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To what extent might N_2 limit dive performance in king penguins?

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Summary

A mathematical model was used to explore if elevated levels of N_2, and risk of decompression sickness (DCS), could limit dive performance (duration and depth) in king penguins (*Aptenodytes patagonicus*). The model allowed prediction of blood and tissue (central circulation, muscle, brain and fat) N_2 tensions (P_{N_2}) based on different cardiac outputs and blood flow distributions. Estimated mixed venous P_{N_2} agreed with values observed during forced dives in a compression chamber used to validate the assumptions of the model. During bouts of foraging dives, estimated mixed venous and tissue P_{N_2} increased as the bout progressed. Estimated mean maximum mixed venous P_{N_2} upon return to the surface after a dive was 4.56±0.18 atmospheres absolute (ATA; range: 4.37–4.78 ATA). This is equivalent to N_2 levels causing a 50% DCS incidence in terrestrial animals of similar mass. Bout termination events were not associated with extreme mixed venous N_2 levels. Fat P_{N_2} was positively correlated with bout duration and the highest estimated fat P_{N_2} occurred at the end of a dive bout. The model suggested that short and shallow dives occurring between dive bouts help to reduce supersaturation and thereby DCS risk. Furthermore, adipose tissue could also help reduce DCS risk during the first few dives in a bout by functioning as a sink to buffer extreme levels of N_2.

Key words: breath-hold diving, decompression sickness, mathematical modeling, aerobic dive limit.

Introduction

King penguins perform extended foraging dive bouts, with long (5 min) and deep (200 m) dives interspersed with short surface intervals (<2 min). The short surface interval is incompatible with paying off a metabolic acidosis that would have accrued if the animals had exceeded their aerobic dive limits (ADL) (Kooyman and Ponganis, 1998). In fact, maximum calculated ADL (cADL, O_2 stores/rate of oxygen consumption $\dot{V}_{O_2}$) is 5.4–7.0 min if $\dot{V}_{O_2}$ during diving is at resting levels (Fahlman et al., 2005; Froget et al., 2004) and O_2 storage capacity between 45 ml O_2 kg^{-1} (Ponganis et al., 1999) and 58 ml O_2 kg^{-1} (Kooyman and Ponganis, 1998). This estimate suggests that >95% of all dives are performed aerobically (Froget et al., 2004). In the king penguin, a surface interval exceeding 15 min has been considered to mark the end of a dive bout (A.S., J.-P. Gendner, C. A. Bost and Y.H., manuscript submitted for publication). Extended surface intervals, or interbout periods, occur more frequently during the night but there are also regular terminations of dive bouts during daylight. Therefore, an important question to pose is why do king penguins stop foraging, sometimes for hours, during a period when food may be readily available? If king penguins seldom exceed their ADL, are there other plausible reasons for dive bout termination besides elevated lactate levels? Could it simply be that the birds are satiated, have lost track of the prey patch, have a gut full of cold prey causing a thermal burden that cannot sustain diving or that they are exhausted? There are probably many reasons causing dive bout termination but what are some of the physiological constraints that may limit extended bouts of repeated dives?

King penguins decrease their body temperatures during diving bouts, returning to normothermic temperatures during the interbout period (Fahlman et al., 2005; Handrich et al., 1997) (A.S., J.-P. Gendner, C. A. Bost and Y.H., manuscript submitted for publication). These re-warming events involve perfusing peripheral tissues with warm blood from the core of the body so long intervals at the surface may be necessary for this thermal process. This would also enable the birds to lay down subcutaneous fat accumulated from prey ingestion (Fahlman et al., 2005). Another possibility is that these surface intervals serve to remove accumulated N_2, which might otherwise limit dive performance.

Boycott et al. (Boycott et al., 1908), using empirical data from studies on goats weighing around 20 kg, suggested that decompression sickness (DCS) risk is negligible unless the difference between the ambient pressure (P_{amb}) and tissue tension exceeds a critical threshold. When extended to human divers, the observations suggested that safe decompressions can be made as long as the ratio between tissue tension and P_{amb}, the supersaturation, never exceeds 2. In other words, a human diver

fully equilibrated with N_2 at 2 atmospheres absolute (ATA; 1 ATA=101 kPa; 2 ATA=10 m depth) can safely return to 1 ATA (Boycott et al., 1908). Air contains ~79% N_2, so safe N_2 levels would be below 1.6 ATA when decompression is made directly to the water surface. Above 1.6 ATA, DCS risk in humans is expected to increase rapidly with increasing tissue and blood N_2 tension (P_{N_2}). In pigs weighing between 17 and 24 kg, none of the animals showed DCS symptoms upon decompression to 1 ATA (0.79 ATA N_2) after being fully equilibrated in an atmosphere containing 2.4 ATA N_2 (3 ATA total pressure), while 50% showed symptoms at 3.1 ATA N_2 (4 ATA total pressure) (Dromsky et al., 2000). As there is a direct relationship between DCS susceptibility and body mass in terrestrial animals it would be expected that a bird of 12 kg would able to withstand a slightly higher pressure reduction before DCS symptoms. However, mixed venous P_{N_2} values above 3 ATA should evince a considerable proportion of symptoms in a 12 kg bird unless it has adaptations that reduce supersaturation and bubble formation or can otherwise tolerate an elevated bubble load.

The aim of this paper is to estimate blood and tissue P_{N_2} in king penguins during extended foraging trips to determine if, after repeated foraging dives, P_{N_2} can reach levels that could cause DCS. If so, we wish to determine possible behavioural or physiological mechanisms that could be used to moderate tissue P_{N_2} and thereby decrease the risk of DCS.

Materials and methods

Model

The model was adapted for king penguins (*Aptenodytes patagonicus* Miller 1778) from models used previously for man and marine mammals (Fahlman et al., 2006; Olszowka and Rahn, 1987). A brief outline of the assumptions and physiological structure of the model are given here and a detailed description can be found elsewhere (Fahlman et al., 2006). Inert gas uptake and removal are regulated by the following equation:

$$\frac{dP_{tiss}}{dt} = \frac{(P_{blood} - P_{tiss})}{\tau_{tiss}}, \quad (1)$$

where P_{tiss} and P_{blood} are inert gas tensions of the tissue and arterial blood, respectively. Inert gas uptake and removal are regulated by the volume of the tissue and the local perfusion rate. The rate of uptake or removal can be expressed as a tissue time constant (τ_{tiss}) that determines the time it takes to reach a new equilibrium after a change in the external pressure. P_{blood} is assumed to follow lung P_{N_2} as long as there is no pulmonary shunt. Thus, N_2 added to blood and tissues during a dive comes from the lungs. τ_{tiss} can be used to compute time to 50% completion ($\tau_{tiss1/2}$) of inert gas uptake or removal for a specific tissue since $\tau_{tiss1/2}=\ln 2 \times \tau_{tiss}$ (Fahlman et al., 2006). Also, $\tau_{1/2}$ describes the time to 50% completion of inert gas uptake or removal. When P_{tiss} reaches P_{blood}, a new steady state is achieved and the tissue is considered saturated. τ_{tiss} is made up of several terms regulating uptake and removal:

$$\tau_{tiss} = \frac{\dot{Q}_{tiss}}{V_{tiss}} \times \frac{S_{blood}}{S_{tiss}}, \quad (2)$$

where $\dot{Q}_{tiss}$ is rate of blood flow through the tissue, V_{tiss} is tissue volume and S_{tiss} and S_{blood} are the solubilities of inert gas of the tissue and blood (Fahlman et al., 2006; Fahlman et al., 2001; Kety, 1951). Consequently, inert gas uptake and removal from a tissue are governed by the volume of the tissue, the solubility of inert gas and the local rate of blood flow. The time it takes for the whole animal to reach a new steady state, or saturation, after an increase in external pressure is determined by cardiac output ($\dot{Q}_{tot}$), the distribution of blood flow and the proportion of tissues with different inert gas solubilities. An animal with a respiratory system that does not collapse during diving only has two ways of altering inert gas uptake and removal: changing $\dot{Q}_{tot}$, or changing its distribution. Consequently, a realistic model to estimate blood and tissue P_{N_2} must have accurate values for blood flow and blood flow distribution.

Measuring P_{N_2} in a diving animal is extremely complex and has only been done successfully in a few studies. In king penguins, heart rate and mixed venous P_{N_2} were measured during forced dives to depth between 34 and 102 m (Ponganis et al., 1999). Despite great differences in the magnitude of cardiovascular change between forced and voluntary dives (Kooyman, 1989) the data reported by Ponganis et al. (Ponganis et al., 1999) make it possible to validate assumptions used in the model. In addition, the physiological description of inert gas flux enables us to identify variables which have a large impact on P_{N_2} levels and that deserve further research.

