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RESEARCH ARTICLE

Acoustic measurements of post-dive cardiac responses in southern elephant seals (*Mirounga leonina*) during surfacing at sea

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ABSTRACT

Measuring physiological data in free-ranging marine mammals remains challenging, owing to their far-ranging foraging habitat. Yet, it is important to understand how these divers recover from effort expended underwater, as marine mammals can perform deep and recurrent dives. Among them, southern elephant seals (*Mirounga leonina*) are one of the most extreme divers, diving continuously at great depth and for long duration while travelling over large distances within the Southern Ocean. To determine how they manage post-dive recovery, we deployed hydrophones on four post-breeding female southern elephant seals. Cardiac data were extracted from sound recordings when the animal was at the surface, breathing. Mean heart rate at the surface was 102.4 ± 4.9 beats min^{-1} and seals spent on average 121 ± 20 s breathing. During these surface intervals, the instantaneous heart rate increased with time. Elephant seals are assumed to drastically slow their heart rate (bradycardia) while they are deep underwater, and increase it (tachycardia) during the ascent towards the surface. Our finding suggests that tachycardia continues while the animal stays breathing at the surface. Also, the measured mean heart rate at the surface was unrelated to the duration and swimming effort of the dive prior to the surface interval. Recovery (at the surface) after physical effort (underwater) appears to be related to the overall number of heart beats performed at the surface, and therefore total surface duration. Southern elephant seals recover from dives by adjusting the time spent at the surface rather than their heart rate.

KEY WORDS: Marine mammal, Heart rate, Dive recovery, Diving behaviour, Passive acoustic monitoring

INTRODUCTION

Diving marine mammals face a strong dilemma: their food resources are located at depth, while they need to restore oxygen supply at the surface. This specificity influences their behaviour because their breath-hold capability limits the time spent foraging. Hence, surfacing is essential to reconstitute oxygen stores by breathing and restoring oxygen levels in muscles and organs. Oxygen stores are higher for diving species than for non-diving ones (Butler and Jones, 1997) and are located in the blood and muscles

(Hassrick et al., 2010; Kooyman et al., 1983). Bradycardia is the common response to diving in marine mammals and diving seabirds (Ponganis, 2015), with, for instance, northern elephant seals, *Mirounga angustirostris*, reducing their heart rate by 64% (Andrews et al., 1997). Regulation of heart rate, cardiac output and the degree of vasoconstriction and blood circulation shutdown is critical to the management and utilisation of oxygen stores.

Measures of physiological data are essential to understand diving mammal metabolism (Butler and Jones, 1997). Ideally, physiological parameters should be recorded from free-ranging animals diving voluntarily (Webb et al., 1998). However, accessing physiological data *in situ*, like cardiac response at the surface, from free-ranging animals in the open ocean remains difficult. The pinnipeds share their time between the sea and the land or ice (Harrison and Kooyman, 1968). This bimodal cycle coupled to their large size makes them a unique system to study physiological adaptations to deep dives, because the deployment and recovery of loggers are eased on land (Costa et al., 2004). Kooyman et al. (1968, 1971, 1973) were the first to access physiological data and calculate metabolic rate using a respiratory chamber on free-ranging Weddell seals, *Leptonychotes weddellii*, with man-made ice-hole experiments under semi-natural conditions.

Because of difficulties in employing this technique *in situ* in other marine species, methods based on heart rate have been used as a reliable indicator of field metabolic costs (Butler et al., 2004; McPhee et al., 2003; Ropert-Coudert et al., 2012). For instance, Weimerskirch et al. (2000) successfully used heart rate as a proxy for energy expenditure and instantaneous effort in flying wandering albatrosses, with the highest heart frequencies observed while albatrosses were walking on land and taking off. This method also provides advantages over the doubly labelled water (DLW) method in pinnipeds, as it provides an estimation of metabolic rate during specific activities, such as a dive cycle (Butler et al., 2004). The electrical approach, which measures the electrical signal of the heart, is the most common way to record heart rate in free-ranging diving mammals (Ropert-Coudert et al., 2012; Webb et al., 1998).

