J. exp. Biol. 127, 349–354 (1987)
Printed in Great Britain © The Company of Biologists Limited 1987

THE EFFECTS OF REDUCED GILL AREA AND HYPEROXIA ON THE OXYGEN CONSUMPTION AND SWIMMING SPEED OF RAINBOW TROUT

BY GARRY G. DUTHIE* AND GEORGE M. HUGHES

Research Unit for Comparative Animal Respiration, University of Bristol, Woodland Road, Bristol BS8 1UG

Accepted 29 July 1986

SUMMARY

Rainbow trout with a cautery-induced 30% reduction in functional gill area showed significant proportional reductions in maximum oxygen consumption ($\dot{V}_{O_2max}$) in comparison to controls, but oxygen consumption at rest and at sub-critical swimming speeds was not affected. This corroborates suggestions that total gill area is utilized for oxygen uptake only under conditions of maximum aerobic demand. During swimming trials, hyperoxia (P_{O_2} 300 mmHg) neither increased $\dot{V}_{O_2max}$ of control fish nor compensated for the reduced $\dot{V}_{O_2max}$ apparent in fish with reduced gill area. Therefore a direct limitation on oxygen uptake at the gills is implied.

INTRODUCTION

When a fish swims at its maximum sustained speed (U_{crit}), its oxygen consumption ($\dot{V}_{O_2}$) approaches a maximum level ($\dot{V}_{O_2max}$) (Brett, 1964) reflecting maximal aerobic requirements of those tissues involved in locomotion (Jones & Randall, 1978). If it is assumed that at $\dot{V}_{O_2max}$ the gills are fully perfused by blood (Randall & Daxboeck, 1982) and that oxygen transfer proceeds at an optimum rate then any reduction in functional gill area will lead to a corresponding reduction in $\dot{V}_{O_2max}$ indicating a direct limitation on oxygen uptake at the level of the gills. However, any deviation from a proportional decrease in $\dot{V}_{O_2max}$ as a result of gill area reduction would indicate limitation of oxygen transport at the circulatory or tissue level.

The purpose of the present study was to ascertain whether oxygen uptake is directly limited by gill area by effecting artificial reduction in respiratory area of rainbow trout and observing the effects on $\dot{V}_{O_2max}$ and swimming performance. Further, in the absence of any means of increasing gill area, fish were exposed to hyperoxia in an effort to increase the rate of oxygen transfer across the gills.

*Present address: Rowett Research Institute, Greenburn Road, Bucksburn, Aberdeen, AB2 9SB, Scotland.

Key words: trout, reduced gill area, hyperoxia, swimming performance.

MATERIALS AND METHODS

Rainbow trout (mean ± S.D.; length 31 ± 1 cm, mass 286 ± 25 g, $N = 48$) were obtained from a fish farm near Nailsworth, Gloucester and maintained in stocktanks at 15 ± 0·2°C for 2 weeks prior to surgical manipulations. Fish were fed every second day with 'Puwoods Silver Cup' trout pellets, supplemented weekly with chopped liver, and starved for 2 days prior to swimming trials. To avoid interbatch variation in swimming performance, the same group of fish was used in all experiments and individuals were used only once to avoid possible training effects (Beamish, 1978).

After anaesthesia (MS222, 0·1 g l^{-1}), fish were placed on the bench on their sides and the opercular flap held open. A hot wire cauterizer was applied smoothly along the length of the anterior hemibranch of either the first or second gill arch at the point of filamentar attachment, resulting in haemostasis with minimal blood loss. The procedure was repeated on the posterior hemibranch of the same arch, and on the corresponding arch on the other side of the fish, thus accounting for about 30 % of gill surface area (Hughes, in Paling, 1968). The operation was completed within 3 min. Mortality rate was 15 % within the first 24 h, thereafter survival was 100 %.

