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Respiration by buried echidnas *Tachyglossus aculeatus*

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Summary

Short-beaked echidnas have an impressive ability to submerge completely into soil or sand and remain there, cryptic, for long periods. This poses questions about how they manage their respiration, cut off from a free flow of gases. We measured the gradient in oxygen partial pressure (P_{O_2}) away from the snouts of buried echidnas and oxygen consumption ($\dot{V}_{O_2}$) in five individuals under similar conditions, in two substrates with different air-filled porosities (f_a). A theoretical diffusion model indicated that diffusion alone was insufficient to account for the flux of oxygen required to meet measured rates of $\dot{V}_{O_2}$. However, it was noticed that echidnas often showed periodic movements of the anterior part of the body, as if such movements were a deliberate effort to flush the tidal air space surrounding their nostrils. These 'flushing movements' were subsequently found to temporarily increase the levels of interstitial oxygen in the soil around the head region. Flushing movements were more frequent while $\dot{V}_{O_2}$ was higher during the burrowing process, and also in substrate with lower f_a. We conclude that oxygen supply to buried echidnas is maintained by diffusion through the soil augmented by periodic flushing movements, which ventilate the tidal airspace that surrounds the nostrils.

Key words: monotreme, burrowing, respiration, gas exchange, oxygen consumption, echidna.

Introduction

Short-beaked echidnas (*Tachyglossus aculeatus*) are famous for many unusual characteristics, among them the ability to avoid capture or predation by 'sinking' into the soil until only the tips of the dorsal spines are visible (Burrell, 1926) and remaining there, holding fast against attempts to dislodge them, for long periods. This behaviour poses questions about how echidnas manage their respiratory gas exchange, cut off from a free flow of gases and threatened by the risk of inhaling particles of soil.

It is clear that echidnas are frequently exposed, at least for short periods, to increased hypoxia and hypercapnia, either when digging into soil or within their burrows (Augee et al., 1971). Previous studies have demonstrated that echidnas are physiologically well suited for burrowing. Augee et al. (Augee et al., 1971) covered echidnas with soil to simulate natural conditions and found them to be very tolerant of high carbon dioxide (CO_2) and low oxygen (O_2) under these conditions.

However, no studies have explored the processes by which O_2 supply is maintained while an echidna is buried, completely surrounded by soil, without any tunnel to facilitate the convective movements of gas. Presumably echidnas re-breathe the interstitial gas around them while buried. The diffusive transport of O_2 to a buried mammal was explored in the Namib Desert golden mole *Eremitalpa granti namibensis* (Seymour and Seely, 1996), which survives long periods buried in sand. By comparing measurements of the P_{O_2} gradient away from the snout of a buried mole to the P_{O_2} gradient predicted from a mathematical model of gaseous diffusion through sand, Seymour and Seely (Seymour and Seely, 1996) explained that golden moles can survive being buried in sand by utilizing O_2 diffusing through sand into the tidal air space that surrounds the snout. However, their model predicted an upper size limit of approximately 200 g for resting mammals able to continue respiration in this manner. Echidnas are commonly 2–4 kg and can reach 7 kg, so the model used to explain sub-sand respiration by the golden mole cannot by itself explain the submergence capabilities observed in echidnas.

In this study we took a similar approach to that of Seymour and Seely (Seymour and Seely, 1996). We measured the P_{O_2} gradient away from the snouts of submerged echidnas and $\dot{V}_{O_2}$ in separate experiments on five individuals under similar conditions, in two media with differing porosity f_a values, with the hope of determining how echidnas can remained buried for long periods of time.

Materials and methods

Echidnas

Five echidnas *Tachyglossus aculeatus* Shaw (2.34–4.18 kg) previously fitted intraperitoneally with calibrated temperature

transmitters were used. Animals had been collected previously from Idalia National Park (latitude 24°53′S, longitude 144°46′E), 113 km WSW of Blackall in Australia's semi-arid zone (Brice et al., 2002), and the Texas area, 50 km SW of Stanthorpe in SE Queensland (latitude 28°43′S, longitude 151°28′E). The animals were held in a free-range enclosure at the University of Queensland's Pinjarra Hills Veterinary Farm.

