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ORIGINAL PAPER

Changes in partial pressures of respiratory gases during submerged voluntary breath hold across odontocetes: is body mass important?

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Abstract Odontocetes have an exceptional range in body mass spanning 10^3 kg across species. Because, size influences oxygen utilization and carbon dioxide production rates in mammals, this lineage likely displays an extraordinary variation in oxygen store management compared to other marine mammal groups. To examine this, we measured changes in the partial pressures of respiratory gases (P_{O_2}, P_{CO_2}), pH, and lactate in the blood during voluntary, quiescent, submerged breath holds in Pacific white-sided dolphins (*Lagenorhynchus obliquidens*), bottlenose dolphins (*Tursiops truncatus*), and a killer whale (*Orcinus orca*) representing a mass range of 96–3,850 kg. These measurements provided an empirical determination of the effect of body size on the variability in blood biochemistry during breath hold and experimentally determined aerobic dive limits (ADL) within one taxonomic group (odontocetes). For the species in this study, maximum voluntary breath-hold duration was positively correlated with body mass, ranging from 3.5 min in white-sided dolphins to 13.3 min for the killer whale. Variation in breath-hold duration was associated with differences in the rate of change for P_{O_2} throughout breath hold; P_{O_2} decreased twice as fast for the two smaller species (-0.6 mmHg O_2 min^{-1}) compared to the largest species (-0.3 mmHg O_2 min^{-1}). In contrast, the rate of increase in P_{CO_2} during breath hold was similar across species. These results demonstrate that large body size in odontocetes facilitates increased aerobic breath-hold capacity as mediated by decreased mass-specific metabolic rates (rates of change in P_{O_2} served as a proxy for oxygen utilization). Indeed the experimentally determined 5 min ADL for bottlenose dolphins was surpassed by the 13.3 min maximum breath hold of the killer whale, which did not end in a rise in lactate. Rather, breath hold ended voluntarily as respiratory gases and pH fell within a narrow range for both large and small species, likely providing cues for ventilation.

Keywords Aerobic dive limit · Blood gas · Blood pH · Plasma lactate · Odontocete · Cetacean

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Introduction

Across mammalian groups, odontocetes (toothed whales) demonstrate one of the broadest ranges in body mass from the 30 kg vaquita (*Phocoena sinus*) to the 70,000 kg sperm whale (*Physeter macrocephalus*; Evans 1987). As a result, the total metabolic rate theoretically increases by over 100-fold across this vertebrate group (calculated from Williams et al. 1993b, 2001) as metabolic rate scales positively and

predictably with mammalian body mass during rest (Kleiber 1975; Williams et al. 1999, 2001) and free-ranging activity (Nagy 1994). Mass-specific metabolic rates and hence the relative rates of oxygen utilization in tissues are the highest in the smallest marine mammal species (Williams et al. 2001). Because metabolic rate is a major determinant of aerobic dive duration, this scaling of metabolism has important implications for dive capacities (Kooyman 1989; Ponganis et al. 1993). Indeed it has been suggested that large body size provides an advantage for diving for aquatic animals in terms of both the absolute size of oxygen stores as well as the relative decrease in mass-specific metabolic rate (Kooyman et al. 1983; Hudson and Jones 1986; Costa 1991; Schreer and Kovacs, 1997; Noren and Williams 2000). Although this provides a theoretical framework to predict the diving capacity of marine mammals, little is known about the rates for oxygen depletion or carbon dioxide production during breath hold within one taxonomic group, especially for one as diverse in size as the odontocetes.

The rates of oxygen depletion and carbon dioxide production are central to understand the underlying mechanisms that dictate breath-hold capacities of marine mammals as well as their ability to endure prolonged apnea during the periods of foraging and locomotion. Yet the difficult logistics of studying free-diving odontocetes precludes these types of measurements in free-diving dolphins and whales. Although aerobic dive limits have been estimated for many diving vertebrates using the calculated aerobic dive limit (cADL; ratio of total body oxygen stores and the rate at which these stores are utilized; Kooyman 1989; Castellini et al. 1992; Boyd and Croxall 1996; Costa et al. 1998, 2004; Noren et al. 2002, 2004, 2005; Williams et al. 2011), experimentally derived aerobic dive limits have only been determined in five marine mammal species. These include three pinnipeds, the Weddell seal (*Leptonychotes weddellii*; Kooyman et al. 1980, 1983; Ponganis et al. 1993), the Baikal seal (*Phoca sibirica*; Ponganis et al. 1997a), and the California sea lion (*Zalophus californianus*; Ponganis et al. 1997b), and two moderately sized cetaceans, the beluga whale (*Delphinapterus leucas*; Shaffer et al. 1997) and the bottlenose dolphin (*Tursiops truncatus*; Williams et al. 1999). This limited data set has precluded the examination of the effect of body size on ADL within a single marine mammal group. In addition, with the knowledge of the changes in the blood for respiratory gases and pH levels, we might better understand the utilization of oxygen stores and the cues for ventilation.

In this study, we examined changes in blood biochemistry during voluntary breath hold in three odontocete species (Pacific white-sided dolphin, bottlenose dolphin, and killer whale). These species represented a 40-fold range of body mass, which enabled us to investigate the effect of body mass on the rate of change in the partial pressure of oxygen in the blood (our proxy for oxygen depletion rates) and the resulting experimentally derived aerobic breath-hold capacity within one marine mammal group, odontocetes. Sequential blood samples were taken throughout voluntary breath hold as the animal rested ventral side up on the water surface so that their blowhole was submerged. The samples were evaluated for blood gases (P_{O_2}, P_{CO_2}), pH, and lactate levels following the methods of Shaffer et al. (1997), Williams et al. (1999) so that direct comparisons could be made to these prior studies on odontocetes. Using this technique, we were able to quantify the physiological effect of body size on breath-hold capabilities without the confounding influence of exercise, because as exercise intensity increases both oxygen consumption and lactate production increase in dolphins (Williams et al. 1992, 1993a). Both of these factors would influence breath-hold duration and the experimentally determined ADL. By analyzing sedentary animals, we were able to standardize the level of activity across animals and provide an analysis of blood gas, pH, and lactate changes during the periods of minimum locomotory muscle metabolism. This should maximize the potential duration of aerobic metabolism in muscle, and contribute to maximum ADL durations.

Materials and methods

Animals

The study included two adult female Pacific white-sided dolphins (*Lagenorhynchus obliquidens*, mass: 96.0 ± 4.2 kg), three adult female bottlenose dolphins (*Tursiops truncatus*, mass: 189 ± 14.1 kg), and one adult male killer whale (*Orcinus orca*, mass 3,850 kg, estimated from girth and length measurements). The white-sided dolphins were housed in an indoor pool (56 × 26 × 9 m deep; John G. Shedd Aquarium, Chicago, IL, USA) with an average T_{water} = 13.0°C. Bottlenose dolphins were maintained in outdoor floating pens (7 × 7 × 3 m deep; Naval Ocean Systems Center, Oahu, HI, USA) at T_{water} = 24.0°C. The killer whale was housed in a large, outdoor pool (46 × 23 × 8 m deep; Oregon Coast Aquarium, Newport, OR, USA) at T_{water} = 10.0°C. All animals that had been held at their respective facility for several years were acclimated to local environmental temperatures, and were fed a mixed fish diet (capelin, herring, squid, mackerel, and smelt) supplemented with vitamins.

