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CUTANEOUS RESPIRATION IN *OCTOPUS VULGARIS*

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Summary

The skin of *Octopus vulgaris* consumes considerable quantities of oxygen *in vitro*, averaging $4.55\times10^{-5}\pm1.80\times10^{-5}$ ml mm^{-2} h^{-1} (mean ± S.D.), if a flow is maintained over the skin sample (*N*=32). The consumption is higher still *in vivo*, $11.36\times10^{-5}\pm2.73\times10^{-5}$ ml mm^{-2} h^{-1} (*N*=8), suggesting an additional net import of oxygen through the skin when the blood system is intact. If a substantial boundary layer is allowed to develop, oxygen uptake *in vitro* falls to $2.09\times10^{-5}\pm0.56\times10^{-5}$ ml mm^{-2} h^{-1} (*N*=15). The proportion of the animals' total oxygen consumption that cutaneous uptake will represent must thus depend on how much of the skin is exposed and how well it is ventilated. Estimates indicate that some 41 % of the total oxygen requirement of an animal at rest might be satisfied in this manner. During exercise, with water flowing over the entire surface of the animal, cutaneous uptake will increase but is nevertheless likely to form a smaller proportion (about 33 %) of the total uptake. In an animal curled up in its den and digesting a substantial meal, cutaneous uptake could shrink to as little as 3 % of the total. Similar results were obtained in a small number of pilot experiments with a range of octopod, sepioid and teuthoid species.

Key words: cutaneous respiration, skin oxygen uptake, cephalopods, *Octopus*.

Introduction

All soft-bodied aquatic animals exchange gases through their skins. Many small invertebrates and some vertebrates (Booth and Feder, 1991) are wholly dependent upon this means of obtaining oxygen. Cephalopods are often large and typically have high metabolic rates. They all have well-developed gills and this has perhaps deflected attention from the possibility that a significant proportion of their oxygen uptake may be through the skin.

Only two published papers have reported attempts to assess cutaneous oxygen uptake in cephalopods. One of these dealt with *Octopus vulgaris*, the subject of most of the experiments to be reported below. In this, Wells and Wells (1983) enclosed the head and arms of octopuses in plastic bags and measured oxygen uptake inside the bag at the same time as oxygen uptake by the rest of the animal in a respirometer. They showed that 10–13 % of the oxygen uptake of the whole animal could be accounted for in this way.

A different approach was adopted by Pörtner (1994) working with *Illex illecebrosus* and *Loligo pealei*. He measured pH in the blood of squid swum to exhaustion against a current in a Brett respirometer. The quantity of carbon dioxide present implied oxygen consumption by the mantle muscle well in excess of the likely arterial oxygen input. The oxygen, he argued, could only have come in through the skin. On this basis, cutaneous uptake must have provided up to 73 % (depending on the assumed respiratory quotient) of the total oxygen consumption by the mantle during jet propulsion.

The skin of cephalopods is a very active tissue containing chromatophores with their intrinsic musculature, nerves, gland cells and blood vessels, although blood vessels appear to be absent in squids (Madan, 1995). In animals such as *Octopus vulgaris*, the skin can be thrown into folds and papillae by a web of muscle fibres. The skin must itself consume oxygen, which could either be supplied from within through the extensive network of blood vessels or, as Pörtner's work implies, it may take up oxygen in excess of its own needs to supply more deeply seated tissues.

The report that follows is an attempt to examine cutaneous respiration in *Octopus vulgaris*. Some results are also reported from other cephalopod species.

Materials and methods

Most of the experiments reported here were carried out using skin from *Octopus vulgaris* Cuvier. The animals ranged in mass from 40 to 1718 g (*N*=37) for *in vitro* and from 673 to 1369 g (*N*=8) for *in vivo* experiments. Other species used were *Eledone cirrhosa* Lamarck (43 and 99 g), *E. moschata* Lamarck (153 and 341 g), *Sepia officinalis* L. (192 and 220 g), *S. orbigyniana* Férussac (44 and 80 g), *Lolliguncula brevis* Blainville (4.4–34 g; *N*=8) and *Sepioteuthis lessoniana*

