THE EXCHANGE OF OXYGEN AND CARBON DIOXIDE ACROSS THE GILLS OF RAINBOW TROUT

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INTRODUCTION

Hughes & Shelton (1962), Hughes (1964) and Rahn (1966) have analysed gas exchange in animals in an aquatic environment, particularly fish, from a theoretical viewpoint. These authors provide equations which permit a quantitative analysis of the gas-exchange process. From these, it is possible to determine the relative effectiveness of the exchange process (Hughes & Shelton, 1962) in a single animal under a variety of conditions, or to compare gas exchange in different animals, independent of the environment in which they live, whether it be aquatic or aerial.

Much of the present information on the circulatory and respiratory systems of fishes is of a fragmentary nature encompassing fish of widely differing habitat, habit and species. This has limited any analysis of gas exchange in fish, as well as any comparison between fish and air-breathing mammals. An effort has been made, however, to measure many of the factors affecting gas exchange across the gills of fish in a simultaneous and integrated manner by Holeton & Randall (1967*a, b*) and by Stevens & Randall (1967*a, b*). These experiments were carried out on intact, unanaesthetized, unrestrained rainbow trout (*Salmo gairdneri*) subjected to two conditions: first, hypoxia (Holeton & Randall, 1967*a, b*), and secondly, moderate exercise (Stevens & Randall, 1967*a, b*). This data is sufficiently extensive to permit an analysis of gas exchange across the gills of a single species of fish.

METHODS

Symbols

The symbols employed are somewhat different from those used by Hughes & Shelton (1962) and Hughes (1964), but are generally those used in mammalian respiration as described in Ruch & Patton (1965), p. 769, and employed by Rahn (1966). The terms used are:

$\dot{V}g$	volume flow of water over gills per unit time
$\dot{V}_{O_2}, \dot{V}_{CO_2}$	O_2 uptake and CO_2 output per unit time
$\dot{Q}g$	volume flow of blood through gills per unit time
P	partial pressure of gas
αw	solubility coefficient in water
αb	solubility coefficient in blood

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Subscripts

i	referring to water in the buccal cavity, i.e. inspired water
e	referring to water in the opercular cavity, i.e. expired water
a	arterial blood leaving gills
v	venous blood entering gills
O_2, CO_2	molecular species

Hughes & Shelton (1962) and Hughes (1964) have compared gas exchange at the gills with known relationships derived from studies upon compact heat exchangers. They show how several heat-exchanger relationships are of use in analysing gas-exchange systems. One such relationship is the effectiveness of transfer. Effectiveness (Hughes & Shelton, 1962) is the ratio of the actual gas transfer ($\dot{V}_{O_2}$, $\dot{V}_{CO_2}$, etc.) to the maximum rate of gas transfer possible, expressed as a percentage.

The effectiveness of oxygen uptake by the blood is expressed by the following equation:

$$\text{effectiveness (\%)} = \frac{\dot{V}_{O_2}}{\dot{Q}g . \alpha b_{O_2} [P_{iO_2} - P_{vO_2}]} \times 100.$$

Maximum oxygen uptake will occur if the blood leaving the gills is at the same partial pressure of oxygen as the inspired water.

The solubility of oxygen in the blood (αb_{O_2}) is not constant but varies with the slope of the oxygen dissociation curve of the blood.

The same form of equation can be used to determine the effectiveness of oxygen removal from water, the effectiveness of carbon dioxide uptake by the water, and the effectiveness of carbon dioxide removal from the blood.

The effectiveness of oxygen removal from the water is

$$\frac{\dot{V}_{O_2}}{\dot{V}g . \alpha w_{O_2}[P_{iO_2} - P_{vO_2}]} \times 100.$$

The effectiveness of carbon dioxide uptake by the water is

$$\frac{\dot{V}_{CO_2}}{\dot{V}g . \alpha w_{CO_2} . [P_{vCO_2} - P_{iCO_2}]} \times 100$$

assuming a respiratory quotient of 1 then $\dot{V}_{O_2} = \dot{V}_{CO_2}$, and assuming that P_{iCO} is zero, then the effectiveness of carbon dioxide uptake by the water is

$$\frac{\dot{V}_{O_2}}{\dot{V}g . \alpha w_{CO_2} . P_{vCO_2}} \times 100.$$

The effectiveness of carbon dioxide removal from the blood is

$$\frac{\dot{Q}g . \alpha b_{CO_2} . [P_{vCO_2} - P_{aCO_2}]}{\dot{Q}g . \alpha b_{CO_2} . [P_{vCO_2} - P_{iCO_2}]} \times 100,$$