Breath-hold capacity and blood sampling

Several months prior to the experimental period, the animals were trained using standard operant conditioning to hold their breath while sedentarily floating ventral side up

on the water surface. This voluntary breath-hold technique was used so that the results could be compared to the previous studies that have used the same methodologies on other species of odontocete (Shaffer et al. 1997; Williams et al. 1999). Once in this position a 19-gauge needle connected to a butterfly catheter was inserted into the arterial plexus on the ventral aspect of the fluke. We cannot validate whether the needle was inserted into a vein or artery, thus we must assume that our blood sampling represents a mixture of venous and arterial blood as in Shaffer et al. (1997), Williams et al. (1999). Nonetheless, a study of blood biochemistry in sedentary, breath-holding Northern elephant seals (*Mirounga angustirostris*) demonstrated that venous P_{O_2} values were indistinguishable from arterial values after the first minute of apnea (Stockard et al. 2007).

Sequential anaerobic blood samples (3–5 ml) were drawn while the animals remained in breath hold. The sample was drawn through a stopcock system so that any air in the system was flushed out before switching the open port on the stopcock to the gas proof plastic syringe for the sample collection. This ensured that the blood samples were drawn anaerobically. Sampling intervals were <30 s for the white-sided dolphins, alternating between anaerobic draws for the blood gas determination and plasma lactate analysis. The sampling intervals were increased to 60 s for the bottlenose dolphins and killer whale. Each animal voluntarily ended the breath-hold session by righting itself and taking a breath, and the entire breath-hold duration (inspiration to inspiration) was timed with a stopwatch. One breath-hold session was conducted every other day over a 2-week period for the white-sided dolphins, once per week over a 6-week period for the bottlenose dolphins, and once a day over 4 days for the killer whale.

Whole blood samples from the white-sided dolphins and killer whale were analyzed immediately for the partial pressure of oxygen (P_{O_2}), partial pressure of carbon dioxide (P_{CO_2}), and pH with a hand-held gas analyzer at 37°C (i-STAT thermal control model, Signal Devices, Inc., Waukesha, WI, USA). Blood samples from the bottlenose dolphins were placed on ice and analyzed within several minutes of collection with a desktop gas analyzer at 37°C (158 pH/blood gas analyzer, Ciba-Corning, Medfield, MA, USA) according to Williams et al. (1999). The temperature of the blood gas analyzers (37°C) mimics the body temperature of dolphins (Noren unpubl. data). Both instruments were calibrated against the standards that spanned expected ranges for P_{O_2} and P_{CO_2}. In addition, an aliquot of blood from a sample that was taken from a bottlenose dolphin was analyzed in both the desktop and hand-held analyzers; the values provided by each machine were identical.

For all trials, blood samples for lactate analyses were placed on ice until centrifuged (10 min at 1,000*g*, 22°C). Plasma was collected, placed in pre-frozen tubes, and maintained at −80°C until the concentration of lactate was measured with a desktop analyzer. An Analox GM7 Lactate Analyzer with GMRD-090 kit (Analox Instruments, Inc., Lunenburg, MA, USA) was used to analyze plasma samples from the white-sided dolphins and killer whale. A YSI Industrial Lactate Analyzer model 27 (Yellow Springs Instrument Co., Yellow Springs, OH, USA) was used for the bottlenose dolphin samples. Both lactate analyzers were calibrated daily with the standards ranging from zero through the span of expected lactate concentrations.

Data analyses

Although data were collected from several animals within a species (with the exception of the killer whale), the purpose of this study was not to examine individual variation within a species, but rather to profile changes in blood chemistry during breath hold within a species and to compare these relationships across species. Since the inclusion of individual variation can only serve to weaken any resulting relationships, this analytical approach reinforces the robustness of the conclusions from the pooled data. This approach is similar to other studies examining blood biochemistry changes during breath hold (i.e. Kooyman et al. 1980; Shaffer et al. 1997; Stockard et al. 2007), where data for multiple breath holds and several individuals within a species were pooled to provide sufficient power to profile a given parameter over the entire breath-hold duration. Once data were pooled, species-specific relationships for time into breath hold versus P_{O_2}, P_{CO_2}, and pH were determined. Changes in blood gases were best described by non-linear regressions, while changes in pH were described by linear regressions as determined by least squares methods. The least squares method was also used to determine if lactate levels increased during breath hold. Significance of the regressions was determined by *F* tests. Differences in the slopes of the regressions across species were determined by one way analysis of variance in combination with Tukey all pairwise comparisons procedure or student's *t* tests. All statistics were according to Sigma Stat (Systat Software, Richmond, CA, USA). All mean values are reported as ±1 SD.

Results

Breath-hold duration

Voluntary breath-hold duration for 96.0 ± 4.2 kg white-sided dolphins ranged from 2.4 to 3.5 min. In comparison, maximum breath-hold durations were nearly 2–4 times longer for the two other odontocete species examined.

Table 1 Ranges for respiratory blood gases (P_{O_2} and P_{CO_2}), pH, and lactate concentrations in the blood of submerged breath-holding bottlenose dolphins, Pacific white-sided dolphins, and a killer whale

Species	P_{O_2} (mmHg)	P_{CO_2} (mmHg)	pH (units)	Lactate ($mmol^{-1}$)
Pacific white-sided dolphin (*Lagenorhynchus obliquidens*)	30.0–59.0	61.0–70.3	7.31–7.37	0.80–3.60 (2.77 ± 0.70)
Bottlenose dolphin (*Tursiops truncatus*)	17.0–53.3	47.0–63.3	7.29–7.44	1.00–3.10
Killer whale (*Orcinus orca*)	31.0–85.0	46.4–75.2	7.21–7.37	1.00–2.35 (1.81 ± 0.33)

Means ± SD are reported in parenthesis only for the parameters that did not change throughout the breath hold

Durations ranged from 3.3 to 6.8 min for 189 ± 14.1 kg bottlenose dolphins and from 9.6 to 13.3 min for a 3,850 kg killer whale.

Blood gases and pH during breath hold

P_{O_2}, P_{CO_2}, and pH levels were quite variable throughout the duration of the breath hold for all three species (Table 1). As expected, P_{O_2} was the highest at the onset of breath hold (Fig. 1a) and corresponded to the lowest levels for P_{CO_2} (Fig. 1b). Meanwhile, pH levels in the blood were the highest at the onset of breath hold (Fig. 1c).

The greatest rate of change in blood gas occurred during the first 3 min of breath hold for all animals. Over the entire breath hold, P_{O_2} in the blood decreased non-linearly with time as described by:

$$P_{O_2} = 58.1 \text{ breath-hold duration}^{-0.6} \quad \left(r^2 = 0.77, F_{1,13} = 43.38, P < 0.0001\right) \quad (1)$$

for Pacific white-sided dolphins,

$$P_{O_2} = 58.8 \text{ breath-hold duration}^{-0.6} \quad \left(r^2 = 0.65, F_{1,19} = 35.56, P < 0.0001\right) \quad (2)$$

for bottlenose dolphins, and

$$P_{O_2} = 83.0 \text{ breath-hold duration}^{-0.3} \quad \left(r^2 = 0.39, F_{1,24} = 15.59, P = 0.0006\right) \quad (3)$$

for the killer whale, where P_{O_2} is in mmHg (1 mmHg = 0.1333 kPa) and breath-hold duration is in min (Fig. 1a). The rate of change in P_{O_2} in the blood during breath hold was significantly different between species ($F_{2,59} = 4.48$, $P = 0.02$). The largest animal had a significantly slower rate of change in P_{O_2} than the smaller animals (Pacific white-sided dolphin vs. killer whale: $P = 0.03$ and bottlenose vs. killer whale: $P = 0.02$). Meanwhile, the smallest animals had similar rates of change for P_{O_2} (Pacific white-sided dolphin vs. bottlenose: $P = 0.85$). The rate of change in P_{O_2} was nearly two times faster in white-sided and bottlenose dolphins compared to that of the larger killer whale (Fig. 1a).