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Blainville (34–322 g, *N*=5). Most of the experiments with *Octopus vulgaris* (and all those with *Eledone* spp.) were carried out at the Laboratoire Arago in Banyuls-sur-Mer, France, a minority with *Octopus vulgaris* at the Station de Recherches Sous-Marines et Océanographiques near Calvi, France; the animals were collected by SCUBA divers or by trawling. Experiments with *Sepia officinalis* and *Sepioteuthis lessoniana* were made using animals reared from eggs at the Marine Biomedical Institute, Galveston, Texas, USA. *Lolliguncula brevis* and *Sepia orbigyniana* were caught by trawling in Galveston and Banyuls respectively. All except *Eledone cirrhosa* and *Sepia orbigyniana* come from shallow warm waters and all were kept and used in experiments at 20–24 °C.

In nearly all of the experiments, a Perspex cell with a capacity of 2 or 6 ml was clamped around an area of skin with the outside surface (or exceptionally in some *in vitro* experiments the inside surface) of the skin facing into the cell. Inside the cell, the tip of a neonatal oxygen catheter detected oxygen levels, signalling to a Neocath 1000 oxygen monitor (Pfizer Biomedical Sensors) and a Goerz Servogor 110 recorder. The basic cell could be used in various configurations, with (6 ml cell) or without (2 ml cell) a magnetic stirrer ('stirred' and 'unstirred' experiments) and with or without a through-flow of sea water. Alternatively, the oxygen electrode could be housed in a separate small cell, connected immediately upstream or downstream of the respirometer in through-flow experiments. Two cells could be clamped together, with the skin forming a membrane between the two to study movement of oxygen through the skin.

In vivo experiments, run only with *Octopus vulgaris*, used a 4 ml cell, with magnetic stirrer, glued onto the skin or bolted through the dorsal mantle muscle.

Circulation where appropriate was created by a Watson–Marlow peristaltic pump. Typical *in vitro* experiments lasted for approximately 60 min, with the skin, which can remain alive for several hours in sea water, still showing chromatophore and other muscular activity at the end of the experiment. In closed-cell and through-flow experiments, measurements were made without skin present to check the biological oxygen demand of the sea water. In most such experiments (and in all through-flow experiments), this demand was negligible compared with the considerable uptake by the skin.

To prevent the animal detaching the cell or the circulation cannulae during the *in vivo* experiments, the anterior and posterior basal lobes of the brain, which are parts of the brain concerned with higher motor control (Young, 1971), were excised. After this operation, carried out under 2.5 % ethanol anaesthesia, the arms still showed local reflexes but little whole-arm coordination so that, while they could still touch the apparatus, they did not grasp and pull on it. Animals treated in this way cannot feed, but they may nevertheless survive for several weeks (Wells, 1978). In our experiments, animals were all used within 2–3 days of operation.

Octopuses (*Octopus vulgaris* and *Eledone* spp.) were kept in holding tanks following collection until it was obvious that they had not been damaged during collection. Skin samples from these and other species were taken immediately after death by decapitation. The skin over the abdomen of octopuses moves freely over the underlying muscles and can be peeled off with minimal use of a scalpel to free the tissue. The skin of *Sepia officinalis* and of *S. orbigyniana* is less mobile than that of the octopuses studied, but can be freed from over the shell in a similar manner. The skin of *Lolliguncula brevis* and *Sepioteuthis lessoniana* is much thinner and more delicate than that of sepiods and octopods; removal of unpunctured sheets large enough to span the 2 cm diameter of the respirometers requires great care.

As described by Madan (1995), skin surface areas were estimated geometrically, on the basis that the abdomen consisted of five rectangles (four sides and an end), the arms long thin cones with cylindrical suckers attached, and so on. A problem arises because the skin of *Octopus vulgaris* is exceedingly flexible, so that it can be thrown into folds or extended to several times its 'resting' area. The interbrachial web can be extended considerably and the arms themselves can show great changes in length. We have tried to adopt dimensions appropriate to an animal sitting in the open, 'at rest', neither extended to grope round rocks nor crammed into a hole.