Subsequent to the operation, fish were transferred to water containing antibiotic (oxytetracycline hydrochloride, 0·05 g l^{-1}) for 5 days prior to being returned to stocktanks for 9–14 days before swimming trials were conducted. By this stage damaged filaments were white in appearance and obviously non-functional (see Fig. 1). Feeding resumed within 48 h of the operation. Control fish were subjected to a sham-operation and identical subsequent treatment.

Relationships between swimming speed (U) and oxygen consumption ($\dot{V}_{O_2}$) were constructed essentially as described in Webb (1971). In brief, fish were transferred to a Brett (1964) respirometer and left overnight in a water flow of 15 cm s^{-1}. The following morning, velocity increments of 0·2 bodylengths per second (L s^{-1}) of 30 min duration were successively imposed until the fish was unable to remove itself from the retaining grid. In trials where hyperoxia was employed oxygen was bubbled through a series of aeration columns supplying the respirometer until a Pw_{O_2} of 300 mmHg was attained. During each velocity increment the respirometer was closed off for 15 min and declining Pw_{O_2} monitored with a Radiometer E5046 oxygen electrode. U_{crit} was calculated as described in Beamish (1978). $\dot{V}_{O_2}$ at U_{crit} was assumed to be $\dot{V}_{O_2max}$. Standard oxygen consumption ($\dot{V}_{O_2stand}$) was estimated by extrapolation of the U–$\dot{V}_{O_2}$ regression to zero U.

RESULTS

An example of necrotic filaments of the second gill arch 2 weeks after the cautery procedure is shown in Figs 1 and 2. Undamaged filaments are present at the isthmus and base of the arch where access by the cauterizer was restricted.

Table 1 gives the U–$\dot{V}_{O_2}$ regressions, $\dot{V}_{O_2max}$, $\dot{V}_{O_2stand}$ and U_{crit} values for all treatments. Estimates of the induced reduction of gill area are calculated from da[illegible] for rainbow trout given in Paling (1968).