While the partial pressure of oxygen declined, the partial pressure of carbon dioxide in the blood increased non-linearly during breath hold according to the relationships:

$$P_{CO_2} = 64.6 \text{ breath-hold duration}^{0.1} \quad \left(r^2 = 0.40, F_{1,14} = 9.46, P = 0.0082\right) \quad (4)$$

for white-sided dolphins,

$$P_{CO_2} = 51.1 \text{ breath-hold duration}^{0.1} \quad \left(r^2 = 0.49, F_{1,19} = 18.07, P = 0.0004\right) \quad (5)$$

for bottlenose dolphins, and

$$P_{CO_2} = 50.1 \text{ breath-hold duration}^{0.1} \quad \left(r^2 = 0.38, F_{1,25} = 15.29, P = 0.0006\right) \quad (6)$$

for the killer whale where P_{CO_2} is in mmHg (1 mmHg = 0.1333 kPa) and breath-hold duration is in min (Fig. 1b). In contrast to P_{O_2}, the rate of change in P_{CO_2} in the blood during breath hold was equivalent for the three species regardless of body size ($F_{2,61} = 0.45$, $P = 0.64$, Fig. 1b).

Blood pH decreased linearly during the course of the breath hold in all three species (Fig. 1c) according to:

$$\text{pH} = 7.347 - 0.008 \text{ breath-hold duration} \quad \left(r^2 = 0.12, F_{1,13} = 1.72, P = 0.21\right) \quad (7)$$

for white-sided dolphins,

$$\text{pH} = 7.364 - 0.008 \text{ breath-hold duration} \quad \left(r^2 = 0.11, F_{1,19} = 2.29, P = 0.15\right) \quad (8)$$

for bottlenose dolphins, and

$$\text{pH} = 7.316 - 0.007 \text{ breath-hold duration} \quad \left(r^2 = 0.34, F_{1,25} = 12.63, P = 0.002\right) \quad (9)$$

for the killer whale where pH is in standard pH units and breath-hold duration is in min. There were no differences across species in the change of pH over the breath hold ($F_{2,60} = 0.02$, $P = 0.98$, Fig. 1c).

Blood pH was also negatively correlated with P_{CO_2} for the three species of odontocete in this study (Fig. 2), with the range of the relationship dependent on the duration of voluntary breath hold. Least squares linear regressions are:

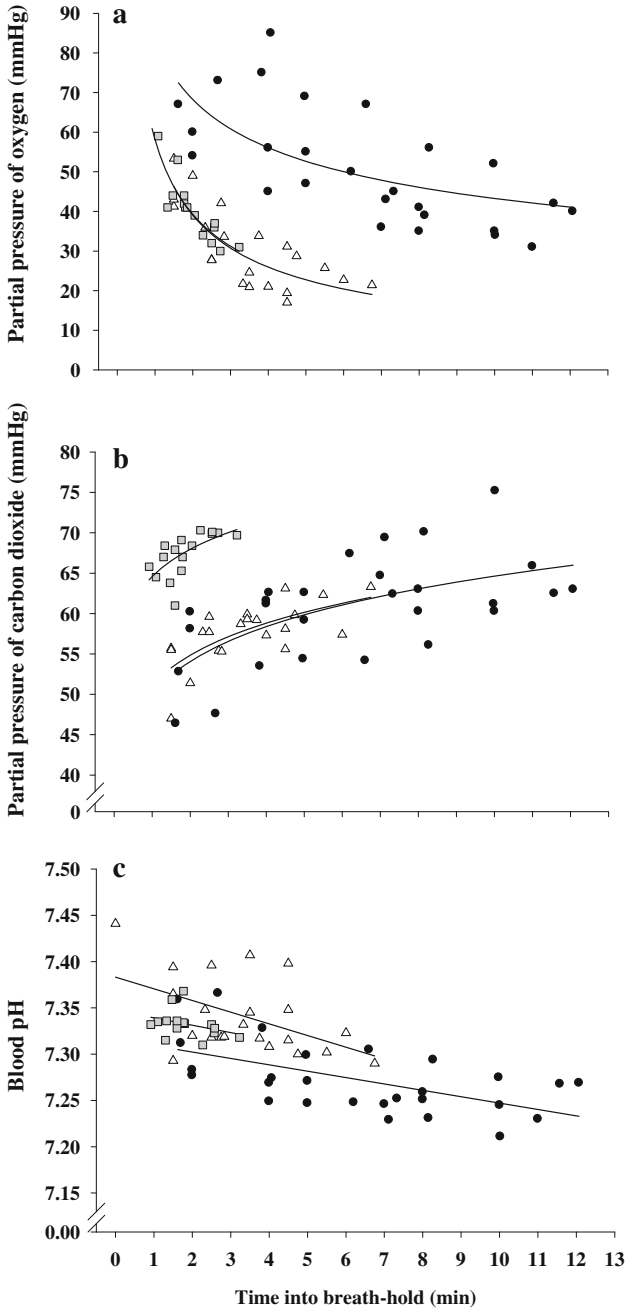

Fig. 1 Partial pressures of blood oxygen (**a**), carbon dioxide (**b**), and pH (**c**) in relation to time into breath hold for three species of odontocete. Each *data point* represents a single blood sample for Pacific white-sided dolphin (*closed squares*), bottlenose dolphin (*open triangles*), and killer whale (*closed circles*). The *lines* represent the non-linear regressions of the relationships for each species as described in the text. 1 mmHg = 0.1333 kPa

$$\text{pH} = 7.549 - 0.003P_{\text{CO2}} \quad (r^2 = 0.33,\ F_{1,12} = 5.93,\ P = 0.03) \qquad (10)$$

for white-sided dolphins,

$$\text{pH} = 7.556 - 0.004P_{\text{CO2}} \quad (r^2 = 0.15,\ F_{1,20} = 3.39,\ P = 0.08) \qquad (11)$$

for bottlenose dolphins, and

$$\text{pH} = 7.603 - 0.005P_{\text{CO2}} \quad (r^2 = 0.89,\ F_{1,25} = 193.45,\ P < 0.001) \qquad (12)$$

for the killer whale where pH is in the standard units and P_{CO_2} is in mmHg (1 mmHg = 0.1333 kPa). The rate of change for pH in response to P_{CO_2} in the blood was significantly different across species ($F_{2,60} = 13.70$, $P < 0.001$, Fig. 2) and appeared to scale with body size; the smallest species had the lowest rate while the largest species had the fastest rate. Indeed the slope of this relationship for white-sided dolphin was significantly lower than that of the bottlenose dolphin ($df = 34, t = -2.520, P = 0.017$) and killer whale ($df = 39$, $t = 2.047$, $P = 0.047$), and the bottlenose dolphin had a significantly lower rate of change than that of the killer whale ($df = 47, t = 4.894, P < 0.001$). Ultimately blood gases and pH for all three species fell within a narrow range at the voluntary termination of breath hold, regardless of body mass or duration of the breath hold, and are in agreement with levels measured at the termination of breath hold in other marine mammals (Table 2).

