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Ontogeny of oxygen stores and physiological diving capability in Australian sea lions

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Summary

1. For air-breathing animals in aquatic environments, foraging behaviours are often constrained by physiological capability. The development of oxygen stores and the rate at which these stores are used determine juvenile diving and foraging potential.

2. We examined the ontogeny of dive physiology in the threatened Australian sea lion *Neophoca cinerea.* Australian sea lions exploit benthic habitats; adult females demonstrate high field metabolic rates (FMR), maximize time spent near the benthos, and regularly exceed their calculated aerobic dive limit (cADL). Given larger animals have disproportionately greater diving capabilities; we wanted to determine the extent physiological development constrained diving and foraging in young sea lions.

3. Ten different mother/pup pairs were measured at three developmental stages (6, 15 and 23 months) at Seal Bay Conservation Park, Kangaroo Island, South Australia. Hematocrit (Hct), haemoglobin (Hb) and plasma volume were analyzed to calculate blood O_2 stores and myoglobin was measured to determine muscle O_2. Additionally, FMR's for nine of the juveniles were derived from doubly-labelled water measurements.

4. Australian sea lions have the slowest documented O_2 store development among diving mammals. Although weaning typically occurs by 17·6 months, 23-month juveniles had only developed 68% of adult blood O_2. Muscle O_2 was the slowest to develop and was 60% of adult values at 23 months.

5. We divided available O_2 stores (37·11 ± 1·49 mL O_2 kg^{-1}) by at-sea FMR (15·78 ± 1·29 mL O_2 min^{-1} kg^{-1}) to determine a cADL of 2·33 ± 0·24 min for juvenile Australian sea lions. Like adults, young sea lions regularly exceeded cADL's with 67·8 ± 2·8% of dives over theoretical limits and a mean dive duration to cADL ratio of 1·23 ± 0·10.

6. Both dive depth and duration appear impacted by the slow development of oxygen stores. For species that operate close to, or indeed above their estimated physiological maximum, the capacity to increase dive depth, duration or foraging effort would be limited. Due to reduced access to benthic habitat and restricted behavioural options, young benthic foragers, such as Australian sea lions, would be particularly vulnerable to resource limitation.

Key-words: cADL, doubly-labelled water, FMR, myoglobin, *Neophoca cinerea*

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Introduction

The question of when physiology limits behaviour is central to the field of physiological ecology (Costa & Sinervo 2004). For reptiles, birds and mammals that have successfully reinvaded aquatic environments, diving capability, which is critical to successful foraging, is often constrained by physiology (Kooyman 1989; Boyd & Croxall 1996; Costa, Gales & Goebel 2001). Not only are physiological limits different across species, but species dive differently with respect to limits (Ridgway & Johnston 1966; Lenfant, Johansen & Torrance 1970; Butler & Jones 1997).

The aerobic component of diving metabolism (DMR) is thought to be the major determinant of

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diving capacity (Kooyman *et al.* 1980; Costa 1993; Ponganis, Kooyman & Castellini 1993). Theoretically, aerobic diving results in increased foraging time by minimizing variations in blood chemistry requiring extended recovery (Castellini, Davis & Kooyman 1988). Although diving anaerobically increases a single dive's duration, total time underwater is reduced as more time must be spent at the surface clearing accumulated lactate (LA) (Kooyman *et al.* 1980, 1983). Many air-breathing vertebrates dive within their limit of estimated O_2 stores for the vast majority of dives (Kooyman *et al.* 1980; Dolphin 1988; Kooyman 1989).

The aerobic dive limit (ADL), or diving lactate threshold (DLT), is experimentally defined as the maximum dive duration without increases in blood LA (Kooyman *et al.* 1980, 1983; Butler 2006). It has only been directly measured in a few species (Kooyman *et al.* 1980, 1983; Williams, Friedl & Haun 1993; Ponganis *et al.* 1997a,b; Ponganis, Kooyman & Winter 1997c; Shaffer *et al.* 1997). However, these studies found ADL could be predicted by dividing available O_2 stores by O_2 consumption rates. This equation, the calculated aerobic dive limit (cADL), is a conceptual tool used as an index of aerobic and physiological capacity (Kooyman 1989).

In some species where adults are known to primarily dive aerobically, juveniles rely on energy produced anaerobically for a larger percentage of dives (Kooyman *et al.* 1983; Burns 1999). This is due to inexperience and a lower ADL. Divers are born with minimal O_2 stores that develop as they mature (Davis 1991; Horning & Trillmich 1997; Ponganis *et al.* 1999). Young animals have intrinsically higher metabolic rates and costs associated with growth (Brody 1945; Schmidt-Nielsen 1984; Thorson & Le Boeuf 1994). They appear unable to regulate heart rate, respiration, vasoconstriction or body temperature as effectively as adults, which would also limit diving ability (Rea & Costa 1992; Ponganis *et al.* 1993; Greaves *et al.* 2005).

Oxygen stores appear particularly important in determining dive potential (Horning & Trillmich 1997; Ponganis *et al.* 1999; Burns *et al.* 2005). Across different species, numerous studies found a positive relationship between total O_2 stores and diving ability (Ridgway & Johnston 1966; Lenfant *et al.* 1970; Keijer & Butler 1982). Muscle myoglobin (Mb) tends to be the most prominent and consistent physiological predictor of breath-hold endurance (Kooyman 1989; Kooyman & Ponganis 1998; Ponganis *et al.* 1999). In previous studies, Mb has been the slowest O_2 store to develop (Thorson & LeBoeuf 1994; Noren *et al.* 2001, Richmond, Burns & Rea 2006). Although few studies have examined otariids, they store proportionately more O_2 in muscle than seabirds and phocids (Kooyman 1989), so extended Mb development may particularly constrain otariid pups.

Australian sea lions *Neophoca cinerea* (Péron) are excellent subjects to study otariid diving ontogeny as they are non-migratory and demonstrate extended dependency, during which pups begin diving. Pups are suckled for 17·6 ± 0·1 months, one of the longest lactation periods in pinnipeds thought to have evolved as an adaptation to a marine environment where resources are limited and show little seasonal fluctuation (Higgins 1993; Gales, Shaughnessy & Dennis 1994). As a result of small population size, small breeding colony size, exposure to human activities and evidence of population declines, Australian sea lions have recently been listed as threatened (EPBC Act 2000).

The Australian sea lion provides an intriguing system as one of few species in which adults regularly exceed cADL's, with almost 80% of dives over predicted limits (Costa *et al.* 2001). Adults spend 58% of time at-sea underwater and exhibit high field metabolic rates (FMR) (Costa & Gales 2003). Given the extreme foraging behaviour of adults and the potentially limited capabilities of younger animals, we wanted to examine diving ability in Australian sea lion pups and juveniles.

We investigated the ontogeny of O_2 stores (hematocrit (Hct), haemoglobin (Hb), plasma volume, Mb), FMR and cADL in Australian sea lions. Although many studies have looked at one or more aspects of cADL development in marine mammals, this is the first to simultaneously measure blood and muscle O_2 stores with FMR. By examining the extent physiology limits dive behaviour in young Australian sea lions, we can answer a central question in physiological ecology for this species, provide insight into its threatened status, and contribute to the emerging field of conservation physiology (Wikelski & Cooke 2006).

Materials and methods

Fieldwork was conducted between June 2001 and August 2003 at Seal Bay Conservation Park, Kangaroo Island, South Australia (35°41′S, 136°53′E). A known-aged cohort of 55 pups (28 males and 27 females) was flipper-tagged in 2001 (Fowler *et al.* 2006). To ensure individuals were only measured once, mothers and pups received a subcutaneous passive microtransponder chip (Destron Fearing Corporation, South St Paul, MN, USA).

Mother/pup pairs were captured simultaneously, sedated with Isoflurane gas anaesthesia, and weighed with a digital scale (± 0·1 kg) (Gales & Mattlin 1998). Pairs were captured during three field seasons: (i) 6-month pups (March 2002); (ii) 15-month pups (November 2002); and (iii) 23-month juveniles (July 2003). Ten different mother/pup pairs were captured each season, with the exception of two 15-month pups and five 23-month juveniles, which were never observed suckling and were captured alone. Adult females suckling young pups were captured in place of their mothers. The remaining 23-month juveniles were observed suckling at least once during the field season, despite the fact weaning usually occurs by 17·6 months (Higgins 1993). In July 2003, only six 23-month juveniles (not sampled in March or November 2002) could be located. Therefore, age for the remaining juveniles was estimated using pelage condition and growth curves constructed from data on mass and standard length (Fowler 2005). One independent juvenile from the previous cohort (aged *c.* 3 years) was also captured and sampled in July 2003.

HAEMATOLOGY

Hematocrit (Hct) declines as the spleen expands under general anaesthesia, so we took initial blood samples using manual restraint (Zapol *et al.* 1989; Ponganis *et al.* 1992; Costa, Gales & Crocker 1998). We measured Hct in quadruplicate the same day of collection in capillary tubes following centrifugation for 5 min at 11 500 r.p.m. For individuals that could not be sampled using manual restraint (one 6-month pup, one 15-month pup, two 23-month juveniles), we used a minimum of three sampling points and linear regression to hindcast Hct before administration of anaesthesia.

To determine whole-blood haemoglobin concentration (Hb), 10 µL whole blood were added to 2·5 mL Drabkins solution (Kit 525A, Sigma Diagnostics, St Louis, MO, USA) and later assayed in duplicate using the cyan-methhaemoglobin photometric method (ICSH 1967). Samples were read at 540 nm (Spectronic 1001, Bausch & Lomb, Rochester, NY, USA) and Hb was determined by comparison with standard dilution curves. Following methods for Hct, linear regression was used to hindcast Hb when necessary. We determined mean corpuscular haemoglobin content (MCHC) using the equation:

$$\text{MCHC} = (\text{Hb} \times 100) \times \text{Hct} - 1.$$

PLASMA VOLUME

Plasma volume was determined using Evans Blue dilution (ICSH 1967). A 10-mL blood sample was drawn from the caudal gluteal vein, followed by an intravenous injection of pre-weighed Evans Blue dye (Sigma Diagnostics) approximating a dosage of 0·6 mg kg^{-1} (Costa *et al.* 1998). The syringe was flushed with blood to ensure injections were intravenous and all dye was administered. Two to three serial samples followed at 10 min intervals.