Heart rate studies on elephant seals with the electrical method showed that during diving, they exhibited bradycardia. Heart rate rapidly decreased by 50–80% at the beginning of the dive and remained low while the seal was submerged (Andrews et al., 1997). Hindell and Lea (1998) recorded extreme bradycardia, with heart rate reaching 2 beats min^{-1} in 23 dives. Heart rate then increased gradually as the seal rose to the surface (Andrews et al., 1997). Bradycardia, apnoea and vasoconstriction of the peripheral system constitute the dive response in pinnipeds (Harrison and Kooyman, 1968). However, the electrical method requires the fixation of an electrode into the body, which can cause complications in the field (Ropert-Coudert et al., 2012).

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Fletcher et al. (1996), using an acoustic approach, provided the first record of respiratory rate at the surface of translocated northern elephant seals (*M. angustirostris*). Between breaths, putative heart beats were distinguished and cardiac frequency extracted. Several acoustic studies confirmed that this method could provide physiological data such as breath frequency or heart rate at the surface (Burgess et al., 1998; Génin et al., 2015; Le Boeuf et al., 2000).

The aim of this study was to investigate the cardiac response at the surface to active dives in free-ranging southern elephant seals. Southern elephant seals are a major predator in the Southern Ocean. At sea, they dive repeatedly to around 500 m during 20–30 min, with surface intervals lasting on average 2 min, but extreme depth records reached over 1800 m (Hindell et al., 1991; McConnell et al., 1992). They come back on land twice a year for mating during the southern spring, and moulting in the summer, with a high site fidelity (Fabiani et al., 2006).

Génin et al. (2015) have shown that the number of breaths is tightly related to surfacing time and mainly explained by dive duration and swimming effort made by southern elephant seals. Yet, in terms of recovery, cardiac function might play a major role. Our intention in this study was to explore the recovery behaviour of southern elephant seals through examining variation in heart frequency. First, we investigated how the instantaneous cardiac frequency evolves at the surface. Second, we studied the relationship between the mean cardiac frequency during the surface interval, and the dive duration and foraging effort performed by southern elephant seals during the previous dive.

MATERIALS AND METHODS
Ethics statement
All animals in this study were treated in accordance with the French Polar Institute (IPEV) ethical and Polar Environment Committee guidelines. All scientific procedures conducted on southern elephant seals had been validated beforehand.

Deployment of devices and data collection
This study is based on data collected from four post-breeding female southern elephant seals (Table 1): two in October 2011 and another two in October/November 2012 on Kerguelen Islands (49° 20′S, 70°20′E). Individuals were captured, then anaesthetised using a 1:1 combination of tiletamin and zolazepam (Zoletil 100), which was injected intravenously (0.8 mg 100 kg^{-1}; McMahon et al., 2000). They were then equipped with two devices, glued to the head or the back of the individual using quick-setting epoxy (Araldite AW 2101, Ciba) after cleaning the fur with acetone. First, an Argos-GPS satellite tag (Splash 10-F, Wildlife Computer, Redmond, WA, USA) was glued to the seal's head. It provided real-time position of the seals through the Argos system and also collected GPS location data. Second, an autonomous acoustic/ accelerometer/magnetometer and pressure logger (Acousonde™, model 3A; Acoustimetrics, Greeneridge Sciences, Inc., Santa Barbara, CA, USA) (Burgess, 2000; Burgess et al., 1998) was

fixed on the dorsal fur in the longitudinal axis, 10 cm behind the scapula. Each Acousonde™ recorded at a sampling frequency of 6.3 kHz in 2011 and 12.2 kHz in 2012, with an acoustic sampling resolution of 16 bits. This difference in sampling rates does not affect our study as cardiac and respiratory events occur in a frequency range of less than 1 kHz. To save battery power and storage space, the device was programmed to record sound for 3 h every 12 h in 2011 and for 4 h every 24 h in 2012. All devices provided measurements of time, location and depth at 1 Hz, as well as the three-dimensional magnetic field strength and acceleration at a 5 Hz frequency. The instruments sampled acoustic data until battery exhaustion, which occurred between 10 and 20 days after deployment. All devices were retrieved once individuals returned ashore to moult after their foraging trip in January/February following deployments. Seals were located on land using their Argos position.

Acoustic data processing
Detection of cardiac occurrences
When the animal is surfacing, the water flow noise produced by swimming ceases and most of the sound is due to breathing. Respiratory signals are contained at frequencies within the 0–700 Hz range. Between two respirations, spectrograms (time–frequency representation) showed putative cardiac occurrences (Fletcher et al., 1996; Le Boeuf et al., 2000). Heart sounds are expected to be dual because of the closure of the mitral and aortic valves (Burgess et al., 1998). The two sounds are indistinguishable, as they occur too close together in time. Hence, cardiac occurrences (a combination of the two valves' sounds) are brief and regular temporal impulsions at frequencies from 0 to 150 Hz (Burgess et al., 1998).