Plasma lactate during breath hold

For the three species examined, only the bottlenose dolphins showed marked changes in plasma lactate concentration during voluntary breath hold (Fig. 3). Plasma lactate

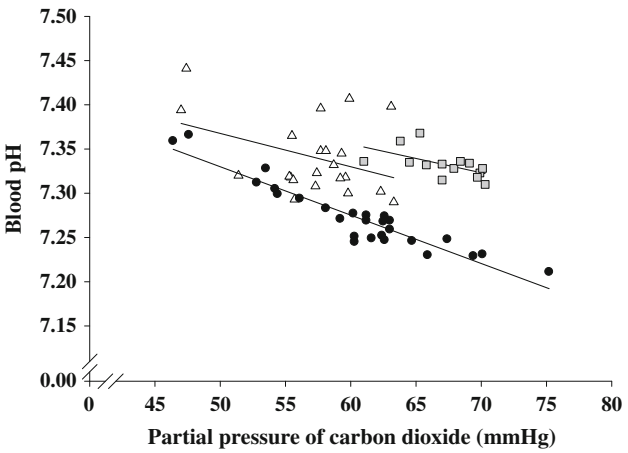

Fig. 2 Blood pH in relation to the partial pressure of carbon dioxide in the blood in three species of odontocete. Each *data point* represents a single blood sample for Pacific white-sided dolphin (*closed squares*), bottlenose dolphin (*open triangles*), and killer whale (*closed circles*). The *lines* represent the least squares linear regressions of the relationships for each species as described in the text. 1 mmHg = 0.1333 kPa

Table 2 Apnea duration and end of apnea blood biochemistry, including partial pressures of oxygen (P_{O_2}) and carbon dioxide (P_{CO_2}) and pH for voluntarily breath-holding marine mammals

Species	Apnea (min)	P_{O_2} (mmHg)	P_{CO_2} (mmHg)	pH (units)
Submerged sedentary breath hold				
Pacific white-sided dolphin[a] (*Lagenorhynchus obliquidens*)	2.4–3.5 (<ADL)	30–37	69.70–70.3	7.31–7.33
Bottlenose dolphin[a]	3.3–4.5 (<ADL)	17–21.7	57.3–63.1	7.31–7.40
(*Tursiops truncatus*)	6–6.8 (>ADL)	21.4–22.7	57.4–63.3	7.29–7.32
Killer whale[a] (*Orcinus orca*)	9.6–13.3 (<ADL)	31–40	63–75.2	7.21–7.27
Beluga whale[b] (*Delphinapterus leucas*)	17	20–23	83	7.17
Sedentary breath-hold				
Weddell seal[c] (*Leptonychotes weddellii*)	Max 4.5 and 8	25	55	7.32
Northern elephant seal[d] (*Mirounga angustirostris*)	3.1–10.9	15–31	55–72	7.26–7.31
Actively diving				
Weddell seal[e]	<17	24.5 ± 2.86	48.0 ± 4.22	7.37 ± 0.027
(*Leptonychotes weddellii*)	>17	19.9 ± 2.05	50.5 ± 1.17	7.34 ± 0.025
Northern elephant seal[f] (*Mirounga angustirostris*)	>10	Venous 2–10 arterial 12–23		

[a] This study where we report the levels at the end of each breath hold

[b] Shaffer et al. 1997 where only the extreme values across the two belugas were reported so levels for short apneustic periods are not represented

[c] Kooyman et al. 1980 where only the extreme values from each of the two seals were reported so levels for short apneustic periods are not represented

[d] Stockard et al. 2007 where only the extreme values from each of the 13 seals were reported so levels for short apneustic periods are not represented (we combined venous and arterial data because they were not different after 1 min of apnea)

[e] Qvist et al. 1986 where levels were reported within 2 min of surfacing for dives <17 min and within 9 min of surfacing for dives >17 min

[f] Meir et al. 2009 provides a range of minimum values over a range of dive durations but emphasizes the minimum values obtained for dives >10 min, which are routine for this species

concentration increased significantly ($df = 1.6$, $r^2 = 0.53$, $F = 6.81, P = 0.04$), particularly over the breath-hold period of 5–6 min for this odontocete. Final plasma lactate levels were 2–3 times higher than initial levels (1.10 mmol l^{-1}), suggesting that the aerobic breath-hold limit (ADL) of bottlenose dolphins is approximately 5 min according to the definition by Kooyman (1989). Although the killer whale demonstrated the longest breath hold, plasma lactate concentration did not increase ($df = 1.12$, $r^2 = 0.12$, $F = 1.65$, $P = 0.22$) and averaged 1.81 ± 0.33 mmol l ($n = 14$; Fig. 3). In view of this, it appears that the killer whale did not surpass its aerobic breath-hold limit during these tests. Likewise, plasma lactate concentration showed no relationship with breath-hold duration ($df = 1.13$, $r^2 = 0.01$, $F = 0.18$, $P = 0.68$) in the white-sided dolphins and averaged 2.77 ± 0.70 mmol l ($n = 15$; Fig. 3).

Discussion

Interrelationships between body mass, metabolic rate and aerobic dive limit

Theoretically, large body size facilitates prolonged dive durations in big odontocetes due to the disproportionate scaling of oxygen stores and metabolic rate with body mass (Noren and Williams 2000). Assuming that the rate of decrease in blood oxygen content can be considered a reliable indication of the depletion of the oxygen resources during suspended breathing (Johansen et al. 1966), then it appears that metabolic demand is a critical factor defining breath-hold duration across odontocetes of varying body size. The three odontocete species examined in this study represented a 40-fold difference in body size and showed variable rates of change for P_{O_2} and accumulation of P_{CO_2} in the blood with the result that there was a fourfold difference in voluntary breath-hold capacity. For example, the small white-sided dolphins had a relatively fast rate of change for P_{O_2} (Fig. 1a) compared to the killer whale. Although the rates of change for P_{CO_2} were indistinguishable between the odontocetes, qualitatively carbon dioxide accumulated earlier into breath hold for the white-sided dolphins compared to the two larger species (Fig. 1b). The inability to detect an effect of body size on carbon dioxide production may be due in part to precision error. For a given change in the partial pressure of oxygen, the corresponding change in the partial pressure of carbon dioxide is much smaller (Boutilier et al. 2001) due to the relatively higher physical solubility of carbon dioxide (≈25 times higher compared to oxygen) and the chemical binding of