The extreme elasticity of the skin will, of course, also affect the area within the cell in the respirometry experiments. In theory, this was always 314 mm^2, but because the muscles in the living skin could be quite variably contracted (the skin of *Octopus vulgaris* is rarely if ever completely smooth), the actual area probably differed somewhat from one experiment to the next. We tried to allow for this by taking care not to stretch the skin unduly when inserting it into the respirometer, but it was impossible always to ensure exactly the same degree of extension. This probably accounts for most of the apparent variation in skin oxygen uptake per unit area.

The statistics given always represent the mean and standard deviation.

Results

In vitro *experiments with the skin of* Octopus vulgaris

In a closed respirometer, skin from the dorsal part of the abdomen of *Octopus* consumed oxygen at a rate that declined with time. This could have been because the skin is an oxygen conformer or because the condition of the skin declines rapidly with time. To resolve this, pairs of skin samples from freshly killed animals of similar size were tested, one of the pair starting with fully oxygenated and the other with oxygen-depleted water (created by dropping the pressure in a syringe until about half of the oxygen was drawn off and discarded). Circulation within the respirometer was ensured by a magnetic stirrer. The results of three such experiments are shown in Fig. 1A–C. Two further experiments were made with skin from the arms, with similar results. Oxygen consumption fell with time, but this appeared to be due