For the model, the body of the penguin was partitioned into five different compartments: blood (arterial and venous) and four tissues (central circulation, brain, fat and muscle). The central circulation compartment included heart, kidney and liver while the fat compartment included subcutaneous and abdominal fat, skin, bone, alimentary tract and connective tissue (Fahlman et al., 2006). This separation divides the body into tissues with a fast (central circulation), intermediate (brain and muscle) or slow (fat) rate of inert gas uptake or removal and makes it possible to look at temporal differences in their contribution to mixed venous P_{N_2} (Fahlman et al., 2006).

The proportion of muscle mass (35.0%) was taken from non-fasted king penguins (Cherel et al., 1994a; Cherel et al., 1994b). Central circulation (heart, liver and kidney) was assumed to be 5.4% of total body mass. Mass proportions for heart (1.3–1.6%) and liver (3.6 %) were taken from non-fasted penguins (Cherel et al., 1994a; Cherel et al., 1994b) and the proportional mass of the kidney (0.4%) was assumed to be equal to the Weddell seal (Davis and Kanatous, 1999). The size of the brain compartment was taken as 26 g (from Ponganis et al., 1999). The mass proportion of the fat compartment was 50.7%. Blood volume (83 ml kg^{-1}) and volume of the respiratory system (lung + trachea + air sacs, 69 ml kg^{-1}) were assumed to be similar to those previously reported for this species (Ponganis et al., 1999). We assumed that the average resting heart rate for king penguins on land (87 beats min^{-1}) (Froget et al., 2004) represented a $\dot{Q}_{tot}$ of 2.4 ml s^{-1} kg^{-1}, which is similar to that of resting emperor penguins on sea ice (Kooyman et al., 1992). In king penguins, average heart rates while diving and at the water surface between dives were 101 beats min^{-1} and 186 beats min^{-1}, respectively (Froget et al., 2004). Diving and post-dive $\dot{Q}_{tot}$ were scaled directly according to changes in heart rate from resting levels on land for king penguins (Table 1). Therefore, diving $\dot{Q}_{tot}$ was 16% higher and post-diving was 114% higher than the

resting value (Table 1). Thus, $\dot{Q}_{tot}$ while diving was assumed to be 50% of the surface value.

Blood flow distribution during diving has been measured in forced (Jones et al., 1979; Stephenson et al., 1994) and voluntarily (Bevan and Butler, 1992) diving ducks and in force dived Weddell seals (Zapol et al., 1979). In the Pekin duck and Weddell seal, blood flow to the brain was approximately 1% of $\dot{Q}_{tot}$ at rest. With an approximate fivefold decrease in heart rate and $\dot{Q}_{tot}$ during diving, brain blood flow increased to 10% of $\dot{Q}_{tot}$ during forced dives in the duck (Jones et al., 1979). We therefore assumed that blood flow to the brain at the surface was 1% of $\dot{Q}_{tot}$ and 10% during forced dives (Table 1). In foraging birds, we assumed that the blood flow rate to the brain increased by a similar amount as during forced dives. Due to the smaller cardiovascular changes in freely diving birds, the proportion of $\dot{Q}_{tot}$ was therefore initially set to 4% (Table 1). Muscle blood flow at the surface was assumed to be 0.35 ml min^{-1} g $tissue^{-1}$ in both forced trials and during a foraging trip, which is similar to those measured in resting ducks (Table 1) (Grubb, 1982; Jones et al., 1979). During forced diving, little or no muscle blood flow was detected in the Pekin duck (Jones et al., 1979), Macaroni (*Eudyptes chrysolophus*) or Gentoo penguin [*Pygoscelis papua* (Scholander, 1940)]. It was therefore assumed that blood flow to the muscle during forced dives decreased tenfold. As a result, 26% of $\dot{Q}_{tot}$ was directed to the muscle during forced diving. This is similar to the assumptions used for the Weddell seal (Davis and Kanatous, 1999). Deep diving penguins, on the other hand, swim actively during portions of the dive (Sato et al., 2002). Therefore, blood flow to muscle cannot be reduced to the same extent as those measured in forced diving birds. However, the reduced work by alternating periods of active swimming with periods of gliding (Sato et al., 2002) reduces the rate of O_2 utilization and myoglobin desaturation and thereby prolongs aerobic metabolism of the muscle. For this reason, the proportion of $\dot{Q}_{tot}$ directed to the muscle was reduced to 13% (Table 1). For central circulation, blood flow at the surface during forced trials was assumed to be similar in proportion to organ mass specific metabolic rate of heart, liver and kidney (~43% of total metabolic rate) in the Weddell seal at rest (Davis and Kanatous, 1999). Heart rate at the surface was 33% higher in foraging as compared with force dived birds. As $\dot{Q}_{tot}$ was directly scaled to heart rate, this led to a 33% increase in $\dot{Q}_{tot}$ for foraging birds at the surface as compared with force dived birds, and we assumed that most of this was directed to the central circulation. Thus, the proportion of $\dot{Q}_{tot}$ directed to central circulation while birds were at the surface during the foraging trip was 56% (Table 1). Blood flow to kidney and liver was significantly reduced while flow to the heart was maintained during forced dives in the Pekin duck (Jones et al., 1979). The proportion of $\dot{Q}_{tot}$ directed to central circulation during forced dives increased slightly with an overall 72% reduction in the local blood flow rate (Table 1). In foraging birds, a 28% reduction in the local blood flow rate to central circulation was the result of a 50% decrease in $\dot{Q}_{tot}$ and a 45% increase in the proportion of $\dot{Q}_{tot}$ directed to this compartment (Table 1). Blood flow to the fat compartment was assumed to be 4% of $\dot{Q}_{tot}$ at the surface and was unchanged in one case during forced dives (see below) or allowed to decrease to 2% during diving in both forced trials and while foraging (Table 1).

Validation of mathematical model using data from forced dives in penguins

Measured venous P_{N_2} data in king penguins reported by Ponganis et al. (Ponganis et al., 1999) for forced dives in a water filled hyperbaric chamber were used to validate the model. Estimated venous P_{N_2} from the model was compared with measured data for dives to three different pressures (11.2 ATA, 7.8 ATA and 4.4 ATA, equal to depths of 102 m, 68 m and 34 m, respectively), assuming a symmetric compression and decompression rate of 1 m s^{-1} (Ponganis et al., 1999). Dive duration, including descent, bottom and ascent duration, was 270 s to a depth of 102 m, 200 s to a depth of 68 m and 134 s to a depth of 34 m. The reported mean diving and post-surface heart rates from forced dives were 30 beats min^{-1} and 141 beats min^{-1}, respectively (Ponganis et al., 1999). Changes in heart rate were assumed to be directly proportional to adjustments in $\dot{Q}_{tot}$ from the resting heart rate on land [87 beats min^{-1} (Froget et al., 2004)]. $\dot{Q}_{tot}$ during forced diving and during the post-dive surface period were, respectively, modelled as 33% and 162% of the resting value of 2.4 ml s^{-1} kg^{-1}. Consequently, $\dot{Q}_{tot}$ during forced diving was 0.8 ml s^{-1} kg^{-1} and the post-diving surface value 3.9 ml s^{-1} kg^{-1}. We assumed body mass (M_b) to be 12 kg, which was within the range of values reported by Ponganis et al. (Ponganis et al., 1999).

Estimated data from foraging penguins

We used time–depth recordings published elsewhere (A.S., J.-P. Gendner, C. A. Bost and Y.H., manuscript submitted for publication) from four king penguins during a complete foraging trip (Table 2). The time–depth recorder measured dive depth every 2 s and allowed detection of pressure changes as small as 0.02 ATA (0.2 m). To correct for baseline noise, only dives deeper than 0.6 m for at least 6 s were considered to be dives. A dive bout was defined as a minimum of three repeated deep dives ($\geqslant$50 m) followed by a surface interval of less than 15 min.

Sensitivity analysis for foraging birds

The model variables of cardiac output, regional blood flow tissue distribution and parabronchial shunt were varied and the model run with each new set of variables for each bird and for each foraging trip (Table 3). Tissue and mixed venous P_{N_2} as the bird surfaced after deep and shallow dives and from the last dive of a dive bout was compared between models with different cardiovascular variables (Table 4). This allowed an assessment of the initial conditions of the model and enabled determination of the sensitivity of estimated blood and tissue P_{N_2} values to changes in physiological variables (Table 4).