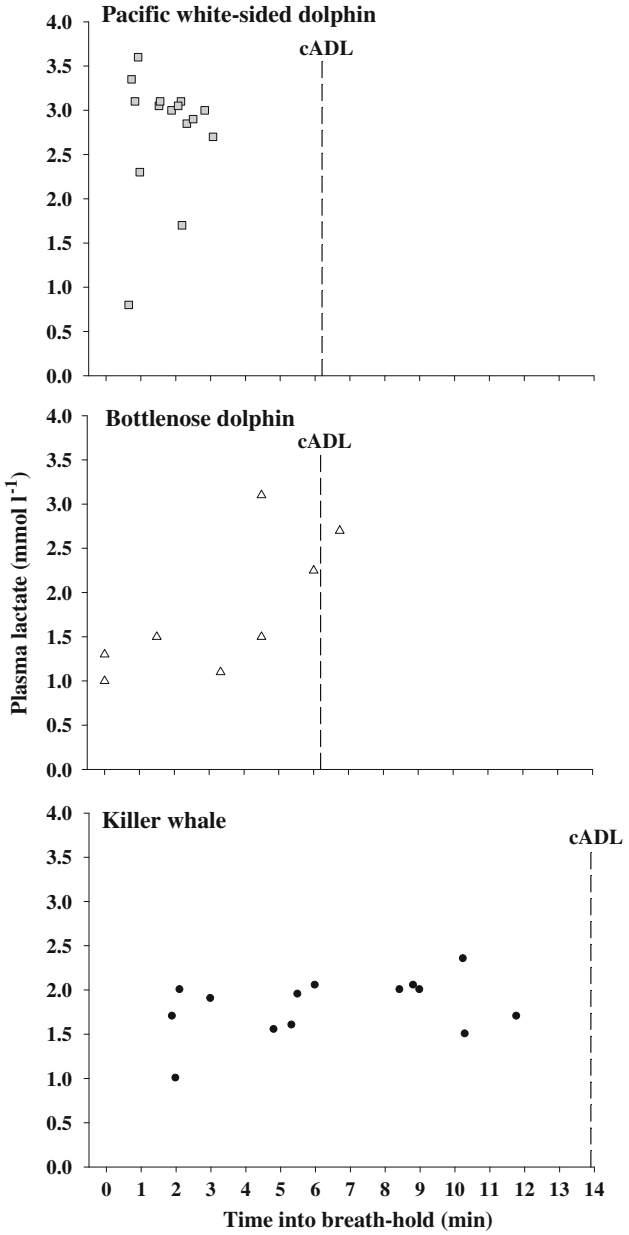

Fig. 3 Plasma lactate in relation to time into breath-hold in three species of odontocete. Each *data point* represents a single blood sample for Pacific white-sided dolphin (*closed squares*), bottlenose dolphin (*open triangles*), and killer whale (*closed circles*). The *dashed line* indicates the resting-surface cADL

carbon dioxide as bicarbonate (Piiper 1990). Admittedly these sedentary breath-hold durations represent a proxy for diving capacity in the wild, but similar trends for body size and dive duration are observed in free-ranging odontocetes. For example, the maximum dive duration for one of the smallest odontocetes, the harbor porpoise (*Phocoena phocoena*), is approximately 6 min (Westgate et al. 1995), while the largest odontocete, the sperm whale, can dive for over 73 min (Watkins et al. 1993). It is not surprising that body mass influences dive performance of odontocetes, as this had been hypothesized for odontocetes in a prior study (Noren and Williams 2000). Here we provide the first empirical data to support this theoretical framework.

Similar to Noren and Williams (2000), we demonstrate that body mass has a profound influence on the diving capacity in odontocetes, but environmental factors may have influenced how oxygen is stored in these animals which can also impact breath-holding capabilities. For example, the previous research on beluga whales by Shaffer et al. (1997) demonstrate a dive duration greater than that of the killer whale, even though the killer whale was nearly five times larger than the belugas. This result may be due to the differences in species-specific oxygen stores because unlike killer whales, belugas must sustain breath hold to forage under ice-cover. Indeed, Noren and Williams (2000) were able to explain 83% of the variation in dive performance across odontocetes species when they included the variation in body size along with the variation in the muscle oxygen store. Thus it is the oxygen store as well as the rate at which it is used that define the limits to breath-hold performance in odontocetes.

To ascertain why the larger killer whale demonstrated a lower breath-holding capability than the belugas, we calculated the total body oxygen stores and the calculated aerobic dive limit (cADL) for the species in this study as well as for belugas. Total body oxygen stores were calculated according to Kooyman (1989), as modified for odontocetes by Shaffer et al. (1997), Noren et al. (2002, 2004), and provided an indication of potential differences in the physiological evolution of body oxygen stores across these species (Table 3). We used the calculation of aerobic dive limit (cADL) to ascertain why lactate accumulation was not found for some of the animals in this study. The cADL is the ratio of total body oxygen stores and the rate at which these stores are utilized (Kooyman 1989) and can provide an estimate for the experimentally determined aerobic dive limit (ADL; Kooyman 1989; Kooyman and Ponganis 1998). The ADL can be accurately predicted by the cADL when estimates of body oxygen stores and metabolic rate are reliable (Ponganis et al. 1997a). We maximized the reliability of our calculations by utilizing species-specific oxygen store data (Table 3) and considering a range of realistic metabolic rates, which represented three activity levels (resting-submerged, resting-surface, and exercising). Details regarding the assumptions that were made for these calculations are provided in the appendix found in the electronic supplementary material.

For the four odontocete species analyzed, there is a 40-fold increase in body size, yet only a twofold increase in dive performance across species (Table 4). Although smaller species have higher mass-specific metabolic demands, enhanced oxygen stores may offset them, allowing for longer dive durations than expected. For

Table 3 Estimated oxygen stores in the lung, skeletal muscle, and blood of four species of odontocete

	Pacific white-sided dolphin	Bottlenose dolphin	Beluga whale	Killer whale
Mass (kg)	96	189	776	3,850
Total lung O_2 (L)	0.9	1.7	6.7	31.1
Mass-specific lung O_2 (ml O_2 kg^{-1})	9.4	9.1	8.6	8.1
Muscle myoglobin [g Mb (100 g muscle)$^{-1}$]	3.45[a]	2.76[a]	3.44[a]	3.07[a]
Muscle mass (%)	24[g]	36[c]	36[c]	36[c]
Total muscle O_2 (L)	1.1	2.5	12.9	57.0
Mass-specific muscle O_2 (ml O_2 kg^{-1})	11.1	13.3	16.6	14.8
Blood hemoglobin [g Hb (100 ml blood)$^{-1}$]	18.85[b]	16.10[b]	21.57[b]	14.63[b]
Blood volume (ml kg^{-1})	108[d]	71[d]	98.9[f]	90[e]
Total blood O_2 (L)	1.9	2.1	20.7	49.2
Mass-specific blood O_2 (ml O_2 kg^{-1})	19.8	11.1	26.7	12.8
Combined total body O_2 (L)	3.9	6.3	40.3	137.4
Combined total mass-specific O_2 (ml O_2 kg^{-1})	40.2	33.5	51.9	35.7

Calculations described in the appendix found in the electronic supplementary material. References: [a] Noren and Williams 2000; [b] Hedrick and Duffield 1991; [c] Goforth 1986; [d] Ridgway and Johnston 1966; [e] Ridgway 1972; [f] Ridgway et al. 1984; [g] S. R. Noren, unpublished

Table 4 The mean mass, maximum voluntary breath-hold duration, calculated aerobic dive limits [cADLs assumed a resting-submerged (RSb), resting-surface (RSf), and exercising (E) metabolism as described in the text], experimentally determined aerobic dive limit (ADL), and maximum observed dive duration for free-ranging animals for four species of odontocete