Blood samples were kept on ice until centrifuged the same day for 10 min at 3400 revs min^{-1}. Plasma was kept frozen for a maximum of 3 months, when samples were thawed and centrifuged for 10 min. Plasma optical densities were determined at 624 and 740 nm following El-Sayed, Goodall & Hainsworth (1995), with modifications by Foldager & Blomqvist (1991). Adjusted absorbances were logarithmically transformed and linear regression used to determine dye concentration at time of injection. If the regression was not significant or the line's slope was positive (two 6-month pups and three 23-month juveniles), adjusted absorbance values were averaged (Jørgensen *et al.* 2001; Arnould *et al.* 2003). Plasma volume was calculated as distributional volume of injected dye (El-Sayed *et al.* 1995).

MYOGLOBIN

Biopsies were collected from the dorsal triceps and pectoralis complex locomotor muscles to analyze Mb. Additionally, we obtained muscle opportunistically from 10 fresh carcasses, ranging in age from 1 week to adult. Samples were frozen at $^{-}80$ °C until analyses. We determined Mb following methodology detailed in Reynafarje (1963), as modified by Castellini & Somero (1981). Buffer blanks and elephant seal muscle of known Mb were used as assay controls.

TOTAL OXYGEN STORES

We determined total available O_2 stores by adding stores in blood, muscle and lungs (Lenfant *et al.* 1970; Kooyman *et al.* 1983; Kooyman 1989). Blood O_2 stores were calculated as the sum of arterial and venous O_2:

$$\text{Arterial } O_2 = Vb \times 0.33 \times Ca_{O_2}$$
$$\text{Venous } O_2 = Vb \times 0.66 \times Ca_{O_2} \times SV_{O_2}$$

where Vb is blood volume, 0·33 is the percentage arterial blood, 0·66 is the percentage venous blood, Ca_{O_2} (capacitance coefficient of O_2 in blood) = Hb × 1·34 mL O_2, and SV_{O_2} is O_2 saturation of venous blood. An O_2 carrying capacity of 1·34 mL O_2 (g^{-1}) was assumed (Kooyman 1989). We also assumed 75% of arterial blood O_2 was available during a dive (with 15% used to maintain vital body and brain functions; 95% O_2 saturation to 20%: Ponganis *et al.* 1997c; Costa *et al.* 2001) and mixed venous blood had an O_2 content 5% by volume less than initial Ca_{O_2} (Ponganis *et al.* 1993), so $SV_{O_2} = [(Ca_{O_2} - 50)\,(Ca_{O_2})^{-1}] \times 100$ (Davis & Kanatous 1999).

Muscle O_2 stores were calculated using the equation:

$$\text{Muscle } O_2 = \text{Mb} \times 1{\cdot}34\ \text{mL } O_2 \times \text{body mass} \times 0{\cdot}3$$

where 0·3 is the fraction of muscle mass in the body (Kooyman *et al.* 1983). This is also identical to the fraction of muscle mass found from complete dissection of 1-month Steller sea lion *Eumetopias jubatus* pups (Richmond *et al.* 2006). For two 23-month-old juveniles for which muscle biopsies were not available, mean Mb determined for the age class was used to calculate muscle O_2.

Following Costa *et al.* (2001), lung O_2 stores were derived from allometric estimates of lung volume for otariids:

$$\text{Lung } O_2 = V_i \times 0{\cdot}15\,\text{FO}_2$$

where V_i (diving lung volume) = $0{\cdot}5 \times 0{\cdot}10 \times \text{mass}^{0{\cdot}95}$ and 0·15 FO_2 is O_2 extracted from air in the lungs (Kooyman *et al.* 1971; Kooyman 1989).

FIELD METABOLIC RATES (FMR)

Concurrent measurements of CO_2 production and diving behaviour were carried out on eight of the 23-month Australian sea lions and the one 3-year-old to determine at-sea metabolism using oxygen-18 doubly-labelled water (Lifson & McClintock 1966; Nagy & Costa 1980; Speakman 1997). For comparison with published adult FMR, methodologies were identical to Costa & Gales (2003).

Pre-injection blood samples were taken to determine isotope background values, followed by intraperitoneal injections of 60–80 mL 15% oxygen-18 water ($H_2{}^{18}O$) and 18·5 MBq/mL tritiated water (HTO) in 3 mL sterile saline. Syringes were weighed ($\pm$ 0·001 g) before and after injections to determine masses injected. After 3 h equilibration, body mass was recorded and a 10 mL blood sample collected to determine isotope concentrations at the start of the experimental period. Juveniles were equipped with Wildlife Computers (Redmond, WA, USA) time/depth recorders (TDR's) and VHF radio transmitters (Sirtrack Ltd, Havelock, New Zealand) (Fowler *et al.* 2006). Juveniles were recaptured after 5–8 days to record body mass, collect final blood samples, and recover TDR's.

Tritium specific activity was determined by scintillation spectrometry (Tri-Carb 2100TR, Packard, Canberra, ACT, Australia) of duplicate aliquots 0·2 mL pure water (distilled from plasma samples) in 10 mL scintillation fluid (Ultima Gold scintillation fluid, Packard Bio Science, Meriden, CT, USA). Specific activity of $H_2{}^{18}O$ was determined by mass ratio spectrometry (Metabolic Solutions, Nashua, NH, USA).

Initial dilutions of $H_2{}^{18}O$ were used to determine total body water (TBW) (Nagy & Costa 1980). Final TBW was calculated by the equation from initial TBW corrected for change in mass. We calculated lean body mass from TBW, assuming a hydration constant of 74·2% reported for California sea lions *Zalophus californianus* (Oftedal, Iverson & Boness 1987), and calculated CO_2 production using Speakman's (1997) two-pool model to correct for errors associated with isotope fractionation. Water influx was calculated using equations (5) and (6) in Nagy & Costa (1980), assuming an exponentially changing body water pool.

As Australian sea lions' diet is not well-known and six juveniles were observed suckling at least once during the field season, we followed calculations for Steller sea lion juveniles (Richmond *et al.* 2006) and used a respiratory quotient (RQ) of 0·76 (19·3 kJ L^{-1} O_2). This assumes a 50 : 50 lipid : protein fuel source intermediate between nursing pups' lipid-rich diet and foraging adult's protein-rich diet (Schmidt-Nielsen 1997; Iverson, Frost & Lang 2002). Assuming a diet of 100% lipid or 100% protein alters the RQ by less than 5%. We divided CO_2 production by RQ to determine O_2 consumption.

Data collected over measurement intervals included variable amounts of onshore FMR and were normalized to estimate metabolism at-sea. Percentages of time spent at-sea were calculated from TDR data and following methods from Costa & Gales (2003) for adult Australian sea lions, we plotted FMR data (containing both at-sea and onshore components) as a function of percentage time spent at-sea. Least squares linear regression was used to predict FMR for each animal at their respective percentage time spent at-sea (Costa & Gales 2003). The difference (residual) between predicted and actual FMR was added to extrapolated FMR where the animal spent 0% time at-sea to determine onshore metabolism. At-sea FMR was calculated from the equation:

$$\text{FMR} = \text{at-sea FMR (\% time at-sea)} + \text{onshore FMR (\% time ashore)}.$$

CALCULATED AEROBIC DIVE LIMIT (CADL)

We calculated cADL's based on equations from Kooyman *et al.* (1980, 1983):

$$\text{cADL (min)} = \text{total } O_2 \text{ (available blood } O_2 + \text{muscle } O_2 + \text{lung } O_2\text{) (at-sea FMR)}^{-1}.$$

Dive behaviour data recorded from these juveniles and reported in Fowler *et al.* (2006) were used to calculate percentages of dives over cADL and ratios of mean dive duration to cADL.

For comparison with at-sea FMR, we substituted estimates of otariid DMR from the literature and recalculated cADL's for juvenile Australian sea lions. Hastie, Rosen & Trites (2006) used metabolic rates of Steller sea lion females trained to dive to depth in the open ocean to construct a model predicting O_2 consumption. Although some studies have shown during prolonged breath-holds adult pinnipeds may lower metabolism to resting (Hurley & Costa 2001; Sparling & Fedak 2004), young pinnipeds have less metabolic control and juvenile Weddell seals *Leptonychotes weddellii* appear unable to do so; cADL's based on resting metabolism (RMR) overestimated ADL's indicated by changes in LA by 60% (Rea & Costa 1992; Ponganis *et al.* 1993; Burns & Castellini 1996). We therefore chose 2 × RMR (determined by multiplying BMR by age-specific scaling factors estimated for Steller sea lions: Winship, Trites & Rosen 2002) to represent minimum cost of transport (Feldkamp 1987; Costa 1991; Arnould & Boyd 1996). Finally, based on the only direct measurements of ADL in an otariid, we used a value of 17·8 mL O_2 min^{-1} kg^{-1} determined by measuring post-submersion blood LA in similarly-sized (41·4 kg) juvenile California sea lions (Ponganis *et al.* 1997c).

Using the different estimates of DMR, surface metabolic rates (MR) were then calculated from:

$$\text{at-sea FMR} = \text{DMR (\% time diving)} + \text{surface MR (\% surface time)}.$$

When statistical differences were determined by one-way analysis of variance, post-hoc comparisons were made using Tukey tests. If transforming data did not achieve normality and equal variances, differences were determined by Kruskal–Wallis one-way analysis of variance on ranks and Dunn's post-hoc test was used.