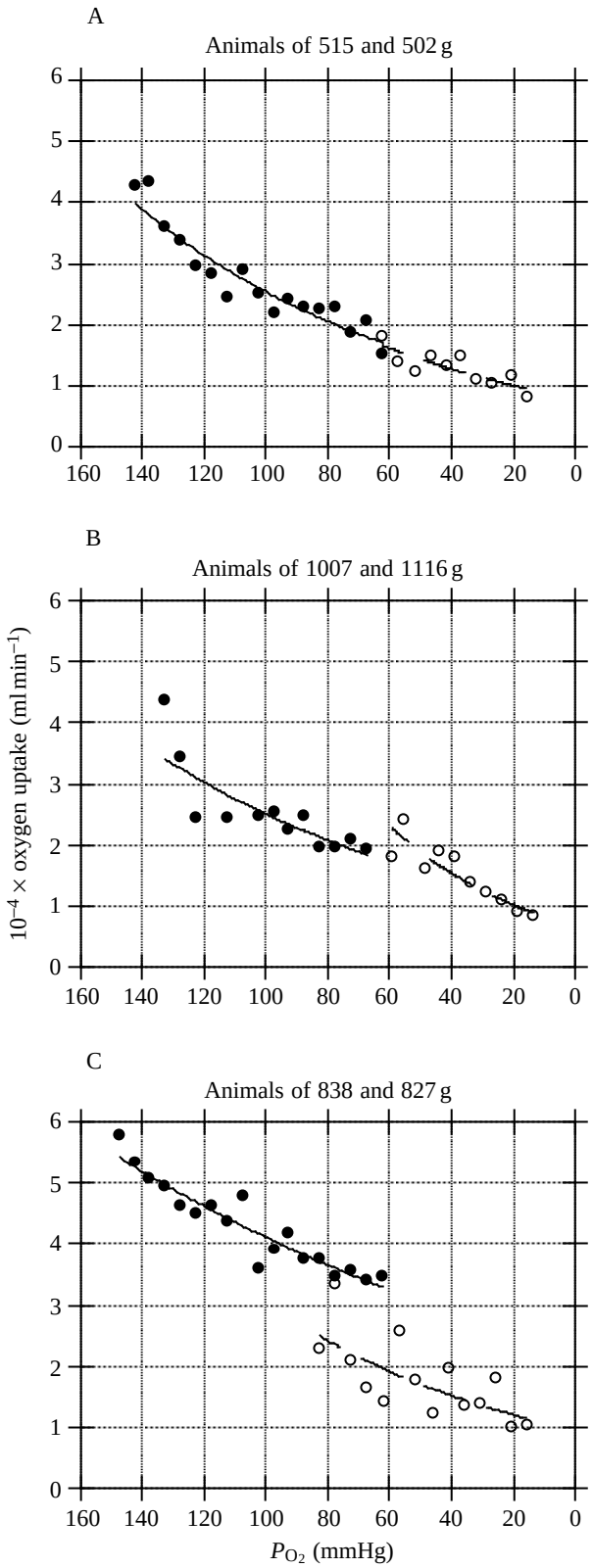

Fig. 1. Three experiments (A–C) on oxygen uptake by the skin of *Octopus vulgaris*. In each case, a skin sample was taken from each of two animals of similar size, immediately following death by decapitation. One sample (filled circles) was placed in normoxic water in a closed respirometer with a magnetic stirrer. The second sample (open circles) was placed in a similar cell filled with partially deoxygenated water at a P_{O_2} that the first sample might be expected to reach in about 60 min. The rate of oxygen uptake plainly depends upon the P_{O_2} and not upon the age of the skin sample. 1 mmHg=0.1333 kPa

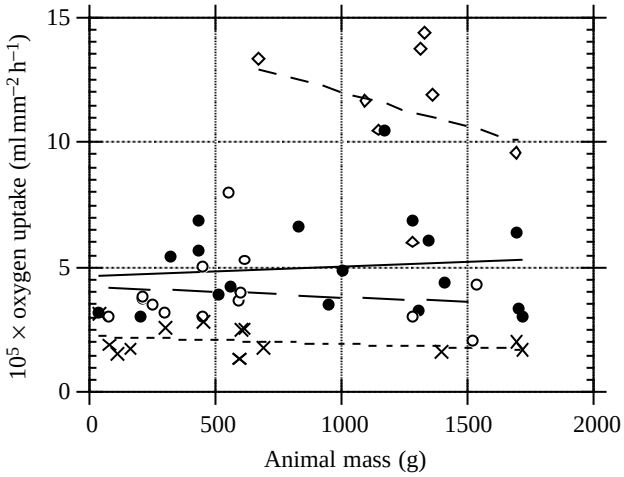

Fig. 2. Cutaneous oxygen uptake by *Octopus vulgaris*. ◇, results from *in vivo* experiments, in which there was a flow of water through a cell bolted to the back of the animal. ● and ○, the oxygen uptakes of skin samples *in vitro* under closed-cell (magnetic stirrer) and through-flow conditions. ×, the results of closed-cell (unstirred) experiments in which a boundary layer was allowed to form over the skin. Continuous water movement over the skin can double oxygen uptake. *In vivo* uptake is two or three times as great as *in vitro*. Linear regressions for each category of experiment indicate no consistent relationship between animal size and skin oxygen uptake.

entirely to the change in P_{O_2}, since fresh skin samples starting at low P_{O_2} values consumed oxygen at the same rate as samples at similar P_{O_2} values that had already been in the respirometer for 60–90 min.

Thirty-two experiments were then made with skin from animals that ranged in mass from 40 to 1718 g. In eighteen of these experiments, the respirometer was closed, with a circulation created by a magnetic stirrer; in each of these experiments, the P_{O_2} started at approximately 155 mmHg and fell progressively to approximately 65 mmHg (1 mmHg= 0.1333 kPa). In the remaining fourteen, a through-flow was maintained at a rate adjusted so that the water entered the chamber at approximately 155 mmHg and left at approximately 130 mmHg. The results are summarised in Fig. 2. Cutaneous uptake averaged $4.95\times10^{-5}\pm1.92\times10^{-5}$ ml mm^{-2} h^{-1} in the closed-circuit experiments and $3.97\times10^{-5}\pm1.48\times10^{-5}$ ml mm^{-2} h^{-1} in the through-flow experiments. There was no detectable change in the rate of skin oxygen uptake with animal size.

Total skin surface area (A) was estimated from 23 sets of measurements from octopuses ranging in mass from 210 to

1705 g. The exponential relationship between skin surface and body mass (m) was found to be:

$$A = 2973m^{0.70}, \quad (1)$$

where A is in mm^2 and m in g.

The resulting estimates of whole-animal cutaneous uptakes were compared with estimates of total whole-animal oxygen uptake ($\dot{V}_{O_2}$ in $ml\,h^{-1}$) calculated on the basis of the formula:

$$\dot{V}_{O_2} = 0.501m^{0.67}. \quad (2)$$

This equation was derived from a total of 341 experiments with 48 animals ranging in mass from 330 to 1200 g, summarised in Wells *et al.* (1983*a*).