Acoustic recordings of surface intervals were visualised and analysed using the software Raven (The Cornell Lab of Ornithology – Bioacoustics Research Program) to generate a spectrogram for each surface interval. The same parameters were used for the computation of all spectrograms: a Hann-type window with a size of 512 samples, an overlap of 50% and a discrete Fourier transform calculated with 512 samples.

Each cardiac beat was determined using visual and auditory cues (Fig. 1). Hence, each occurrence is characterised by its temporal abscissa. When two beats are consecutive, an instantaneous cardiac
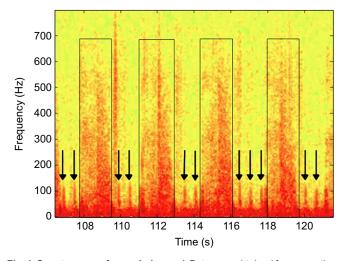

Fig. 1. Spectrogram of recorded sound. Data were obtained from a southern elephant seal (seal 1) during a surface interval, and show four respiratory cycles (rectangles in the 0–700 Hz range) with cardiac occurrences between breaths (black arrows in the 0–150 Hz range).

Table 1. Descriptive information for the four post-breeding female southern elephant seals at deployment

Seal	Acousonde ID	Body mass (kg)	Size (cm)	Departure date
1	A031	255	254	10 Oct. 2011
2	A032	245	238	28 Oct. 2011
3	626019	230	232	28 Oct. 2012
4	626040	292	225	01 Nov. 2012

frequency ($f_{H,inst}$, beats min^{-1}) is calculated using the temporal difference between the two occurrences (Eqn 1):

$$f_{H,inst} = \frac{60}{x_{t+1} - x_t} \, .$$
(1)

Some surface intervals contained high-amplitude noise from external sources (water splashes possibly due to rough weather conditions or due to certain animal behaviours such as grooming). A noisy acoustic environment prevents access to heart data. As such, a surface interval was only kept for further analyses when a minimum of five cardiac beats were measured, so that the cardiac frequency could be estimated with confidence. Cardiac frequency for a surface interval (f_H) was computed as the average of instantaneous heart frequency ($f_{H,inst}$; Eqn 1) during each surface interval.

Surface interval duration

The beginning, end and duration of each surface interval, to the nearest second, were determined using visual and auditory cues. A surface interval begins with the first respiration and ends with the last one.

Dive cycle

A dive cycle is composed of a dive followed by a surface interval. Southern elephant seals were considered to be diving when they reached depths greater than 15 m, to avoid considering subsurface movements as dives. Each dive was divided into three phases: descent, bottom and ascent. Each phase was determined using a vertical speed criterion. Vertical speed was modelled with a fourth-order polynomial function adjusted to instantaneous vertical speed using a custom-written Matlab code (Matlab software 8.1, The MathWorks, Natick, MA, USA). Ascent and descent phases were identified as periods before or after surfacing where the modelled vertical speed exceeded 0.75 m s^{-1}. Bottom phases were identified as periods between ascent and descent phases where the modelled vertical speed remained below 0.75 m s^{-1} (Jouma'a et al., 2016; Vacquié-Garcia et al., 2015).

Dive parameters

For each surface interval where f_H (beats min^{-1}) was measured, data from the previous dive were extracted from the Acousonde™ at 5 Hz resolution. Statistics on each dive were then calculated to give: maximum depth reached (m), dive duration (min), descent, bottom and ascent duration (min) and the location (latitude and longitude) when the southern elephant seal reached the surface. Elephant seals perform dives where they passively descend through the water column over a long period of time (Crocker et al., 1997). These passive 'drift' dives were identified based on the method designed by Dragon et al. (2012) using the package 'rbl' (https://github.com/SESman/rbl) in R. A dive was considered to be a passive drift dive when passive phases were detected with the following parameters: a minimum duration of 50 s, an absolute roll greater than 90 deg and a drift rate (i.e. an absolute vertical speed) ranging between −0.4 and 0.6 m s^{-1}.

Another important dive parameter is total acceleration. It can be divided into two types: static and dynamic. Static acceleration is caused by the Earth's gravitational pull whereas dynamic acceleration results from the animal's movements (body waves, tail strokes, head motions). The static component corresponds to low frequencies and the dynamic one to higher frequencies (Génin et al., 2015; Richard et al., 2014).