Species	Mass (kg)	Max breath hold (min)	cADL RSb (min)	cADL RSf (min)	cADL E (min)	ADL (min)	Max dive (min)
Pacific white-sided dolphin	96.0 ± 4.2[a]	3.5[a]	10.2[a]	6.2[a]	2.1[a]	–	6.2[c]
Bottlenose dolphin	189 ± 14.1[a]	6.8[a]	10.1[a]	6.2[a]	2.1[a]	5[a]	8.0[f]
Beluga whale	776 ± 185.3[b]	17[b]	22.2[a]	13.6[a]	4.5[a]	11[b]	18.3[d]
Killer whale	3,850[a]	13.3[a]	22.8[a]	13.9[a]	4.6[a]	–	10.0[e]

References: [a] present study; [b] Shaffer et al. 1997; [c] Black 1994; [d] Martin et al. 1993, [e] Heyning and Dahlheim 1988; [f] Ridgway and Harrison 1986

example, the calculated mass-specific oxygen stores of Pacific white-sided dolphins (40.2 ml kg^{-1}) were elevated compared to those of bottlenose dolphins (33.5 ml kg^{-1}) and killer whale (35.7 ml kg^{-1}; Table 3). This relatively large oxygen store for the smaller Pacific white-sided dolphins provided for a cADL identical to the two times larger bottlenose dolphin (Table 4). Furthermore, the beluga whale, a species with exceptionally large mass-specific oxygen stores (51.9 ml O_2 kg^{-1}; Table 3), had a cADL similar to that of the five times larger killer whale (Table 4). The primary enhancement in oxygen storage capacity for Pacific white-sided dolphins and beluga whales was in the blood. For these species, approximately 50% of their total body oxygen reserves were represented in the blood (Table 3). In contrast, for bottlenose dolphins and killer whales, approximately 41% of their total body oxygen reserves were found in the skeletal muscle (Table 3). Maintaining relatively high blood oxygen reserves by Pacific white-sided dolphins and beluga whales is similar to New Zealand sea lions (*Phocarctos hookeri*; Costa et al. 1998) and phocid seals (Kooyman 1989). Across these groups, high blood oxygen storage capacity is primarily accomplished by enhancing blood volume (Ridgway and Johnston 1966; Kooyman 1989; Hedrick and Duffield 1991; Costa et al. 1998; Table 3). The relatively large blood oxygen reserves in these animals, in turn, facilitate exceptional dive durations (Kooyman 1989; Costa et al. 1998; current study).

The cADLs were compared to the maximum breath-hold durations and ADLs of the experimental animals as well as the dive performance of wild animals (Table 4) to (1) ascertain which metabolic assumption provided the best prediction of breath-holding capabilities and (2) determine how the performance of the experimental animals compared to their physiological capacities. The resting-submerged cADL of dolphins (10.1 min), which assumed a 39% reduction in metabolic rate to account for metabolic suppression of a dive response (Table 4), overestimated the measured ADL for bottlenose dolphins (Fig. 3). This result suggests that breath-holding dolphins do not rely on whole body metabolic suppression, despite the observation that the dolphins elicited a dive response during experimental breath holds as evident by significant reductions in heart rate (Williams et al. 1999). A similar result was

demonstrated for free-ranging diving Northern elephant seals (Meir et al. 2009). Meanwhile, when the metabolic demand of exercise was added, the resulting exercising cADLs underestimated the ADLs measured for odontocetes, as well as the actual dive performance of free-ranging odontocetes (Table 4). The depletion of muscle oxygen stores, as indicated by an increase in lactate concentration, occurred at 5 min for bottlenose dolphins (Fig. 3), and this was the best approximated by a cADL that assumed a resting-surface metabolic rate equivalent to two times of the Kleiber (1975)-predicted basal metabolic rate for terrestrial mammals of similar body size (Williams et al. 1993b, 2001). This cADL also provided the best approximation of the dive performance of free-ranging odontocetes (Table 4; Williams et al. 1999). Interestingly, there is agreement across taxonomic groups, as the ADL for shallow diving emperor penguins (*Aptenodytes forsteri*; Ponganis et al. 2010) and free-diving Weddell seals (Castellini et al. 1992, Ponganis et al. 1993) was also best approximated by a cADL assuming an oxygen consumption rate twice of Kleiber's. Thus, if we assume that a cADL based on a metabolic rate twice of Kleiber's most closely predicts the ADL of the relatively short duration divers in this study, we find that the maximum breath-hold duration of the killer whale and especially the Pacific white-sided dolphins fell short of these limits (Table 4). This suggests that these animals did not perform to their fullest physiological potential and may explain the inability to detect a rise in blood lactate levels as breath hold progressed in these species.

Cues for initiating ventilation

Values for blood gases and pH at the onset of breath hold in Pacific white-sided dolphins, bottlenose dolphins, and the killer whale (Fig. 1a, b, c) were consistent with the levels measured at the onset of breath hold in beluga whales (P_{O_2} average = 63.5 (max 79) mmHg, P_{CO_2} = 61 mmHg and pH = 7.26; Shaffer et al. 1997) and pre-dive levels in Weddell seals (P_{O_2} = 78.1 ± 12.9 mmHg, P_{CO_2} = 42.5 ± 1.63 mmHg and pH = 7.37 ± 0.010; Qvist et al. 1986). In addition, all aquatic animals examined to date demonstrate a non-linear decline in blood P_{O_2} during sedentary breath hold (Fig. 1a; Kooyman et al. 1980; Shaffer et al. 1997; Stockard et al. 2007) or diving (Qvist et al. 1986; Meir et al. 2009; Ponganis et al. 2010) with the most marked decline in the partial pressure of oxygen in the blood occurring during the first half of the breath hold (Fig. 1a). For belugas, P_{O_2} decreased by 65% during the first 8 min of breath hold and only declined further by 8% as breath hold proceeded past 10 min (calculated from Shaffer et al. 1997). Likewise, after comparatively short duration dives, P_{O_2} in seal blood declined by 69% and only declined further by 6% with dive durations that exceeded 17 min (calculated from Qvist et al. 1986). This dramatic decrease in blood oxygen at the onset of the dive is likely due to the combined influences of cells utilizing these oxygen reserves directly as well as oxygen being shuttled from the blood oxygen carrier (hemoglobin) to the higher affinity muscle oxygen carrier (myoglobin) as the muscle oxygen stores become depleted.

In these breath-holding odontocetes, Northern elephant seals (*Mirounga angustirostris*; Stockard et al. 2007), and Weddell seals (Kooyman et al. 1980) as well as free-diving (Qvist et al. 1986) Weddell seals, reductions in blood P_{O_2} were accompanied by a concomitant increase in blood P_{CO_2} and decrease in blood pH (Table 2). These final oxygen tensions in marine mammals indicate hypoxemic tolerance, as these same values correspond to the threshold at which humans lose consciousness (25–30 mmHg; Ferretti et al. 1991; Ferrigno and Lundgren 2003). This tolerance may be necessary to allow for more effective utilization of the blood oxygen store (Stockard et al. 2007). Indeed, blood gases at the termination of dives for freshwater turtles (*Rheodytes leukops*) also fell within this range (Gordos et al. 2004), while emperor penguins (*Aptenodyte forsteri*) show greater reductions, where final venous P_{O_2} values for these consummate divers were less than 20 mmHg in 29% of dives (Ponganis et al. 2007). In general, the minimum levels of O_2 or the maximum levels of CO_2 may be cueing ventilation across these broad taxonomic groups, as has been previously suggested (Kooyman et al. 1971; Pasche 1976a, b; Craig and Pasche 1980).