Results

HAEMATOLOGY

Australian sea lions had fully developed adult Hct and Hb by 15 months (Table 1). Six-month pups were the only age class with significantly lower Hct and Hb (Hct: $H_3 = 28{\cdot}85$, $P < 0{\cdot}001$; Hb: $H_3 = 24{\cdot}21$, $P < 0{\cdot}001$). Values for adult females agree closely with published values for this species (Costa *et al.* 2001). As there were no significant differences between sexes within age classes, data were combined (t-test: $t_7 = 0{\cdot}11$, $P = 0{\cdot}92$). There were no significant differences between age classes for MCHC ($F_{3,46} = 2{\cdot}19$, $P = 0{\cdot}10$), which remained relatively constant throughout development and was 34·6 ± 0·5 g/dL at 6 months, 35·4 ± 0·9 g/dL at 15 months, 38·5 ± 0·6 g/dL at 23 months and 35·8 ± 1·1 g/dL for adult females.

PLASMA VOLUME

Plasma and blood volumes were slower to develop (Table 1). Well beyond the age of average weaning, 23-month juveniles demonstrated mass-specific blood volumes only 68% of adult blood volumes. Pups had significantly lower mass-specific plasma and blood volumes than adult females (plasma volume: $F_{3,20} = 6{\cdot}89$, $P = 0{\cdot}002$; blood volume: $F_{3,20} = 10{\cdot}03$, $P < 0{\cdot}001$) and although plasma volumes for 23-month juveniles were lower than adult values, this was not significant ($P = 0{\cdot}08$). Plasma volume was 10% of lean body mass in both juvenile and adult Australian sea lions.

MYOGLOBIN

Muscle Mb increased linearly with age ($r^2 = 0{\cdot}82$, $P < 0{\cdot}001$) and mass ($r^2 = 0{\cdot}80$, $P < 0{\cdot}001$). However, development was comparatively slow: 23-month juveniles only developed Mb to 60% of adult capacities (Table 1). Muscle Mb was significantly different across age classes ($F_{4,16} = 49{\cdot}63$, $P < 0{\cdot}001$), with the exception of values between 15 and 23 months ($P = 0{\cdot}07$). The measured Mb for adult females (2·7 ± 0·1 g/100 g wet tissue) agrees closely with the published value (2·8 g%) and the difference is within the assay's resolution (Costa *et al.* 2001).

TOTAL OXYGEN STORES

Mass-specific blood, muscle and total O_2 stores were significantly higher in older animals (blood: $F_{3,20} = 19{\cdot}28$, $P < 0{\cdot}001$; muscle: $F_{3,14} = 58{\cdot}46$, $P < 0{\cdot}001$;

Table 1. Summary of O_2 storage parameters for different age classes of Australian sea lions (mean ± SE); * = values significantly different from adult. Ranges of ages are reported in parentheses following mean ages and *n* is given in parentheses below other values. Although hematocrit and haemoglobin were fully developed by 15 months, plasma volume, blood volume, and muscle myoglobin were slower to develop

Age (months)	Mass (kg)	Hematocrit (%)	Haemoglobin (g dL^{-1})	Plasma volume (L)	Plasma volume (mL kg^{-1})	Blood volume (L)	Blood volume (mL kg^{-1})	Myoglobin (g%)
6·1 ± 0·2 (5·4–7·1)	30·0 ± 1·7* (10)	39·3 ± 1·0* (10)	13·6 ± 0·5* (10)	1·6 ± 0·1* (7)	52·4 ± 5·4* (7)	2·6 ± 0·2* (7)	83·5 ± 7·5* (7)	0·8 ± 0·2* (4)
14·5 ± 0·2 (13·4–15·7)	44·5 ± 2·0* (10)	51·4 ± 0·7 (10)	18·2 ± 0·4 (10)	2·0 ± 0·2* (10)	45·6 ± 3·4* (10)	4·2 ± 0·4* (10)	93·8 ± 7·1* (10)	1·3 ± 0·1* (5)
22·6 ± 0·2 (22·1–22·9)	48·3 ± 2·6* (9)	49·5 ± 1·0 (9)	19·0 ± 0·4 (9)	2·8 ± 0·3* (8)	60·1 ± 4·1 (8)	5·7 ± 0·6* (8)	120·9 ± 8·6* (8)	1·6 ± 0·2* (6)
3 years	65·0 (1)	51·6 (1)	19·6 (1)	4·4 (1)	68·0 (1)	9·1 (1)	140·6 (1)	2·2 (1)
Adult	88·2 ± 2·1 (21)	51·7 ± 0·5 (21)	18·6 ± 0·6 (21)	6·7 ± 1·3 (2)	83·7 ± 12·5 (2)	14·2 ± 3·0 (2)	178·3 ± 30·9 (2)	2·7 ± 0·1 (3)

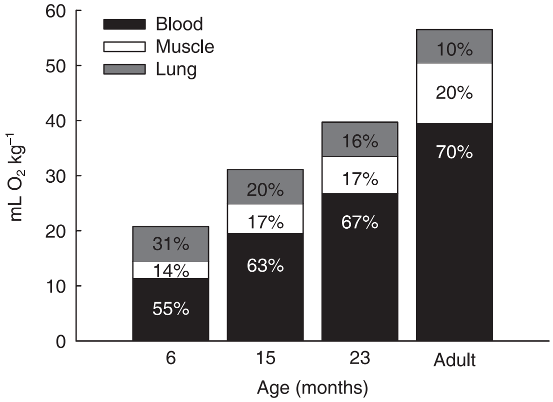

Fig. 1. Mean mass-specific O_2 stores for different age classes of Australian sea lions. Each bar's height represents total stores and bars are divided into three shades to depict different storage compartments. The percentage of total O_2 represented by each compartment is written in the bar.

total: $F_{3,27} = 56{\cdot}73$, $P < 0{\cdot}001$) and had not reached adult capacities by 23 months (Fig. 1). Even by 3 years, mass-specific blood O_2 was only 29·0 mL O_2 kg^{-1}, muscle O_2 was 8·7 mL O_2 kg^{-1} and total stores were 43·8 mL O_2 kg^{-1} (78% of adult total stores). Total available mass-specific O_2 stores increased significantly with age ($r^2 = 0{\cdot}66$, $P < 0{\cdot}001$) and mass ($r^2 = 0{\cdot}70$, $P < 0{\cdot}001$).

Mean and maximum dive depth (Fig. 2a,b), and mean ($r^2 = 0{\cdot}51$, $P < 0{\cdot}001$), and maximum dive duration ($r^2 = 0{\cdot}56$, $P < 0{\cdot}001$) increased with increasing O_2 stores (Fowler *et al.* 2006).

FIELD METABOLIC RATES (FMR)

Mean FMR for 23-month juveniles was 15·44 ± 1·30 mL O_2 min^{-1} kg^{-1} (see Appendix for FMR data). Field metabolism for the 3-year-old was 18·25 mL O_2 min^{-1} kg^{-1}. Mass-specific FMR's were slightly higher for juveniles than values calculated for adult Australian sea lions using Speakman's (1997) two-pool model (Costa & Gales 2003), although this was not significant ($t_{27} = 0{\cdot}26$, $P = 0{\cdot}80$). At-sea FMR was calculated to be 15·78 ± 1·29 mL O_2 min^{-1} kg^{-1} at 23 months and 18·41 mL O_2 min^{-1} kg^{-1} at 3 years.

CALCULATED AEROBIC DIVE LIMIT (CADL)

By dividing total available O_2 by at-sea FMR, we found juvenile Australian sea lions developed a cADL of 2·33 ± 0·24 min by 23 months (Table 2). Like adults, juveniles exceeded cADL's on the majority of dives (Table 2). Furthermore, there were no significant differences between ratios of mean dive duration to cADL for adults and juveniles ($t_{17} = -1{\cdot}29$, $P = 0{\cdot}90$), indicating both groups exceeded cADL's to similar extents.

There were no significant differences between DMR's determined from FMR, at-sea FMR, predicted O_2 consumption, 2 × RMR, and post-submersion LA ($F_{4,38} = 1{\cdot}41$, $P = 0{\cdot}25$: Table 3). The mass-specific value predicted for Steller sea lions diving to 50 m was remarkably similar to FMR and at-sea FMR measurements for juvenile Australian sea lions diving to depths of 44 ± 4 m (Fowler *et al.* 2006; Hastie *et al.* 2006; Table 3). There were also no significant differences between percentages of dives over cADL ($F_{4,33} = 0{\cdot}44$, $P = 0{\cdot}78$), ratios of mean dive duration to cADL ($F_{4,38} = 1{\cdot}16$, $P = 0{\cdot}35$), or surface MR ($F_{4,30} = 0{\cdot}10$, $P = 0{\cdot}98$; Table 3).

There were increases in minimum post-dive surface intervals (PDSI) as Australian sea lion juveniles approached cADL's (Fig. 3a,b), with minimum PDSI's 2·2 times longer for dives over cADL's than dives under (Fowler *et al.* 2006).

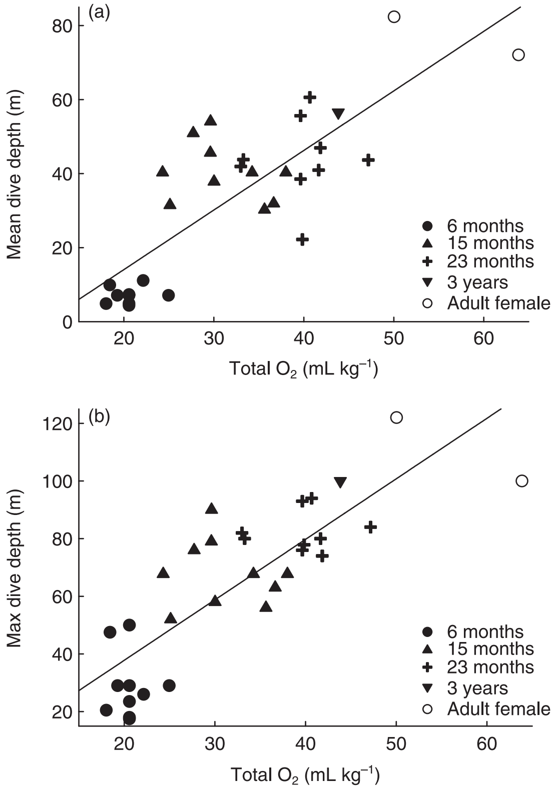

Fig. 2. Total mass-specific O_2 stores in relation to dive depth for different age classes of Australian sea lions (Fowler *et al.* 2006). Symbols represent individual animals. (a) Mean ($r^2 = 0{\cdot}67$, $P < 0{\cdot}001$) and (b) maximum ($r^2 = 0{\cdot}68$, $P < 0{\cdot}001$) dive depth increased significantly with O_2 stores.