Mean swimming effort index

Data provided by the accelerometers were used to calculate absolute and mean swimming effort index for each dive phase and for the entire dive (Eqn 2). The lateral axis of the accelerometer contains information on putative turning and rolling movements (static acceleration) and on flipper stroke (dynamic acceleration) (Richard et al., 2014). Swimming effort was obtained by summing the absolute values of the local extrema of the lateral axis of the acceleration filtered with a band-pass filter which cuts frequencies bellow 0.44 Hz and above 1 Hz (Jouma'a et al., 2016; Richard et al., 2014). Mean swimming effort index of the dive was then calculated by dividing the absolute swimming effort by the duration (Δt) spent underwater:

$$\text{Mean swimming effort index} = \frac{\Sigma \, \text{peaks}}{\Delta t} \, .$$
(2)

Putative capture rate

Putative capture rate (s^{-1}) is the number of prey encounter events (PEEs) divided by the bottom duration of a dive. PEEs were extracted from the acceleration signal which contained head movements by adapting the method developed by Viviant et al. (2009): on each axis, a high-pass filter with a cut-off frequency of 0.33 Hz was applied. Standard deviation was then calculated with a 1 s fixed window and then a 5 s moving window. Significant peaks in this filtered signal were considered as PEEs when they were detected simultaneously on all three axes (for details, see Vacquié-Garcia et al., 2015).

Data design and statistical analyses

Analyses were conducted at two scales: surface interval level and dive cycle level. For the surface interval scale, the statistical unit is a measure of $f_{H,inst}$ associated with temporal abscissa in seconds (0 corresponding to the beginning of the surface interval). For the dive cycle scale, the statistical unit is a dive cycle. We aimed to explain the mean f_H measured at the surface using dive and surface interval parameters.

$f_{H,inst}$ at the surface

The aim of this part of the experiment was to study the evolution of $f_{H,inst}$ within a surface interval. The effect of time on $f_{H,inst}$ was examined using linear regression (lm function in 'stats' R package; R Development Core Team, 2015). The regressions were conducted for each surface interval of each individual. First-order effects were selected over second-order effects based on an Akaike information criterion (AIC) selection (Zuur, 2009). Estimated slopes were used to determine the nature of $f_{H,inst}$ variations. Then, a linear mixed model computed with the 'nlme' package (version 3.1-122, https://CRAN.R-project.org/package=nlme) was run on all the surface intervals of the four individuals at the same time to explore the importance of time for $f_{H,inst}$ (standardised values; n=2978). Individual was set as a random factor. To take into account temporal correlation, an auto-correlation structure was included in our models, using an auto-regressive correlation structure of order 1 (with the function corAR1 from the 'nlme' package, version 3.1-122; https://CRAN.R-project.org/package=nlme). The most relevant model, between random effect model, random intercept model or without random effect model, was selected based on AIC (Zuur, 2009).

Mean f_H and number of beats at the surface

To investigate the contribution of previous diving behaviours to mean f_H at the surface and the number of cardiac beats at the surface, we used linear mixed-effects models, also from the package 'nlme'.

Table 2. Surface interval parameters

Seal	No. of dive cycles	Surface interval (s)	f_H (beats min^{-1})
1	102	132±19	102.4±4.6
2	85	107±13	101.0±3.7
3	70	122±13	105.7±5.2
4	28	121±18	99.0±4.7

f_H, heart rate. Data are means±s.d.

Values outside the 1.5 interquartile range were removed from the data. All explanatory variables were centred and standardised at the population scale to keep individual differences and allow comparisons between slope estimates. Time (in days) was included in the model as we assumed a possible impact on the mean f_H. To test the linear and quadratic effects of time, both variables were included in the model. Individual was set as a random factor, and an auto-regressive correlation structure was included (corAR1). As previously, the most parsimonious model, between a random effect model, random intercept model or without random effect model, was selected based on AIC (Zuur, 2009). The same models and same protocol were used to explain the number of beats at the surface.

All statistical analyses were conducted using the R software package (R Development Core Team, 2015). For each model, normal distributions of the explained variable and of the residuals, and homogeneity of residuals were checked up. All results are expressed as means±s.d. for single parameters. The significance level was set at $P=0.05$.