but P_{iCO_2} = zero, therefore the effectiveness of carbon dioxide removal from the blood, is

$$\frac{(\alpha b_{CO_2} . P_{vCO_2}) - (\alpha b_{CO_2} . P_{aCO_2})}{\alpha b_{CO_2} . P_{vCO_2}},$$

that is

$$\frac{CO_2 \text{ content of venous blood} - CO_2 \text{ content of arterial blood}}{CO_2 \text{ content of venous blood}} \times 100.$$

The solubility coefficient for carbon dioxide in blood varies with the P_{CO_2}. The carbon dioxide content and not the P_{CO_2} must therefore be used in the above equation.

As pointed out by Hughes & Shelton (1962) and Hughes (1964) the effectiveness of gas exchange will be altered by a change in the capacity-rate ratio of blood to water, that is

$$\frac{\dot{Q}g \cdot \alpha b_{O_2}(P_{aO_2} - P_{vO_2})}{\dot{V}g \cdot \alpha w_{O_2}(P_{iO_2} - P_{eO_2})}.$$

The maximum effectiveness of oxygen uptake will occur if the blood is stationary in the gills. As blood flow increases less time is available to saturate a unit volume of blood with oxygen, and the effectiveness of gas exchange will fall. An analysis of any changes in capacity-rate ratio of blood and water at the gills may produce an explanation for any concurrent changes in effectiveness.

The rate of gas exchange will also be affected by the area available for gas exchange, and by the diffusion distance between water and blood. The transfer factor is a measure of the relative ability of the respiratory surface to exchange gases, and is affected by changes in surface area available for exchange, as well as the diffusion distance between blood and water. The oxygen transfer factor T_{O_2}, is defined as

$$T_{O_2} = \frac{\dot{V}_{O_2}}{\frac{1}{2}(P_{iO_2} + P_{eO_2}) - \frac{1}{2}(P_{aO_2} + P_{vO_2})}.$$

An increase in the transfer factor will result in a more effective exchange of gases across the gills.

These equations were applied to data reported by Holeton & Randall (1967a, b) and Stevens & Randall (1967a, b). These data were obtained from rainbow trout (*Salmo gairdneri*) in fresh water. The experiments of Holeton & Randall (1967) were carried out at $15 \pm 1^\circ$ C., whereas those of Stevens & Randall (1967b) were at 4–8° C. The solubility of oxygen in blood was determined from oxygen dissociation curves constructed in this laboratory (Randall, Beaumont & Holeton, unpublished) unless otherwise stated.

Changes in the effectiveness of gas exchange, capacity-rate ratio of blood and water, and the oxygen transfer factor, during hypoxia and during moderate exercise, were calculated.

RESULTS AND DISCUSSION

The effectiveness of blood oxygenation is very near 100 % in the resting rainbow trout at environmental temperatures of approximately 5 and 15° C. Moderate exercise produced only slight decrease in effectiveness (Fig. 1); but a reduction in the oxygen content of the water resulted in a marked decrease in effectiveness, reaching values of 30–40 % at environmental P_{O_2} of 40 mm. Hg (Fig. 2). Removal of oxygen was 30 % effective in experiments carried out at approximately 15° C. (Fig. 2) but was only 11 % at temperatures of 5–8° C. (Fig. 1). Hypoxia and moderate exercise had very little effect on the effectiveness of oxygen removal from the water.

These results illustrate the very high effectiveness of gas exchange across the gills of fishes. Only during hypoxia did the effectiveness of loading the blood with oxygen fall much below 95–100 %.