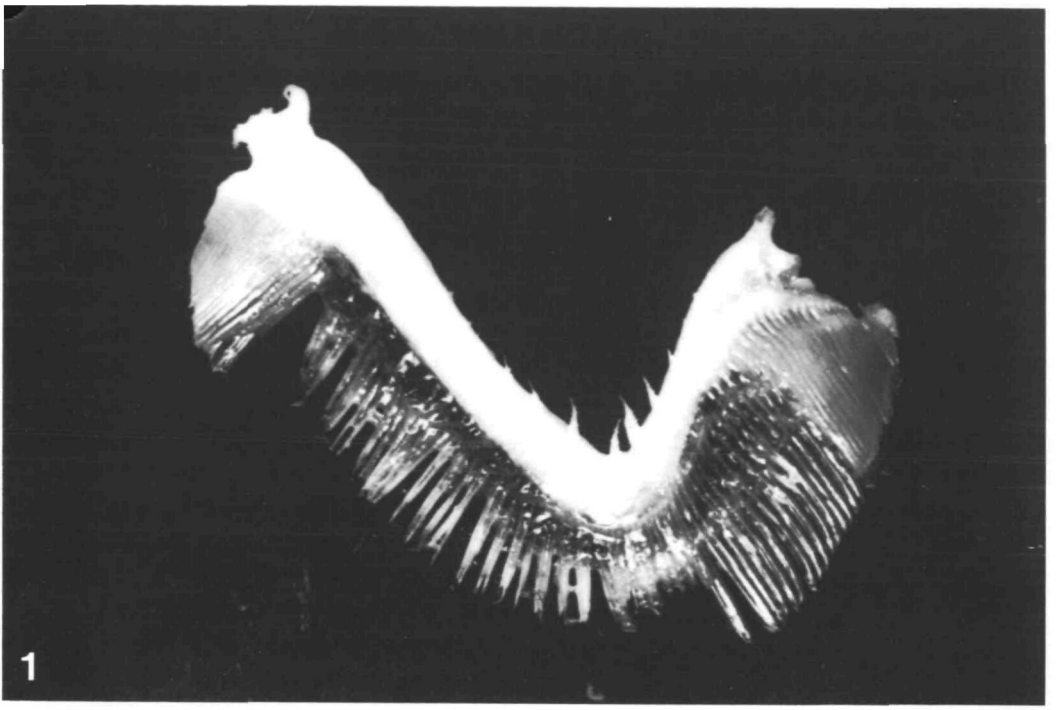

Fig. 1. The effects of the cautery procedure on the gill filaments of the second gill arch. Undamaged filaments are present at the bases of the arch.

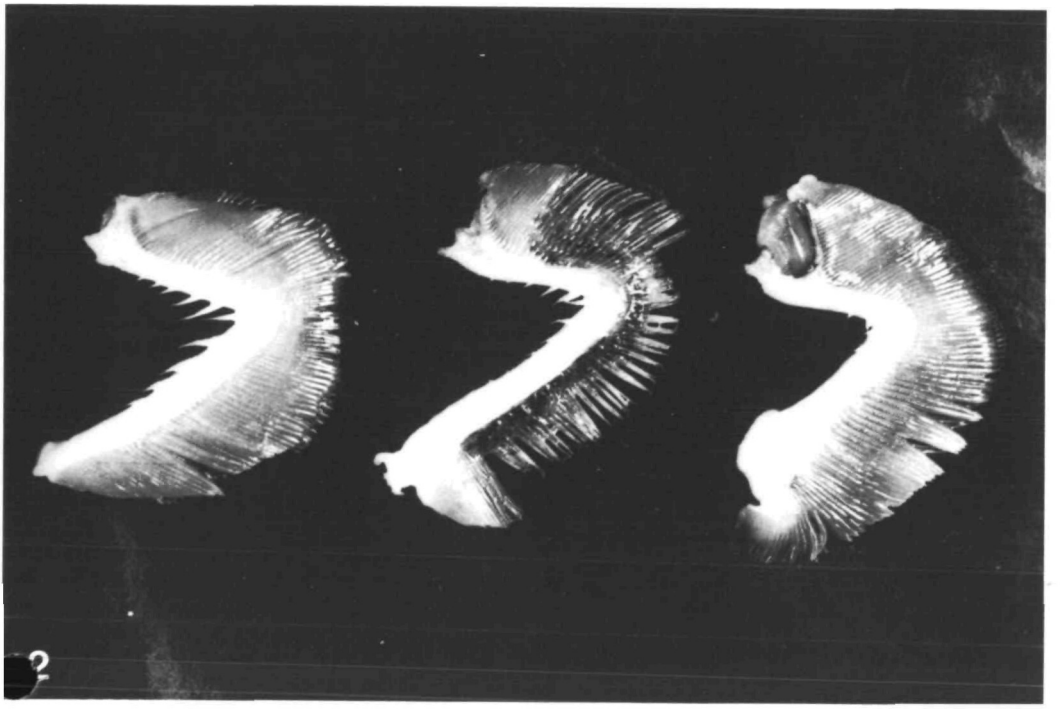

Fig. 2. Intact gill arches (first and third) shown for comparison with the arch containing damaged filaments.

Table 1. *Results from swimming trials for controls and fish with cautery-induced reduction in gill area*