RESULTS

Foraging trips and overall diving behaviour

Each southern elephant seal travelled eastward of Kerguelen Islands. Acousondes™ provided data for the first days of foraging trips. We obtained 296 h of sound recorded in 84 files, of which 15 files (53 h) were immediately put aside because the animal was still on land or data were too bad to be exploited. Of the 243 h left, there were 688 dive cycles and we kept the cycles that counted more than five heart beats: 284 dive cycles were kept for this study. On average, seals dived for 18.4±3.7 min with a mean depth of 546±159 m. Time spent at the surface recovering averaged 121±19 s (i.e. 2 min 1 s; Table 2) with a maximum of 208 s (i.e. 3 min 28 s). Consequently, seals were submerged on average 90.1%
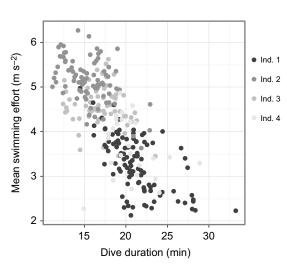
Table 3. Dive parameters of the four female southern elephant seals

Seal	Maximum dive depth (m)	Dive duration (min)	No. of PEEs	Putative capture rate (s^{-1})	Mean swimming effort index (m s^{-2})
1	611.5±164.4	21.2±3.1	4.9±3.4	0.015±0.023	3.3±0.6
2	434.9±114.9	15.5±2.5	6.4±5.0	0.014±0.011	5.2±0.5
3	575.3±138.5	17.0±2.4	8.9±5.4	0.026±0.020	4.6±0.4
4	579.7±128.2	20.5±2.8	6.6±5.7	0.015±0.015	3.7±0.7

PEEs, prey encounter events. Data are means±s.d.

of the time, ranging from 89.3% for seal 3 to 91.1% for seal 4. The four individuals showed differences in their diving strategies. Seal 1 performed deep long dives whereas seal 2 performed shallower and shorter dives. Seal 3 had a greater number of PEEs and a higher putative capture rate compared with the other three (Table 3). The longest (33 min 12 s) and the deepest (938.2 m) dives were both performed by seal 1.

There was a strong negative relationship between dive duration and mean swimming effort index per dive across the four individuals (Pearson's correlation coefficient=−0.74, $P\leq0.001$; Fig. 2).

$f_{H,inst}$ during surface intervals

There was a positive relationship between $f_{H,inst}$ and time in more than 90% of the 284 dives. Mean R^2 (the percentage of the variance explained by the models) was 0.32±0.26 with a minimum of 6×10^{-5} and a maximum of 0.97. Linear models showed that $f_{H,inst}$ increased significantly with time spent at the surface (Fig. 3, estimate=0.009± 0.001, $t=7.41$, $P\leq0.001$) with no individual effect.

Mean f_H at the surface and underwater

At the surface, mean f_H was 102.4±4.9 beats min^{-1} with significant differences between seals (Kruskal–Wallis test: $\chi^2_3=42.8$, $P\leq0.001$; Table 2). The mean f_H for individual seals ranged from 99.0±4.7 (seal 4) to 105.7±5.2 beats min^{-1} (seal 3) while they were breathing at the surface.

In most dives, flow noise generated by seal movements prevented the detection of heart beats. However, quiet recording conditions observed during the passive drift phases allowed detection of beats (e.g. short impulse signals) in the frequency range between 0 and
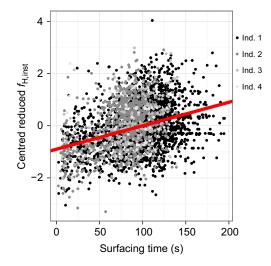
Fig. 2. Relationship between dive duration and mean swimming effort for the four southern elephant seals studied.

Fig. 3. Linear relationship between time and instantaneous cardiac frequency ($f_{H,inst}$) for the four seals. $f_{H,inst}=-0.9+0.009\times$surfacing time.

Table 4. Results of linear models looking at mean surface f_H or the number of beats in relation to dive parameters and time spent at sea

Model	Parameter	Estimated coefficient (estimate±s.d.)	P-value
$f_H \sim$	Intercept	94.2±1.5	≤0.001
(GLS model	Mean swimming effort (m s^{-2})		NS
correlation $\sim$ time (s) | individual)	Dive duration (min)		NS
	Putative capture rate (s^{-1})	0.3±0.1	0.02
	Time (days)	1.5±0.4	≤0.001
	Time^2	−0.05±0.02	0.02
No. of beats $\sim$	Intercept	225.2±22.5	≤0.001
(LME model	Mean swimming effort (m s^{-2})	20.4±8.4	0.01
random $\sim$ slope | individual	Dive duration (min)	14.9±7.5	0.04
correlation $\sim$ time (s) | individual)	Putative capture rate (s^{-1})		NS
	Time (days)		NS
	Time^2		NS

GLS, generalized least squares; LME, linear mixed-effects.
Data are means±s.d. NS, not significant.