The effectiveness of oxygen removal from water was low (Figs. 1, 2). This was associated with a low percentage utilization of oxygen passing over the gills (Holeton & Randall, 1967 b; Stevens & Randall, 1967 b,). The essential difference between percentage utilization and the effectiveness of oxygen removal from water is that effectiveness takes into account the oxygen tension of the venous blood. The rela-
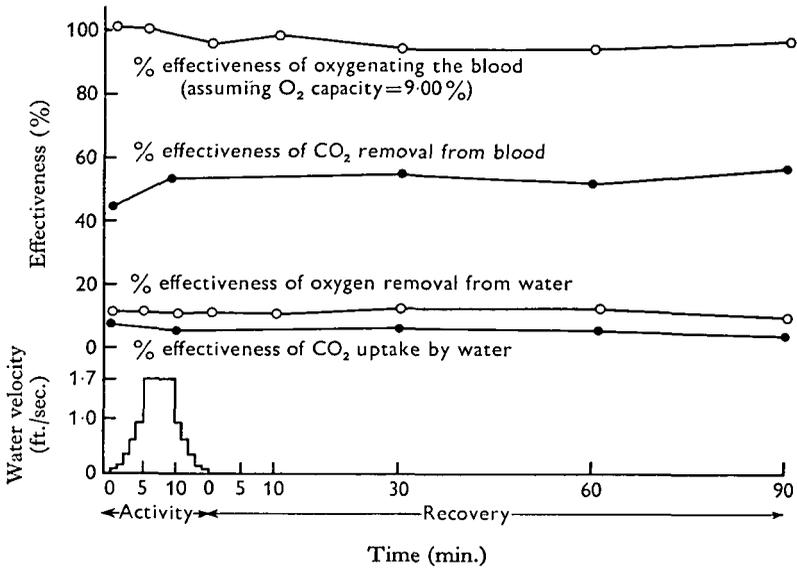

Fig. 1. The effectiveness of the exchange of oxygen and carbon dioxide between blood and water across the gills of rainbow trout during moderate exercise.
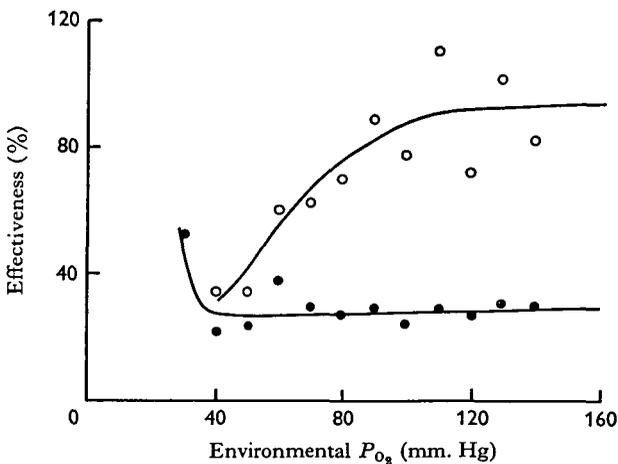

Fig. 2. The effectiveness of the exchange of oxygen between blood and water across the gills of rainbow trout during hypoxia. O, Oxygen loading by blood; ●, Oxygen removal from respiratory water.

tionship between percentage utilization of oxygen from the water (% *U*) and the effectiveness of oxygen removal from the water is described by the following equation:

$$\text{effectiveness of O}_2 \text{ removal} = \%U\left[\frac{P_{iO_2}}{P_{iO_2} - P_{vO_2}}\right].$$

When P_{vO_2} is zero the effectiveness of oxygen removal will equal the percentage utilization of oxygen, but as P_{vO_2} increases the difference between effectiveness and utilization will increase. When P_{vO_2} is small, however, a low percentage utilization will be associated with a low effectiveness of oxygen removal from water. The rainbow trout in these studies faced upstream into flowing water. It has been suggested (Holeton & Randall, 1967*b*) that this water flow aided breathing, resulting in a high ventilation volume and a low percentage utilization (Hughes 1966). This would also produce a low effectiveness of oxygen removal from the water. If the fish was in still water, the cost of respiration would undoubtedly be higher (Schumann & Piiper, 1966) and ventilation volume somewhat lower than reported by Holeton & Randall (1967*b*) and Stevens & Randall (1967*b*). Under these conditions it is possible that the percentage utilization of oxygen would be high, perhaps of the order of 70–80 %, and the effectiveness of oxygen removal would approach 100 %. This is to be expected when the cost of moving such a dense medium is such a large component of the resting oxygen consumption (Schumann & Piiper, 1966).

The differences in the effectiveness of oxygen removal from the water, in the resting fish, reported in Figs. 1 and 2 are probably related to differences in the velocity of water flow. The calculated values reported in Fig. 1 were obtained from data collected from fish exposed to water velocities of about 7 cm./sec. whereas the calculated values reported in Fig. 2 were from fish exposed to water velocities of 2 cm./sec.

