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Developmental changes in cardiorespiratory patterns associated with terrestrial apnoeas in harbour seal pups

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Summary

During the nursing period seals undergo several physiological and behavioural changes. A key component of development is increased cardiorespiratory control, fundamental for breath-holding and thus diving. This study focused on the ontogenetic changes in cardiac responses to respiration in quietly resting, pre-weaned harbour seal pups (*Phoca vitulina*). During periods of quiet rest, breathing became episodic, eupnoea interspersed with periods of apnoea. Little change was observed in respiration (~35 breaths min^{-1}) and eupnoeic heart rate (~160 beats min^{-1}) throughout the nursing period. However, apnoea duration increased (from ~20 to 40 s), while apnoeic heart rate decreased with age (from ~150 to 90 beats min^{-1}). The observed decline in apnoeic heart rate resulted from an increase in cardiorespiratory control as pups approached weaning, evident by the ability to maintain a lower heart rate more consistently. Similar changes in cardiorespiratory patterns have been reported for elephant and Weddell seals. Due to the early onset of independent foraging, however, the rate of cardiorespiratory control development was more rapid in harbour seals. Our findings suggest that by 1 month of age, harbour seal pups possess the cardiorespiratory control necessary to sustain long-duration apnoeas, fundamental for proficient diving and successful foraging upon weaning.

Key words: apnoea, eupnoea, respiration, heart rate, bradycardia, sleep, ontogeny, cardiorespiratory control, harbour seal, *Phoca vitulina*.

Introduction

The obligation to surface periodically and replenish oxygen stores limits the underwater foraging capacity of marine mammals. To compensate for this need, most marine mammals possess a suite of physiological adaptations which, together, maximize foraging time while minimizing surface intervals (Butler and Jones, 1997). The ability to perform long apnoeas, however, not only facilitates underwater foraging, but also aids in conserving water and energy during terrestrial fasts (Ortiz et al., 1978; Costa and Ortiz, 1980; Huntley, 1984; Blackwell, 1996). Seals at rest, whether hauled-out or submerged, exhibit long-duration, sleep-associated apnoeas (Bartholomew, 1954; Ridgway et al., 1975; Castellini et al., 1994a), which elicit physiological responses similar to those observed while diving (Castellini, 1991, 1994; Andrews et al., 1997).

The physiological factors that limit both diving and sleep apnoea duration are correlated with animal size and age. In adults, total body oxygen stores are proportional to body mass (M_b; Kooyman, 1989), whereas metabolic rate scales to $M_b^{0.75}$ (Kleiber, 1961). Thus, apnoea durations are shorter for smaller species. In addition, pups and juveniles have reduced breath-holding capacities because young, growing animals have higher mass-specific metabolic rates and lower mass-specific total body oxygen stores, compared to adults of similar size (Brody, 1945; Kleiber, 1961; Poczopko, 1979; Lavigne et al., 1986). Furthermore, the ability to regulate physiological processes such as heart rate, respiration, body temperature and vasoconstriction is not fully developed at birth (Cherepanova et al., 1993; Castellini et al., 1994b; Thorson and Le Boeuf, 1994; Burns et al., 1996; Hansen and Lavigne, 1997; Falabella et al., 1999).

The rate of physiological development appears to be closely linked to the onset of independent foraging (Castellini et al., 1994b; Burns et al., 1996; Castellini, 1996; Burns, 1997; Falabella et al., 1999; Noren et al., 2001). To survive this transition, seal pups must develop adequate swimming and diving skills before their energy reserves are depleted. A key component of neonatal development is increased cardiorespiratory control (Castellini, 1996). Prior to their first foraging trip, weaned elephant seals (*Mirounga angustirostris* and *M. leonina*) spend several months on land, which represents a critical period for the maturation of cardiorespiratory control mechanisms (Castellini et al., 1994b; Castellini, 1996; Falabella et al., 1999). Older seals not only exhibit a stable breathing pattern, consisting of extended apnoeas followed by short periods of eupnoea, they also display a lower and less variable heart rate during apnoea, and a well-developed sinus arrhythmia during eupnoea (Castellini et al., 1994b; Castellini, 1996; Falabella et al., 1999).

Harbour seals *Phoca vitulina*, unlike most other phocid neonates, enter the water within hours of birth (Newby, 1973) and are increasingly aquatically active throughout the nursing period (Bekkby and Bjørge, 2000; Jørgensen et al., 2001). Therefore, the acquisition of swimming and diving skills is temporally separated from the onset of independent foraging. As such, the degree of cardiorespiratory control may be greater at birth, and/or the rate of maturation more rapid in this precocial species, compared to phocid species that delay aquatic activity until after weaning (Castellini, 1995, 1996; Burns, 1997). Thus, the aim of this study was to describe the ontogenetic changes in cardiorespiratory patterns associated with terrestrial apnoeas in harbour seal pups from birth to weaning.