Discussion

Juvenile Australian sea lions well beyond the age of average weaning had not developed adult O_2 stores. Juveniles were also not reaching adult dive depths or durations, suggesting they are limited by physiological capability. Like adults, juveniles appeared to be operating close to, or above their estimated physiological maximum, with the majority of dives over cADL's. These factors suggest juvenile Australian sea lions have limited ability to compensate for environmental fluctuations, restricted potential foraging habitat and restricted behavioural options.

Table 2. Calculated aerobic dive limits (cADL) for Australian sea lions (mean ± SE). Values for adult females from Costa *et al.* (2001); * = values significantly different from adult. Masses are presented as means from initial captures and recaptures. At-sea field metabolic rates (FMR) were calculated using equations from Speakman (1997) and corrected for percentage of time spent onshore. Juvenile total available O_2 stores were significantly lower than adult ($t_{19} = -7{\cdot}72$, $P < 0{\cdot}001$), but juveniles exceeded cADL's as often ($t_{16} = 1{\cdot}81$, $P = 0{\cdot}09$)

Animal ID	Age (months)	Mean mass (kg)	Total available O_2 (mL)	At-sea FMR (mL O_2 min^{-1})	cADL (min)	Mean dive duration (min)	Dives > cADL (%)	Mean dive duration (cADL)$^{-1}$
AD	22·1	58·5	1456	780·25	1·87	2·96	73·5	1·59
ED	22·4	43·9	1907	659·05	2·89	2·79	63·8	0·96
EL	22·8	40·8	1281	771·07	1·66	2·50	67·4	1·50
FI	22·9	59·8	2421	944·78	2·56	2·50	55·2	0·98
GO	22·2	45·8	1449	526·53	2·75	2·80	67·8	1·02
LE	22·6	50·1	1698	1005·45	1·66	2·61	78·2	1·58
WI	22·8	41·5	1619	468·87	3·45	3·70	69·5	1·07
MA	22·9	41·1	1519	834·47	1·82	2·14	66·9	1·18
Juvenile mean	22·6 ± 0·1	47·7 ± 2·7*	1668·8 ± 126·2*	748·81 ± 66·67*	2·33 ± 0·24	2·75 ± 0·16*	67·8 ± 2·8	1·23 ± 0·10
SC	3 years	67·7	2849	1246·57	2·29	2·75	68·70	1·20
Cow mean	Adult	77·8 ± 3·8	3656·0 ± 179·0	1589·91 ± 101·53	2·34 ± 0·11	3·14 ± 0·16	79·4 ± 4·8	1·38 ± 0·11

BLOOD OXYGEN STORES

Hematocrit (Hct) and Hb increased while MCHC remained constant, suggesting although red blood cells mature early, young pups have fewer cells. However, Hct and Hb were fully developed by 15 months, so lower blood O_2 stores in older pups resulted from differences in mass-specific blood volumes (Table 1). Age-specific differences in plasma and blood volumes may be largely due to changes in body composition and hydration. We found, like harbour seals *Phoca vitulina* and Steller sea lions, although mass-specific plasma and blood volumes fluctuated throughout development, ratios of plasma volume to lean body mass remained relatively constant (Burns *et al.* 2005; Richmond *et al.* 2006).

MUSCLE OXYGEN STORES

Increases in Mb and muscle O_2 stores lagged behind development of blood O_2 stores in Australian sea lions. In previous studies on diving vertebrates, Mb had also not reached adult levels by onset of independent foraging (Weber, Hemmingsen & Johansen 1974; Thorson & LeBoeuf 1994; Noren *et al.* 2001). As Mb is the slowest O_2 storage parameter to mature in species studied to date, its development appears to be a major limiting factor on diving ability in juveniles.

Because Australian sea lions have to meet physiological demands of benthic diving at an early age, they may deal with prolonged development in muscle by storing proportionately more O_2 in blood and less in muscle than other pinnipeds (Fowler *et al.* 2006). On average, adult otariids store 51% of total O_2 in blood and 33% in muscle (Kooyman 1989; Richmond *et al.* 2006) and phocids store 65% in blood and 25% in muscle (Kooyman 1989; Burns *et al.* 2005; Noren, Iverson & Boness 2005). Australian sea lion adults store 70% in blood and 20% in muscle (Costa *et al.* 2001), which more closely resembles stores of deeper diving phocids than otariids. In Australian sea lion juveniles, blood O_2 stores accounted for 1·53 ± 0·20 min (62·2%) of cADL's and muscle O_2 stores accounted for 0·47 ± 0·06 min (19·7%).

TOTAL OXYGEN STORES

Protracted blood volume and Mb maturation, as well as low body mass contributed to low total available O_2 stores in pups and juveniles. Fowler *et al.* (2006) reported young Australian sea lions dived benthically

Table 3. Summary of different diving metabolic rates (DMR) used to calculate aerobic dive limits (cADL) in juvenile Australian sea lions ($n = 8$, mean ± SE). Field metabolism (FMR) was determined using doubly-labelled water (Lifson & McClintock 1966); at-sea FMR (**bold**) was corrected for percentage of time spent onshore. For comparison, predicted O_2 consumption for Steller sea lions (SSL) diving to 50 m (Hastie *et al.* 2006), 2 × resting metabolism (RMR) representing minimum cost of transport, and DMR determined by measuring post-submersion blood lactate (LA) in similarly-sized juvenile California sea lions (CSL) (Ponganis *et al.* 1997c) are presented. There were no significant differences between alternate cADL's ($H_4 = 3{\cdot}22$, $P = 0{\cdot}52$)

DMR method	Diving MR (mL O_2 min^{-1} kg^{-1})	cADL (min)	Dives > cADL (%)	Mean dive duration (cADL)$^{-1}$	Surface MR (mL O_2 min^{-1} kg^{-1})
FMR	15·44 ± 1·30	2·39 ± 0·25	66·7 ± 2·8	1·21 ± 0·10	16·54 ± 1·27
at-sea FMR	**15·78 ± 1·29**	**2·33 ± 0·24**	**67·8 ± 2·8**	**1·23 ± 0·10**	**15·78 ± 1·29**
SSL predicted	15·40	2·30 ± 0·11	67·7 ± 4·4	1·25 ± 0·09	16·25 ± 4·29
2 × RMR	17·53 ± 0·21	2·02 ± 0·11	71·4 ± 3·8	1·42 ± 0·11	11·45 ± 4·39
CSL post-submersion LA	17·80	1·99 ± 0·11	71·5 ± 3·8	1·44 ± 0·11	10·97 ± 4·42

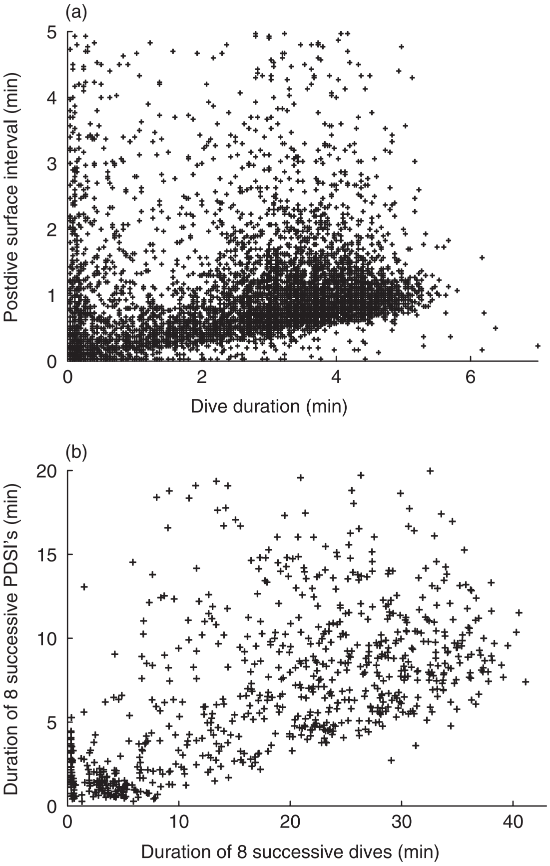

Fig. 3. (a) Dive durations and post-dive surface intervals (PDSI) for 23-month Australian sea lions. (b) Duration of eight successive dives and PDSI's, following methods of Horning & Trillmich (1997). There were slights increases in both individual (a) and cumulative PDSI's (b), but no evidence of disproportionate increases. Even when dives durations exceeded 4 min, 90% of PDSI's remained under 2 min.

like adults, but did not achieve adult dive depths or durations. By 23 months, dive behaviour and dive physiology developed to similar percentages of adult capacity: maximum depth was 76% adult depth, maximum duration was 77% adult duration, and total mass-specific O_2 stores were 70% adult stores (Fowler *et al.* 2006). The positive relationship between ontogeny of O_2 stores and ontogeny of dive performance suggests slow physiological development may constrain foraging capacities (Fig. 2).

Australian sea lions demonstrated the slowest development of total O_2 stores documented to date in mammalian divers and only developed 55% of adult female mass-specific total O_2 before average weaning. In comparison, 9-month Steller sea lions develop 81% (Richmond *et al.* 2006), 12-week northern elephant seals *Mirounga angustirostris* develop 74% (Thorson & LeBoeuf 1994), 41-day grey seals *Halichoerus grypus* develop 67% (Noren *et al.* 2005) and Weddell seals aged 41–50 days develop 64% (Burns & Castellini 1996) prior to onset of independent foraging. Although exact ages were not available, juvenile New Zealand sea lions *Phocarctos hookeri* developed an estimated 87% (Costa *et al.* 1998) and juvenile California sea lions developed 59% (Kuhn *et al.* 2006).