Model simulation details

M_b was assumed to be 12 kg during the entire foraging trip, which was the average M_b of the four birds before and after the foraging trip (Table 2). Forced diving birds show a more pronounced diving bradycardia and presumably greater changes

Table 1. *Size of each compartment in relation to whole body, cardiac output and tissue blood flow distribution values used for estimating tissue and blood N_2 tension for a 12 kg king penguin*

	Mass (kg)	Volume (l)	Mass proportion of WB (%)	Proportion of $\dot{Q}_{tot}$ (%)				Estimated blood flow (ml g^{-1} tissue min^{-1})			
				Surface		Diving		Surface		Diving	
				Forced trials	Foraging trips	Forced trials	Foraging trips	Forced trials	Foraging trips	Forced trials	Foraging trips
WB	12.0	11.22	100	100	100	100	100	0.234	0.312	0.047	0.156
CC	0.648[1,2,3]	0.611	5.40	43	56	60	81	1.863	3.236	0.520	2.340
M	4.200[1,2]	3.962	35.0	52	39	26	13	0.348	0.348	0.035	0.058
B	0.026[3]	0.028	0.22	1	1	10	4	1.080	1.440	2.160	2.880
F	6.080	6.623	50.7	4	4	4	2	0.018	0.025	0.004	0.006
Bl	1.046[3]	0.996	8.71	–	–	–	–	–	–	–	–

WB, whole body; $\dot{Q}_{tot}$, cardiac output.

CC, central circulation (including heart, liver, kidney and alimentary tract); M, muscle; B, brain; F, fat (subcutaneous and abdominal fat, skin, bone and connective tissue); Bl, blood. The brain and fat compartments were assumed to have a density equal to fat (0.918 kg l^{-1}), muscle and central circulation a density of 1.060 kg l^{-1} (Kayar et al., 1997) and blood a density of 1.050 kg l^{-1} (Berne and Levy, 1997). The values for $\dot{Q}_{tot}$ and blood flow distributions for a foraging bird are the same as that for model A in Table 3.

Superscripts refer to the following references: [1](Cherel et al., 1994a), [2](Cherel et al., 1994b), [3](Ponganis et al., 1999), [4](Davis and Kanatous, 1999).

Table 2. *Summary of dive data used for estimation of tissue and blood N_2 tension during foraging dives in king penguins*

Bird number	M_b (kg)		Foraging duration (days)	Total number of dive bouts	Dive bout duration (min)		Dives			Dive duration (s)			Dive depth (m)			Mean surface interval in a bout (s)
	Pre-foraging	Post-foraging			Mean	Max	Total number	Max number in a bout	Number >50 m	Mean	Mean in a bout	Max	Mean max	Mean max in a bout	Max	
1	12.3	13.9	15	24	432	1648	6381	1099	1411	87.1	117	306	25.0	44.7	184	34
2	10.6	12.6	15	27	329	1094	5075	977	1121	88.7	145	394	29.8	66.4	200	55
3	11.6	13.1	19	40	283	1046	7501	1121	1570	78.9	138	374	25.2	53.7	240	40
4	10.4	11.9	18	36	277	1099	4955	595	1176	102.4	133	400	30.5	49.5	237	53
Mean ± s.d.	11.2±0.9	12.9±0.8	17±2	32±8	330±72	1222±285	5978±244	948±244	1320±209	89.3±9.8	133±12	368±43	27.6±2.9	53.6±9.3	215±28	46±10

All birds were males.

Values were taken from Schmidt et al. (A.S., J.-P. Gendner, C. A. Bost and Y.H., manuscript submitted for publication).

Table 3. *Different models used in a sensitivity analysis to determine the effect of changes in cardiovascular and respiratory variables on compartment N_2 tensions shown in Table 4*

	Blood flow distribution during diving (% of $\dot{Q}_{tot}$)							$\tau_{tiss1/2}$ (min)							
								Diving				Surface			
Model	CC	M	B	F	Surface $\dot{Q}_{tot}$ (ml kg^{-1} s^{-1})	Diving bradycardia (% of surface $\dot{Q}_{tot}$)	Pulmonary shunt	CC	M	B	F	CC	M	B	F
0	60	26	10	4	3.9	20	None	1.26	18.8	1.73	1022	0.35	1.88	3.46	204
A	81	13	4	2	5.2	50	None	0.28	11.3	1.30	613	0.20	1.88	2.59	153
B	81	13	4	2	5.2	0	None	0.14	5.64	0.65	307	0.20	1.88	2.59	153
C	81	13	4	2	5.2	50	100%, 100 m	0.28	11.3	1.30	613	0.20	1.88	2.59	153
D	56	39	1	4	5.2	50	None	0.40	3.76	5.18	307	0.20	1.88	2.59	153
E	34	60	4	2	5.2	50	None	0.67	2.45	1.30	613	0.20	1.88	2.59	153
F	83	13	2	2	5.2	50	None	0.27	11.3	2.59	613	0.20	1.88	2.59	153
G	81	13	4	2	5.2	50 (below 30 m depth)	None	0.28	11.3	1.30	613	0.20	1.88	2.59	153
H	81	13	4	2	5.2	50	None	0.28	11.3	1.30	613	0.35	1.22	0.65	153
I	85	13.5	0.5	1	5.2	50	None	0.27	10.9	10.4	1226	0.20	1.88	2.59	153

CC, central circulation; M, muscle; B, brain; F, fat; $\tau_{tiss1/2}$, tissue half-time for each compartment shown for the dive and surface period.

Model A is a model with initial conditions similar to the foraging bird in Table 1 and is the model to which all other models are compared.

Model 0 gives the values for the forced diving validation simulation.

The proportion of $\dot{Q}_{tot}$ at the surface was constant for all models except H and was 56% for central circulation (CC), 39% for muscle (M), 1% for brain (B) and 4% for the fat compartment.

For model B, there was no diving bradycardia.

For model C gas exchange ceased below 100 m (100% pulmonary shunt).

Model D examined the effect of end bout P_{N_2} in the case of no changes in tissue blood flow distribution during diving and model E how end bout P_{N_2} is affected by a higher blood flow to central circulation and with a reduction to muscle.

Model F shows how changes in $\dot{Q}_{tot}$ at the surface affect end bout P_{N_2}.

Model H surface blood flow distribution was 32% to central circulation, 60% to muscle, 4% to brain and 4% to fat.

Model I attempts to minimize P_{N_2} levels in all compartments, based on the results in Table 4 and Figs 2 and 3.

Table 4. *Estimated mean compartment P_{N2} levels for the four birds during an entire foraging trip at the time the bird reaches the surface at the end of a dive bout or after deep or shallow dives*

	End bout P_{N_2} (ATA)					End dive P_{N_2} (ATA)									
						Deep dives (>50 m)					Shallow dives (⩽50 m)				
Model	CC	M	B	F	MV	CC	M	B	F	MV	CC	M	B	F	MV
A	1.55±0.02	1.55±0.13	2.43±0.15	1.43±0.10	1.56±0.07	3.92±0.07	2.57±0.07	5.11±0.27	1.31±0.06	3.30±0.06	1.02±0.08	1.23±0.05	1.47±0.16	1.28±0.05	1.12±0.06
B	1.22±0.03	1.85±0.16	2.17±0.12	1.89±0.15	1.50±0.08	2.43±0.04	3.38±0.12	4.67±0.17	1.72±0.08	2.80±0.04	0.95±0.05	1.35±0.08	1.30±0.15	1.54±0.07	1.14±0.06
C	1.51±0.04	1.42±0.15	2.19±0.22	1.28±0.12	1.48±0.09	3.71±0.03	2.19±0.13	4.26±0.11	1.19±0.08	3.02±0.06	1.02±0.08	1.14±0.08	1.36±0.18	1.14±0.06	1.07±0.08
D	1.61±0.02	2.05±0.17	2.13±0.17	1.90±0.15	1.80±0.08	4.24±0.09	3.96±0.17	3.77±0.15	1.72±0.08	4.03±0.11	1.03±0.09	1.42±0.11	1.46±0.10	1.54±0.07	1.21±0.09
E	1.66±0.01	2.20±0.01	2.41±0.16	1.42±0.10	1.87±0.08	4.59±0.16	4.46±0.21	5.03±0.24	1.31±0.06	4.41±0.17	1.03±0.09	1.44±0.13	1.45±0.16	1.27±0.05	1.21±0.10
F	1.54±0.02	1.55±0.13	2.36±0.16	1.43±0.10	1.55±0.07	3.89±0.07	2.47±0.07	4.60±0.24	1.31±0.06	3.28±0.06	1.02±0.08	1.23±0.05	1.52±0.14	1.28±0.05	1.12±0.06
G	1.40±0.03	1.41±0.03	2.68±0.19	1.40±0.08	1.42±0.05	3.34±0.07	2.34±0.08	5.34±0.28	1.30±0.04	2.89±0.06	0.98±0.06	1.12±0.08	1.66±0.16	1.20±0.02	1.05±0.07
H	1.63±0.06	1.32±0.11	1.90±0.13	1.43±0.10	1.45±0.09	4.01±0.08	2.26±0.06	4.73±0.23	1.31±0.06	2.87±0.05	1.06±0.10	1.11±0.04	1.24±0.12	1.28±0.05	1.11±0.06
I	1.54±0.02	1.57±0.13	1.81±0.15	1.13±0.06	1.54±0.07	3.86±0.07	2.62±0.07	2.93±0.09	1.06±0.04	3.26±0.05	1.02±0.08	1.24±0.05	1.35±0.06	1.06±0.03	1.11±0.06