Modelling the diffusive exchange of respiratory gases

The diffusion model (Seymour and Seely, 1996) assumes that the buried animal is surrounded completely by a medium that permits O_2 to diffuse radially towards it from all directions. This assumption is supported by empirical data (Seymour and Seely, 1996; Wilson and Kilgore, 1978; Withers, 1978). In a steady state, the amount of O_2 diffusing radially through a given spherical shell is equal to the rate at which it is consumed. As O_2 diffuses radially toward the animal, the volume through which it passed decreases and, therefore, the changes in ambient P_{O_2} shell by shell, with decreasing distance to the animal, can be calculated from the equation (Seymour and Seely, 1996):

$$\dot{V}_{O_2} = K_{O_2}(P_o - P_i)4\pi r_o r_i / r_o - r_i \,, \quad (1)$$

where $\dot{V}_{O_2}$ is the rate of oxygen consumption ($cm^3\ min^{-1}$), K_{O_2} is the diffusion coefficient of oxygen in substrate ($cm^2\ min^{-1}\ kPa^{-1}$), P_o and P_i are the oxygen partial pressures (kPa) at the outer (r_o) and inner (r_i) radii of a given spherical shell (cm). The following additional assumptions were made for the model: (1) K_{O_2} was the product of the binary diffusion coefficient of oxygen in air (D_{O_2}=12.1$cm^2\ min^{-1}$ at 25°C) (Nobel, 1983), the O_2 capacitance of air β_{O_2}=–0.0098$cm^3\ cm^{-3}\ kPa^{-1}$ (Seymour and Seely, 1996) and the air-filled porosity coefficient=$f_a^{1.5}$ (Marshall, 1959; Seymour and Seely, 1996). K_{O_2} was therefore taken as 0.032 $cm^2\ min^{-1}\ kPa^{-1}$ in coarse sand of f_a=0.42, and 0.053 $cm^2\ min^{-1}\ kPa^{-1}$ in kitty litter (a water absorbent granular substance made from recycled newspaper) of f_a=0.580 (see later). (2) P_o was assumed in the earlier study (Seymour and Seely, 1996) to be atmospheric at r_o=100 cm. In our case, the echidna was in a large plastic bin and the surface through which diffusion was possible was therefore constrained. The environment of this experiment is therefore more inimical to gaseous diffusion than that on which the model is based, and that needs to be kept in mind when interpreting the results. (3) The internal radius was the radius of a sphere of sand containing a volume of interstitial gas equal to the tidal volume of the animal's breath (Seymour and Seely, 1996). This assumption is based on the premise that a volume of interstitial air in the sand, equal to tidal volume, was constantly being re-breathed and mixed by the animal. The radius of this 'tidal space' was calculated using the equation (Seymour and Seely, 1996):

$$r_i = (3V_T/4\pi f_a)^{1/3} \,, \quad (2)$$

in which V_T is the tidal volume. V_T (cm^3) was calculated from animal mass (kg) using an equation for echidnas (Bech et al., 1992), V_T=8.96 ml kg^{-1}.

To apply the model to echidnas, $\dot{V}_{O_2}$ values and P_{O_2} gradients away from buried animals were measured separately, and the assumption made that the $\dot{V}_{O_2}$ measured while submerged was indicative of the $\dot{V}_{O_2}$ during measurement of the P_{O_2} gradient, as was done previously with the golden mole (Seymour and Seely, 1996).

Measurement of $\dot{V}_{O_2}$

$\dot{V}_{O_2}$ was measured using a flow-through respirometry system, at an ambient temperature of 25°C. Each animal was placed in an air tight, 50 cm deep and 40 cm in diameter, cylindrical chamber ¾-filled with a test medium into which the echidnas could burrow. Gas entered the chamber into an air space above the burrowing medium and was extracted from below the burying medium through a wire mesh-covered hole at the base of the chamber. Two media with differing f_a values were used and each animal was exposed to each medium. The f_a of each of the two media was measured by filling a 1000 ml graduated cylinder with the medium and slowly adding it to 1000 ml of water in a 2000 ml cylinder, avoiding any bubbles (Seymour and Seely, 1996). The kitty litter, which would otherwise have absorbed water, was first sprayed with a water repellent spray (Motortech, Balchan International, Australia). While this method discounted any porosity of the kitty litter itself, we consider this to have been negligible.