Galapagos fur seals *Arctocephalus galapagoensis* are the only pinniped with a typical lactation interval longer than Australian sea lions (Trillmich 1986). Horning & Trillmich (1997) found similar development of Hct and Hb in Galapagos fur seals, but did not collect data on blood volume or Mb. However, in the divers studied, species developing total O_2 stores earlier tend to wean at younger ages (Arnould *et al.* 2003).

FIELD METABOLIC RATES (FMR)

Field metabolic rates (FMR) for 23-month Australian sea lions were 3·95 times basal metabolic rates (BMR) predicted for adult terrestrial animals of equivalent size (Kleiber 1975). Although this is the first study to measure FMR in a juvenile sea lion, results fall within the range of rates reported for young pinnipeds (phocids: 2·7–6·0 times BMR (Reilly 1991; Kretzmann, Costa & LeBoeuf 1993; Lydersen & Hammill 1993; Lydersen & Kovacs 1996; Lydersen *et al.* 1996), otariids: 3·25–3·84 times BMR (Arnould *et al.* 2003)) and were only slightly higher than FMR's measured for adult Australian sea lions using equivalent methods (Costa & Gales 2003).

This is the second study to calculate at-sea FMR or DMR for an immature pinniped. While we estimated DMR for juvenile Australian sea lions ($n = 8$) to be 4·03 times BMR, Lydersen & Hammill (1993) used a value of 5·88 times BMR for ringed seal *P. hispida* pups ($n = 3$). This difference cannot be accounted for by differences in body mass (18·6 ± 0·3 *vs* 47·7 ± 2·7 kg), as basal metabolism was scaled to $\text{mass}^{0\cdot75}$ in both studies. However, ringed seals were studied at a younger age and their higher DMR may reflect immature metabolic control, faster growth rates and/or increased thermoregulatory demands in their arctic environment (Kleiber 1975; Rea & Costa 1992; Lydersen & Hammill 1993).

CALCULATED AEROBIC DIVE LIMIT (CADL)

Given juvenile Australian sea lions were found to exceed theoretical aerobic limits on 67·8% of dives, there are three possibilities: (i) cADL's were miscalculated; (ii) Australian sea lions are highly adapted to deal with LA; or (iii) this species is operating at or near its physiological limit in a marginal foraging environment (Costa & Gales 2003; Chilvers *et al.* 2006).

(i) cADL's were miscalculated

To accurately determine cADL's, available O_2 stores and O_2 use underwater must be known. Although body mass, Hb, Hct, blood volume and Mb were measured, muscle mass and respiratory volumes were estimated from the literature (Kooyman *et al.* 1971, 1983; Richmond *et al.* 2006). We assumed 75% of arterial O_2 and 100% of muscle O_2 was available during a dive. Our calculations should be conservative and over-estimate ADL as Davis & Kanatous (1999) reported only 63% of total O_2 stores may be accessible and not all muscle O_2 can be utilized aerobically.

Our approach is supported by an earlier study on California sea lions that measured increases in LA in freely-diving trained animals (Ponganis *et al.* 1997c). Researchers found a close agreement between measured ADL's and cADL's based on estimates of total O_2 stores similar to presented here (Ponganis *et al.* 1997c). Furthermore, FMR's derived from doubly-labelled water measurements on wild adult females (Costa, Antonelis & DeLong 1990), such as presented here, gave more accurate predictions of ADL than calculations using metabolic rates estimated from flumes.

It is important to note at-sea FMR integrates costs of highly variable surface swimming, diving, digestion and resting. Juvenile DMR may be higher or lower. Behavioural modifications, such as burst and glide locomotion (Skrovan *et al.* 1999; Crocker, Gales & Costa 2000; Williams *et al.* 2000), drift diving (Crocker, LeBoeuf & Costa 1997; Page *et al.* 2005) and 'wait and ambush' hunting tactics (Thompson & Fedak 1993), or physiological adaptations, such as hypometabolism (Castellini *et al.* 1985; Butler & Jones 1997; Butler 2006) and regional hypothermia (Andrews *et al.* 1994; Handrich *et al.* 1997; Bevan *et al.* 2002) may help vertebrates reduce O_2 consumption while diving. During deeper benthic dives, changes in buoyancy and ground effect may lead to reduced stroke frequency and lower DMR (Ponganis & Kooyman 2000).

However, we compared at-sea FMR to a number of different estimates for otariids and found no differences between DMR's and cADL's (Table 3). Furthermore, DMR estimates from the literature resulted in slightly lower cADL's, with juveniles consequently exceeding cADL more often and to a greater extent, so calculations using at-sea FMR appear conservative. We therefore feel until direct measurements can be made, at-sea FMR provides a useful DMR estimate for juvenile Australian sea lions. Additionally, at-sea FMR was used to determine cADL's in adult Australian sea lions, as well as other adult pinnipeds, allowing for internally consistent comparisons (Costa *et al.* 2001, 2004).

(ii) Australian sea lions are highly adapted to deal with LA

Weddell seals can remain submerged for durations three times their ADL, indicating sufficient O_2 left to supply the nervous system and, to at least some extent, locomotor muscles (Kooyman *et al.* 1980; Butler 2004). Likewise, muscle has probably begun proportional energy production by anaerobic glycolysis at the point of net production of LA (Kooyman 2006). These factors complicate assumptions about O_2 store management which must be made to calculate cADL. Without measurements of post-dive LA in additional species, or a better understanding of how divers manage and use O_2, it is impossible to draw definitive conclusions about LA levels solely from cADL.

There were increases in minimum PDSI's as Australian sea lion juveniles approached maximum dive durations (Fig. 3a, b), which may indicate LA production. However, there was no evidence of extended PDSI's or cumulative effects predicted to clear accumulated LA (G.L. Kooyman & T.G. Kooyman 1995). Lack of extended surface recovery has also been documented in a number of adult penguins and pinnipeds found to dive beyond their cADL, including king penguins *Aptenodytes patagonicus* (Handrich *et al.* 1997), northern elephant seals (Le Boeuf *et al.* 1989), and New Zealand (Crocker *et al.* 2000; Chilvers *et al.* 2006) and Australian sea lions (Costa & Gales 2003). Although these data indicate some species use more aerobic capacity than others, none of these studies were able to measure changes in LA. Results highlight the potentially problematic nature of assuming aerobic dives if PDSI's are not protracted and further research into whether or how these animals exceed their ADL is required. Until direct measurements of post-dive LA are taken, other indirect measurements of effort, such as heart rate or acceleration, would be instructive (Butler *et al.* 1992, 1995; Sato *et al.* 2002).

If Australian sea lions were relying on anaerobic metabolism for the majority of dives, they must be able to buffer, recycle and/or burn LA. As juveniles only exceeded cADL's by a mean factor of 1·23, this would lead to a minor metabolic imbalance of acidosis. Using the Weddell seal as a model, exceeding its 20 min ADL by a factor of 1·23 is equivalent to a 25 min dive resulting in a blood LA concentration of *c.* 2 μmol and requiring less than 10 min surface recovery (Kooyman *et al.* 1980). Likewise, resting LA turnover rates (26·2 μmol min^{-1} kg^{-1}) measured in the more similarly-sized harbour seal (40·3 kg) would require 9 min recovery (Davis *et al.* 1991). Yet, only 1·1% of PDSI's reached 9 min for juvenile Australian sea lions. However, relative differences were similar: for 25 min dives, Weddell seals require PDSI's two to three times longer than aerobic dives, while minimum PDSI's for Australian sea lions were 2·2 times longer for dives over cADL's than dives under.

Alternatively, juvenile Australian sea lions could use accumulated LA as substrate on following dives (Castellini *et al.* 1988). One molecule of ATP is produced for each molecule of LA through glycolysis, yet 17 molecules of ATP are produced by subsequent oxidation of one molecule of LA (Fedak & Thompson 1993). Therefore, in the unlikely event juveniles were producing energy exclusively anaerobically during the 0·42 min (25 s) beyond cADL's, it would take 7·10 min ($0{\cdot}42 \times 17$) of aerobic diving using LA as substrate until blood LA returned to normal (Butler 2004). However, harbour seals swimming at 35% VO_{2max} increase LA turnover to 39·7 μmol min^{-1} kg^{-1}, which would decrease recovery time for a blood LA concentration

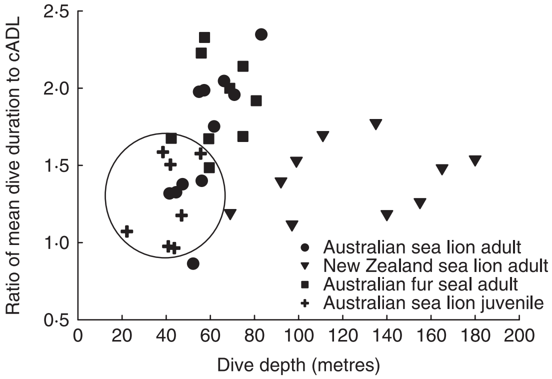

Fig. 4. Dive performance defined as ratios between mean dive duration and cADL as a function of mean dive depth for benthic foragers. Symbols represent individual animals. Juvenile Australian sea lions (circled) were added to a figure modified from Costa *et al.* (2004) and did not reach depths typical of other benthic foragers.

of 2 µmol to 5 min; swimming at 50% VO_{2max} would require a 4 min recovery (Davis *et al.* 1991). With mean dive durations of 2·75 min, juveniles would have to perform 1–3 dives within the cADL following every dive over (Fowler *et al.* 2006). As 67·8% of dives were over, juvenile Australian sea lions may use additional or alternative methods to deal with LA.

