

## SYMPOSIUM

### Do Bar-Headed Geese Train for High Altitude Flights?

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**Synopsis** Exercise at high altitude is extremely challenging, largely due to hypobaric hypoxia (low oxygen levels brought about by low air pressure). In humans, the maximal rate of oxygen consumption decreases with increasing altitude, supporting progressively poorer performance. Bar-headed geese (*Anser indicus*) are renowned high altitude migrants and, although they appear to minimize altitude during migration where possible, they must fly over the Tibetan Plateau (mean altitude 4800 m) for much of their annual migration. This requires considerable cardiovascular effort, but no study has assessed the extent to which bar-headed geese may train prior to migration for long distances, or for high altitudes. Using implanted loggers that recorded heart rate, acceleration, pressure, and temperature, we found no evidence of training for migration in bar-headed geese. Geese showed no significant change in summed activity per day or maximal activity per day. There was also no significant change in maximum heart rate per day or minimum resting heart rate, which may be evidence of an increase in cardiac stroke volume if all other variables were to remain the same. We discuss the strategies used by bar-headed geese in the context of training undertaken by human mountaineers when preparing for high altitude, noting the differences between their respective cardiovascular physiology.

#### Introduction

Migratory birds undertake some of the most challenging feats of exercise of any vertebrate animals on the planet. For example, arctic terns (*Sterna paradisaea*) have been recorded making annual migration round trips of >80,000 km (Egevang et al. 2010), bar-tailed godwits (*Limosa lapponica*) have been recorded making non-stop flights of 11,000 km in a

little more than a week (Gill et al. 2009), and great snipe (*Gallinago media*) can maintain flight speeds of 80 km/h for several days (Klaassen et al. 2011). Flight is one of the cheapest forms of locomotion per unit of distance, but flapping flight is extremely costly per unit of time, requiring 10-times or more the resting basal metabolic rate (Bishop 1999; Butler 2016). Some large birds (such as raptors) have therefore

evolved behaviors such as soaring, gliding, and thermalling to reduce flight costs, requiring them to flap very little to remain aloft (Duriez et al. 2014; Sherub et al. 2016; Nourani and Yamaguchi 2017). Avian flight muscle is consequently highly adapted, and has two- to five-times greater mitochondrial density than most mammalian skeletal muscle, leading to a much higher mass specific ATP turnover rate (Kuzmiak-Glancy and Willis 2014). Avian flight muscle also generates the highest mass specific power outputs (up to 433 W/kg) of any cyclically contracting vertebrate muscle (Askew and Marsh 2002).

Some species of birds not only migrate long distances but also travel to high altitudes en-route to their breeding or wintering grounds (Prins and Namgail 2017). As altitude increases, barometric pressure decreases, leading to reduced partial pressure of oxygen and consequently to hypoxic conditions. For example, at the mean altitude of the Tibetan Plateau (mean altitude ~4800 m), there is ~50% of the oxygen at sea level (Cerretelli 1976). The challenge of supplying sufficient oxygen for aerobic metabolism in flight is therefore magnified. Bar-headed geese (*Anser indicus*) migrate across the Tibetan Plateau, and while flying northwards in spring make the fastest recorded sustained rates of climb over the Himalayas, flying from approximately sea-level to 4500 m altitude in under 8 h, with no opportunity to acclimatize (Hawkes et al. 2011). By contrast, human athletes show reduced metabolic rates and increased lactate production at altitude (Chapman et al. 2015; Smith et al. 2015), with  $\dot{V}O_{2MAX}$  declining by about 6–8% per 1000 m of altitude (Wehrlein et al. 2016). One might therefore imagine, if considering the context of human climbers, that bar-headed geese would undergo considerable preparation for their annual migration not only in training for distance but also for altitude exposure (but see Tucker 1968).

### Training in humans

In humans, athletic training usually involves either endurance exercise (e.g., long distance running or cycling), exercise for varying durations close to  $\dot{V}O_{2MAX}$  (the maximal sustainable rate of oxygen consumption), or short, intense “explosive strength training” sessions. Such training exercise produces a variety of physiological responses that enhance capacity for efficient work output and use of oxygen (Jones and Carter 2000, Joyner and Coyle 2008; Hellsten and Nyberg 2011; Hoffman 2014). For example, training can increase the volume and thickness of the left ventricle of the heart,

leading to an increase in stroke volume (the amount of blood ejected per heart beat) (Weiner et al. 2015). Training can increase blood hemoglobin content, red cell count, and plasma volume (Mairbäurl 2013) and can decrease lactate production during exercise and increase the rate of lactate removal when it is produced (Jones and Carter 2000).