The lid of the respirometry chamber was transparent, so lung ventilation movements could be directly observed during these experiments even when the animal was completely buried (the surface of the substrate moved slightly with each breath). The behaviour of an animal on introduction to the respirometry chamber, whether for $\dot{V}_{O_2}$ or P_{O_2} gradient measurements, was to burrow immediately vertically downward until submerged completely in the substrate, a behaviour identical to that of echidnas burying themselves in the wild. $\dot{V}_{O_2}$ was measured while the animal remained buried. Resting metabolic rate was considered to have been reached when the animal had been submerged and resting for 4 h and the fractional concentration of O_2 in the excurrent air was stable. Air from the chamber was passed through small diameter tubing to a CO_2 absorbent (Soda Lime) and then a desiccant (Drierite™) before passing into a mass flow meter (MFS-1; Sable Systems, Las Vegas, NV, USA). The mass flow meter pulled air though the chamber and its exhalent air was sampled *via* an oxygen analyser (Sable Systems PA-1B) calibrated with oxygen-free gas (0.00% O_2) and room air (20.95% O_2). The flow rate through the respirometry chamber was 750 ml min^{-1} in all cases. Body temperature T_b was monitored throughout the measurement period using the implanted temperature transmitter and a radio receiver connected to a pulse meter. The output voltage from the O_2 analyser and pulse meter were fed into PowerLab hardware (ADInstruments, Sydney, NSW, Australia) connected to a computer running Chart5 software (v5.0.1. ADInstruments).

Measurement of P_{O_2} at different distances from the snout in buried echidnas

Measurements of P_{O_2} within the substrate surrounding buried echidnas were performed in a large cylindrical plastic bin (measurements given above) at an ambient temperature of 25°C. Experiments were commenced by placing the animal on top of the substrate and, in all cases, echidnas burrowed immediately until they were completely submerged.

Gas samples from the snout region were collected through silicone tubing (1 mm i.d.) after flushing dead space from the tubing into 3 ml plastic syringes. The tip of this tube was secured onto the nose of the animal above the nostril using a combination of medical glue (collodian) and micropore tape. To measure the P_{O_2} gradient within the medium away from the snout region, further lengths of silicon tubing were attached at 2 cm intervals along the initial tubing, up to 10 cm away from the snout tip.

Gas samples (2 ml) were analysed for O_2 content with a thermally stabilised Clarke Oxygen Electrode (DOX, Analytical Sensors, Inc., Sugarland, TX, USA), connected to a Radiometer PHM73 gas analyser (Copenhagen, Denmark), calibrated with outside air (20.95% O_2) and oxygen-free gas.

Gas samples were taken immediately on submergence of the echidna, and further samples were taken at 15 min intervals for a 5 h period. Each animal was measured individually in each of the media. T_b was also monitored throughout these experiments using the implanted temperature transmitter.

Measurement of P_{O_2} and movement

During trial measurements of O_2 tensions around buried echidnas, the animals would move periodically. Such movements would be followed by a rise in P_{O_2} levels in the substrate, seemingly as a result of these movements. To determine if there was a causal relationship between the movements of echidnas in different substrates and the P_{O_2} measured in the snout region while submerged, a piezoelectric movement sensor (Sigma Delta technologies, Perth, Western Australia) was attached to the echidnas. This sensor was wrapped in electrical tape and glued to a dorsal spine on the shoulder region of the animal. The output voltage from the movement sensor was recorded, simultaneously with T_b and measured P_{O_2} samples, using PowerLab hardware connected to a computer running Chart5 software (v5.0.1. ADInstruments). Respiration rate and larger 'flushing movements' were detected by the movement sensor (Fig. 1).

Statistics

All results are presented as means ± s.d. Differences between empirical and theoretical values of P_{O_2} in substrate surrounding buried echidnas were tested using paired *t*-tests. Significance was assumed at $P<0.05$.

Results

Data were collected on $\dot{V}_{O_2}$ of each echidna, the P_{O_2} in the snout region, and movement and activity level while submerged in two different substrates, coarse sand (f_a=0.42) and kitty litter (f_a=0.58).

$\dot{V}_{O_2}$ measurements while submerged

$\dot{V}_{O_2}$ measurements were taken at a chamber temperature of 25°C. As would be expected, $\dot{V}_{O_2}$ was greater during burrowing than resting, and substrate type did not influence resting rate of $\dot{V}_{O_2}$ (Table 1). T_b and respiration rate always decreased during the course of $\dot{V}_{O_2}$ measurements. T_b of echidnas at the start of the experiments ranged from 27.0 to 34.5°C and, on average, decreased 2.2±1.3°C (N=5) during a trial. Respiration decreased from 12.0±0.5 to 4.6±2.2 breaths min^{-1} over the 5 h measurement period.