Regime	N	$\dot{V}_{O_2stand}$ (mg O_2 kg^{-1} h^{-1})	$\dot{V}_{O_2max}$ (mg O_2 kg^{-1} h^{-1})	U_{crit} (L s^{-1})	U–$\dot{V}_{O_2}$ regression	r	P	% reduction in gill area
Normoxia								
Control	11	97 ± 11	476 ± 66	1·76 ± 0·11	$\log\dot{V}_{O_2} = 0{\cdot}40U + 1{\cdot}99$	0·968	<0·001	0
First arch	8	88 ± 6	340 ± 36*	1·33 ± 0·09*	$\log\dot{V}_{O_2} = 0{\cdot}43U + 1{\cdot}95$	0·973	<0·001	27
Second arch	8	91 ± 15	349 ± 50*	1·34 ± 0·07*	$\log\dot{V}_{O_2} = 0{\cdot}44U + 1{\cdot}95$	0·979	<0·001	30
Hyperoxia								
Control	9	103 ± 9	474 ± 47	1·77 ± 0·11	$\log\dot{V}_{O_2} = 0{\cdot}38U + 2{\cdot}01$	0·962	<0·001	0
First arch	7	98 ± 13	366 ± 64†	1·44 ± 0·14†	$\log\dot{V}_{O_2} = 0{\cdot}40U + 1{\cdot}98$	0·966	<0·001	27
Second arch	5	89 ± 22	326 ± 51†	1·30 ± 0·11†	$\log\dot{V}_{O_2} = 0{\cdot}47U + 1{\cdot}91$	0·936	<0·01	30

Trials conducted under normoxia or hyperoxia (300 mmHg).
$\dot{V}_{O_2}$ and U_{crit} data are presented as means ± S.D.
Percentage reduction of gill area was estimated from values of gill areas in Paling (1968).
* Significant difference from controls for trials in normoxia.
† Significant difference from controls in hyperoxic regimes.
L is body length.

In normoxia, there was no significant difference between slopes or elevations of the U–$\dot{V}_{O_2}$ regressions ($P > 0{\cdot}05$; ANCOVA, Snedecor & Cochran, 1972) when fish with reduced gill area were compared to controls, and consequently the extrapolated estimates of $\dot{V}_{O_2 stand}$ did not differ significantly ($P > 0{\cdot}05$; Student's t-test). However, reduction in functional gill area of 27 % as a result of cautery of the first arches and 30 % due to cautery of the second arches produced significant reductions in $\dot{V}_{O_2 max}$ of 29 % and 27 %, respectively, in comparison to controls. U_{crit} values were significantly reduced ($P < 0{\cdot}05$) as a consequence of gill area reduction.

Hyperoxia did not significantly increase $\dot{V}_{O_2 max}$ or U_{crit} of control fish compared to those values obtained when swimming trials were conducted in normoxia. In addition, no significant increases in $\dot{V}_{O_2 max}$ or U_{crit} were observed in hyperoxia in fish with reduced gill area when compared to corresponding values obtained in normoxia.

DISCUSSION

Artificial reduction of functional gill area induced a directly proportional reduction in $\dot{V}_{O_2 max}$ indicating that the fish were unable to make major compensatory adjustments in circulation to enhance oxygen transport. Under conditions of maximum aerobic demand, all available gill area would appear to be fully perfused. As $\dot{V}_{O_2 stand}$ and $\dot{V}_{O_2}$ at lower swimming speeds were unaffected by the imposed gill reduction, under resting or routine conditions total gill area is likely to be incompletely utilized, agreeing with suggestions of Booth (1978) and Hughes (1984).

The present experiments corroborate suggestions that the gas exchange surface of fish relates to maximum oxygen uptake capacity rather than to that incurred under resting conditions (Hughes, 1984). Both $\dot{V}_{O_2 max}$ and gill area scale to increasing body mass by an allometric coefficient of 0·9 (Brett & Glass, 1973; Hughes, 1980). As $\dot{V}_{O_2 stand}$ scales to body mass by approximately 0·75 (Winberg, 1956), no direct link between gill area and $\dot{V}_{O_2 stand}$ should be expected and it is not apparent in the present study.

Attempts to increase the rate of oxygen transfer at the gills by hyperoxic exposure failed to increase $\dot{V}_{O_2 max}$ in controls or in fish with reduced gill area, suggesting a direct limit on oxygen uptake at the gills independent of environmental P_{O_2} when elevated above normoxia.