Lower Hct, lower blood viscosity and potentially higher lung turn-over rates (due to high flow rates and tidal volumes: Irving, Scholander & Grinnell 1941; Olsen, Hale & Elsner 1969; Matthews 1977) may enable otariids to shorten O_2 loading and LA clearance times, and recover from anaerobic dives faster than phocids (Kooyman 2006). Smaller, younger animals, such as otariid pups, with higher mass-specific metabolic rates may be able to process LA faster (Brody 1945; Schmidt-Nielsen 1984; Thorson & Le Boeuf 1994). Finally, as Kooyman (2006) suggested for adult Australian sea lions, juveniles spend 32% of time at-sea on the surface, which is a relatively large percentage (potentially clearing LA) compared to other species which regularly exceed cADL's such as southern elephant seals *M. leonina* and only spend 10% of time on the surface (Hindell, Slip & Burton 1991; Fowler *et al.* 2006).

(iii) This species is operating at or near its physiological limit in a marginal foraging environment

Regardless of potential problems, cADL can be a useful conceptual tool for determining foraging effort and efficiency in relation to environmental quality, as well as providing comparisons between different ages, sexes and species (Costa *et al.* 2004; Kooyman 2006). Animals operating well within their physiological capacity are able to draw on reserves to pursue prey deeper, dive longer or extend foraging bouts if necessary (Boyd *et al.* 1994). Both Australian sea lion adults and juveniles appear to operate at or near their physiological maximum in their present habitat, so the capacity to increase dive depth, duration, or foraging effort would be limited (Costa *et al.* 2001). Additionally, young sea lions do not reach adult dive depths or durations (Fowler *et al.* 2006). Restricted behavioural options would make juveniles even more susceptible to shifts in prey availability.

The ability to exploit different dive depths is important in determining potential foraging habitat, especially for benthic divers such as Australian sea lions (Costa & Gales 2003; Fowler *et al.* 2006). One limitation of benthic foraging is restricted available habitat due to the finite extent of continental shelves. This limitation is even more severe for pups that cannot or do not reach adult depths. Although juvenile Australian sea lions exceeded cADL's on the majority of dives and to similar extents as adult Australian sea lions, New Zealand sea lions and Australian fur seals *A. pusillus doriferus*, they did not achieve depths typical of other benthic foragers (Costa *et al.* 2004; Fig. 4).

IMPLICATIONS

Relative to many other species, which exceed theoretical ADL's on 4%–10% of dives, Australian sea lions use more metabolic scope to forage (Gentry, Kooyman & Goebel 1986; Feldkamp, Long & Antonelis 1989; Boyd & Croxall 1996). Additionally, Australian sea lions demonstrate the slowest development of O_2 stores during one of the longest lactation periods, strongly supporting the idea that rates of O_2 development are related to investment duration in pinnipeds (Arnould *et al.* 2003). With restricted access to benthic habitat and limited behavioural options, juvenile Australian sea lions appear particularly susceptible to resource limitation. Shallower dive depths would also restrict juveniles to near-shore waters, which would increase interaction risks with fisheries. These results have potential implications for the conservation and management of other species, especially those that forage benthically, such as New Zealand, Steller, and southern sea lions *Otaria flavescens* and Australian fur seals (Werner & Campagna 1995; Merrick & Loughlin 1997; Costa *et al.* 2004), as well as for benthically foraging seabirds such as penguins and cormorants (Croxall *et al.* 1991; Bost, Puetz & Lage 1994; Gremillet *et al.* 1998; Tremblay & Cherel 2000). Although development of oxygen stores have not been well studied in these species, ontogeny may constrain diving behaviour and limit potential foraging habitat, making young divers more susceptible to resource limitation.

Successful juvenile recruitment is critical for a long-lived species with a low reproductive rate, small total population and small breeding colony size (Environmental Protection and Biodiversity Conservation Act 1999). The recent listing of Australian sea lions as threatened was in response to poor population performance across their range, in stark contrast to the rapidly recovering sympatric New Zealand fur seal

A. forsteri (Gales, Haberley & Collins 2000). Lack of physiological reserves for juvenile Australian sea lions may be a major limitation on population potential. Our results suggest slow development of oxygen stores limits diving ability in juvenile Australian sea lions, and similar to adults, juveniles are working at or near their physiological capacity.

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References

Andrews, R.D., Jones, D.R., Williams, J.D., Crocker, D.E., Costa, D.P. & Le Boeuf, B.J. (1994) Thermoregulation and metabolism in freely diving northern elephant seals. *FASEB* **8**, A2.

Arnould, J.P.Y. & Boyd, I.L. (1996) The relationship between foraging behaviour and energy expenditure in Antarctic fur seals. *Journal of Zoology London* **239**, 769–782.

Arnould, J P.Y., Luque, S.P., Guinet, C., Costa, D.P., Kingston, J. & Shaffer, S.A. (2003) The comparative energetics and growth strategies of sympatric Antarctic and subantarctic fur seal pups at Îles Crozet. *Journal of Experimental Biology* **206**, 4497–4506.

Bevan, R.M., Butler, P.J., Woakes, A.J. & Boyd, I.L. (2002) The energetics of gentoo penguins, *Pygoscelis papua*, during the breeding season. *Functional Ecology* **16**, 175–190.

Bost, C.A., Puetz, K. & Lage J. (1994) Maximum diving depth and diving patterns of the gentoo penguin *Pygoscelis papua* at the Crozet Islands. *Marine Ornithology* **22**, 237–244.

Boyd, I.L., Arnould, J.P.Y., Barton, T. & Croxall, J.P. (1994) Foraging behaviour of Antarctic fur seals during periods of contrasting prey abundance. *Journal of Animal Ecology* **63**, 703–713.

Boyd, I.L. & Croxall, J.P. (1996) Dive durations in pinnipeds and seabirds. *Canadian Journal of Zoology* **74**, 1696–1705.

Brody, S. (1945) *Bioenergetics and Growth with Special Reference to the Efficiency Complex in Domestic Animals*. Hafner Press, New York.

Burns, J.M. (1999) The development of diving behaviour in juvenile Weddell seals: pushing physiological limits in order to survive. *Canadian Journal of Zoology* **77**, 737–747.

Burns, J.M. & Castellini, M.A. (1996) Physiological and behavioural determinants of the aerobic dive limit in Weddell seal (*Leptonychotes weddellii*) pups. *Journal of Comparative Physiology B* **166**, 473–483.

Burns, J.M., Costa, D.P., Frost, K. & Harvey, J.T. (2005) Development of body oxygen stores in harbour seals: effects of age, mass, and body composition. *Physiological and Biochemical Zoology* **78**, 1057–1068.

Butler, P.J. (2004) Metabolic regulation in diving birds and mammals. *Respiratory Physiology and Neurobiology* **141**, 297–315.

Butler, P.J. (2006) Aerobic dive limit. What is it and is it always used appropriately? *Comparative Biochemistry and Physiology, Part A* **145**, 1–6.

Butler, P.J., Bevan, R.M., Woakes, A.J., Croxall, J.P. & Boyd, I.L. (1995) The use of data loggers to determine the energetics and physiology of aquatic birds and mammals. *Brazilian Journal of Medical and Biological Research* **28**, 1307–1317.

Butler, P.J. & Jones, D.R. (1997) The physiology of diving of birds and mammals. *Physiological Reviews* **77**, 837–899.

Butler, P.J., Woakes, A.J., Boyd, I.L. & Kanatous, S. (1992) Relationship between heart rate and oxygen consumption during steady-state swimming in California sea lions. *Journal of Experimental Biology* **170**, 35–42.

Castellini, M.A., Davis, R.W. & Kooyman, G.L. (1988) Blood chemistry regulation during repetitive diving in Weddell seals. *Physiological Zoology* **61**, 379–386.

Castellini, M.A., Murphy, B.J., Fedak, M.A., Ronald, K., Gofton, N & Hochachka, P.W. (1985) Potentially conflicting metabolic demands of diving and exercise in seals. *Journal of Applied Physiology* **58**, 392–399.

Castellini, M.A. & Somero, G.N. (1981) Buffering capacity of vertebrate muscle: correlations with potentials for anaerobic function. *Journal of Comparative Physiology B* **143**, 191–198.

Chilvers, B.L., Wilkinson, I.S., Duignan, P.J. & Gemmell, N.J. (2006) Diving to extremes: are New Zealand sea lions (*Phocarctos hookeri*) pushing their limits in a marginal habitat? *Journal of Zoology* **269**, 233–240.

Costa, D.P. (1991) Reproductive and foraging energetics of pinnipeds: implications for life history patterns. *Behaviour of Pinnipeds* (ed. D. Renouf), pp. 300–344. Chapman & Hall, London.

Costa, D.P. (1993) The secret life of marine mammals: novel tools for studying their behaviour and biology at sea. *Oceanography* **6**(3), 120–128.

Costa, D.P., Antonelis, G.P. & DeLong, R.L. (1990) Effects of El Niño on the foraging energetics of the California sea lion. *Effects of El Niño on Pinnipeds* (eds F. Trillmich & K. Ono), pp. 156–165. Springer-Verlag, Berlin.

Costa, D.P. & Gales, N.J. (2003) Energetics of a benthic diver: seasonal foraging ecology of the Australian sea lion, *Neophoca cinerea*. *Ecological Monographs* **73**(1), 27–43.

Costa, D.P., Gales, N.J. & Crocker, D.E. (1998) Blood volume and diving ability of the New Zealand Sea Lion, *Phocarctos hookeri*. *Physiological Zoology* **71**(2), 208–213.

Costa, D.P., Gales, N.J. & Goebel, M.E. (2001) Review: aerobic dive limit: how often does it occur in nature? *Comparative Biochemistry and Physiology, Part A* **129**, 771–783.

Costa, D.P., Kuhn, C.E., Weise, M.J., Shaffer, S.A. & Arnould, J.P.Y. (2004) When does physiology limit the foraging behaviour of freely diving mammals? *International Congress Series* **1275**, 359–366.

Costa, D.P. & Sinervo, B. (2004) Field Physiology: physiological insights from animals in nature. *Annual Review Physiology* **66**, 209–238.