Details of the four birds are given in Table 2. CC, central circulation; M, muscle; B, brain; F, fat; MV, mixed venous blood; P_{N_2}, N_2 partial pressure.

Values are means ± s.d. (N=4).

The initial conditions for each model are indicated in Table 3.

in $\dot{Q}_{tot}$ than freely diving birds. As $\dot{Q}_{tot}$ was scaled to heart rate, we adjusted $\dot{Q}_{tot}$ for a free-ranging bird. $\dot{Q}_{tot}$ while diving was assumed to be 50% of its value at the surface (2.6 *vs* 5.2 ml s^{-1} kg^{-1}, Table 1).

We looked at the N_2 flux during an interbout period to evaluate if the short (<60 s) and shallow (<30 m) dives that commonly occur between diving bouts can help to remove N_2 while concurrently protecting against DCS. We ran the model twice, once with the complete data set and the other with short shallow interbout dives removed.

Results

Validation of mathematical model using data from forced dives in penguins

Estimated mixed venous P_{N_2} of a 12 kg king penguin was compared with measured values presented by Ponganis et al. (Ponganis et al., 1999). The resulting N_2 uptake or removal $\tau_{tiss1/2}$ for central circulation, muscle, brain and fat were 0.35, 1.88, 3.46 and 204 min while at the surface and 1.26, 18.8, 1.73 and 1022 min while diving (Model 0, Table 3). In Fig. 1, each observed mixed venous P_{N_2} value at depth was obtained in a separate trial. The solid black line represents estimated mixed venous P_{N_2} when assuming a regional blood flow distribution during diving as in Model 0 in Table 3. The dotted black line represents estimated mixed venous P_{N_2} when the regional blood flow to central circulation, muscle and fat were changed to 36%, 52%, and 2%, respectively, of $\dot{Q}_{tot}$. Each experiment varied in compression and decompression sequences and the average profile for each dive depth is given in the figure. In all trials, maximum venous P_{N_2} occurred after the return to the surface (Fig. 1). The immediate drop in mixed venous P_{N_2} occurring as the bird returned to the surface, is caused by the sudden change in blood flow distribution to the various tissues upon surfacing. An immediate change in tissue-specific blood flow affects the amount of N_2 removed from each tissue and pooling into the mixed venous blood, thereby initiating the decline. Blood perfusion changes upon reaching the surface are immediate in some species (Butler and Jones, 1982), and this characteristic could therefore be a physiological reality. For Model 0 (Table 3), maximum estimated venous P_{N_2} was 4.2 ATA during a compression to 11.2 ATA, 3.0 ATA for a compression to 7.8 ATA and 1.7 ATA for a compression to 4.4 ATA.

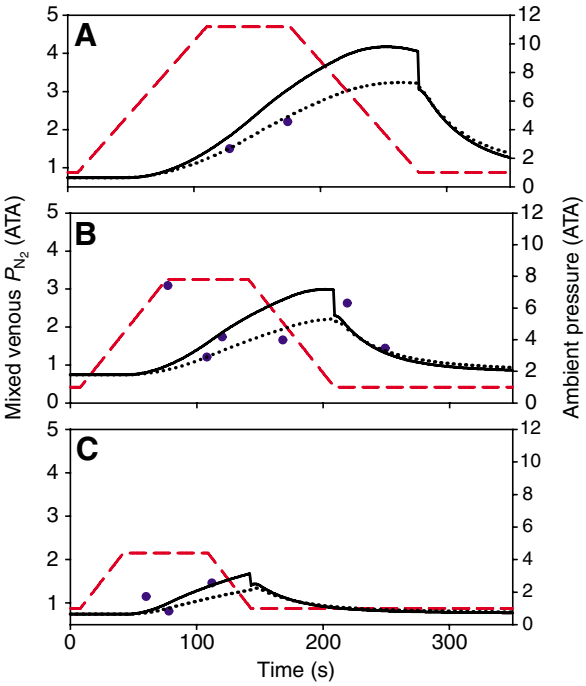

Fig. 1. Ambient pressure (red broken lines), measured (solid blue circles) (Ponganis et al., 1999) and estimated (solid and dotted black lines) mixed venous N_2 tensions (P_{N_2}) during hyperbaric exposures to (A) 102 m, (B) 68 m and (C) 34 m depth. Estimated P_{N_2} assumed a blood flow distribution of 60% to central circulation, 26% to muscle, 10% to brain and 4% to fat, as in model 0 in Table 3 (solid lines) and 36%, 52%, 10% and 2%, respectively (dotted lines).

Sensitivity analysis

Cardiovascular variables were altered sequentially and the model simulation repeated for each of the four birds (Table 3, Table 4). For models A–G, the distribution of $\dot{Q}_{tot}$ was 56% to central circulation, 39% to muscle, 1% to brain and 4% to the fat compartment while the bird was at the surface. Model A was the same as that used for foraging birds in Table 1 and was used as our initial model, to which we compared all other models. The effect of diving bradycardia was explored in model B. The effect of a 100% parabronchial shunt, i.e. no gas exchange, established at 100 m of depth, was explored in model C. Models D–F and H explored the effect of regional changes in blood flow distribution. In model G we determined how the pre-surface tachycardia that is reported to occur in king penguins would affect tissue and blood P_{N_2} (Table 3). The results from models A–G were used to select blood flow distributions for a final model used in the remainder of the analysis (Model I, Table 3).

Without a diving bradycardia, P_{N_2} decreased in tissues where $\tau_{tiss1/2}$<1.3 min (central circulation and brain) and also by as much as 15% for mixed venous P_{N_2} during deep dives (Table 3, Table 4, models A *vs* B). In other words, increasing $\dot{Q}_{tot}$ reduced P_{N_2} at the end of the dive for fast tissues while it increased P_{N_2} in slow tissues. Termination of gas exchange at 100 m reduced N_2 levels in all compartments. Brain decreased by as much as 17% while mixed venous P_{N_2} decreased by 8% (Table 4, models A *vs* C). Central circulation P_{N_2} increased by 4% at the end of dive bouts and by 8% at the end of deep dives when blood flow decreased by 31% (models A *vs* D). A further 39% decrease in blood flow increased P_{N_2} an additional 3% (1.61 ATA, model D *vs* model E). Brain P_{N_2}, on the other hand, decreased by 1–26% with a 75% decrease in blood flow while fat P_{N_2} increased by 20–33% with a 100% increase in blood flow (models A *vs* D). With a 200% increase in blood flow, muscle P_{N_2} increased by 15–54% (models A *vs* D). There were no apparent trends in the data and under certain conditions an increase or reduction in blood flow had similar effects on tissue and blood P_{N_2} levels as the bird surfaced. To explore this further, end bout or end dive P_{N_2} was estimated by sequentially varying $\tau_{tiss1/2}$ for each tissue either while diving (Fig. 2) or while at the surface (Fig. 3). The results suggested that very high or very low blood flows during diving reduced P_{N_2}. A reduction in blood flow at the surface, on the other hand, reduced N_2 removal and increased P_{N_2} at the end of dive bouts and dives (Fig. 3). The data in Fig. 2 was used to adjust blood flow distribution to minimize tissue P_{N_2} (model I, Table 3, Table 4).