Materials and methods

This study was conducted near Bic (48°24′N, 68°51′W) and Métis (48°41′N, 68°01′W), QC, Canada, on the south shore of the St Lawrence River Estuary, from May to July of 2001 and 2002. Harbour seals *Phoca vitulina* L. were captured in the water using a dip net and an inflatable boat, and subsequently transferred to a larger boat where all handling took place. Prior to analysis, seals were weighed (to ±0.5 kg), tagged, and their sex noted (Dubé et al., 2003). Additionally, as part of a concurrent study, blood samples were collected from each animal prior to release (Clark, 2004). Animals were recaptured opportunistically throughout the nursing period and re-monitored when the interval between consecutive recordings exceeded 5 days. When possible, weaned pups and adult females (mothers of pre-weaned pups) were also captured and monitored in a similar manner to younger pups.

Data collection

Due to the precocial nature of this species, and the logistic constraints imposed by working on small boats, measurements could not be obtained from unrestrained animals. As a result, during the 2001 field season, pups were manually restrained during measurements: with a pup positioned in ventral recumbency, a single handler, facing in the opposite direction, knelt and straddled the pup at midbody, pinioned the foreflippers to the side of the animal, and then leaned forward and applied pressure to the hindflippers. In 2002, pups were physically restrained using a custom built adjustable wooden V-board and canvas harness. Adult females were covered with a net and manually restrained by a single handler. Electrocardiogram (ECG), heart rate and respiration data were collected using a portable multi-channel physiological recorder (BioTach with Serial Linked Interface Component Software, Model 2121/3R-SP, UFI, Morro Bay, CA, USA) connected to custom designed electrodes (25 G × 1.5 inch hypodermic needles soldered to 8 m lengths of 4-conductor 20 G microphone cable; PrecisionGlide® #305127, Becton-Dickinson, Franklin Lakes, NJ, USA; #8424, Belden Inc., Richmond, IN, USA) and a portable computer. Electrodes were inserted subdermally in the mid-dorsal region of the animal and then anchored to the fur using cyanoacrylate adhesive and accelerator (Superbonder® 422 Instant Adhesive and Tak Pak® 7452 Accelerator, Loctite Canada Inc., Mississauga, ON, Canada). The negative electrode was placed over the left scapula, the positive electrode at a diagonal equidistant from the heart on the opposite side, and the ground to the left of the positive electrode. The BioTach monitor converted changes in thoracic impedance into a respiratory signal; therefore any movement in addition to that of the ribcage (e.g. the seal, handler, boat, etc.) was also detected, resulting in erroneous values. As a result, a camcorder (Sony Handycam®, CCD-TRV57, and Rain Jacket, LCR-TRX3, Sony Ltd., Toronto, ON, Canada) was also used to record respiration visually and to obtain additional behavioural data.

Analysis and statistics

Rest was defined as a period of time during which an animal was lying quietly with its eyes closed (Blackwell and Le Boeuf, 1993; Castellini et al., 1994b). However, it should be noted that even though an animal is lying motionless with its eyes closed, it may not be sleeping; such a state has also been exhibited during periods of quiet wakefulness (Mukhametov et al., 1982). Eupnoea was categorized by regular opening and closing of a pup's nostrils, while apnoea was observed as a breath-hold, during which nostrils remained closed for >10 s following an exhalation (Bacon et al., 1985; Blackwell and Le Boeuf, 1993; Falabella et al., 1999). Since electroencephalographic activity was not recorded during this study, respiratory pauses are referred to as terrestrial apnoeas rather than sleep apnoeas (Castellini et al., 1994b).

Video recordings were viewed and the percentage of time spent resting was calculated using the camcorder on-screen clock display. The occurrence of each respiration was determined from the video recordings using an elapsed timing program designed in MatLab 6.1® (The Mathworks Inc., Natick, MA, USA). Subsequently, the time interval between consecutive breaths was converted to an instantaneous breathing rate (breaths min^{-1}). The precise onset and cessation of each apnoea was verified by the respiration trace obtained by the physiological monitor. For instances where no corresponding video recording was obtained, apnoeas were selected using a combination of information from field notes, event markers, and the respiration trace. The time interval between successive heartbeats was measured using an R-wave peak detection program designed in Mathcad 2001 Professional® (MathSoft Engineering & Education Inc.