The capacity-rate ratio of blood to water at the gills is between 0·2 and 0·3 in the resting rainbow trout (Figs. 3 and 4). Moderate exercise resulted in a small increase, whereas hypoxia produced a doubling of capacity-rate ratio. As Hughes & Shelton (1962) have pointed out, the capacity-rate ratio can have a strong influence upon effectiveness. During hypoxia the capacity-rate ratio increased and as might be expected the effectiveness of gas transfer decreased. The increase in the capacity rate for blood during hypoxia must be related to an increased solubility of oxygen in blood, because cardiac output does not alter appreciably (Holeton & Randall, 1967*b*). The changes in the solubility of oxygen in blood are described by the oxygen dissociation curve, which in turn is dependent upon the characteristics of haemoglobin. The decrease in effectiveness during hypoxia is related therefore to the nature of the oxygen dissociation curve of the blood. A carp, having a haemoglobin with a very high affinity for oxygen (Black, 1940) should be able to maintain a high level of effectiveness of gas transfer even at very low environmental oxygen levels. This presumably confers an advantage to the carp over other fishes, in a hypoxic environment.

There is a marked increase in the transfer factor of the gills of trout in response to hypoxic conditions in the environment (Fig. 5); during exercise the transfer factor increased almost fivefold (Fig. 3), indicating that during both hypoxia and exercise there is an increase in the effective exchange area, or a decrease in diffusion distance, or both.
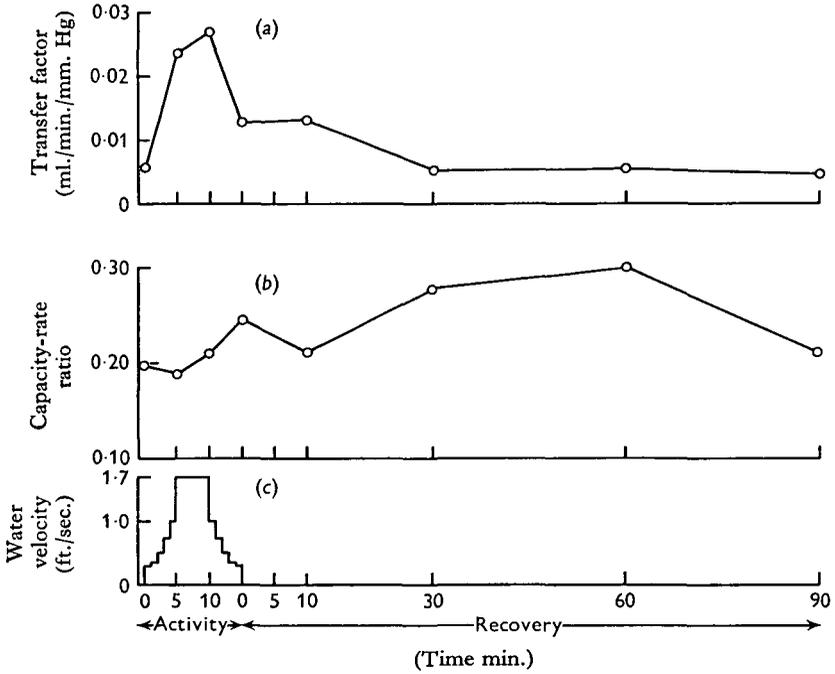
Fig. 3. Changes in the capacity-rate ratio and the transfer factor of the gills for oxygen during moderate exercise.

$$(a) \quad T_{O_2} = \frac{\dot{V}_{O_2}}{\frac{1}{2}(P_{iO_2}+P_{eO_2})-\frac{1}{2}(P_{aO_2}+P_{vO_2})} ;$$

$$(b) \quad \text{Capacity-rate ratio} = \frac{\text{blood}}{\text{water}} = \frac{\dot{Q}g.\alpha b_{O_2}(P_{aO_2}-P_{vO_2})}{\dot{V}_g.\alpha w_{O_2}(P_{iO_2}-P_{eO_2})}$$
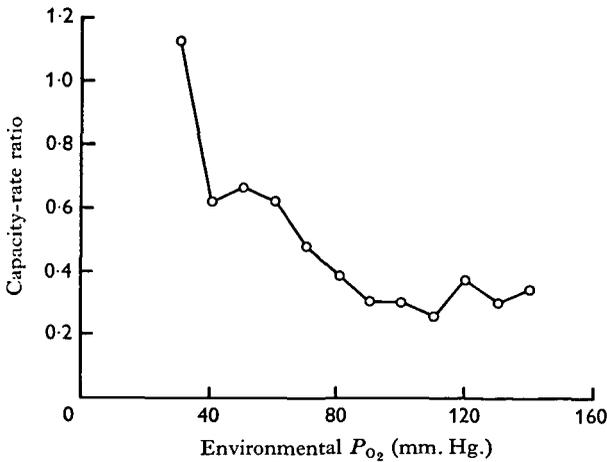