Cutaneous uptake, estimated as above, varied from 21 to 79 % of the total expected oxygen uptake of the animal in the closed-circuit experiments and from 15 to 58 % in the through-flow experiments. Both estimates assume that the entire skin surface of the animal is exposed to moving water and that the whole skin surface of the animal takes up oxygen at the same rate as the skin from the dorsal part of the abdomen.

In further experiments to investigate the validity of this second assumption, samples from ten animals were taken both from the dorsal part of the abdomen and from the interbrachial web joining the arms. The rate of oxygen uptake from the dorsal mantle samples averaged $4.55\times10^{-5}\pm1.41\times10^{-5}\,ml\,mm^{-2}\,h^{-1}$, that from the interbrachial web samples averaged $3.94\times10^{-5}\pm0.41\times10^{-5}\,ml\,mm^{-2}\,h^{-1}$. It was concluded that the two areas took up oxygen at the same rate.

The hundreds of suckers in *Octopus vulgaris* provide a very high proportion (35 %) of the total skin area of the animal. The suckers are very active and well supplied with blood vessels. We were unable to dissect off samples of skin with suckers still attached to test *in vitro*. The best we could manage was to cut off an arm and measure the oxygen uptake of this in a small closed-circuit respirometer with a magnetic stirrer. Excised arms remain active for an hour or more *in vitro*. The skin area of the arms and suckers was estimated in each case from arms of similar mass cut from a preserved specimen. Three experiments were carried out. The oxygen uptake averaged $7.45\times10^{-5}\pm2.96\times10^{-5}\,ml\,mm^{-2}\,h^{-1}$. It should be remembered that these preparations included the arm musculature as well as skin and that many of the blood vessels were probably still active; there is much local peristalsis even in excised arms and oxygen may have been carried to underlying tissues, adding to uptake by the skin itself.

Under some conditions, when an octopus is sitting quietly in its hole for example, large areas of the skin will have little or no flow of water passing over them. We tried to mimic this condition *in vitro* by omitting the magnetic stirrer. The cutaneous oxygen uptake of the dorsal mantle skin of five animals under these conditions averaged $1.66\times10^{-5}\pm0.22\times10^{-5}\,ml\,mm^{-2}\,h^{-1}$. The corresponding mean uptake of the total of 32 skin samples tested under the closed stirred cell (N=18) and through-flow (N=14) conditions was $4.55\times10^{-5}\pm1.80\times10^{-5}\,ml\,mm^{-2}\,h^{-1}$.

In a further ten experiments, two skin samples were taken from each animal; one sample was tested in the unstirred condition, the other in the stirred (or through-flow) condition. Unstirred uptakes from these ten animals averaged $2.25\times10^{-5}\pm0.61\times10^{-5}\,ml\,mm^{-2}\,h^{-1}$. Stirred or through-flow values averaged $4.24\times10^{-5}\pm1.13\times10^{-5}\,ml\,mm^{-2}\,h^{-1}$. The mean oxygen uptake for the total of 15 unstirred samples was $2.09\times10^{-5}\pm0.56\times10^{-5}\,ml\,mm^{-2}\,h^{-1}$.

It is concluded that cutaneous oxygen uptake is much reduced if substantial boundary layers of slower-moving or stagnant water are allowed to form over the skin surface.

In vivo *experiments with the skin of* Octopus vulgaris

Octopuses with the basal lobes of the brain removed will live for many weeks; ventilation and circulation appear to be normal, but the animals cannot feed or move about in an integrated manner (Wells, 1978). If a respirometer is bolted onto the back, through the muscle to a plate in the dorsal mantle space, and the nuts are not done up too tightly, circulation to the skin in the respirometer is maintained. With a through-flow circulation and the oxygen electrode transferred at intervals upstream or downstream of the experimental cell covering the skin, cutaneous oxygen uptake can be studied in the living animal under near-normal conditions.