Cambridge, MA, USA). Subsequently, each R–R interval was converted to an instantaneous heart rate (beats min^{-1}).

Pups weighing ⩽11.1 kg at first capture were considered newborn (pup age = 1 day; Dubé et al., 2003). For pups not captured on the day of birth, age (days) was calculated by subtracting the published mass at birth (11.1±0.22 kg) from the mass at first capture (in kg) and dividing by the observed growth rate (kg day^{-1}) (Dubé et al., 2003). The lactation period for this population of harbour seal pups has been estimated to last 34±1.8 days (Dubé et al., 2003). Pups approaching this age were considered weaned if they were observed to be alone and their blood plasma was clear (i.e. non-lipaemic; Clark, 2004; Bowen et al., 1985).

To achieve statistical independence, and to optimize age distribution, only one data file was selected from individuals monitored multiple times throughout the nursing period. Effects of year and sex were assessed using a two-way analysis of variance (ANOVA); age relationships were examined using least-squares linear regression; and paired *t*-tests were used to compare heart rate during apnoea and eupnoea (Systat 9 for Windows®, SPSS Inc., Chicago, IL, USA; SAS System for Windows 8.02®, SAS Institute Inc., Cary, NC, USA). Significance level was set at 0.05. Data are presented as means ± S.E.M.

Results

Electrocardiogram, heart rate and respiration records (Fig. 1) were analyzed from 45 pre-weaned harbour seal pups weighing 9–32 kg and ranging from 1–33 days of age. Due to inclement weather only 39 corresponding videos were obtained and analyzed. Observation time averaged 30.4±1.3 min.

Pre-weaned pups spent 57.0±3.0% of the observation time resting; during this time, breathing became episodic, consisting of eupnoea interspersed with periods of apnoea. Breathing frequency did not change significantly over the course of the nursing period; on average, pups breathed at a rate of 35.0±0.9 breaths min^{-1} (Fig. 2). Eupnoea episodes lasted 2.5±0.2 min. Pups spent 12.5±1.7% of the time resting in apnoea. Pups exhibited significantly fewer apnoeas during the 2001 field season compared to 2002 ($F_{27,18}$=7.704, P=0.008); the mean number of apnoeas was 2.7±0.5 (range 0–9) and 5.2±0.8 (range 0–12), respectively. Mean apnoea duration increased significantly during the nursing period (Fig. 3). However, maximum apnoea duration (43.4±2.8 s, range 11–78 s) did not increase with age. The longest breath-hold observed occurred in a 19 day-old pup and lasted for 78 s.

Heart rate was significantly lower during apnoea than during eupnoea (t_{31}=–16.05, P<0.0001). Mean eupnoeic heart rate was 159.6±1.9 beats min^{-1} and did not change significantly during the nursing period (Fig. 4). Mean apnoeic heart rate decreased significantly with age (Fig. 5A). The distribution of apnoeic heart rate was bimodal, with a minimum number of observations around 100 beats min^{-1}. Heart rates were categorized as ‘low’ (<100 beats min^{-1}) or ‘high’ (>100 beats min^{-1}; Fig. 6). High apnoeic heart rate decreased significantly with age, while the low apnoeic heart rate remained stable at 75.7±1.0 beats min^{-1} (Fig. 5B). The proportion of time spent in each mode changed throughout the nursing period. The low mode dominated as age increased (Figs 5C, 6, 7). The lowest instantaneous heart rate recorded, 9 beats min^{-1}, occurred in a 19 day-old pup during a 78 s apnoea. The highest instantaneous heart rate recorded was 213 beats min^{-1} and occurred during eupnoea in three pre-weaned pups ranging in age from 4–28 days. No sex differences were observed for any of the parameters reported above.