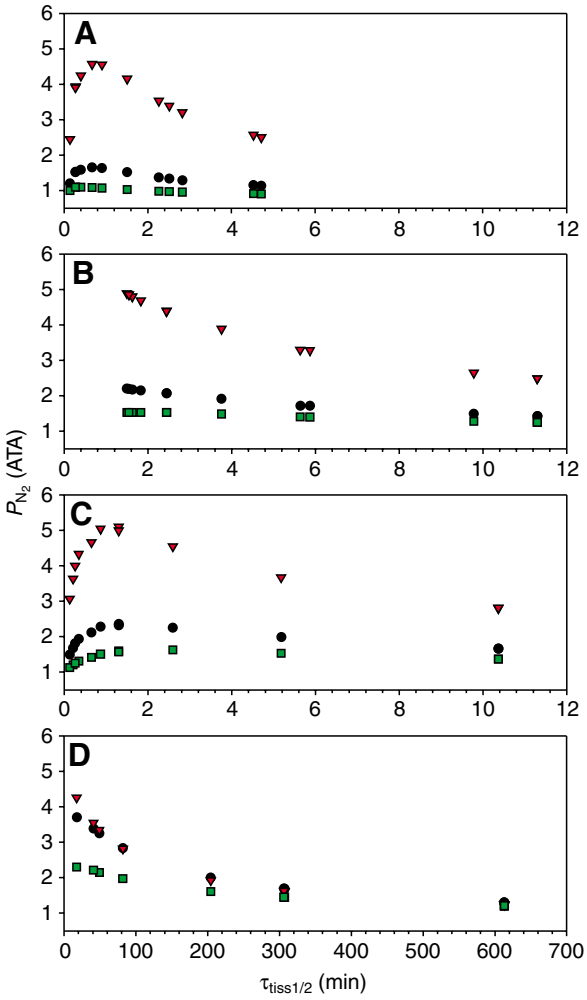

Fig. 2. Model sensitivity analysis comparing (A) central circulation, (B) muscle, (C) brain and (D) fat P_{N_2} levels against diving tissue time constant ($\tau_{tiss1/2}$). Results shown are predicted compartment values at the time the bird reaches the surface after the last dive in a dive bout (black circles), or after deep (>50 m, red triangles) or shallow (⩽50 m, green squares) dives.

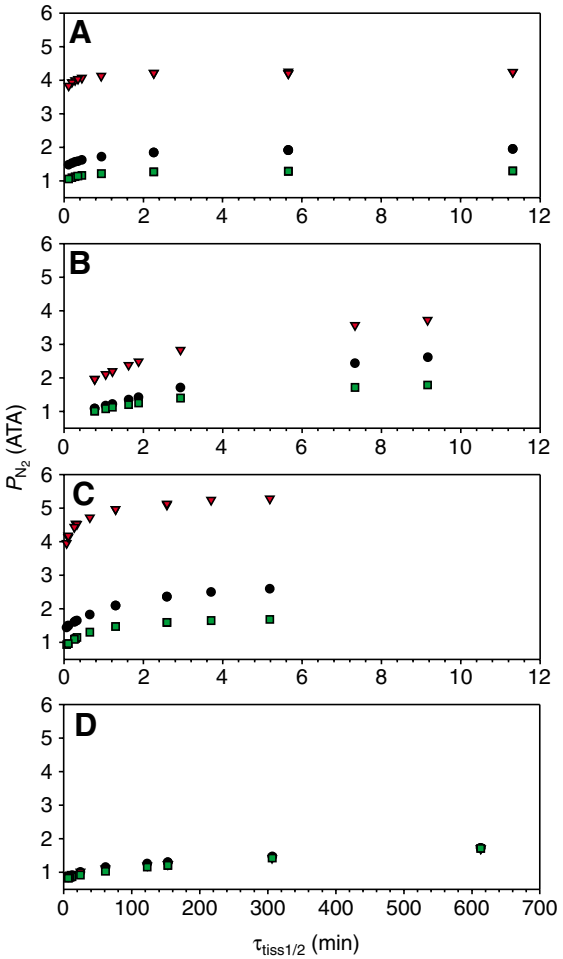

Fig. 3. Model sensitivity analysis comparing (A) central circulation, (B) muscle, (C) brain and (D) fat P_{N_2} levels against surface tissue time constant ($\tau_{tiss1/2}$). Results shown are predicted compartment values at the time the bird reaches the surface after the last dive in a dive bout (black circle) or after deep (>50 m, red triangle) or shallow (⩽50 m, green square) dives.

In particular, blood flow to brain was reduced to 25% of its surface value, which reduced P_{N_2} by 8–43%. Similarly, a reduction in fat blood flow by 50% reduced P_{N_2} by 17–21% (Table 4, models A *vs* I).

Estimated P_{N_2} levels during foraging

In the remainder of the analysis, the cardiovascular variables detailed in model I were used. Fig. 4 shows changes occurring in estimated venous N_2 levels during and after a dive by a king penguin. Central circulation P_{N_2} rapidly increased leading to a concomitant increase in mixed venous P_{N_2} (Fig. 4). At 101 s, 7 s after the start of ascent, mixed venous P_{N_2} exceeded lung P_{N_2}. At this time N_2 flux was reversed and re-distribution of available N_2 continued according to P_{N_2} gradients within the body. Central circulation P_{N_2} exceeded lung and arterial P_{N_2} 3 s after ascent was initiated. Despite this, central circulation P_{N_2} continued to increase until 11 s after the start of the ascent. This delay is caused by blood transit time (11 s arterial transit time during diving). Similarly, P_{N_2} in muscle and fat continued to increase throughout the entire dive and even after the bird had returned to the surface (Fig. 4). At the end of the 46 s surface interval, 96% of N_2 taken up was removed from the central circulation (max P_{N_2}=4.45 ATA, P_{N_2} at end of surface interval=0.884 ATA). Thus, central circulation P_{N_2} was still higher than surface P_{N_2}, i.e. ambient lung P_{N_2}=0.741 ATA after correction for water vapour. Likewise, P_{N_2} in muscle (1.03 ATA) and brain (1.06 ATA) at the end of the surface interval were still higher than the pre-dive ambient values by 39% and 44%, respectively. Fat P_{N_2} increased only by 0.5% during the dive to 0.745 ATA and decreased insignificantly (<0.01%) during the surface interval. A surface interval duration exceeding dive duration by at least fivefold would be required to remove excess N_2 from the mixed venous blood (data not shown). The fact that no tissue returned to ambient P_{N_2} levels indicates that repeated diving will lead to a build-up of P_{N_2}.

Estimated tissue and blood P_{N_2} using model I (Table 3) for

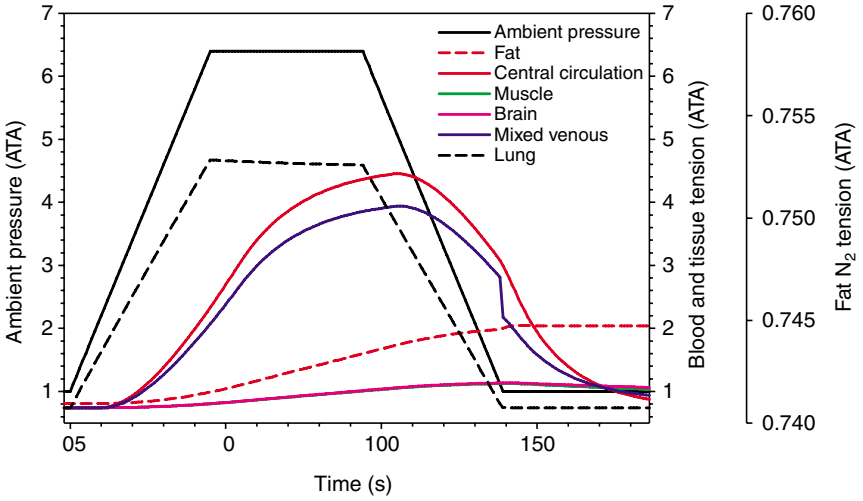

Fig. 4. Estimated blood and tissue N_2 tensions (P_{N_2}) during a single dive for a 12 kg king penguin. Dive duration (133 s), dive depth (54 m) and surface interval (46 s) were mean values of all dives during the foraging bout. Note that P_{N_2} for the fat compartment is on a different scale. Initial conditions are those of model I in Table 3.