The details of a typical experiment are shown in Fig. 3. Fig. 2 includes a summary of the results of this and of seven further similar experiments. In the first three of these, the cell was stuck (with cyanoacrylate glue) onto the skin; in the last five, including the case illustrated in Fig. 3, the cell was bolted

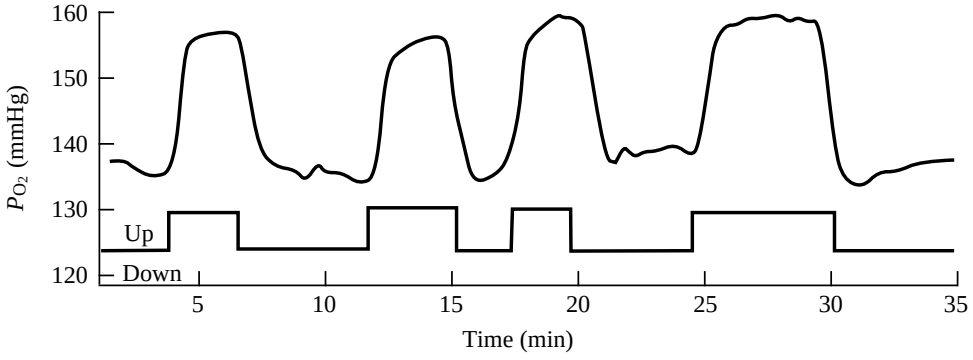

Fig. 3. Details of an *in vivo* experiment with *Octopus vulgaris*. A cell was bolted onto the dorsal mantle surface of an animal with the basal lobes of the brain removed. The oxygen electrode was temporarily transferred from downstream to upstream of the respirometer on four occasions. Through-flow was at a rate of 1 ml in 1.45 min. The results of this and of seven further similar experiments are included in the summary given in Fig. 2. 1 mmHg=0.1333 kPa.

on. Bolts were more satisfactory than glue, because the animal secretes mucus under the glue and it is only a matter of time before the cell begins to peel off the skin and leak around the edges.

Oxygen uptake under these conditions was 2–3 times greater than that found under stirred or through-flow conditions *in vitro*. The eight *in vivo* animals averaged $11.36\times10^{-5}\pm 2.73\times10^{-5}$ ml mm^{-2} h^{-1}, compared with the uptake of $4.55\times10^{-5}\pm1.80\times10^{-5}$ ml mm^{-2} h^{-1} observed *in vitro*.

If these very high rates of uptake were typical of the whole animal, the estimated cutaneous uptake would average 86±21 % of the predicted whole-animal uptake.

Can Octopus vulgaris *import oxygen through the skin?*

The difference between the *in vivo* and the *in vitro* results could arise because the skin is in better condition if the blood supply is intact. It could also mean that the intact blood vessels carry away oxygen to deeper tissues, increasing the diffusion gradient across the thickness of the skin in the cell. Alternatively or additionally, oxygen might be passing through the whole thickness of the skin to underlying muscle and other tissues.

To assess this latter possibility, *in vitro* experiments were carried out with the skin from the dorsal mantle forming a membrane between two respirometer cells. The lower one of these, over the outside surface of the skin, had fully oxygenated water flowing through it. The upper cell contained water that had been partially degassed so that the water on the inside of the skin had a P_{O_2} of 20–40 mmHg, a little lower than the partial pressure range likely for the venous drainage from the mantle. This closed cell had a magnetic stirrer.

In a preliminary experiment, a test membrane was made from a surgical rubber glove and placed between the two cells. Oxygen diffused through this quite rapidly; after 100 min, the P_{O_2} in the upper chamber had risen from 40 to 100 mmHg.

In total, 12 experiments were run with skin forming a membrane between two cells. In three of these, the P_{O_2} rose a little (up to 20 mmHg h^{-1}) in the upper, initially low-P_{O_2} chamber. In the remaining nine, the P_{O_2} fell at similar rates. Far from allowing the passage of oxygen down the P_{O_2} gradient, the skin was generally removing oxygen from the low-oxygen chamber as well as from the chamber containing normoxic water.

The skin-membrane experiments did not, of course, perfectly mimic the *in vivo* situation because the blood vessels *in vitro* were not able to carry away oxygen to other parts of the body. Diffusion through the full thickness of the skin could be slow because of the thick layer of connective tissue on the inside surface. To evaluate this, *in vitro* (closed-circuit, stirred) experiments were made comparing uptake through the inner and outer surfaces of the skin from seven animals. Outer surfaces averaged $5.89\times10^{-5}\pm2.24\times10^{-5}$ ml mm^{-2} h^{-1}, inner surfaces $4.99\times10^{-5}\pm0.97\times10^{-5}$ ml mm^{-2} h^{-1}.