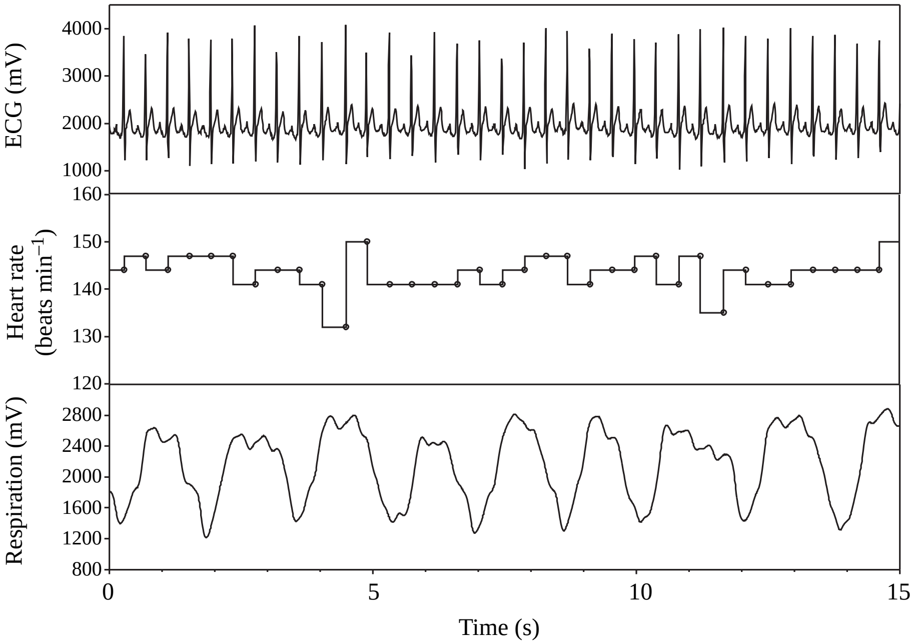

Fig. 1. A 15 s segment of the electrocardiogram, instantaneous heart rate (beats min^{-1}), and respiratory impedance (inspiration upward) trace from a quietly resting 16 day-old male harbour seal pup in eupnoea.

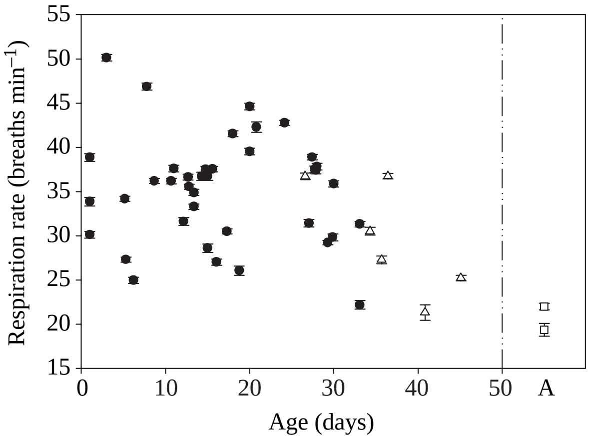

Fig. 2. Respiration rate (mean ± S.E.M.) of quietly resting pre-weaned harbour seal pups (circles; N=38), weaned pups (triangles; N=6) and adult females (A; squares; N=2).

Discussion

Harbour seal pups demonstrated the ability to regulate heart rate during terrestrial apnoeas from birth. This skill progressed with age, such that by weaning the degree of cardiorespiratory control was similar to that exhibited by older animals. These findings suggest that the rate of maturation of cardiorespiratory control is more rapid in harbour seals than in other previously studied phocid species. By 1 month of age, harbour seal pups possess the cardiorespiratory control necessary to sustain long-duration apnoeas, fundamental for proficient diving and successful foraging upon weaning.

The terrestrial breathing pattern of resting, pre-weaned harbour seal pups was irregular, consisting of apnoeas and eupnoeas of varying durations. Breathing frequency did not change throughout the nursing period and fell within the range observed previously for juvenile harbour seals (18–53 breaths min^{-1}; Harrison and Tomlinson, 1960; Påsche and Krog, 1980; Skinner and Milsom, 2001). Terrestrial

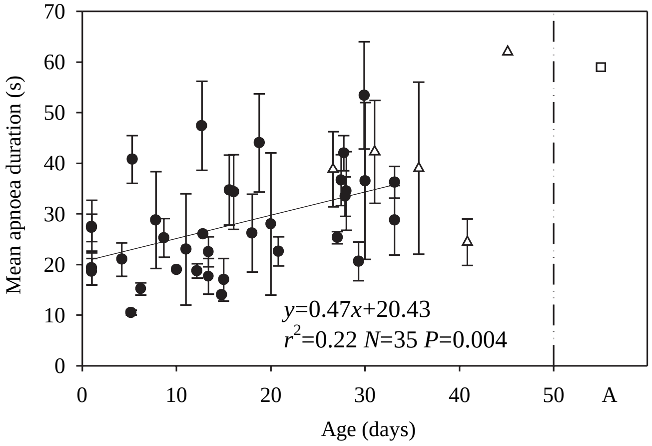

Fig. 3. Terrestrial apnoea duration (mean ± S.E.M.) of quietly resting pre-weaned harbour seal pups (circles; N=35), weaned pups (triangles; N=5) and an adult female (A; square; N=1).