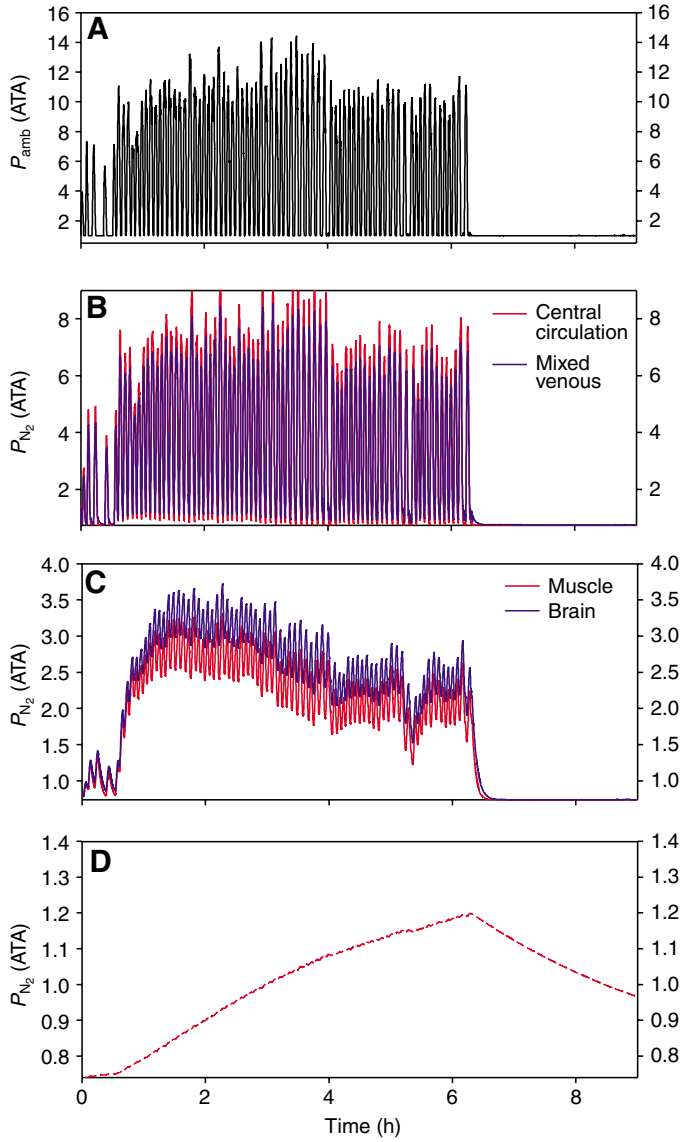

Fig. 5. (A) Ambient pressure (P_{amb}), and (B–D) estimated N_2 tensions (P_{N_2}) for (B) central circulation and mixed venous blood, (C) brain and muscle and (D) fat in a 12 kg king penguin. Data used to estimate P_{N_2} are from an actual dive bout for bird 3 (see Table 2). Initial conditions are those for model I (Table 3).

a diving bout for bird 3 (Table 2) are shown in Fig. 5, consisting of 92 dives with an average duration of 180 s (range 6–254 s), an average surface interval of 69 s (range 10–452 s), an average depth for each dive of 52 m (range 0.2–85 m) and an average maximum depth of 87 m (0.5–143 m). During the bout, estimated P_{N_2} increased in all tissues. For a fast tissue (central circulation) the pattern of change in P_{N_2} more or less followed P_{amb} (Fig. 5A *vs* B). For tissues with an intermediate time constant (brain and muscle), an initial rapid increase was followed by a slow decline to values that became more or less stable (Fig. 5C). For a slow tissue (fat), there was a continuous increase throughout the entire bout (Fig. 5D). Maximum estimated P_{N_2} at bout termination was 3.81 ATA for central circulation, 2.24 ATA for muscle, 2.56 ATA for brain, 1.23 ATA for fat and 3.54 ATA for mixed venous blood (Fig. 5, model I, Table 3A). Average estimated mixed venous P_{N_2} for all birds when surfacing at the end of a dive bout was 1.54±0.07 ATA (range 1.47–1.62 ATA) with maximum values ranging between 3.26 ATA and 3.62 ATA (model I, Table 4). During the extended surface interval following a dive bout, venous P_{N_2} initially decreased as N_2 was removed from the fast tissue (central circulation). P_{N_2} then followed removal of N_2 from the intermediate tissues and finally was held at a supersaturated state for an extended period as fat P_{N_2} slowly decreased (Fig. 5D). Note that at the end of the ~2.3 h (160 min) interbout period, subcutaneous fat P_{N_2} was still higher than ambient surface P_{N_2}.

Estimated fat P_{N_2} levels for each animal throughout an entire foraging trip are shown in Fig. 6. Average estimated fat P_{N_2} (1.13±0.06 ATA; range: 1.05–1.23 ATA, Table 4, model I) at the end of a dive bout did not vary significantly between birds (P>0.1). In addition, fat P_{N_2} very seldom reached levels higher than 2.0 ATA (Fig. 6). For the brain, average estimated P_{N_2} at the end of a dive bout was 1.81±0.15 ATA (range: 1.67–1.98 ATA, Table 4, model I). The amount of N_2 taken up by adipose tissue was positively correlated with the duration of the dive bout (P<0.01, Fig. 6).

Predicted mixed venous supersaturation [$(P_{N_2venous}–P_{N_2amb})/P_{N_2amb}$] during an interbout interval for a bird either performing short and shallow dives or resting at the surface is shown in Fig. 7. Supersaturation initially exceeds 1.4 as the bird returns

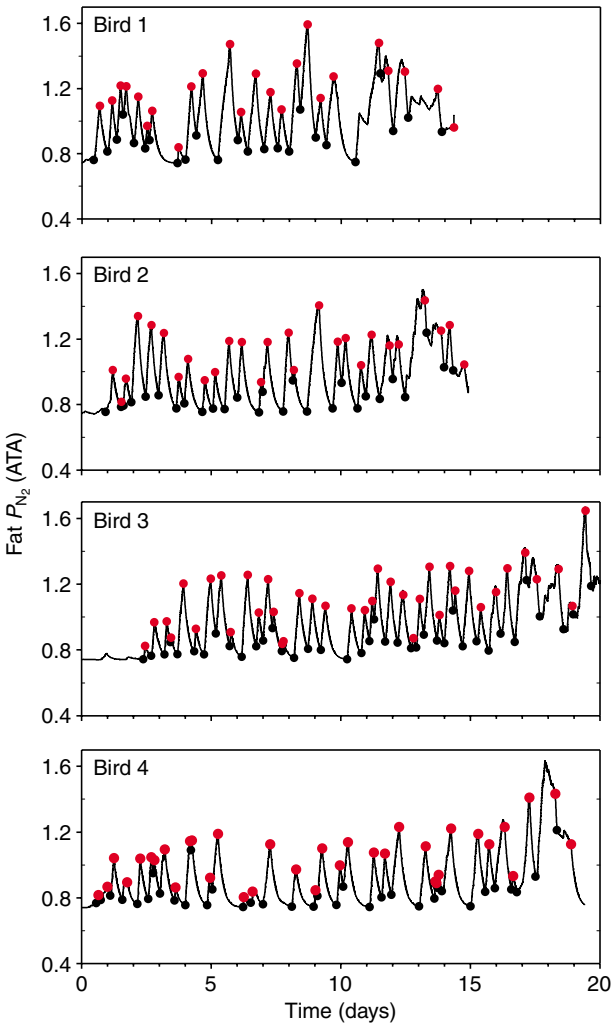

Fig. 6. Estimated subcutaneous fat N_2 tensions (P_{N_2}) for birds 1–4 during an entire foraging trip with several diving bouts. Black and red circles show, respectively, the start and end of a dive bout. Initial conditions are those for model I (Table 3).