Crocker, D.E., Gales, N.J. & Costa, D.P. (2000) Swimming speed and foraging energetics of New Zealand sea lions, *Phocarctos hookeri*. *Journal of Zoology London* **254**, 267–277.

Crocker, D.E., LeBoeuf, B.J. & Costa, D.P. (1997) Drift diving in female northern elephant seals: implications for food processing. *Canadian Journal of Zoology* **75**, 27–39.

Croxall, J.P., Naito, Y., Kato, A., Rothery, P. & Briggs D.R. (1991) Diving patterns and performance in the Antarctic blue-eyed shag *Phalacrocorax atriceps*. *Journal of Zoology London* **225**, 177–200.

Davis, B.J. (1991) Developmental changes in the blood oxygen transport system of Kemp–Ridley sea turtles, *Lepidochelys kempi*. *Canadian Journal of Zoology* **69**(10), 2660–2666.

Davis, R.W., Castellini, M.A., Williams, T.M. & Kooyman, G.L. (1991) Fuel homeostasis in the harbour seal during submerged swimming. *Journal of Comparative Physiology B* **160**, 627–635.

Davis, R.W. & Kanatous, S.B. (1999) Convective oxygen transport and tissue oxygen consumption in Weddell seals during aerobic dives. *Journal of Experimental Biology* **202**, 1091–1113.

Dolphin, W.F. (1988) Foraging dive patterns of humpback whales, *Megaptera novaeangliae*, in southeast Alaska: a cost-benefit analysis. *Canadian Journal of Zoology* **66**, 2432–2441.

El-Sayed H., Goodall, S.R. & Hainsworth, R. (1995) Re-evaluation of the Evans blue dye dilution method of plasma volume measurement. *Clinical Laboratory Haematology* **17**, 189–194.

Fedak, M.A. & Thompson, D. (1993) Behavioural and physiological options in diving seals. *Symposium Zoological Society London* **66**, 333–348.

Feldkamp, S.D. (1987) Swimming in the California sea lion: morphometrics, drag, and energetics. *Journal of Experimental Biology* **131**, 117–135.

Feldkamp, S.D., Long, R.L. & Antonelis, G.A. (1989) Diving patterns of California sea lions, *Zalophus californianus*. *Canadian Journal of Zoology* **67**, 872–883.

Foldager, N. & Blomqvist, C.G. (1991) Repeated plasma volume determination with the Evans blue dye dilution technique: the method and a computer program. *Computational Biological Medicine* **21**(1/2), 35–41.

Fowler, S.L. (2005) *Ontogeny of diving in the Australian sea lion. PhD thesis*, University of California, Santa Cruz.

Fowler, S.L., Costa, D.P., Arnould, J.P.Y., Gales, N.J. & Kuhn, C.E. (2006) Ontogeny of diving behaviour in the Australian sea lion: trials of adolescence in a late bloomer. *Journal of Animal Ecology* **72**(2), 358–367.

Gales, N.J., Haberley, B. & Collins, P. (2000) Changes in the abundance of New Zealand fur seals, *Arctocephalus forsteri*, in Western Australia. *Wildlife Research* **26**, 165–168.

Gales, N.J. & Mattlin, R.H. (1998) Fast, safe, field-portable gas anaesthesia for otariids. *Marine Mammal Science* **14**, 355–361.

Gales, N.J., Shaughnessy, P.D. & Dennis, T.E. (1994) Distribution, abundance, and breeding cycle of the Australian sea lion, *Neophoca cinerea* (Mammalia: Pinnipedia). *Journal of Zoology* **234**, 353–370.

Gentry, R.L., Kooyman, G.L. & Goebel, M.E. (1986) Feeding and diving behaviour of northern fur seals. *Fur Seals: Maternal Strategies on Land and at Sea* (eds R.L. Gentry & G.L. Kooyman), pp. 61–78. Princeton University Press, Princeton.

Greaves, D.K., Schreer, J.R., Hammill, M.O. & Burns, J.M. (2005) Diving heart rate development in postnatal harbour seals, *Phoca vitulina*. *Physiological and Biochemical Zoology* **78**(1), 9–17.

Gremillet, D., Argentin, G., Schulte, B. & Culik, B.M. (1998) Flexible foraging techniques in breeding cormorants *Phalacrocorax carbo* and shags *Phalacrocorax aristotelis*: benthic or pelagic feeding. *Ibis* **140**, 113–119.

Handrich, Y., Bevan, R.M., Charrassin, J.B., Butler, P.J., Pütz, K., Woakes, A.J., Lage, J. & LeMaho, Y. (1997) Hypothermia in foraging king penguins. *Nature* **388**, 64–67.

Hastie, G.D., Rosen, D.A.S. & Trites, A.W. (2006) The influence of depth on a breath-hold diver: predicting the diving metabolism of Steller sea lions (*Eumetopias jubatus*). *Journal of Experimental Marine Biology and Ecology* **336**, 163–170.

Higgins, L.V. (1993) The nonannual, nonseasonal breeding cycle of the Australian sea lion, *Neophoca cinerea*. *Journal of Mammalogy* **74**(2), 270–274.

Hindell, M.A., Slip, D.J. & Burton, H.R. (1991) The diving behaviour of adult male and female southern elephant seals *Mirounga leonina* (Pinnipedia, Phocidae). *Australian Journal of Zoology* **39**(5), 595–619.

Horning, M. & Trillmich, F. (1997) Development of haemoglobin, hematocrit and erythrocyte values in Galapagos fur seals. *Marine Mammal Science* **13**(1), 100–113.

Hurley, J.A. & Costa, D.P. (2001) Standard metabolic rate at the surface and during trained submersions in adult California sea lions (*Zalophus californianus*). *Journal of Experimental Biology* **204**, 3273–3281.

ICSH (1967) International Committee for Standardization in Haematology. *Journal of Clinical Pathology* **31**, 139.

Irving, L., Scholander, P.F. & Grinnell, S.W. (1941) The respiration of the porpoise (*Tursiops truncatus*). *Journal of Cellular and Comparative Physiology* **17**, 145–168.

Iverson, S.J., Frost, K. & Lang, S.L.C. (2002) Fat content and fatty acid composition of forage fish and invertebrates in Prince William Sound, Alaska: factors contributing to among and within species variability. *Marine Ecology Progress Series* **241**, 161–181.

Jørgensen, C., Lydersen, C., Brix, O. & Kovacs, K.M. (2001) Diving development in nursing harbour seal pups. *Journal of Experimental Biology* **204**, 3993–4004.

Keijer, E. & Butler, P.J. (1982) Volumes of the respiratory and circulatory systems in tufted and mallard ducks. *Journal of Experimental Biology* **101**, 213–220.

Kleiber, M. (1975) *The Fire of Life*. Krieger Publishing, New York.

Kooyman, G.L. (1989) Diverse divers: physiology and behaviour. *Zoophysiology Series, vol. 23* (eds K. Johansen & D.S. Farner). Springer-Verlag, Berlin.

Kooyman, G.L. (2006) Mysteries of adaptation to hypoxia and pressure in marine mammals. *Marine Mammal Science* **22**(3), 507–526.

Kooyman, G.L., Castellini M.A., Davis R.W. & Maue R.A. (1983) Aerobic diving limits of immature Weddell seals. *Journal of Comparative Physiology* **151**, 171–174.

Kooyman, G.L., Kerum, D.H., Campbell, W.B. & Wright, J.J. (1971) Pulmonary function in freely diving Weddell seals (*Leptonychotes weddellii*). *Respiratory Physiology* **12**, 271–282.

Kooyman, G.L. & Kooyman, T.G. (1995) Diving behaviour of emperor penguins nurturing chicks at Coulman Island, Antarctica. *Condor* **97**, 536–549.

Kooyman, G.L. & Ponganis, P.J. (1998) The physiological basis of diving to depth: birds and mammals. *Annual Review Physiology* **60**, 19–32.

Kooyman, G.L., Wahrenbrock, E.A., Castellini, M.A, Davis, R.W. & Sinnet, E.E. (1980) Aerobic and anaerobic metabolism during voluntary diving in Weddell seals: evidence of preferred pathways from blood chemistry and behaviour. *Journal of Comparative Physiology B* **138**, 335–346.

Kretzmann, M.B., Costa, D.P. & LeBoeuf, B.J. (1993) Maternal energy investment in elephant seal pups: evidence for sexuality? *American Naturalist* **141**, 466–480.

Kuhn, C.E., Aurioles-Gamboa, D., Weise, M.J. & Costa, D.P. (2006) Oxygen stores of California sea lion pups: implications for diving ability. *Proceedings of the 22nd Wakefield*

Fisheries Symposium, Sea Lions of the World: Conservation and research in the 21st century (eds A.W. Trites, S.K. Atkinson, D.P. DeMaster, L.W. Fritz, T.S. Gelatt, L.D. Rea & K.M. Wynne), pp. 31–44. Alaska Sea Grant Program, Alaska.

Le Boeuf, B.J., Naito, Y., Huntley, A.C. & Asaga, T. (1989) Prolonged, continuous, deep diving by northern elephant seals. *Canadian Journal of Zoology* **67**, 2514–2519.

Lenfant, C., Johansen K. & Torrance J.D. (1970) Gas transport and oxygen storage capacity in some pinnipeds and the sea otter. *Respiratory Physiology* **9**, 277–286.

Lifson, N. & McClintock, R. (1966) Theory of use of the turnover rates of body water for measuring energy and material balance. *Journal of Theoretical Biology* **12**, 46–74.

Lydersen, C. & Hammill, M.O. (1993) Activity, milk intake and energy consumption in free-living ringed seal (*Phoca hispida*) pups. *Journal of Comparative Physiology B* **163**, 433–438.

Lydersen, C. & Kovacs, K.M. (1996) Energetics of lactation in harp seals (*Phoca groenlandica*) from the Gulf of St. Lawrence, Canada. *Journal of Comparative Physiology B* **116**, 295–304.