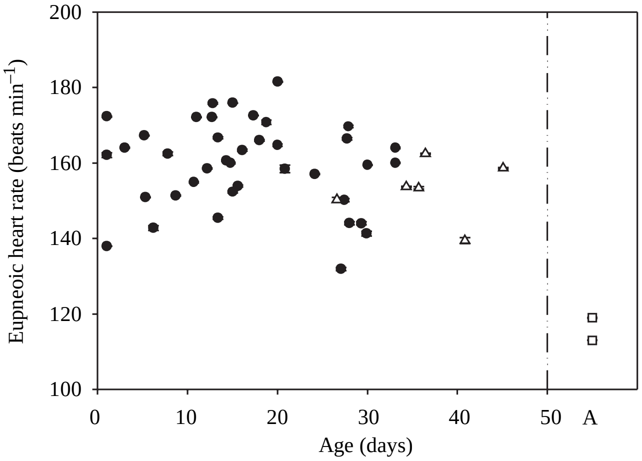

Fig. 4. Eupnoeic heart rate (mean ± S.E.M.) of quietly resting pre-weaned harbour seal pups (circles; N=39), weaned pups (triangles; N=6) and adult females (A; squares; N=2).

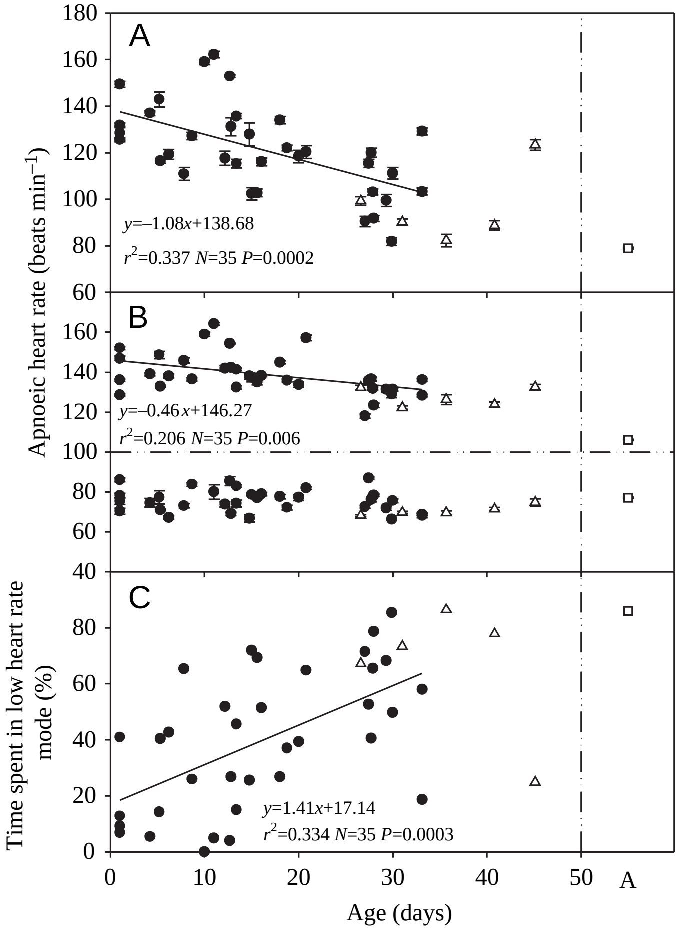

Fig. 5. (A) Apnoeic heart rate (mean ± S.E.M.) of quietly resting pre-weaned harbour seal pups (circles), weaned pups (triangles) and an adult female (A; square). (B) Apnoeic heart rate (mean ± S.E.M.) separated and categorized as 'high' (>100 beats min^{-1}) or 'low' (<100 beats min^{-1}). (C) Proportion of time spent in the low heart rate mode.