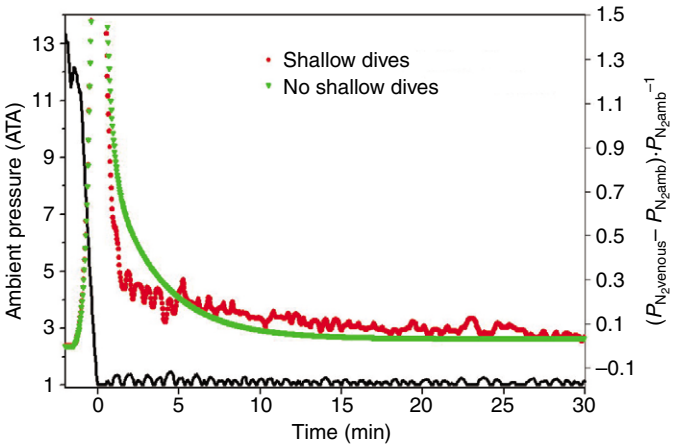

Fig. 7. Ambient pressure (P_{amb}, ATA) and estimated mixed venous supersaturation $[(P_{N2venous}–P_{N2amb})\cdot P_{N2amb}^{-1}]$ for a bird performing short and shallow dives (red dots) or resting at the surface (green dots) during an interbout interval. Initial conditions are those for model I (Table 3).

to the surface. However, in the case where an animal performs short and shallow dives, supersaturation quickly drops to around 0.2 and remains around this level for the next 30 min. This level of supersaturation is much below the level predicted to result in DCS in a human. After approximately 5 min, these short and shallow decompression dives becomes less deep. This helps to improve N_2 removal by increasing the partial pressure gradient but still keeps supersaturation around 0.2. For a bird staying at the surface, supersaturation during the first 5 min is as much as 300% higher than in a bird performing short and shallow dives. After 5 min, the supersaturation is similar to the bird performing decompression dives and declines slowly towards 0 (Fig. 7).

Discussion

The mathematical model estimated values of mixed venous P_{N_2} similar to measured values for forced dives (Ponganis et al., 1999). However, changes in heart rate during diving are very different in freely diving birds (Froget et al., 2004) compared with those that are forced to dive (Ponganis et al., 1999). Assuming no pulmonary diffusion limitation, the most important variable affecting rate of N_2 uptake and removal is $\dot{Q}_{tot}$ and regional blood flow. When 26% of $\dot{Q}_{tot}$ was directed to muscle, mixed venous P_{N_2} during a dive to 34 m or 67 m showed reasonable agreement with observed values. However, during a dive to 102 m mixed venous P_{N_2} was overestimated by 22% and 38% at 120 s and 166 s into the dive, respectively (Fig. 1, solid black lines). With a 100% increase in muscle blood flow during diving, or 52% of $\dot{Q}_{tot}$ directed to the muscle, there was good agreement between measured and observed P_{N_2} for the dive to 102 m (Fig. 1, dotted lines). The reasonable fit between observed and estimated mixed venous P_{N_2} suggests that cardiovascular changes throughout each dive could explain deviations between predicted and observed P_{N_2}. For example, a stressed animal exhibiting tachycardia during the initial stages of the dive explains the one outlying data point during simulated dives to 68 m (Fig. 1B). Parabronchial shunt, caused by engorgement of lung capillaries (Ponganis et al., 1999), could be another explanation for the slight deviation between observed and predicted mixed venous P_{N_2} during the dive to 102 m (Fig. 1A). However, as mixed venous P_{N_2} increased during the bottom phase of the dive a certain level of gas exchange must occur even at 100 m. Therefore, assuming that gas exchange is prevented at 100 m, this would provide the most conservative estimate of tissue and blood P_{N_2}.

This modeling approach allows prediction of tissue and blood P_{N_2} based on different initial conditions. Admittedly, any selected initial conditions are likely to be wrong, at least during parts of the foraging trip, and this could affect our conclusions. In addition, no information exists on $\dot{Q}_{tot}$ and regional blood flow distribution in freely diving birds. It was therefore important to assess how changes in $\dot{Q}_{tot}$, regional blood flow distribution and parabronchial shunt affected blood and tissue P_{N_2}. The slight deviance in observed *versus* predicted mixed venous P_{N_2} during a forced dive to 102 m indicate either that we chose an incorrect $\dot{Q}_{tot}$ or regional blood flow distribution, or that there was a certain level of parabronchial shunt. The geometry and size of the air tubule makes it unlikely that it

would be able to collapse without structural damage and thereby be able to re-expand as the pressure is reduced. Thus, the respiratory system of a diving bird is the opposite of the mammalian respiratory system, where the parabronchi (or lung) is a rigid structure and the air sacs are highly compliant. We assumed that the parabronchi are at least 5 times as rigid as the air sacs and that the parabronchial volume is 10% of the total volume of the respiratory system (Powell and Hopkins, 2004). Using this model of the respiratory system, complete collapse of the air sacs occurs at ~100 m (Denison and Kooyman, 1973; Stephenson, 2005). Increasing the ambient pressure at the time the air sacs have collapsed could produce relatively negative pressures in the parabronchi [the squeeze (Francis and Denison, 1999)], possibly leading to barotrauma. It has been suggested that engorgement of the capillaries reduces parabronchial volume, thereby preventing barotrauma. However, engorgement of the capillaries could impair gas exchange by reducing the diffusion rate. As this question is still unresolved, we decided to test how tissue and blood P_{N_2} would be affected if gas exchange ceased at 100 m. We therefore ran the model assuming a 100% pulmonary shunt for depths exceeding 100 m. For the four birds, maximum P_{N_2} in muscle decreased by 28%, brain by 40%, fat by 20% and central circulation by 45% (models A *vs* C, Table 3, Table 4). However, mean P_{N_2} upon reaching the surface at the end of a dive bout only decreased by 8% for muscle, 10% for brain, 10% for fat, 3% for central circulation and 5% for mixed venous (Table 4, models A *vs* C). Thus, even if gas exchange terminates at depths beyond 100 m, tissue and blood P_{N_2} levels approach those causing DCS symptoms in terrestrial animals of similar size.

It has previously been suggested that the dive response may be a useful mechanism to reduce N_2 uptake during diving and thereby minimize DCS risk (Fahlman et al., 2006; Ponganis et al., 1999). It was therefore surprising when an increase in blood flow resulted in an increase in end bout P_{N_2} in some tissues and a decrease in others. For example, diving bradycardia caused a substantial reduction in brain and central circulation P_{N_2}, while muscle and fat P_{N_2} increased (Table 4, models A *vs* B). These data suggested that an increase in the regional blood flow when $\tau_{tiss1/2} \leqslant 1.30$ min results in a reduction in P_{N_2} as the bird returns to the surface (Table 3, Table 4). In contrast, P_{N_2} increased with a reduction in blood flow when $\tau_{tiss1/2} > 11.3$ min. In addition, P_{N_2} increased in both central circulation and muscle when the regional flow to the muscle was increased and the flow to central circulation reduced. This shows that the diving bradycardia does not simply reduce N_2 levels during repeated diving, as previously suggested (Fahlman et al., 2006; Ponganis et al., 1999). To investigate these unexpected results further we plotted diving $\tau_{tiss1/2}$ (Fig. 2) or surface $\tau_{tiss1/2}$ (Fig. 3) for each tissue against end bout or end dive P_{N_2}, for a series of different regional blood flow distributions. At the surface, an increase in tissue blood flow, or a reduction in surface $\tau_{tiss1/2}$, reduced P_{N_2} at the end of the dive (Fig. 3). For blood flow during diving, on the other hand, the data suggested that end dive P_{N_2} increases to a maximum and then decreases with increasing diving $\tau_{tiss1/2}$. Maximum P_{N_2} occurred at a diving $\tau_{tiss1/2}$ around 1–1.5 min for all compartments. The relationship was particularly pronounced for dives >50 m. Consequently, to reduce blood and tissue P_{N_2} a diving animal can either increase or decrease $\tau_{tiss1/2}$ below this maximum. However, the circulatory system delivers O_2 and removes CO_2, so blood flow changes to each specific tissue is a trade-off between the need to exchange metabolic gases and reducing DCS risk. Due to the predicted extremely high brain P_{N_2} at the end of a bout or a dive for models A–H, regional blood flow was adjusted in an attempt to minimize tissue and blood P_{N_2}. The distributions were chosen to agree with the re-distribution of blood flow during diving according to the dive response. Compared with model A, blood flow was slightly increased to central circulation and muscle and reduced to brain and fat (Table 3). This resulted in an 87.5% decrease in brain blood flow during diving. This is contrary to the common belief that brain blood flow is maintained in freely diving mammals and birds (Butler and Jones, 1982) as the brain is believed to be hypoxia intolerant. The cardiovascular values used for Model I substantially reduced brain and fat P_{N_2} but had little effect on central circulation, muscle or mixed venous P_{N_2}. Despite this marked reduction in brain P_{N_2}, end of dive and bout P_{N_2} remained above 1.6 ATA (Table 4, models A–H *vs* I). Consequently, models A–I provide us with reasonable estimates of the range of blood and tissue P_{N_2} that these birds experience during a foraging trip. The question is to what extent deep diving mammals and birds can reduce blood flow to hypoxia sensitive organs to reduce extreme P_{N_2} without ischemic injury. This will certainly be an intriguing area of research in the future.