40 Hz, which may be attributed to heart beats. Drift dives were mainly observed in seal 1. The low-frequency pattern appeared during the whole passive drift event. A simple calculation of the frequency of occurrence based on drift events exhibited a mean of 20.2±5.1 beats min^{-1}, which represents an 80.3% reduction in f_H compared with surface measurements for that individual.

Mean surface f_H in relation to dive parameters
The most appropriate model in order to explain mean f_H at the surface with dive parameters and time was the one without individual effect. The mean surface f_H was found to be positively correlated with both the number of days elapsed since departure from Kerguelen Island and the putative capture rate (Table 4). The quadratic time term significantly contributed to changes in the relationship. Its estimated coefficient was negative, which means that the relationship between f_H and time was directed towards a concave shape. Therefore, f_H increased with time spent at sea, followed by a 'plateau' effect
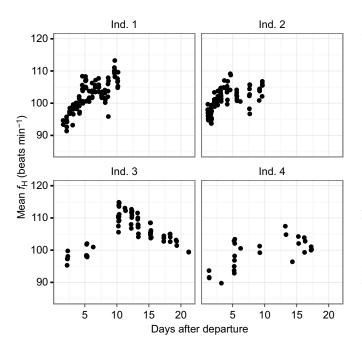

Fig. 4. Relationship between time spent at sea and mean heart rate (f_H) for the four seals. Mean f_H measured during the post-dive interval at the surface is plotted against time spent at sea after the breeding season on Kerguelen Island.

(Fig. 4). Dive duration and swimming effort did not influence f_H at the surface for the seals studied (Table 4).

Total number of heart beats at the surface in relation to dive parameters
To explore the variation in the number of beats counted during surface intervals, we used the same explanatory variables as above. As expected, a strong correlation between the number of heart beats and surface duration was found (Pearson's coefficient=0.96, $P \leq 0.001$). In this case, there were random effects (on the slope and the intercept) across individuals. The total number of heart beats during the surface interval was positively related to both the mean swimming effort index and dive duration. However, the total number of cardiac beats at the surface was unrelated to putative prey capture rate and time (Table 4).

DISCUSSION
Measuring f_H through acoustic records
This study provides one of the very few datasets of f_H simultaneously with breathing rate (Génin et al., 2015) for free-ranging post-breeding female southern elephant seals. Previous studies carried out on f_H used mainly captive or translocated animals (Andrews et al., 1997; Burgess et al., 1998; Fletcher et al., 1996; Le Boeuf et al., 2000). Acoustic records offer the possibility of accessing free-ranging southern elephant seal f_H during post-dive surface intervals, although records show heart sounds underwater only when the flow noise stops (Burgess et al., 1998). This condition of quiet soundscape is satisfied when southern elephant seals are passively drifting through the water column. Therefore, f_H could not be quantified while the seals were actively swimming or gliding underwater because of the associated flow noise. In our study, mean f_H at the surface measured in the four voluntarily diving post-breeding females was 102.4±4.9 beats min^{-1}. With northern elephant seals, previous acoustic studies recorded a mean f_H at the surface of 86 beats min^{-1} for adult males (Le Boeuf et al., 2000) and a range of 106–121 beats min^{-1} for juveniles (Andrews et al., 1997; Burgess et al., 1998; Fletcher et al., 1996; Le Boeuf et al., 2000). All these studies demonstrate the reliability of the acoustic method to analyse the cardiovascular system. In our data, f_H was detected only at the surface. Hindell and Lea (1998), using an electrical approach, extracted f_H at the surface of one post-breeding female southern elephant seal over a 50 day period, and found a f_H ranging between 65 and 95 beats min^{-1}. They estimated that this value was underestimated by 10–15% as a result of sampling bias. Hence, the two measures are of the same order of magnitude.