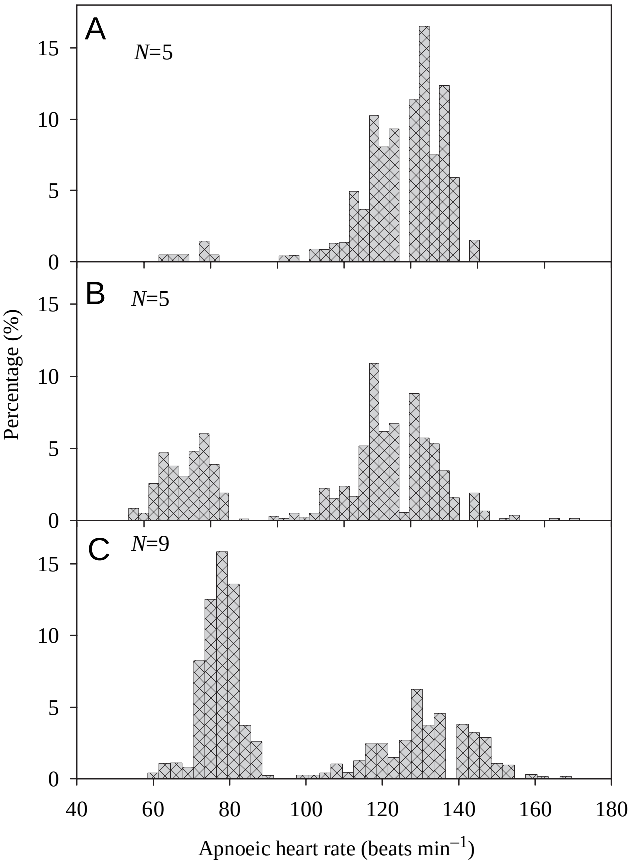

Fig. 6. Frequency histograms illustrating the bimodality of the apnoeic heart rate for a (A) 4 day, (B) 16 day, and (C) 31 day-old quietly resting harbour seal pup. *N*=number of apnoeas.

apnoea duration increased during the lactation period, paralleling an increase in dive duration (Greaves et al., in press a), and values were similar to those obtained for resting, captive harbour seal pups less than 2 weeks of age (31 s; Bacon et al., 1985).

Upon weaning, however, several changes occurred. Respiration rate decreased (Fig. 2), approaching adult levels (8–22 breaths min^{-1}; Harrison and Tomlinson, 1960; Bacon et al., 1985). Consistent with dive duration (Greaves et al., in press a), the length of terrestrial apnoeas continued to increase post-weaning (Fig. 3). Terrestrial apnoea durations were similar to those obtained previously for juvenile harbour seals (19–104 s; Irving et al., 1935; Dykes, 1974; Påsche and Krog, 1980; Jobsis et al., 2001), but less than those displayed by adults (225 s; Williams and Bryden, 1993). Additionally, upon weaning, the alternating apnoeic–eupnoeic breathing pattern began to stabilize. For example, a 41 day-old weanling that appeared to be sleeping displayed a regular apnoea–eupnoea cycle until it awoke. Irving et al. (1935) observed a similar breathing pattern in quietly resting 4 month-old harbour seals, describing it as a regularly maintained sequence of 6–12 respirations followed by an apnoea of 30–60 s. The observed changes, from birth to adulthood, can be attributed in part to an increase in total body oxygen stores (Kodama et al., 1977; Ponganis et al., 1993; Thorson and Le Boeuf, 1994; Burns et al., 1999; Clark, 2004) and a decrease in metabolic rate (Miller and Irving, 1975; Ashwell-Erickson and Elsner, 1981; Rea and Costa, 1992; Hansen, 1995) that occurs with increasing size and age. Additionally, improved cardiovascular control also contributes to the increased breath-holding capacity observed (Castellini et al., 1994b; Castellini, 1996).

In phocids, while diving or sleeping, heart rate follows the pattern of bradycardia during apnoea and tachycardia during eupnoea (Kooyman and Campbell, 1972; Andrews et al., 1997). This bimodal or two-speed heart rate pattern begins to develop *in utero* (Liggins et al., 1980; Bacon et al., 1985). During the last third of gestation, foetal harbour seals exhibited a ‘slower’ heart rate (79 beats min^{-1}) that was distinct from the ‘faster’ embryonic rate (125 beats min^{-1}) first observed early in gestation (Bacon et al., 1985). The amount of bradycardia steadily increased as parturition approached (Bacon et al., 1985). This bimodal heart rate pattern continues to develop *postpartum* (Table 1).

Eupnoeic heart rate remained stable during the lactation period and was similar to the surface heart rate of free-ranging, diving harbour seal pups (~155 beats min^{-1}; Greaves et al., in press a), but greater than the mean rate obtained for resting, captive harbour seal pups (138 beats min^{-1}; Bacon et al., 1985). Eupnoeic heart rate presumably begins to decrease post-weaning once animals start to accumulate body mass (~4–6 weeks post-weaning; Muelbert et al., 2003), and then gradually declines (Fig. 4) as animals grow into adulthood (Fig. 4; Stahl, 1967; Castellini and Zenteno-Savin, 1997). Terrestrial apnoeic heart rate decreased throughout the nursing period, paralleling a similar decrease in mean diving heart rate (from ~140 to 70 beats min^{-1}; Greaves et al., in press a). However, Bacon et al. (1985) observed a lower mean terrestrial apnoeic heart rate for harbour seal pups less than 2 weeks of age (84 beats min^{-1}). Similar to eupnoeic heart rate, further decreases in mean apnoeic heart rate (Fig. 5) would be expected as animals grow into adulthood (Stahl, 1967; Castellini and Zenteno-Savin, 1997).