P_{O_2} gradient in substrate while submerged

P_{O_2} measurements were taken at an ambient temperature of 25°C. The mean atmospheric P_{O_2} was 19.7 kPa (range 19.3–19.9 kPa) in water-saturated air. The P_{O_2} level in each

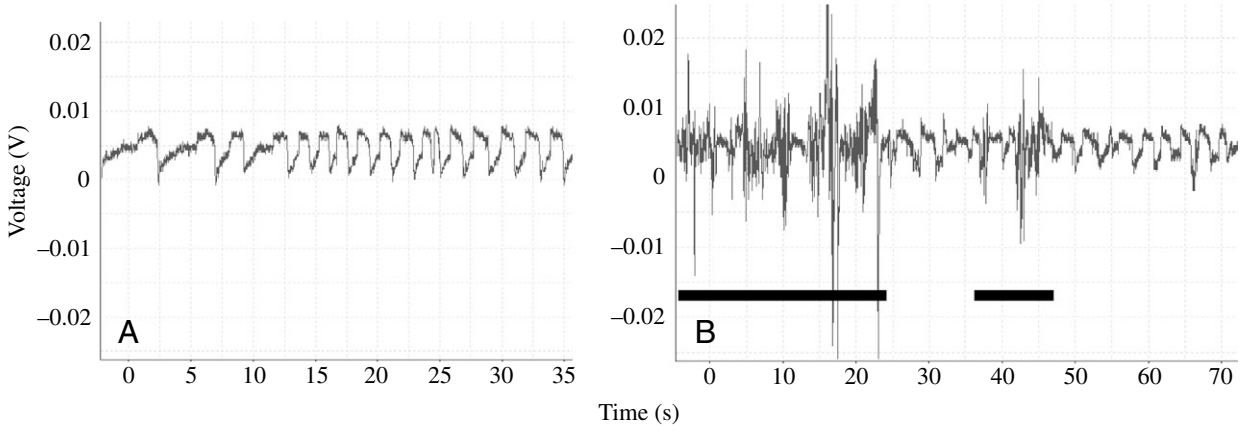

Fig. 1. Voltage output from the movement sensor when attached to the shoulder region of a buried echidna. (A) Lung ventilation movements; increase in voltage represents inspiration, and decrease, expiration. (B) 'Flushing movements', indicated by dark horizontal bars as well as lung ventilation movements.

Table 1. *$\dot{V}_{O_2}$ measurements from each echidna buried in sand and kitty litter substrates*

		$\dot{V}_{O_2}$ (ml g^{-1} h^{-1})			
		Burrowing		Resting	
Echidna	Mass (g)	Sand	Kitty litter	Sand	Kitty litter
E1	4175	0.194	0.110	0.161	0.097
E2	2950	0.114	0.202	0.096	0.137
E3	2600	0.190	0.196	0.102	0.204
E4	2420	0.197	0.217	0.028	0.154
E5	2360	0.271	0.292	0.155	0.223
Mean	2901±748	0.193±0.056	0.203±0.065	0.108±0.054	0.163±0.051

Sand, f_a=0.42; kitty litter, f_a=0.58.
Mean values are ± s.d. (N=5).
A one-way ANOVA indicated that resting $\dot{V}_{O_2}$ was not significantly different in different substrates (P=0.102).

substrate, without an echidna, was atmospheric. P_{O_2} levels of gas samples from near the snout of echidnas buried in the medium were always below the atmospheric level, even immediately after burial. The mean minimum P_{O_2} values at the tip of the echidna's snout during burrowing in each substrate were 12.1±1.4 kPa in coarse sand (f_a=0.42, N=5) and 12.3±1.4 kPa in kitty litter (f_a=0.58, N=5). Gas samples taken at a series of distances away from the snout revealed a P_{O_2} gradient within the substrate (Table 2).

T_b and respiration rate always decreased over time, independent of medium, during experiments that measured P_{O_2} gradients. T_b of echidnas at the start of the experiments ranged from 28.0 to 35.0°C and, on average, decreased 1.2±0.5°C (N=5) during a trial. Respiration decreased from 12.0±0.5 to 4.7±3.3 breaths min^{-1} over the 5 h measurement period.

Comparison of measured P_{O_2} values with those predicted by modelling diffusive exchange of oxygen

Measured $\dot{V}_{O_2}$ data and published values for tidal volume (Bech et al., 1992) were used to generate theoretical diffusion gradients away from the snout for each of the substrates, using Eqn 1. These theoretical P_{O_2} values at distances away from the snout were compared with empirically measured P_{O_2} values around buried echidnas (Figs 2, 3).

