not the case in hypoxia-acclimated animals, since a significant increase in SMR was noted at water P_{O_2} of 40 mmHg. Ott, Heisler & Ultsch (1980) also noted a similar increase in non-acclimated rainbow trout during hypoxic exposure. In our experiments, SMR in control fish remained stable at reduced water P_{O_2} of 40 mmHg. According to Ott *et al.* (1980) SMR in rainbow trout increased between 20 and 100 % at this P_{O_2} before falling when the ambient P_{O_2} was lowered below the critical point (22 mmHg at 15 °C) and oxygen uptake became dependent on P_{O_2}. It is difficult to understand, however, why in our study the increase in SMR was apparent only in the hypoxia-acclimated fish and not in the control group.

Oxygen consumption of normoxia-acclimated fish at each swimming speed remained remarkably independent of environmental P_{O_2}. This is, however, in agreement with previous work on less or minimally active rainbow trout exposed to hypoxic water levels of 90 mmHg (Smith & Jones, 1982) and 30 mmHg (Holeton & Randall, 1967). It contrasts, however, with the large increase in $\dot{V}_{O_2}$ resulting from a decreased ambient P_{O_2} (90 mmHg) which was reported by Hughes & Saunders (1970).

Oxygen uptake of hypoxia-acclimated trout in normoxia was similar to that of control fish at all swimming speeds in normoxia. With the onset of hypoxia, a significant increase ($P < 0·001$) in oxygen uptake at 5·5 cm s^{-1} was recorded but did

not persist at the remaining swimming speeds since $\dot{V}_{O_2}$ at 21·8 cm s^{-1} and 38·3 cm s^{-1} were the same as in normoxia. $\dot{V}_{O_2}$ measured in hypoxia (P_{O_2} = 40 mmHg) at the lowest swimming speed could increase because of an oxygen debt incurred during the preceding swimming trials in normoxia. We believe, however, this to be unlikely since there was no significant elevation of $\dot{V}_{O_2}$ in control fish when swimming in 60 or 40 mmHg at 5·5 cm s^{-1}. This was further substantiated by the results of tests conducted to determine the possibility of an oxygen debt resulting from the experimental protocol followed in the normoxia experiments. These tests showed that oxygen consumption in normoxia, measured at 5·5 cm s^{-1} before and after the swimming protocol used in the experiment, were the same. Finally, a similar change in $\dot{V}_{O_2}$ was noted when 'prenormoxia' $\dot{V}_{O_2}$ was compared to normoxic $\dot{V}_{O_2}$. In this case, when fish were taken from an ambient P_{O_2} of 40 mmHg to normoxic levels, rather than *vice versa*, $\dot{V}_{O_2}$ decreased 31 %. Despite the opposite direction in the change of ambient P_{O_2}, in both cases $\dot{V}_{O_2}$ was higher in hypoxia than in normoxia. An increased $\dot{V}_{O_2}$ of hypoxia-acclimated fish in hypoxia at low levels of activity, relative to non-acclimated fish, was reported in carp (Lomholt & Johansen, 1979) and flounder (Kerstens, Lomholt & Johansen, 1979). Oxygen consumption of acclimated carp was 40 % higher while acclimated flounders maintained $\dot{V}_{O_2}$ at 100 % above controls in hypoxia. It should be noted, however, that the increased $\dot{V}_{O_2}$ values were significantly lower than $\dot{V}_{O_2}$ recorded in normoxia, even though ambient P_{O_2} was above the critical P_{O_2} of 21 mmHg earlier reported for carp (Ott *et al.* 1980).

The acclimation history of the fish in our study may have influenced the results by giving an excitement or stress factor not present in control animals. Hypoxia-acclimated animals experienced two dramatic changes in P_{O_2} during the course of the experiment. Following 3 weeks of acclimation to P_{O_2} of 40 mmHg, water tension was raised to air saturation and returned to 40 mmHg 4 h later. Such changes in P_{O_2} of water may affect the physiological responses (blood O_2 transport properties) of hypoxia-acclimated fish differently from fish facing a single change in ambient water conditions (Soivio & Nikinmaa, 1981).

An increased oxygen uptake in response to decreasing P_{O_2} recorded in lamprey, carp, goldfish, brook trout and rainbow trout has been attributed by many authors to increased cost of ventilation during certain phases of exposure to hypoxia (Beamish, 1964; Marvin & Heath, 1968; Hughes & Saunders, 1970; Nikinmaa & Weber, 1984). Gill water flow ($\dot{V}_G$) measured directly in trout (Randall & Jones, 1973; Smith & Jones, 1982) was shown to double when environmental P_{O_2} was reduced from normoxia to 90 mmHg, while a reduction to 40 mmHg doubled $\dot{V}_G$ in plaice and flounder (Steffensen, Lomholt & Johansen, 1982) and increased it six times in carp (Lomholt & Johansen, 1979). It is tempting to apply the same argument to explain the decreased $\dot{V}_{O_2}$ in hypoxia-acclimated trout when P_{O_2} was raised to normoxic levels. Perhaps, in response to suddenly increased P_{O_2}, hypoxia-acclimated fish respond with a reduction in ventilation volume similar to the reduction in gill water flow recorded in carp, goldfish, and trout, when exposed to hyperoxia (Dejours, 1973). A potential lowering in metabolism from a lowered $\dot{V}_G$, however, is not supported by the $\dot{V}_{O_2}$ values measured in control fish. Ventilation volume of control fish in hypoxia (60 or 40 mmHg) should be much higher than in normoxia, yet $\dot{V}_{O_2}$ at the lowest swimming speed (5·5 cm s^{-1}) was unchanged.

Blood respiratory properties

The differences in blood respiratory properties between the normoxic and hypoxic groups obtained from *in vitro* analysis in our study confirm those from earlier studies (Wood & Johansen, 1972; Tetens & Lykkeboe, 1981).

The overall consequence of the blood changes for blood O_2 transport must be that gill blood O_2 loading is favoured in the hypoxia-acclimated fish as a result of the O_2 affinity increase caused by the reduced red cell organic phosphate concentration. Why the O_2-Hb affinity increase measured in this study is less than predictable from the reduction in red cell ATP concentration is not clear. In this respect, it may be that sampling *via* caudal vessels may affect O_2 binding properties of the red blood cells as a result of swelling (Nikinmaa, 1981).

It is possible that exercise as such causes rapid alterations in the blood respiratory function (increase in blood oxygen affinity etc.) (Nikinmaa, 1983; Nikinmaa, Cech & McEnroe, 1984) which result in maximal and similar O_2 loading in both normoxic and hypoxia-acclimated fish. Overall, however, it seems clear that the swimming performance gauged by the experimental conditions does not in any crucial way depend on altered blood respiratory properties brought about by hypoxia acclimation. While a slight shift in the oxygen dissociation curve will increase maximal O_2 delivery to the tissues, it may be that the back-up internal O_2 transport capacity depends more heavily on an increase in cardiac output and tissue perfusion.

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