The electrical method also allowed the detection of heart beats while the southern elephant seal was swimming underwater. During a dive, an elephant seal exhibits pronounced bradycardia (Elsner et al., 1966), a finding confirmed in free-ranging seals (Burgess et al., 1998). f_H during diving of free-ranging southern elephant seals decreases from 40 beats min^{-1} for dives less than 13 min to 14 beats min^{-1} for dives lasting between 13 and 37 min (Hindell and Lea, 1998). In this study, we estimated f_H during one drift dive to be 20.2±5.1 beats min^{-1}, which is consistent with the values found by Hindell and Lea (1998; see their fig. 3) and represents an 80% reduction of f_H relative to the surface value, which tends to be more important, compared with the mean 64% reduction found in northern elephant seals for all dives combined (Andrews et al., 1997). This higher reduction might be related either to drift dives, which are assumed to represent a recovery behaviour (Crocker et al., 1997), or possibly to the longest duration of drift dives, which are on average 25% longer than foraging/travelling dives (C.G., unpublished data). The high standard deviation calculated here indicates a high variation between inter-beat intervals, suggesting that bradycardia might be unstable at depth. This is consistent with the hypothesis of Williams et al. (2015), who found that both depth and exertion distort bradycardia in Weddell seals (*L. weddellii*) and bottlenose dolphins (*Tursiops truncatus*). However, this diving f_H obtained by acoustics has to be validated in laboratory condition, using electrocardiogram methods, for example.

Post-dive recovery and $f_{H,inst}$

During surface interval periods, $f_{H,inst}$ increases with time: it is significantly higher at the end of the surface interval than at the beginning. Additionally, all seals exhibited the same pattern. This result probably represents a part of the dive response of the southern elephant seal. Indeed, after reaching very low values when the seal chases at the bottom of its dive, f_H increases gradually while the seal ascends towards the surface (Harrison and Kooyman, 1968). Andrews et al. (1997) found that the rate of increase of tachycardia was most marked just prior to surfacing, during approximately the last 15 s of ascent. Therefore, increasing f_H observed during the surface interval in this study is likely to correspond to the decelerating phase of the tachycardia, which reaches its maximum value at the end of the surface interval, prior to diving. Periods of tachycardia enable rapid oxygen loading at the surface, in both blood and muscle stores, and elimination of carbon dioxide accumulated during the previous dive (Reed et al., 1994). Associated with high breathing frequency, high f_H eases quick gas exchange at the surface and a more efficient recovery (Fedak et al., 1988; Le Boeuf et al., 2000). Hence, surface duration is minimised and submergence times are maximised. Indeed, the four seals studied here spent about 90% of their time underwater, enabling this central place predator to take full advantage of its underwater prey.

Post-dive recovery and physical effort exerted by the southern elephant seal

The costs associated with diving are a central component of a marine mammal's energy budget (Maresh et al., 2015). This budget can be divided into oxygen-consuming additive elements: basal metabolic costs, locomotor costs, feeding costs and thermoregulatory costs (Costa and Williams, 1999). In this study, exertion levels during diving were evaluated via three parameters: dive duration, mean swimming effort index and putative capture rate. Contrary to our expectation that, as in terrestrial mammals, f_H would increase with increasing foraging effort, no relationship was found between mean f_H at the surface and dive duration or

swimming effort. However, a positive relationship was found with both putative prey capture rate and the number of days elapsed since departure from Kerguelen Islands. Dive duration may not reflect the exertion level during a dive as it could be biased because southern elephant seals reduce their relative mean swimming effort with increasing dive duration (Fig. 2). Swimming effort, i.e. movements of the hind flippers, appears to be a reliable indicator of costs due to locomotion (Williams et al., 2004; Wilson et al., 2006). Putative capture rate can easily be linked to foraging costs during a dive. Feeding events are responsible for an increase of 44.7% of the energetic budget in Weddell seals (Williams et al., 2004). Nevertheless, interpretations should be made with caution as putative capture rate is calculated from prey capture attempts and not effective catches. Indeed, around 90% of the dive cycles analysed in this study had at least one capture attempt (failed or successful).