In kitty litter, which had the highest porosity (f_a=0.58), the theoretically calculated P_{O_2} values away from the snout were not significantly different from the measured values (Fig. 2), implying that diffusion into the tidal space was sufficient in this porous medium to accommodate the oxygen requirements of a resting, buried echidna.

In the natural substrate, coarse sand (f_a=0.42) the theoretical calculated values were significantly different from the measured values (Fig. 3). In coarse sand, supply of oxygen by diffusion alone was apparently insufficient to account for the measured rates of $\dot{V}_{O_2}$.

P_{O_2} in relation to body movements

A behavioural pattern was observed in the experiments that measured P_{O_2} profiles in the substrate surrounding buried echidnas. P_{O_2} at any measurement distance from the snout did

Table 2. *P_{O_2} profiles surrounding echidnas buried in kitty litter and course sand*

		Distance from snout (cm)											
		Minimum P_{O_2} (kPa)						Equilibrium P_{O_2} (kPa)					
Echidna	Mass (g)	0	2	4	6	8	10	0	2	4	6	8	10
Kitty litter													
E1	4175	12.56	15.3	16.37	18.1	18.77	19.4	15.44	16.3	17.6	18.4	19	19.24
E2	2950	12.2	14.5	14.95	15.6	16.79	17.5	14.2	15.6	16.1	17.2	18	18.4
E3	2600	12.87	15.6	16.99	17.1	17.27	18	14.11	15.2	16.2	16.6	16.8	17.8
E4	2420	13.86	16.4	17.83	18.2	18.34	18.7	14.49	17.3	18.4	18.7	18.7	19.16
E5	2360	10.07	15.5	17.21	18.3	18.54	18.9	12.98	15.3	17.1	18.2	18.6	19.15
Sand													
E1	4175	11.52	13	15	16.5	19.43	19.7	14.8	17.5	18	19	19.8	19.82
E2	2950	11.47	13.4	15.25	16.6	17.07	18.5	15.57	17	17.3	17.9	18.6	19.05
E3	2600	14.6	16	16.63	16.8	16.85	17.7	17.32	18	18.8	19	19	19.5
E4	2420	13.56	15	17.51	18.4	19.1	19.2	15.8	16.3	17.3	18.5	19.1	19.75
E5	2360	9.27	14	14.68	15.9	16.66	19.6	11.15	15.8	16.7	17.5	18.3	19.34

Sand, f_a=0.42; kitty litter, f_a=0.58.

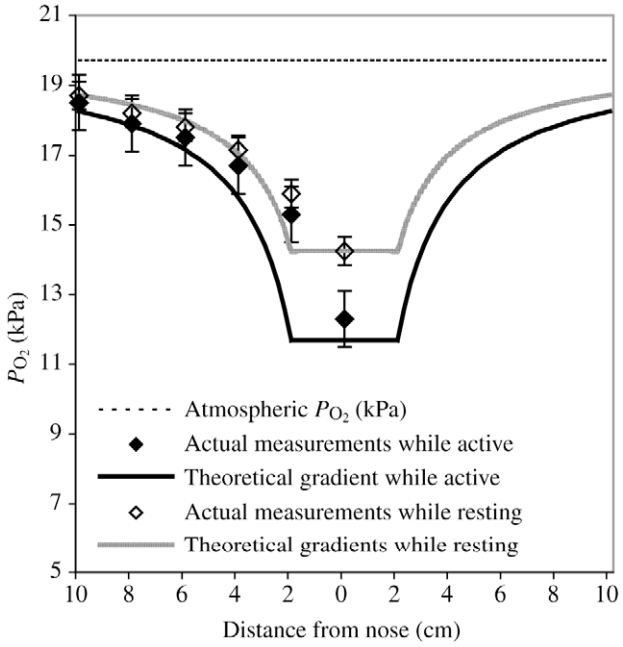

Fig. 2. The relationship between theoretical values, as predicted by the model (Seymour and Seely, 1996), and mean empirical measurements, from five echidna buried in kitty litter (f_a=0.58). The curves are calculated from an equation for echidna tidal volume (Bech et al., 1992) and $\dot{V}_{O_2}$ of a 2.9 kg echidna buried in kitty litter. The upper curve represents an individual with a resting metabolic rate at steady state. The lower curve represents an individual with higher metabolic rate while actively burrowing into the substrate. Theoretical and empirically measured values were compared using paired Student *t*-test (minimum values, *P*=0.067; steady state values, *P*=0.241).