The connective tissue layer cannot therefore be the reason why oxygen does not pass through the skin in the *in vitro* experiments, because the uptake is similar in both orientations of the mantle skin.

Experiments with other cephalopod species

Similar *in vitro* experiments were made with dorsal mantle skin from *Eledone cirrhosa* and *E. moschata*. Four experiments were carried out with, and five without, a magnetic stirrer. Calculations of the total skin surface area of the animals were made in the same manner as for *Octopus vulgaris*. Uptake was $5.53\times10^{-5}\pm3.13\times10^{-5}$ ml mm^{-2} h^{-1} in the stirred condition and $3.81\times10^{-5}\pm2.59\times10^{-5}$ ml mm^{-2} h^{-1} in the unstirred (results from the two species were very similar and have been combined here). These figures are comparable with the *Octopus vulgaris* stirred and unstirred values of $4.55\times10^{-5}\pm1.80\times10^{-5}$ ml mm^{-2} h^{-1} and 2.09±0.56 ml mm^{-2} h^{-1}.

Values for two *Sepia orbigyniana* where skin samples were in cells with magnetic stirrers were 3.49×10^{-5} and 5.67×10^{-5} ml mm^{-2} h^{-1}, and for two *Sepia officinalis* rates of uptake in the absence of stirring were 1.44×10^{-5} and 2.36×10^{-5} ml mm^{-2} h^{-1}.

Eight experiments were made with *Lolliguncula brevis* and five with *Sepioteuthis lessoniana*, all with the skin in the unstirred condition. The results obtained, $2.93\times10^{-5}\pm0.89\times10^{-5}$ and $3.00\times10^{-5}\pm1.11\times10^{-5}$ mm^{-2} h^{-1}, are comparable with those obtained from *Octopus vulgaris*, *Eledone* spp. and *Sepia officinalis*.

Discussion

Does the skin import oxygen?

Observations that oxygen does not readily penetrate the full thickness of *Octopus vulgaris* skin show only that direct diffusion through the skin is unlikely to be the way in which oxygen penetrates to deeper tissues in this species. The fact that uptake *in vivo*, with the cutaneous blood vessels intact, is 2–3 times as rapid as *in vitro* suggests that substantial amounts of oxygen may be carried to deeper tissues by the blood. The few experiments carried out with excised arms again indicate, in a situation where it is still possible that blood vessels are carrying oxygen away from the skin, that there is a greater uptake per mm^2 of skin surface than in *in vitro* tests. Some caution is needed in making this interpretation, however, because the presence of an intact circulation might be expected to improve the condition of the skin and perhaps add to oxygen consumption because of the activities of the blood vessels themselves, all of which exhibit active peristalsis.

The situation in squids remains unresolved; no experiments on oxygen flux through the skin were made. Unlike the skin of octopuses and sepiods, squid skin has no blood vessels. But the skin is very much thinner, so that direct diffusion to the blood vessels and muscles beneath seems likely. On the basis of the metabolites found in the veins draining the mantle, Pörtner (1994) has estimated that as much as 73 % of the oxygen consumed by the mantle musculature may be entering through

the skin. Further experiments are needed because the skin is so very different from that found in octopuses.

Cutaneous oxygen uptake in the life of Octopus vulgaris

An octopus spends much of its life curled up in a hole. At other times, it walks or swims about its environment, with most or perhaps all of its skin surface area exposed to moving water. When in its hole, the only part of the animal exposed to moving water (equivalent to the 'stirred' condition for the skin *in vitro* or the through-flow *in vivo*) would be the internal mantle surface and perhaps also the surface of the head and abdomen, and some part of the upper surfaces of the arms. A rough estimate suggests that half of the total surface of the animal might be available for gas exchange in these circumstances.