Fig. 4. Changes in capacity-rate ratio of blood to water at the gills during hypoxia

$$\frac{\text{Blood}}{\text{Water}} = \frac{\dot{Q}g.\alpha b_{O_2}(P_{aO_2}-P_{vO_2})}{\dot{V}_g.\alpha w_O(P_{iO_2}-P_{eO_2})} .$$

Hughes & Grimstone (1965) and Steen & Kruyse (1964) have reported morphological evidence indicating that the effective exchange area of the gills can be increased. Steen & Kruysse (1964) demonstrated the presence of alternate circulatory pathways in the gills of the eel and ten other teleosts, including the rainbow trout. They demonstrated that changes in the pattern of blood flow through the gills could affect the degree of oxygenation of the blood of the eel. Holeton & Randall (1967a) concluded that there was an increase in vascular resistance to blood flow through the gills during hypoxia. It appears probable that the change in vascular resistance was due to a change in the pattern of blood flow through the gills, presumably shunting blood closer to the surface of the gills which, in turn, would augment gas exchange. Hughes & Grimstone (1965) suggested that contractile elements in the secondary lamellae could alter the thickness of the lamellae, and thus alter the conditions for diffusion of gases between blood and water.
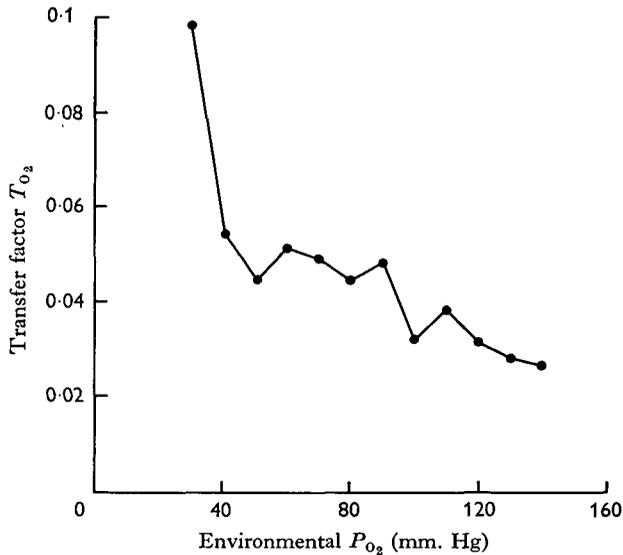

Fig. 5. Changes in the transfer factor of the gills for oxygen during hypoxia

$$T_{O_2} = \frac{O_2 \text{ uptake ml./min./kg}}{\text{Mean } O_2 \text{ gradient water/blood (mm.Hg)}}.$$

An increase in the volume of blood in the respiratory circulation, the shunting of blood closer to the surface of the gills and a decrease in the thickness of the lamellae would all tend to increase the oxygen transfer factor of the gills. All may be operative during hypoxia.

The effectiveness of carbon dioxide removal from the blood did not change markedly during exercise, and was of the order of 50 % (Fig. 1). The carbon dioxide content of blood afferent and efferent to the gills was estimated using the P_{CO_2} data of Stevens & Randall (1967b) and a carbon dioxide dissociation curve was constructed from data on the carbon dioxide content of the blood of rainbow trout (Black, Kirkpatrick & Tucker, 1966; Ferguson & Black, 1941). The effectiveness of carbon dioxide uptake by the water varied between 4 and 7 % (Fig. 1). These values were obtained assuming a

respiratory quotient of 1, and a solubility coefficient for carbon dioxide in water of 1·8 ml./l./mm. Hg P_{CO_2}. The low effectiveness of carbon dioxide uptake by the water is a result of the high capacity of water for carbon dioxide.

The ratio of diffusion coefficients for oxygen and carbon dioxide in tissues is about 1:20. The mean gradient driving oxygen into the blood is approximately 70 mm. Hg. One would expect, therefore, the mean gradient for carbon dioxide between blood and water to be about 3 to 4 mm. Hg. The P_{CO_2} of the arterial blood is of the order of 2·5 mm. Hg. This is probably determined by the diffusion characteristics of the gills. Holeton & Randall (1967b) recorded slightly lower P_{CO_2} values than Stevens & Randall (1967b), (1·5 and 2·5 mm. Hg respectively). This may be a reflexion of differences in diffusion characteristics of the gills due to differences in temperature (15 ± 1° C. and 4–8° C. respectively).