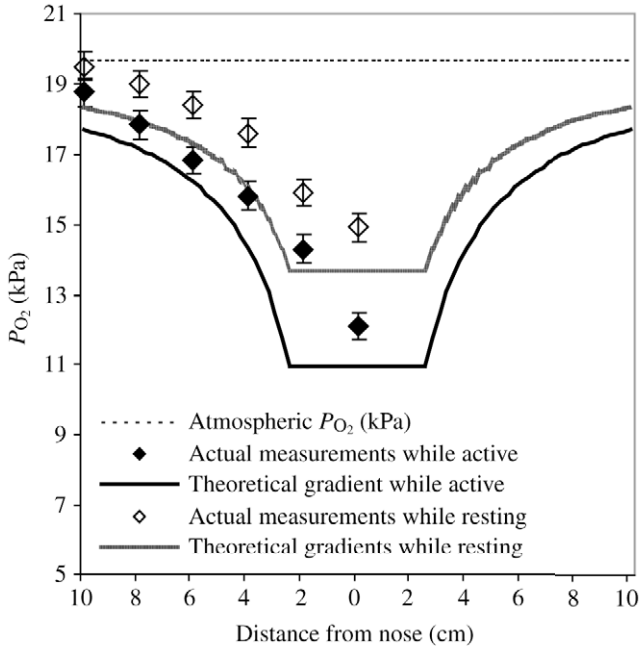

Fig. 3. The relationship between theoretical values, as predicted by the model (Seymour and Seely, 1996), and mean empirical measurements from five echidna buried in coarse sand (f_a=0.42). The curves are calculated from an equation for echidna tidal volume (Bech et al., 1992) the $\dot{V}_{O_2}$ of a 2.9 kg echidna buried in sand. The upper curve represents an individual with a resting metabolic rate at steady state. The lower curve represents an individual with higher metabolic rate while actively burrowing into the substrate. Theoretical and empirically measured values were compared using paired Student *t*-test (minimum values, *P*=0.010; steady state values, *P*=0.001).

not remain constant, it fluctuated in a cyclic manner and mean values tended to increase as the experiment progressed (Fig. 4). When first introduced to the burrowing medium, echidnas immediately dug down until completely encased in the substrate. At this stage, animals had a relatively high $\dot{V}_{O_2}$, T_b and respiration rate, and the minimum P_{O_2} near the snout was always lowest during this early phase of an experiment. The minimum P_{O_2} rose over time as $\dot{V}_{O_2}$ fell to a resting rate (Fig. 4). Periodic movements of the anterior body were more common early in these experiments when oxygen consumption rates were higher, and these movements were followed soon after by an increase in P_{O_2} close to the snout and further away from the snout (Fig. 4) indicating that such 'flushing movements' caused the convective movement of oxygen from the atmosphere to the snout area.

The ability of echidnas to respire while submerged in different media

The frequency of flushing movements varied between kitty litter and course sand (Fig. 4). In kitty litter there were approximately two movements h^{-1} for the first hour and then one movement h^{-1} for the rest of the experiment (Fig. 4A), while in course sand there were on average 6.2 movements h^{-1} for the first hour, 4.6 movements h^{-1} for the second hour, and two movements h^{-1} for the rest of the trial (Fig. 4B).

Table 3. *Resting $\dot{V}_{O_2}$ values for T. aculeatus from different studies*

Mass* (kg)	Range (kg)	$\dot{V}_{O_2}$ (ml g^{-1} h^{-1})	Reference
2.901±0.75	2.34–4.18	0.108	Present study
3	2.5–3.5	0.217	Schmidt-Nielsen et al., 1966
2.3	2–2.8	0.18	Parer and Hodson, 1974
	2.64–4.22	0.132	Dawson et al., 1979
2.73±0.85	1.54–4.27	0.174	Bech et al., 1992
3.126±0.633		0.206	Frappell et al., 1994

*Values are means ± s.d.

Discussion

Modelling of diffusive exchange of respiratory gases

Comparison of measured and theoretical P_{O_2} gradient

It appeared that echidnas were able to maintain their O_2 supply while encased in substrate, including while they were digging in, by a combination of diffusion through the substrate