In Table 1, we have estimated possible cutaneous uptake on this basis. If all of the skin exposed were well-ventilated, cutaneous oxygen uptake could account for some 41 % of the total oxygen uptake, assuming figures derived from the *in vivo* experiments with flow over the skin. If we assume, at the other extreme, that substantial boundary layers are allowed to form over the exposed parts of the body and adopt figures from the unstirred *in vitro* experiments, this value falls to approximately 8 %. The first assumption is more or less certain to lead to an overestimate of cutaneous uptake, because the whole of the exposed area of the skin is unlikely to be ventilated as thoroughly as in the stirred or through-flow *in vivo* experiments. The second assumption will underestimate cutaneous uptake because the internal mantle surface at least will be well-ventilated. The true figure must lie somewhere between the two extremes.

When the animal leaves its den to forage or to seek for mates, a much higher proportion of the skin surface will be exposed to moving water. In most cases, however, not all of the skin area will be exposed because the animal typically crawls along the bottom with the lower surfaces of the arms and suckers applied to the substratum for much of the time and so unavailable for gas exchange. Only when the animal is swimming (it can jet backwards or forwards but spends only small proportion of each day doing this; Mather and O'Dor, 1991), will the whole skin surface be available for oxygen uptake.

The proportion of total oxygen consumption taken up through the skin may nevertheless fall when the animal is active. An octopus running in an exercise wheel consumes about two and a half times as much oxygen as a resting animal (Wells *et al.* 1983*b*; Houlihan *et al.* 1986). Even assuming *in vivo* levels, cutaneous uptake could supply only 33 % of this. Again, there is an unknown factor; blood vessels in the skin may expand or open up new capillaries when oxygen demand rises, as seems to be the case with the gills (Eno, 1987).

A third condition is envisaged in Table 1. When the animal is digesting a meal, oxygen uptake can again rise to levels comparable with those found during exercise (Wells *et al.* 1983*c*). Food is often taken home to be eaten (Mather and O'Dor, 1991) and the animal will then be curled up in its den, with minimal surface exposure. In these circumstances, cutaneous respiration could drop to as little as 3 % of the total, but is likely to be a little higher because of the ventilation of the internal mantle surface.

Table 1. *A summary of the results of* in vivo *and* in vitro *cutaneous oxygen uptake experiments with* Octopus vulgaris

	In vivo stirred	*In vitro* stirred	*In vitro* unstirred
Skin uptake ($ml\,mm^{-2}\,h^{-1}$)	11.36×10^{-5} $\pm2.73\times10^{-5}$ (N=8)	4.55×10^{-5} $\pm1.80\times10^{-5}$ (N=32)	2.09×10^{-5} $\pm0.56\times10^{-5}$ (N=15)
Total skin uptake for the whole animal ($ml\,h^{-1}$)	26.56	10.64	4.89
Resting			
Whole-animal $\dot{V}_{O_2}$ ($ml\,h^{-1}$)*	32.22	32.22	32.22
Total estimated skin uptake as a percentage of total $\dot{V}_{O_2}$	82.42	33.02	15.18
Half skin surface area available; uptake as a percentage of total	**41.21**	16.51	**7.59**
Active (or feeding)			
Whole-animal $\dot{V}_{O_2}$ ($ml\,h^{-1}$)*	80.55	80.55	80.55†
Skin uptake as a percentage of total $\dot{V}_{O_2}$ during exercise or feeding	**32.97**	13.21	**3.04**†

Estimates of the proportion of the animal's total oxygen uptake that skin uptake would represent under a variety of conditions are also given.

Calculations are made for a 500 g animal.

*These $\dot{V}_{O_2}$ values are obtained using equation 2 in text.

†During exercise, the boundary layer would always be stirred. During feeding, when $\dot{V}_{O_2}$ can also increase by 2.5-fold (Wells *et al.* 1983*c*), the animal would be sitting with about half of the skin area exposed, much of it unventilated.

At present we can say very little about cutaneous uptake by other species. Sepioids typically dig into sand or mud when not actively searching for food or mates and, in this condition at least, could not rely upon significant cutaneous respiration, a situation that would change as soon as they became active. Squids are nearly always moving (*Lolliguncula brevis* sometimes rests on the bottom but never digs in) and are typically surrounded by well-aerated water. Cutaneous respiration could be important here, as Pörtner's (1994) biochemical studies suggest, despite their very high metabolic rates.

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