Our simulations showed that the average surface interval after a representative dive is too short to remove all the N_2 taken up (Fig. 4). For all birds, end bout mixed venous P_{N_2} averaged 1.54 ATA with values exceeding 3.68 ATA (Table 4, model I). These P_{N_2} values exceed those that cause a predicted 15% DCS rate in mammals of similar size (Dromsky et al., 2000). The maximum P_{N_2} values are dependent on N_2 uptake and removal from fast tissues (central circulation, brain and muscle). The magnitude of mixed venous P_{N_2} upon surfacing depends on the behaviour of the bird during the ascent phase, as N_2 is rapidly removed from tissues with high blood flow. Consequently, these tissues contribute only transiently to DCS risk. Removal of N_2 from fat, on the other hand, is much slower and leads to prolonged elevated mixed venous P_{N_2} (Fig. 5C). As fat P_{N_2} rises throughout the dive bout it increasingly contributes to DCS risk as compared with fast tissues. Average fat P_{N_2} values when surfacing at the end of a dive bout ranged between 1.05 ATA to 1.20 ATA (model I, Table 4) while average maximum end bout values were between 1.43 ATA to 1.65 ATA (Fig. 6). Consequently, during an extended surface interval between two dive bouts (interbout interval) the likelihood of bubble formation and growth may be dependent on delayed N_2 removal from tissues with poor circulation and the extent to which they have reached complete pressure equilibration (saturation). To avoid DCS the bird may be forced to end a dive bout and stay at the surface for an extended surface interval to remove N_2 from the adipose tissues. This could explain why these birds sometimes end dive bouts during daylight hours when food should be plentiful and why they re-warm the periphery during the interbout interval (A.S., J.-P. Gendner, C. A. Bost and Y.H., manuscript submitted for publication). If so, this suggests that during prolonged diving adipose tissues may become a liability and could contribute more to the risk of decompression sickness as compared with other tissues (Tikuisis and Gerth, 2003).

Interestingly, bout duration increased for each bird throughout the foraging trip, causing an increase in end bout fat P_{N_2} throughout the foraging trip (Fig. 6). The increase in bout duration could be due to increased aerobic fitness or foraging efficiency. Alternatively, as subcutaneous fat stores increase throughout the trip so does the N_2 buffering capacity, allowing longer bouts before dangerous N_2 levels are reached.

The high N_2 capacitance of adipose tissues was suggested to act as a N_2 absorbent and reduce bubble formation during deep and short duration dives (Behnke et al., 1935). During these dives, P_{N_2} in fast tissues are elevated while P_{N_2} in slow tissues remain low. Supersaturation of mixed venous blood during decompression from such a dive is therefore minimal and of short duration. Consequently, the likelihood of bubble formation and growth is unlikely. In king penguins, this resembles P_{N_2} distribution during dives performed early during a dive bout. This is in contrast to the elevated adipose P_{N_2} at the end of a bout that would force the bird to undertake a long surface interval. Consequently, adipose tissue could help buffer P_{N_2} at the beginning of a dive bout but be a liability after a long bout.

If dive bout terminations during the day are, at least in part, caused by elevated tissue P_{N_2} levels, it would be expected that P_{N_2} levels would be higher than those during bout termination events at night. There were no differences in tissue P_{N_2} levels during interbout intervals occurring at day or night, suggesting that elevated N_2 is not the prime reason that birds stop deep diving. It is possible that the diurnal vertical distance migrated through the water column is particularly extensive on clear days and that prey descend too deep and it becomes too energetically costly for birds to continue foraging. However, this is a complex subject that depends on light availability, the time available for travel to the prey patch and the horizontal and vertical abundance of prey. In any case, it is clear that king penguins may experience extremely high levels of N_2 in blood and tissues and it is likely that they live with tissue and blood P_{N_2} levels that would cause DCS in similarly sized terrestrial animals. While slow tissues may become a liability over the course of an extended dive bout, dive behaviour may significantly alter mixed venous P_{N_2} at the end of individual dives due to its important effect on gas exchange in fast tissues.

A large proportion of dives by king penguins are performed to depths that are probably too shallow to be foraging dives. The function of these dives is currently unexplained. One suggestion is that these dives represent travel between prey patches. Another possibility postulated (Kooyman, 1989) is that these dives help remove accumulated anaerobic by-products after a series of deep and long dives that have exceeded the ADL. Gas exchange in general would benefit from these short duration dives. During these dives, as compared with staying at the surface, parabronchial and arterial P_{O_2} would increase, enhancing diffusion of O_2 into depleted blood and tissues and aiding in removal of CO_2 by the Haldane effect. Thus, these dives would improve uptake of O_2 and removal of CO_2 and help remove anaerobic by-products (Castellini et al., 1988). The amount of N_2 removed from the tissues during these shallow dives depends on the depth (ambient pressure), duration and $\dot{Q}_{tot}$. As compared with staying at the surface, these dives reduce the pressure difference between tissue and lung P_{N_2} and therefore the rate of N_2 removal. However, staying at the surface increases supersaturation $[(P_{N_2tiss}-P_{N_2amb})/P_{N_2amb}]$ and therefore the probability of bubble formation and growth (Tikuisis and Gerth, 2003). To reduce DCS risk, the shallow decompression dives need to be deep enough to reduce supersaturation but shallow enough to allow removal of N_2. According to the estimated P_{N_2} of approximately 1.5–4.0 ATA, this depth would be between 10 and 30 m ($P_{amb}\approx$2–4 ATA). This is close to the actual depth observed for shallow dives that occur between deep foraging dives of king penguins. Fig. 7 shows the effect on mixed venous supersaturation in a bird either performing a series of these short, shallow (<30 m) decompression dives between two dive bouts or resting at the surface. In the case where the bird performs short and shallow interbout dives, supersaturation seldom exceeds 0.2. For a bird resting at the surface, on the other hand, supersaturation during the initial 5 min after the last dive is as much as 300% higher. These initial high levels may increase the likelihood of bubble formation and growth and could increase the risk of DCS in a bird resting at the surface.

If birds end a dive bout due to elevated tissue and blood P_{N_2} how do they sense or detect risky levels of N_2? It is possible that the animals can sense a low amount of bubbles without serious symptoms, e.g. ‘niggles’. By diving to a shallow depth, bubble size and numbers are reduced and the symptoms are alleviated. During repeated dives, the depth becomes shallower as more N_2 is removed during each dive, until there are no more symptoms at the surface (see Fig. 7). Thus, if the short and shallow dives are used to safely remove N_2 we would expect them to decrease in depth throughout the interbout period. Even if birds become aware that a certain bubble load has been reached, we cannot exclude that termination of a dive bout is nothing but a behaviour that occurs when the bird is exhausted or when the prey patch has scattered. Regardless of the mechanism for bout termination, the data presented in the present paper suggest that tissue and blood P_{N_2} at the end of a dive is a complex function of the need to supply O_2 to central organs while at the same time reducing uptake of N_2. The seminal work by Ponganis et al. (Ponganis et al., 1999), suggests that gas exchange does occur to some extent at 102 m or more in king penguins. We lack important information about $\dot{Q}_{tot}$ and blood flow distribution in freely diving penguins and these variables are vital for accurately determining tissue and blood P_{N_2} levels in breath-hold diving animals. However, our mathematical model is a useful tool that can be used to create alternative explanations for some of the unusual behaviours seen in deep diving birds. The data presented here provide a range of tissue and blood P_{N_2} estimates that can be expected during foraging in king penguins. It is clear that king penguins may experience extremely high levels of N_2 in blood and tissues and it is difficult to see how they avoid symptoms of DCS. The results also raise the question as to what extent gas exchange continues during deep diving in penguins, and does P_{N_2} affect diving marine mammals?

List of symbols and abbreviations

ADL	aerobic dive limit
ATA	atmospheres absolute
B	brain

cADL	calculated ADL
CC	central circulation
DCS	decompression sickness
F	fat
M	muscle
MV	mixed venous blood
P_{amb}	ambient pressure
P_{N_2}	partial pressure of nitrogen
P_{O_2}	partial pressure of oxygen
P_{tiss}, P_{blood}	pressure of inert gas in tissue and arterial blood, respectively
$\dot{Q}_{tot}$	total cardiac output
S_{tiss}, S_{blood}	solubility of inert gas in tissue and blood, respectively
τ_{tiss}	tissue time constant
$\dot{V}_{O_2}$	rate of oxygen consumption

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