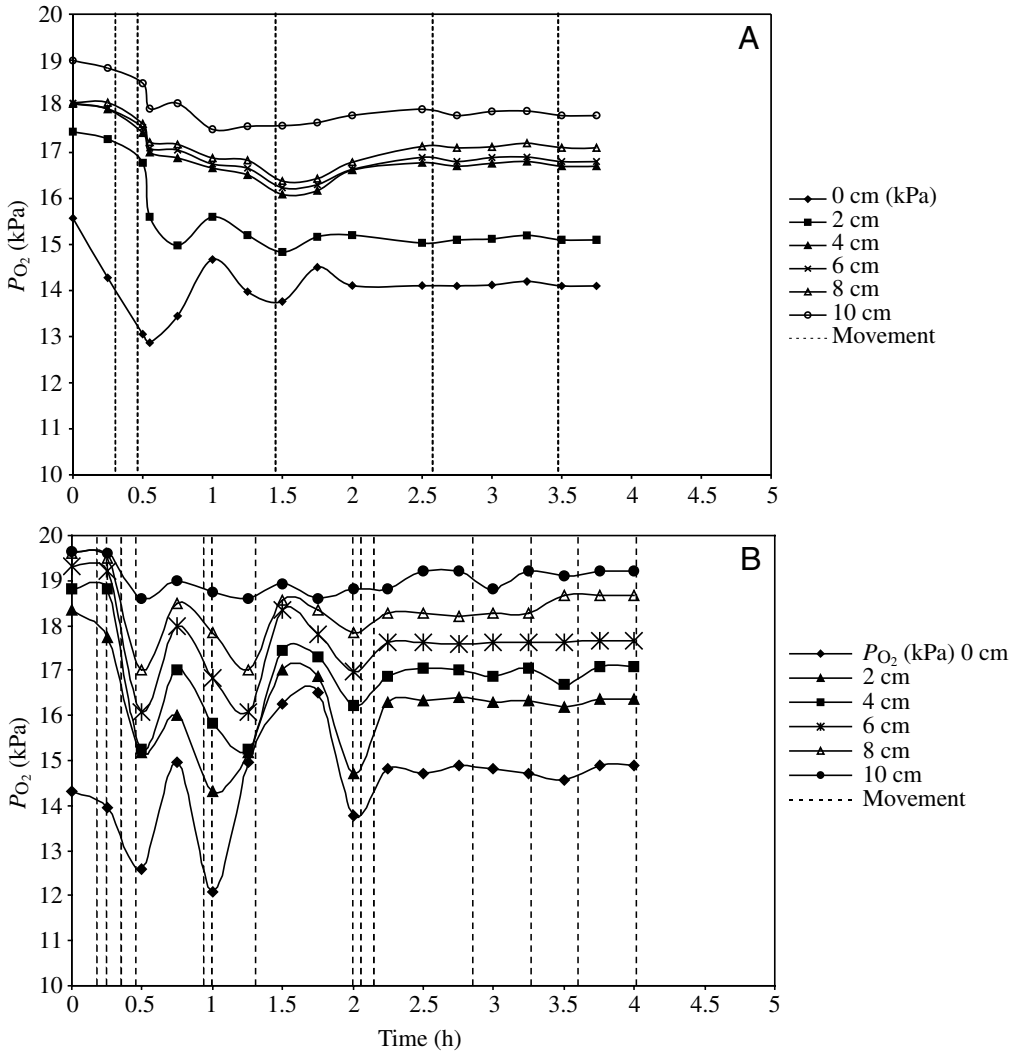

Fig. 4. The relationship between flushing movements and the P_{O_2} profiles surrounding echidnas while buried in two different media. Movements, shown by the dotted vertical lines, are associated with fluctuations in P_{O_2} until a steady state is reached where the P_{O_2} levels stay constant over time. Different symbols represent the P_{O_2} values in the medium away from the snout region of the echidna. (A) Typical echidnas buried in kitty litter. (B) Typical echidnas buried in coarse sand.

augmented by periodic movements, which flush the interstitial air space around the nose. Measured $\dot{V}_{O_2}$ and published values for tidal volume (Bech et al., 1992) were used to generate theoretical P_{O_2} profiles away from the snout region, with which measured P_{O_2} values could be compared. In kitty litter, at rest, there was no significant difference between the measured and the predicted gradients, suggesting that diffusion was sufficient to meet the O_2 requirements when buried in this high porosity medium, as for golden moles buried in fine sand (Seymour and Seely, 1996). Even in this high porosity medium, however, resting echidnas chose to make periodic movements, albeit infrequently.

In the natural substrate, empirically measured and theoretical P_{O_2} values were significantly different. The actual mean minimum P_{O_2} values at the snout region and along the gradient were significantly greater than predicted by the model, suggesting that diffusive oxygen transport through the sand alone was insufficient to account for the measured $\dot{V}_{O_2}$. The higher than predicted P_{O_2} values appear to result from the periodic flushing movements during submergence, which caused temporarily increased interstitial O_2 levels closer to the snout.