Similar to Greaves et al. (in press a), our findings suggest that refinement of the regulatory mechanisms, which brought about the foetal bradycardia observed by Bacon et al. (1985), continues after birth. Terrestrial apnoeic heart rate, like diving heart rate (Greaves et al., in press a), was bimodal with a trough around 100 beats min^{-1}. The high terrestrial apnoeic heart observed in this study was comparable to the ‘diving tachycardia’ (~150 beats min^{-1}), and the low terrestrial apnoeic heart rate equivalent to the ‘diving bradycardia’ (~70 beats min^{-1}), described by Greaves et al. (in press a) for pre-weaned harbour seal pups. In both studies, the decline in mean apnoeic heart rate was driven primarily not by a change in the mean value for each mode but by the proportion of time spent in each mode. Most likely an increase in vagal tone with age (Katona et al., 1980) enabled older animals to spend a greater proportion of time in the lower heart rate mode. In younger pups, the ability to attain a lower heart rate was periodic, whereas in older pups, this lower heart rate was reached but not strictly maintained, as evident from periodic escapes in vagal control to the higher mode. Greaves et al. (in press b) determined that young pups attempted to reduce their heart rate every 3–10 s during a dive. Consistent with Greaves et al. (in press a), by the time pups were weaned (~4 weeks of age), they exhibited a degree of cardiovascular control comparable to that of adults (Fig. 5).

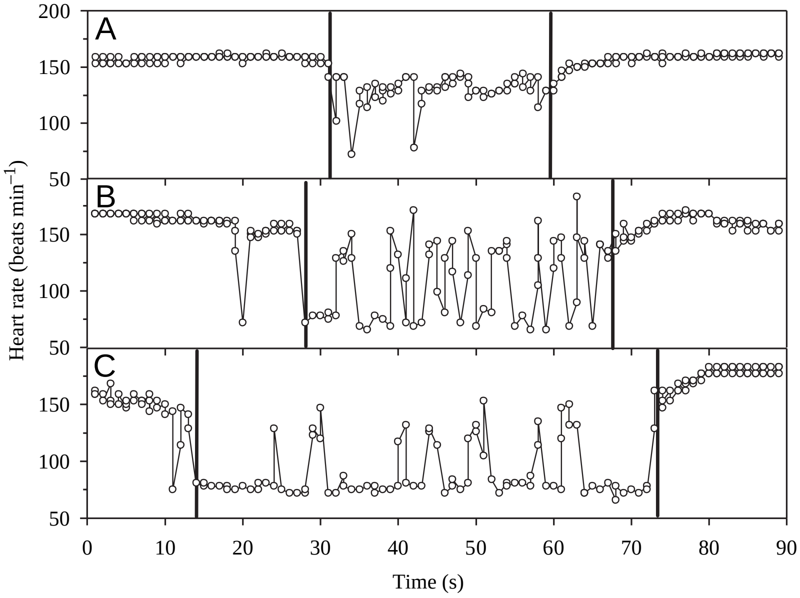

Fig. 7. Instantaneous heart rate trace of a eupnoea–apnoea–eupnoea transition in a (A) 4 day, (B) 16 day, and (C) 31 day-old quietly resting harbour seal pup. Vertical black lines represent the commencement and termination of the apnoea.

As first hypothesized by Castellini (1995), the rate of cardiorespiratory control development was quicker in the harbour seal, compared to elephant and Weddell seals. Although similar changes in cardiorespiratory patterns have been reported for these species (Table 1; Kenny, 1979; Blackwell and Le Boeuf, 1993; Castellini, 1996; Burns et al., 1996), the maturation of cardiorespiratory control mechanisms typically takes longer to develop because of their extended lactation and/or post-weaning fast periods (Castellini et al., 1994a; Burns et al., 1996; Castellini, 1996; Falabella et al., 1999). Unlike harbour seals that enter the water the day they are born (Newby, 1973) and begin foraging before (J.L.L., personal observation) or within days of weaning (Muelbert et al., 2003), northern elephant seals are essentially land bound for 4 months (Thorson and Le Boeuf, 1994). It is during this extended terrestrial fast that cardiorespiratory patterns stabilize (Castellini et al., 1994b; Castellini, 1996; Falabella et al., 1999). Weddell seals begin serious diving around 2 months of age, and as such exhibit a rate of development intermediate to that of harbour and elephant seals

Table 1. *Changes in eupnoeic and apnoeic heart rate with age in four phocid species*