Lydersen, C., Kovacs, K.M., Hammill, M.O. & Gjertz, I. (1996) Energy intake and utilization by nursing bearded seal (*Erignathus barbatus*) pups from Svalbard, Norway. *Journal of Comparative Physiology B* **166**, 405–411.

Matthews, R. (1977) *Pulmonary mechanics of California sea lions, Zalophus californianus. MS thesis*, San Diego State University.

Merrick, R.L. & Loughlin, T.R. (1997) Foraging behaviour of adult female and young-of-the-year Steller sea lions in Alaskan waters. *Canadian Journal of Zoology* **75**, 776–786.

Nagy, K.A. & Costa, D.P. (1980) Water flux in animals: analysis of potential errors in the tritiated water method. *American Journal of Physiology* **238**, R454–R465.

Noren, S.R., Iverson, S.J. & Boness, D.J. (2005) Development of the blood and muscle oxygen stores in grey seals (*Halichoerus grypus*): implications for juvenile diving capacity and the necessity of a terrestrial postweaning fast. *Physiological and Biochemical Zoology* **78**(4), 482–490.

Noren, S.R., Williams, T.M., Pabst, D.A., McLellan, W.A. & Dearolf, J.L. (2001) The development of diving in marine endotherms: preparing the skeletal muscles of dolphins, penguins, and seals for activity during submergence. *Journal of Comparative Physiology B* **171**, 127–134.

Oftedal, O.T., Iverson, S.J. & Boness, D.J. (1987) Milk and energy intakes of suckling California sea lion (*Zalophus californianus*) pups in relation to sex, growth, and predicted maintenance requirements. *Physiological Zoology* **60**, 560–575.

Olsen, C., Hale, F. & Elsner, R. (1969) Mechanics of ventilation in the pilot whale. *Respiratory Physiology* **7**, 137–149.

Page, B., McKenzie, J., Hindell, M.A. & Goldsworthy, S.D. (2005) Drift dives by male New Zealand fur seals (*Arctocephalus forsteri*). *Canadian Journal of Zoology* **83**, 293–300.

Ponganis, P.J. & Kooyman, G.L. (2000) Diving physiology of birds: a history of studies on polar species. *Comparative Biochemistry and Physiology A* **126**, 143–151.

Ponganis, P.J., Kooyman, G.L. & Castellini, M.A. (1993) Determinants of the aerobic dive limit of Weddell seals: Analysis of diving metabolic rates, postdive end tidal *PO_2's* and blood and muscle oxygen stores. *Physiological Zoology* **66**, 732–749.

Ponganis, P.J., Kooyman, G.L., Baranov, E.A., Thorson, P.H. & Stewart, B.S. (1997a) The aerobic submersion limit of Baikal seals, *Phoca sibirica*. *Canadian Journal of Zoology* **75**, 1323–1327.

Ponganis, P.J., Kooyman, G.L., Starke, L.N., Kooyman, C.A. & Kooyman, T.G. (1997b) Post-dive blood lactate concentration in emperor penguins, *Aptenodytes forsteri*. *Journal of Experimental Biology* **200**, 1623–1626.

Ponganis, P.J., Kooyman, G.L., Sartoris, D. & Jobsis, P. (1992) Pinniped splenic volumes. *American Journal of Physiology* 262, R322-R325.

Ponganis, P.J., Kooyman, G.L. & Winter, L.M. (1997c) Heart rate and plasma lactate responses during submerged swimming and trained diving in California seal lions *Zalophus californianus*. *Journal of Comparative Physiology* **167**, 9–16.

Ponganis, P.J., Starke, L.N., Horning, M. & Kooyman, G.L. (1999) Development of diving capacity in emperor penguins. *Journal of Experimental Biology* **202**, 781–786.

Rea, L.D. & Costa, D.P. (1992) Changes in standard metabolism during long-term fasting in northern elephant seal pups (*Mirounga angustirostris*). *Physiological Zoology* **65**, 97–111.

Reilly, J.J. (1991) Adaptation to prolonged fasting in free-living weaned grey seal pups. *American Journal of Physiology* **260**, R267–R272.

Reynafarje, B. (1963) Simplified method for the determination of myoglobin. *Journal of Laboratory Clinical Medicine* **61**, 138–145.

Richmond, J.P., Burns, J.M. & Rea, L.D. (2006) Ontogeny of total body oxygen stores and aerobic dive potential in Steller sea lions (*Eumetopias jubatus*). *Journal of Comparative Physiology B* **176**, 535–545.

Ridgway, S.H. & Johnston, D.G. (1966) Blood oxygen and ecology of porpoises of three genera. *Science* **151**, 456–458.

Sato, K., Naito, Y., Kato, A., Niizuma, Y., Watanuki, Y., Charrassin, J.B., Bost, C.A., Handrich, Y. & Le Maho, Y. (2002) Buoyancy and maximal diving depth in penguins: do they control inhaling air volume? *Journal of Experimental Biology* **205**, 1189–1197.

Schmidt-Nielsen, K. (1984) *Scaling: Why is Body Size So Important?* Cambridge University Press, Cambridge.

Schmidt-Nielsen, K. (1997) *Animal Physiology: Adaptation and Environment*. Cambridge University Press, Cambridge.

Shaffer, S.A., Costa, D.P., Williams, T.M. & Ridgway, S.H. (1997) Diving and swimming performance of white whales, *Delphinapterus leucas*: an assessment of plasma lactate, blood gas levels, and respiratory rates. *Journal of Experimental Biology* **200**, 3091–3099.

Skrovan, R.C., Williams, T.M., Berry, P.S., Moore, P.W. & Davis, R.W. (1999) The diving physiology of bottlenose dolphins (*Tursiops truncatus*) II. Biomechanics and changes in buoyancy at depth. *Journal of Experimental Biology* **202**, 2749–2761.

Sparling, C.E. & Fedak, M.A. (2004) Metabolic rates of captive grey seals during voluntary diving. *Journal of Experimental Biology* **207**(10), 1615–1624.

Speakman, J.R. (1997) *Doubly-labelled Water: Theory and Practice*. Chapman & Hall, London.

Thompson, D. & Fedak, M.A. (1993) Cardiac responses of grey seals during diving at sea. *Journal of Experimental Biology* **174**, 139–164.

Thorson, P.H. & LeBoeuf, B.J. (1994) Developmental aspects of diving in northern elephant seal pups. *Elephant Seals: Population Ecology, Behaviour, and Physiology* (eds B.J. LeBoeuf & R.M. Laws), pp. 271–289. University of California Press, Berkeley.

Tremblay, Y. & Cherel, Y. (2000) Benthic and pelagic dives: a new foraging behaviour in rockhopper penguins. *Marine Ecology Progress Series* **204**, 257–267.

Trillmich, F. (1986) Maternal investment and sex-allocation in the Galápagos fur seal, *Arctocephalus galapagoensis*. *Behavioural Ecology and Sociobiology* **19**, 157–164.

Weber, R.E., Hemmingsen, E.A. & Johansen, K. (1974) Functional and biochemical studies of penguin myoglobin. *Comparative Biochemistry and Physiology B* **49**, 197–214.

Werner, R. & Campagna, C. (1995) Diving behaviour of lactating southern sea lions (*Otaria flavescens*) in Patagonia. *Canadian Journal of Zoology* **73**, 1975–1982.

Wikelski, M. & Cooke, S.J. (2006) Conservation physiology. *TREE* **21**, 38–46.

Williams, T.M., Davis, R.W., Fuiman, L.A., Francis, J., LeBoeuf, B.J., Horning, M., Calambokidus, J. & Croll, D.A. (2000) Sink or swim: strategies for cost-efficient diving by marine mammals. *Science* **288**, 133–136.

Williams, T.M., Friedl, W.A. & Haun, J.E. (1993) The physiology of bottlenose dolphins (*Tursiops truncatus*): heart rate, metabolic rate and plasma lactate during exercise. *Journal of Experimental Biology* **179**, 31–46.

Winship, A.J., Trites, A.W. & Rosen, D.A.S. (2002) A bioenergetic model for estimating the food requirements of Steller sea lions *Eumetopias jubatus* in Alaska, USA. *Marine Ecology Progress Series* **229**, 291–312.

Zapol, W.M., Liggins, G.C., Schneider, R.C., Qvist, J., Snider, M.T., Creasy, R.K. & Hochachka, P.W. (1989) Regional blood flow during simulated diving in the conscious Weddell seal. *Journal of Applied Physiology* **47**, 968–973.

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Appendix

Summary of FMR data for Australian sea lions (mean ± SE). Equations from Nagy & Costa (1980) were used to calculate total body water (TBW) determined by ^{18}O and water influx. We used the two-pool model presented in Speakman (1997) to calculate CO_2 production

Animal ID	Age (months)	Initial mass (kg)	Final mass (kg)	TBW ^{18}O (L)	Interval (days)	Time at-sea (%)	Water influx (mL kg^{-1} d^{-1})	CO_2 (mL g^{-1} h^{-1})
AD	22·1	58·8	58·2	33·8	5·06	25·8	41·8	0·596
ED	22·4	43·4	44·4	26·0	7·76	28·9	49·2	0·669
EL	22·8	41·2	40·4	24·2	6·70	28·5	45·8	0·845
FI	22·9	62·0	57·5	33·3	5·88	22·9	54·7	0·708
GO	22·2	46·2	45·4	22·1	5·90	12·9	33·9	0·506
LE	22·6	49·8	50·4	30·4	6·79	31·9	100·0	0·902
WI	22·8	41·4	41·6	22·8	6·72	17·0	35·6	0·496
MA	22·9	41·0	41·2	24·1	5·96	24·4	44·1	0·908
Juvenile mean	22·6 ± 0·1	48·3 ± 2·6	47·8 ± 2·3	27·6 ± 1·6	6·35 ± 0·29	26·1 ± 2·8	50·6 ± 7·5	0·704 ± 0·059
SC	3 years	65·0	70·4	42·6	6·22	46·8	89·7	0·832