The effect of periodic movement on the P_{O_2} profiles surrounding buried echidnas

Periodic movements were more frequent in coarse sand, particularly in the earlier phase of an experiment when $\dot{V}_{O_2}$ was highest. The higher frequency of 'flushing' movements at the beginning of the burying trials enabled the echidna to stay submerged while experiencing higher O_2 demand, but as O_2 demand decreased later in trials, the frequency of flushing movements decreased. Once a resting rate of $\dot{V}_{O_2}$ was achieved, a steady state was achieved and flushing movements were regular but less frequent and the P_{O_2} profiles became more or less constant.

$\dot{V}_{O_2}$ and T_b of buried echidnas

Monotremes are characterized by metabolic rates and T_b that are lower than those typical of eutherian mammals (Bech et al., 1992; Griffiths, 1978; Schmidt-Nielsen et al., 1966) and this was confirmed in our study. Basal metabolic rates of echidnas have been reported to be only 25–50% of that predicted for eutherian mammals (Bech et al., 1992; Dawson et al., 1978; Dawson et al., 1979; Schmidt-Nielsen et al., 1966). However,

echidnas are notoriously difficult subjects in which to measure resting $\dot{V}_{O_2}$, usually being very restless and attempting to escape while in classical respirometry chambers. This is the first study to measure $\dot{V}_{O_2}$ in echidnas buried in substrate. Indeed, further work in our laboratory has shown that providing echidnas with even a small quantity of material in the respirometer, into which they can bury their head, makes it much easier to achieve measurements at apparently resting levels (P. H. Brice, G. C. Grigg and L. A. Beard, unpublished observations). Accordingly, we think that buried echidnas are more relaxed than echidnas in a respirometer without anywhere to 'hide' and are more likely to provide good data on resting metabolic rates.

The $\dot{V}_{O_2}$ measured from echidnas in this study were also somewhat more variable than might be expected for metabolic rate measured in a typical mammal at rest. The variability may be explained in terms of their heterothermy. Echidnas have the advantages of endothermy, including the capacity for impressive homeothermic endothermy during incubation (Grigg et al., 2004). The modal T_b of active echidnas is 32°C (Grigg et al., 2004). However, they are very relaxed about using thermoregulatory mechanisms to maintain a stable T_b and periods of rest are typically accompanied by a drop in T_b. Accordingly, cyclic changes in daily T_b of 3–6°C are routine. They also show both short- and long-term torpors (Grigg et al., 2004). In our study, T_b at the start of a trial differed between echidnas and this is likely to account for much of the variability in measured $\dot{V}_{O_2}$. The declines in T_b during each experimental trial reflect the expected drops, which occur in echidnas at rest after a period of activity.

The magnitude of the decrease in ambient P_{O_2} surrounding burrowed echidnas

The P_{O_2} of the immediate O_2 environment of the burrowing echidna showed a decrease in ambient oxygen to about 11 kPa, substantially below atmospheric (21 kPa). This is a much greater drop than was found in golden moles buried in sand (Seymour and Seely, 1996), where ambient P_{O_2} values were about 20 kPa. Kuhnen (1986) summarized published data on the burrow O_2 and CO_2 levels for 13 species of burrowing mammals. Typically, these species were exposed to P_{O_2} values between 17–20 kPa, but values down to 10 kPa have occasionally been recorded (Kuhnen, 1986). The results from the present study showed that the immediate O_2 environment of buried echidna near the snout was 12.3±0.2 kPa while active and 14.9±0.2 kPa while resting. A P_{O_2} of 11 kPa was recorded (Augee et al., 1971) in an echidna encased in substrate for a period of 4 h at a depth of 20 cm, which is in good agreement with our study. Bentley et al. (Bentley et al., 1967) found a P_{O_2} of 13.9 kPa in an echidna completely encased in crushed corncobs at a depth of 30–60 cm. The O_2 environment tolerated by buried echidnas seems to put them at the extreme end among fossorial and semi-fossorial mammals.

Behavioural adaptations of echidnas while submerged in different substrates

O_2 supply is one aspect of survival under soil, but the mechanical problem of breathing in loose material also needs to be explored. The porosity of coarse sand was low due to a discontinuity of pore spaces and the smaller particles filling in the void spaces between the larger particles. The echidna apparently used this discontinuity to stay submerged and was able to use its snout to create a small air space and prevent the inhaling of soil particles into the nose.

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