				Heart rate[b] (beats min^{-1})		
Species	Age class[a]	Age	Mass (kg)	Eupnoeic	Apnoeic	Source
Harbour seal	NP	<2 weeks	6.5–10.5	138	84	1
	J	4 months–2 years	25–33	105–145	53–70	2, 3, 4, 5
	A	6 years	78–114	120–125	60–61	1, 6
Weddell seal	WP	6 weeks	106–130	94	46	7
	A		360–430[c]	55	45	8
Northern elephant seal	NP[d,e]	1→8 weeks	40→134	130→85	120→60	9, 10, 11, 12
	WP	8–16 weeks	74–95	84–94	47–54	7, 12
	J	10–34 months	117–198	65–73	45–54	11, 12, 13, 14
	A			55	44	15
Southern elephant seal	NP	<3 weeks		110	89	16
	WP	3–11 weeks		90	61	16
	A			65[f]	45[f]	17

[1]Bacon et al., 1985; [2]Dykes, 1974; [3]Påsche and Krog, 1980; [4]Jobsis et al., 2001; [5]Skinner and Milsom, 2001; [6]Jones et al., 1973; [7]Zenteno-Savin and Castellini, 1998; [8]Kooyman, 1968; [9]Huntley, 1984; [10]Castellini et al., 1994b; [11]Blackwell, 1996; [12]Kohin et al., 1999; [13]Bartholomew, 1954; [14]Andrews et al., 1997; [15]Castellini et al., 1994a; [16]Falabella et al., 1999; Hindell and Lea, 1998.

[a] NP, nursing pups; WP, weaned pups; J, juveniles; A, adults.

[b] Observed range of values (min–max) from various studies; developmental trends denoted by an arrow (→)

[c] Taken from Kooyman and Campbell (1972).

[d] Note: Bartholomew (1954) did not observe a difference between apnoeic and eupnoeic heart rate (~100 beats min^{-1}) in newborn pups.

[e] Also contains weaned, fasting pups.

[f] Surface and diving heart rate.

(Castellini, 1995, 1996). Despite their different developmental strategies, elephant, Weddell and harbour seal pups appear to be capable divers, physiologically, by the time they must begin to forage independently (Castellini et al., 1994b; Burns et al., 1996; Castellini, 1996; Falabella et al., 1999). Species that begin to forage independently prior to 1 month of age, such as the bearded seal (Gjertz et al., 2000), may show even more rapid cardiorespiratory control development.

Unfortunately, harbour seal pups could not be studied while resting in their natural habitat. Unlike other phocids that haul-out on land or ice for extended periods of time and can be approached relatively easily while sleeping (Castellini, 1991; Blackwell and Le Boeuf, 1993; Castellini et al., 1994b; Falabella et al., 1999), harbour seals haul-out on coastal reefs or isolated rocks that flood with the rising tides (Lesage et al., 1995). They are very vigilant and wary (Venables and Venables, 1955; Terhune and Brillant, 1996), fleeing to the water immediately upon approach (J.L.L., personal observation; Terhune and Almon, 1983; Henry and Hammill, 2001). In addition, harbour seal pups are extremely agile and capable of coordinated movement from a very young age (Dubé et al., 2003); therefore, restraint was necessary during our observations. However, we are confident that the effects of our experimental protocol were minimal, as many of the physiological measurements made in this study were quantitatively similar to those obtained by Greaves et al. (in press a) from free-ranging, diving harbour seal pups.

From electrophysiological studies on a variety of captive phocid species, it is apparent that prolonged terrestrial apnoeas are associated with sleep, and that profound cardiovascular and respiratory adjustments occur during the sleep cycle (Ridgway et al., 1975; Huntley, 1984; Castellini et al., 1994a; Skinner and Milsom, 2001). Lyamin et al. (1993) studied sleep in harp seal pups and found that there are developmental changes in sleep parameters. Therefore, in addition to the changes in cardiorespiratory patterns observed in harbour seal pups throughout this study, it is likely that sleep ontogenesis was also occurring during this time. Future studies incorporating both sleep state and apnoea physiology during this critical period of development would be of great interest, as Castellini (1996) has suggested.

In conclusion, developmental changes in the cardiorespiratory patterns associated with terrestrial apnoeas in harbour seal pups were comparable to those reported for elephant and Weddell seals (Castellini et al., 1994b; Castellini, 1996; Falabella et al., 1999). However, due to the early onset of independent foraging, maturation of cardiorespiratory control mechanisms occurred more rapidly in harbour seals. This parallels the development of diving activity (Greaves et al., in press a) and total body oxygen stores (Burns et al., 1999; Clark, 2004), which together enable harbour seal pups to be proficient divers and successful foragers upon weaning.

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