Table 1. *A comparison of the effectiveness of gas exchange in resting man and rainbow trout*

Effectiveness (%)	Man %	Rainbow trout (%)
O_2 uptake by blood	90–100	95–100
CO_2 removal from blood	9	50–60
O_2 removal from medium	33	11–30
CO_2 uptake by medium	60	4–6

The calculated values for the percentage effectiveness of gas exchange in man were based on the following:

	P_{O_2} (mm. Hg)	P_{CO_2} (mm. Hg)
Inspired air	159	0·3
Expired air	120	27
Mixed venous blood	40	45
Mixed arterial blood	95	40

An increase in venous P_{CO_2} would probably not result in an increase in dorsal aortic P_{CO_2}. The added load on the exchange system could be dealt with easily, because of the high capacity of water for carbon dioxide relative to oxygen. Thus an increase in the effectiveness of carbon dioxide removal from the blood would normally result from an increase in venous P_{CO_2}, the arterial P_{CO_2} remaining fairly constant.

A comparison of gas exchange in man and rainbow trout indicates that in both cases oxygen uptake by the blood approaches 100 % effectiveness (Table 1). This is due to the presence of haemoglobin in the blood. Oxygen removal from the medium is of the same order of magnitude in both aerial and aquatic respiration (Table 1), although, as emphasized previously, the value for the effectiveness of oxygen removal from water reported here is probably minimal rather than maximal, and may approach 100 % under certain environmental conditions. In general it is probably higher than that for aerial respiration.

The major difference between aerial and aquatic respiration is related to the effectiveness of carbon dioxide removal from the blood (Table 1). This results in high carbon dioxide levels in arterial blood of terrestrial animals compared with the low carbon dioxide levels recorded in fish. The low effectiveness of carbon dioxide removal from blood in man is related to the tidal nature of breathing. A second factor influencing carbon dioxide removal from blood is that equal volumes of oxygen and carbon dioxide exert the same pressure in air, whereas the ratio of their solubility

coefficients in water is between 1:24 and 1:35 depending on the temperature (Rahn, 1966). As the respiratory quotient is about 1, the differences in the capacities of the two media for carbon dioxide relative to oxygen result in differences in the percentage effectiveness of carbon dioxide uptake by air and water. Effectiveness of carbon dioxide uptake in air is 60 % of maximum, whereas carbon dioxide uptake by the water is only 4–6 % of maximum. Because of the large capacity of water for carbon dioxide compared with oxygen, removal of carbon dioxide from the blood is not a problem.

In man a compromise is made between removal of carbon dioxide and water loss. Tidal ventilation is possible because of the low density and high oxygen content of air compared with water. This minimizes water loss, but reduces the percentage effectiveness of carbon dioxide removal.

In fish there are similar osmotic problems, but tidal ventilation is not practical because of the high density and low oxygen content of the medium. Those factors which increase gas exchange will augment exchange of ions and water. The uptake of water at the gills in fresh water is offset by a high blood pressure and ionic losses are reduced by special cells which actively transport ions from the water into the blood.

SUMMARY

1. The effectiveness of oxygen uptake by the blood of rainbow trout (*Salmo gairdneri*) approaches 100 %, whereas that for the removal of oxygen from water was only 11–30 %.

2. Most of the carbon dioxide is removed from the blood as it passes through the gills, but the effectiveness of carbon dioxide uptake by water is very low, because of the high capacity of water for carbon dioxide compared with oxygen.

3. Moderate exercise had little effect on the effectiveness of gas exchange across the gills. The increased oxygen uptake was facilitated by an increase in the transfer factor of the gills for oxygen. There were small increases in the capacity-rate ratio of blood to water at the gills during moderate exercise.

4. Hypoxia resulted in a marked decrease in the effectiveness of oxygen uptake by the blood, but had little effect on oxygen removal from the water. Gas exchange was facilitated during hypoxia by an increase in transfer factor of the gills, but hindered by an increasing capacity-rate ratio of blood to water at the gills.

5. Gas exchange in an aquatic environment was compared with that in an aerial environment.

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