The partial pressure of carbon dioxide in the arterial blood and brain tissue play a dominant role in the regulation of breathing in mammals (Phillipson et al. 1981). Previous studies of diving animals have suggested that elevated carbon dioxide levels in the blood (Butler 1982) serve as a physiological stimulus to initiate ventilation after periods of apnea. Carbon dioxide may act directly via chemoreceptors in blood vessels and indirectly via receptors in the respiratory muscles to stimulate breathing in diving birds and mammals (Butler 1982). As a result, changes in this blood gas have been associated with the termination of diving in mammals. For example, increased levels of carbon dioxide in the blood have limited the dive durations of manatees (Gallivan 1980). Similarly, elevated blood P_{CO_2} prevented trained dolphins from completing repetitive dives (Ridgway et al. 1969).

Alternatively, rising lactate levels during breath hold may contribute to a decrease in plasma pH levels, and these changes in pH could subsequently stimulate ventilation (Kooyman et al. 1971). Although blood lactate increased during breath hold in bottlenose dolphins (Fig. 3), it is

difficult to separate the effect of lactate from the effect of P_{CO_2} since the accumulation of either contributes to a decrease in blood pH. Interestingly, despite differences in plasma lactate concentration, final pH levels fell within a narrow range after breath holding or diving across a range of marine mammals (Table 2). Thus, there may be another factor influencing blood pH, such as P_{CO_2}.

We found that P_{CO_2} and pH levels in the blood were significantly correlated for bottlenose dolphins, as well as for Pacific white-sided dolphins and the killer whale (Fig. 2). Changes in blood pH have also been attributed to changes in P_{CO_2} in sedentary apneustically breathing Northern elephant seals (Stockard et al. 2007). In view of this, carbon dioxide accumulation and the accompanying decrease in pH may act synergistically with any lactate-induced pH changes to promote ventilation after an apneustic period in cetaceans.

Interestingly, for all values of P_{CO_2}, the smallest odontocete (white-sided dolphin) showed the highest corresponding pH, while the largest species (killer whale) showed the lowest corresponding pH. Indeed there were variable rates of change in pH in relation to P_{CO_2} across the species, with the small Pacific white-sided dolphin having the slowest rate of change and the large killer whale having the greatest rate of change. Meanwhile, the blood lactate levels were the highest in the Pacific white-sided dolphin which should act to further lower blood pH levels. Although Pacific white-sided and bottlenose dolphins have similar buffering capacities in the muscle that counteract changes in pH associated with the accumulation of lactic acid (Noren 2004), there may be differences in the buffering capacity of the blood across these odontocete species and this warrants further investigation.

In summary, the effect of body size on aerobic breath-hold capacity (and by inference dive performance) in odontocetes appeared to be mediated primarily through differences in metabolic demands. Difference in apneustic duration across a large-scale body size range were associated with variable rates of change in blood chemistry, such that faster rates of change for P_{O_2} in the bottlenose and white-sided dolphins reduced breath-hold capacities compared to that of the 20–40 times larger killer whale. Nonetheless, changes in the partial pressure of carbon dioxide in the blood and in the pH of the blood may have also played a role in cueing the ventilation of these small and large odontocetes. This research adds to the limited data on the biochemical changes in the blood during the prolonged breath holds of aquatic animals, particularly odontocetes.

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References

Black NA (1994) Behavior and ecology of Pacific white-sided dolphins (*Lagenorhynchus obliquidens*) in Monterey Bay, California. M.S. Thesis, San Francisco State University, San Francisco, CA

Boutilier RG, Reed JZ, Fedak MA (2001) Unsteady-state gas exchange and storage in diving marine mammals: the harbor porpoise and gray seal. Am J Physiol Regulatory Integrative Comp Physiol 281:R490–R494

Boyd IL, Croxall JP (1996) Dive durations in pinnipeds and seabirds. Can J Zool 74:1696–1705

Butler PJ (1982) Respiratory and cardiovascular control during diving in birds and mammals. J Exp Biol 100:195–221

Castellini MA, Kooyman GL, Ponganis PJ (1992) Metabolic rates of freely diving weddell seals: correlations with oxygen stores, swim velocity and diving duration. J Exp Biol 165:181–194

Costa DP (1991) Reproductive and foraging energetics of high latitude penguins, albatrosses and pinnipeds: implications for life history patterns. Am Zool 31:11–130

Costa DP, Gales NJ, Crocker DE (1998) Blood volume and diving ability of the New Zealand sea lion, *Phocarctos hookeri*. Physiol Zool 71(2):208–213

Costa DP, Kuhn CE, Weise MJ, Shaffer SA, Arnould JPY (2004) When does physiology limit the foraging behaviour of freely diving mammals? Int Congr Ser 1275:359–366

Craig AB, Pasche A (1980) Respiratory physiology of freely diving harbor seals (*Phoca vitulina*). Physiol Zool 53:419–432

Evans PGH (1987) The natural history of whales and dolphins. Facts on File, Inc, New York, p 343

Ferretti C, Costa M, Ferrigno M, Grassi B, Marconi C, Lundgren CEG, Cerretelli P (1991) Alveolar gas composition and exchange during deep breath-hold diving and dry breath holds in elite divers. J Appl Physiol 70:794–802

Ferrigno M, Lundgren CE (2003) Breath-hold diving. In: Brubakk AO, Neuman TS (eds) Physiology and medicine of diving. Saunders, Edinburgh, pp 153–180

Gallivan GJ (1980) Hypoxia and hypercapnia in the respiratory control of the Amazonian manatee (*Trichechus inunguis*). Physiol Zool 53:254–261

Goforth HW (1986) Glycogenetic responses and force production characteristics of a bottlenose dolphin (*Tursiops truncatus*) while exercising against a force transducer. Ph.D. Thesis. University of California, Los Angeles

Gordos MA, Franklin CE, Limpus CJ, Wilson G (2004) Blood-respiratory and acid–base changes during extended diving in the bimodally respiring freshwater turtle *Rheodytes leukops*. J Comp Physiol B 174:347–354