Damage to the filaments of either the first or second arches had a similar effect on $\dot{V}_{O_2}$. Davis (1971) found that trout with first arch ligation had a reduced arterial P_{O_2} and incurred pseudobranch necrosis, although no similar effect occurred if other arches were ligated. Such observations have led to suggestions for the localized presence of oxygen receptors within the region of the first gill arch (Randall, 1982). No pseudobranch necrosis was observed in the present study. Therefore such receptors may not be exclusively present in the area of the first arch and/or sufficient oxygen receptors to regulate ventilation and maintain arterial P_{O_2} at lower activity levels are present on the remaining filaments of the first arch which escaped cautery (Fig. 1).

These experiments indicate that trout can survive with at least a 30% reduction in gill area. Gill damage may be of ecological importance only at higher levels of sustained activity. However, other energetic costs such as reproduction and digestion may require a relatively greater proportion of the reduced aerobic scope that results from gill damage, thus altering the relationship between activity levels and mortality (Priede, 1977).

The authors are grateful to NERC for post-doctoral funding (grant no. GR3/4235) and express appreciation to Dr I. G. Priede for advice on revisions of the manuscript.

REFERENCES

BEAMISH, F. W. H. (1978). Swimming capacity. In *Fish Physiology*, vol. 7 (ed. W. S. Hoar & D. J. Randall), pp. 161–187. New York: Academic Press.

BOOTH, J. H. (1978). The distribution of blood flow in the gills of fish: application of a new technique to rainbow trout (*Salmo gairdneri*). *J. exp. Biol.* **73**, 119–129.

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

BRETT, J. R. & GLASS, N. W. (1973). Metabolic rates and critical swimming speeds of sockeye salmon (*Oncorhynchus nerka*) in relation to size and temperature. *J. Fish. Res. Bd Can.* **30**, 379–387.

DAVIS, J. C. (1971). Circulatory and ventilatory responses of rainbow trout (*Salmo gairdneri*) to artificial manipulation of gill surface area. *J. Fish. Res. Bd Can.* **28**, 1609–1614.

HUGHES, G. M. (1980). Morphometry of gas exchange organs in relation to their respiratory function. In *Environmental Physiology of Fishes* (ed. M. A. Ali), pp. 33–56. New York: Plenum Publishing Co.

HUGHES, G. M. (1984). Scaling of respiratory areas in relation to oxygen consumption of vertebrates. *Experientia* **40**, 519–524.

JONES, D. R. & RANDALL, D. J. (1978). The respiratory and circulatory systems. In *Fish Physiology*, vol. 7 (ed. W. S. Hoar & D. J. Randall), pp. 425–501. New York: Academic Press.

PALING, J. E. (1968). A method of estimating the relative volumes of water flow over the different gills of a freshwater fish. *J. exp. Biol.* **48**, 533–544.

PRIEDE, I. G. (1977). Natural selection for energetic efficiency and the relationship between activity level and mortality. *Nature, Lond.* **267**, 610–611.

RANDALL, D. J. (1982). The control of respiration and circulation in fish during exercise and hypoxia. *J. exp. Biol.* **100**, 275–288.

RANDALL, D. J. & DAXBOECK, C. (1982). Cardiovascular changes in the rainbow trout (*Salmo gairdneri* Richardson) during exercise. *Can. J. Zool.* **60**, 1135–1140.

SNEDECOR, G. W. & COCHRAN, W. G. (1972). *Statistical Methods*, 6th edn. Iowa: Iowa State University Press.

WEBB, P. W. (1971). The swimming energetics of trout. II. Oxygen consumption and swimming efficiency. *J. exp. Biol.* **55**, 521–540.

WINBERG, G. G. (1956). (1) Rate of metabolism and food requirements of fishes. (2) New information on metabolic rate in fishes. *Fish. Res. Bd. Can.* (translation series 194 and 362).