Our results indicate that southern elephant seals manage their recovery by increasing the duration of the post-dive interval and therefore by increasing the total number of cardiac beats (i.e. the duration of the tachycardia) rather than acting on f_H while at the surface. Indeed, the total number of heart beats was highly correlated with the time spent at the surface, breathing. Dive duration and mean swimming effort were the two parameters that positively influenced time spent at the surface. Hence, a long dive and/or a dive where the southern elephant seal gave a high quantity of large tail movements implies a long surface duration. This is in accordance with previous results obtained by Génin et al. (2015) with southern elephant seals. In Weddell seals, *L. weddellii*, the energy expenditure approximated by the number of flipper strokes taken is highly correlated with oxygen consumption (Williams et al., 2004). Maresh et al. (2014) showed that with an artificially increased cost of locomotion, northern elephant seals, *M. angustirostris*, spent more time breathing and thus recovering. The relationship between dive duration and surface interval duration has previously been demonstrated in several marine mammal species such as northern elephant seals (Andrews et al., 1997; Le Boeuf et al., 2000) and grey seals, *Halichoerus grypus* (Thompson and Fedak, 1993), alongside diving birds including thick-billed murres, *Uria lomiva* (Croll et al., 1992).

Putative capture rate positively influenced mean f_H at the surface. This result might indicate that seals recover from a foraging dive with a higher f_H. Hence, the increase of surface mean f_H after putative prey capture attempts could be explained by factors unrelated to swimming effort, such as the added energy required for prey warming and digestion. This hypothesis is consistent as prey assimilation affects both resting and diving metabolic rate (Williams et al., 2004).

This study strongly supports that the time spent at the surface and therefore the total number of breaths and heart beats, rather than f_H, appears to be the main driver of the post-dive recovery behaviour in southern elephant seals. As such, the cardio-respiratory system as a whole needs to be considered to understand the southern elephant seal recovery strategy. Nonetheless, it is critical to bear in mind the complexity of the cardiac responses observed: cardiac regulation is controlled by neural drivers which themselves react to multiple factors such as the environment or a change in behaviour (Williams et al., 2015).

This study also revealed that the mean f_H at the surface for the four seals varied with the time spent at sea with a non-linear relationship (Fig. 4). In the first 5–10 days of trip, the mean f_H at the surface increased with time. The mean f_H then might decrease (seal 3) or remain stable (seal 1, 2 and 4). Females left Kerguelen Island after the breeding season in poor body condition as they had lost 25–50%

of their original mass (McCann et al., 1989). Increasing mean f_H could reflect the adaptation or a response to cardio-vascular training during the first few days spent at sea after 1 month on land. By assuming that the higher the f_H is during surface breathing, the faster gas exchange should be, leading to a more efficient recovery, we suppose that an 'optimal' f_H exists (linked with individual characteristics such as body mass or composition) to maximise gas exchange at the surface. Therefore, future analyses with access to larger datasets and more individuals observed over a longer time period should be able to investigate this hypothesis.

Conclusion

Our findings indicate that southern elephant seals manage their post-dive recovery by modulating the post-dive surface duration, and therefore the number of breaths and heart beats, rather than through changes in their breathing rate (Génin et al., 2015) or their f_H (this study).

Sound recording can be a powerful tool as it provides the simultaneous detection of breathing and f_H, allowing investigation of the cardio-respiratory system in its entirety (Génin et al., 2015; Le Boeuf et al., 2000). Both sets of physiological data are essential to study post-dive recovery of marine mammals and seabirds. However, the main limitation is that we access f_H only when southern elephant seals are breathing at the surface. A study on harbour seals (*Phoca vitulina*) suggested that the mean f_H of the complete dive cycle (i.e. dive and surface) could be easily explained by the percentage dive time and links to oxygen consumption (Fedak et al., 1988). An improvement in data collection is essential to fully exploit the possibilities of the acoustic method. A major breakthrough would be to trigger audio recording based on external events of interest (e.g. using acceleration data). This would save battery and allow long-term datasets to be obtained. Examples of interest include recording acoustic data when the animal is at the surface (to study surface f_H) and/or during drift dives (to study underwater f_H).

In addition, sound records can be used to explore other aspects of elephant seal behaviour and environment. They allow the collection of abiotic sounds, such as those generated by wind and rain, which are of great interest to oceanographers as the Southern Ocean is difficult to observe. Acoustics offer many possibilities, and non-invasive bio-logging data collection could easily be improved by cooperation between users and research teams in the future.

Author contributions
Conceptualisation: C.G.; Methodology: L.D., J.J. and J.B.; Validation: J.B. and J.J.; Formal analysis and investigation: L.D. and J.J.; Writing – original draft preparation: L.D; Writing – review and editing: L.D., J.J., C.G. and J.B.; Resources: C.G. and J.B.; Supervision: C.G. and J.B.; Funding acquisition: C.G.

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