Hedrick MS, Duffield DA (1991) Haematological and rheological characteristics of blood in seven marine mammal species:

physiological implications for diving behavior. J Zool Lond 225:273–283
Heyning JE, Dahlheim ME (1988) Orcinus orca. Mamm Species 304:1–9
Hudson DM, Jones DR (1986) The influence of body mass on the endurance to restrained submergence in the Pekin duck. J Exp Biol 120:351–367
Johansen K, Lenfant C, Grigg GC (1966) Respiratory properties of blood and responses to diving of the platypus *ornithorhynchus anatinus* (shaw). Comp Biochem Physiol 18:597–608
Kleiber M (1975) The fire of life: an introduction to animal energetics. R.E. Krieger Publ. Co, Huntington, p 453
Kooyman GL (1989) Diverse Divers. Springer-Verlag Berlin Heidelberg, Germany
Kooyman GL, Ponganis PJ (1998) The physiological basis of diving to depth: birds and mammal. Annu Rev Physiol 60:19–32
Kooyman GL, Kerem DH, Campbell WB, Wright JJ (1971) Pulmonary function in freely diving Weddell seal, *Leptonychotes weddellii*. Respir Physiol 12:271–282
Kooyman GL, Wahrenbrock EA, Castellini MA, Davis RW, Sinnett EE (1980) Aerobic and anaerobic metabolism during voluntary diving in Weddell seals: evidence for preferred pathways from blood chemistry and behavior. J Comp Physiol B 138:335–346
Kooyman GL, Castellini MA, Davis RW, Mave RA (1983) Aerobic diving limits of immature Weddell seals *Leptonychotes weddellii*. J Comp Physiol B 151:171–174
Martin AR, Smith TG, Cox OP (1993) Studying the behaviour and movements of high Arctic belugas with satellite telemetry. Symp Zool Soc Lond 66:195–210
Meir JU, Champagne CD, Costa DP, Williams CL, Ponganis PJ (2009) Extreme hypoxemic tolerance and blood oxygen depletion in diving elephant seals. Am J Physiol Regul Intergr Comp Physiol 297:R927–R939
Nagy K (1994) Field bioenergetics of mammals: what determines field metabolic rate? Aust J Zool 42:43–53
Noren SR (2004) Muscle acid buffering capacities in cetaceans: influences of diving performance, swimming performance, body size, and postpartum development. Mar Mamm Sci 20(4): 808–822
Noren SR, Williams TM (2000) Body size and skeletal muscle myoglobin of cetaceans: adaptations for maximizing dive duration. Comp Biochem Physiol 126(A):181–191
Noren SR, Lacave G, Wells RS, Williams TM (2002) The development of blood oxygen stores in bottlenose dolphins (*Tursiops truncatus*): implications for diving capacity. J Zool Lond 258:105–113
Noren SR, Cuccurullo V, Williams TM (2004) The development of diving bradycardia in bottlenose dolphins (*Tursiops truncatus*). J Comp Physiol B 174:139–147
Noren SR, Iverson SJ, Boness DJ (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. Physiol Biochem Zool 78(4):482–490
Pasche A (1976a) Hypoxia in freely diving hooded seal, *Cystophora cristata*. Comp Biochem Physiol 55A:319–322
Pasche A (1976b) The effect of hypercapnia on respiratory characteristics and diving behaviour of freely diving seals. Respir Physiol 26:183–194
Phillipson EA, Duffin J, Cooper JD (1981) Critical dependence of respiratory rhythmicity on metabolic CO_2 load. J Appl Physiol 50:45–55
Piiper J (1990) Modeling of gas exchange in lungs, gills and skin. In: Boutilier RG (ed) Advances in comparative and environmental physiology, vol 6. Springer Verlag, Berlin, pp 15–44
Ponganis PJ, Kooyman GL, Castellini MA (1993) Determinants of the aerobic dive limit of Weddell seals: analysis of diving metabolic rates, postdive end tidal P_{O2}'s and blood and muscle oxygen stores. Physiol Zool 66:732–749
Ponganis PJ, Kooyman GL, Baranov EA, Thorson PH, Steward BS (1997a) The aerobic submersion limit of Baikal seals, *Phoca sibirica*. Can J Zool 75:1323–1327
Ponganis PJ, Kooyman GL, Winter LM, Starke LN (1997b) Heart rate and plasma lactate responses during submerged swimming and trained diving in California sea lions, *Zalophus californianus*. J Comp Physiol 167:9–16
Ponganis PJ, Stockard TK, Meir JU, Williams CL, Ponganis KV, van Dam RP, Howard R (2007) Returning on empty: extreme blood O_2 depletion underlies dive capacity of emperor penguins. J Exp Biol 210:4279–4285
Ponganis PJ, Meir JU, Williams CL (2010) Oxygen store depletion and the aerobic dive limit in emperor penguins. Aquat Biol 8:237–245
Qvist J, Hill RD, Schneider RC, Falke KJ, Liggins GC, Guppy M, Elliott RL, Hochachaka PW, Zapol WM (1986) Hemoglobin concentrations and blood gas tensions of free-diving Weddell seals. J Appl Physiol 61:1560–1569
Ridgway SH (1972) Mammals of the sea biology and medicine. Charles C Thomas Publisher, Springfield
Ridgway SH, Harrison RJ (1986) Diving dolphins. In: Bryden MM, Harrison RJ (eds) Research on dolphins. Clarendon Press, Oxford, pp 33–58
Ridgway SH, Johnston DG (1966) Blood oxygen and ecology of porpoises of three genera. Science 151:456–457
Ridgway SH, Scronce BL, Kanwisher J (1969) Respiration and deep diving in the bottlenose porpoise. Science 166:1651–1654
Ridgway SH, Bowers CA, Miller D, Schultz ML, Jacobs CA, Dooley CA (1984) Diving and blood oxygen in the white whale (*Delphinapterus leucas*). Can J Zool 62:2349–2351
Schreer JF, Kovacs KM (1997) Allometry of diving capacity in air-breathing vertebrates. Can J Zool 75:339–358
Shaffer SA, Costa DP, Williams TM, Ridgway SH (1997) Diving and swimming performance of white whales, *Delphinapterus leucas*: an assessment of plasma lactate and blood gas levels and respiratory rates. J Exp Biol 200:3091–3099
Stockard TK, Levenson DH, Berg L, Fransioli JR, Baranov EA, Ponganis PJ (2007) Blood oxygen depletion during rest-assoicated apneas of northern elephant seals (*Mirounga angustirostris*). J Exp Biol 210:2607–2617
Watkins WA, Daher MA, Fristrup KM, Howard TJ, Di Sciara GN (1993) Sperm whales tagged with transponders and tracked underwater by sonar. Mar Mamm Sci 9(1):55–67
Westgate AJ, Read AJ, Berggren P, Koopman HN, Gaskin DE (1995) Diving behaviour of harbour porpoise, *Phocoena phocoena*. Can Fish Aquat Sci 52:1064–1073
Williams TM, Friedl WA, Fong ML, Yamada RM, Sedivy P, Haun JE (1992) Travel at low energetic cost by swimming and wave-riding bottlenose dolphins. Nature 355:821–823
Williams TM, Friedl WA, Haun JE (1993a) The physiology of bottlenose dolphins (*Tursiops truncatus*): heart rate, metabolic rate, and plasma lactate concentration during exercise. J Exp Biol 179:31–46
Williams TM, Friedl WA, Haun JE, Chun NK (1993b) Balancing power and speed in bottlenose dolphins (*Tursiops truncatus*). Symp Zool Soc Lond 66:383–394
Williams TM, Haun JE, Friedl WA (1999) The diving physiology of bottlenose dolphins (*Tursiops truncatus*) I. Balancing the demands of exercise for energy conservation at depth. J Exp Biol 202:2739–2748
Williams TM, Haun J, Davis RW, Fuiman LA, Kohin S (2001) A killer appetite: metabolic consequences of carnivory in marine mammals. Comp Biochem Physiol A 129(4):785–796
Williams TM, Noren SR, Glenn M (2011) Extreme physiological adaptations as predictors of climate-change sensitivity in the narwhal, *Monodon monoceros*. Mar Mamm Sci 27(2):334–349