Training can also alter the relative proportions of muscle fiber types, increase the storage of intramuscular lipids, and increase their relative use during exercise (Dubé et al. 2016) as well as increase muscle capillarity and myoglobin concentration (Jones and Carter 2000). Finally, it can increase the size, number, and function of mitochondria (Bishop et al. 2014). Human training for high altitude usually involves repeated exposure to hypobaria (either simulated or at moderate altitudes), which produces a range of physiological adaptations for enhancing oxygen supply to the tissues (e.g., erythropoiesis, increase in ventilation and heart rate, Bärtsch and Saltin 2008; increase in mitochondrial volume density, Jacobs et al. 2016). Most mountaineering expeditions to very high altitude acclimatize over a series of staged ascents over several weeks, recovering at a base camp in between. Training and acclimatization therefore has a significant impact on the physiological capacity for exercise and high altitude tolerance in humans.

### Training in non-human animals

Undoubtedly, selection pressures on wild animals to be optimally physically fit in their respective environments are very different from those acting on modern, largely sedentary humans (Caldwell 2016). Wild animals must constantly be able to forage efficiently and evade predation, and for many species the ability to move between seasonally changing habitats is key to survival (Chapman et al. 2014). For the purposes of the present study, therefore, the context of human training is unrealistic, but there is no alternate framework for how to improve physical fitness without exercise training. It would appear that there is no existing evidence to suggest that any wild animals undertake voluntary activity to “keep fit” (Portugal et al. 2011; Halsey 2016), but there are observations of wild mice (and even a wild frog) running freely on exercise wheels when they are available to them (Meijer and Robbers 2014). Captive mice and rats, subjected to training, have shown similar improvements in exercise capacity to those detailed above for humans (Kemi et al. 2004; Waters et al. 2004; Goutianos et al. 2015). Butler and Turner (1988) trained captive tufted ducks (*Aythya*

*fuligula*) to swim on a water flume in captivity and showed that training enhanced muscle capillarity and reduced heart rate for a given rate of oxygen consumption, which suggests an accompanying increase in cardiac stroke volume. Training, therefore, clearly produces an effect in non-human animals. There is also evidence that wild animals can respond to high altitude exposure. For example, lowland resident deer mice (*Peromyscus maniculatus*) exposed to hypoxia equivalent to 4300 m for 6–10 weeks increased their tidal volume (Ivy and Scott 2015), increased mitochondrial respiratory capacity and decreased the production of reactive oxygen species (ROS; Mahalingam et al. 2017). Given that migratory animals probably need to increase their physical fitness, from sprint-burst type fitness sufficient for chasing prey and evading predators, to endurance type fitness required to support long-distance migration, it may seem surprising, at least from an anthropomorphic point of view, that there is not more evidence for training in wild animals.

### Training for migration in birds

The phenomenon of “*zugunruhe*” (migratory restlessness) has been well described in captive birds (Gwinner 1968). At times of the year when birds in captivity would otherwise be migrating, they can exhibit agitated movements in specific directions that correspond to their natural migratory pathways, providing an opportunity for study (for a review and new directions, see Muheim et al. (2014)). However, in the wild songbirds apparently switch into migratory mode without previous locomotory preparations (Zúñiga et al. 2016). At the same time as this restlessness, avian muscle also undergoes significant biochemical changes, for example, increase in fatty acid binding proteins (FABP; improving transport and delivery of fatty acids to flight muscles), and citrate synthase activity (which is indicative of muscle oxidative capacity) (Saunders and Klemm 1994; Guglielmo et al. 2002). Several studies have provided evidence to suggest that such changes are likely endogenous and not brought about by training (Bishop et al. 1998; Dietz et al. 1999; Bauchinger and Biebach 2006; Vézina et al. 2007; McFarlan et al. 2009; Portugal et al. 2009; Price et al. 2011). It has also been suggested, however, that flight muscle hypertrophy in wild birds may be brought about as a consequence of pre-migratory mass gain which would increase wing loading and therefore the relative workload in flight (so called “power training”, Marsh and Storer (1981)). There is evidence for flight muscle hypertrophy from a number of studies

on captive birds (Marsh and Storer 1981; Marsh 1984; Butler and Turner 1988; Lindstrom et al. 2000).

For wild birds, the picture is less clear. In eared grebes (*Podiceps nigricollis*), an increase in body mass does not appear to be correlated with flight muscle hypertrophy, and “conspicuous flapping exercise” has been noted during recovery from wing moult, which might represent training (Gaunt et al. 1990). Bishop et al. (1998) demonstrated that citrate synthase activity in the pectoral muscles of barnacle geese (*Branta leucopsis*) was higher in wild than captive barnacle geese, and Pelters et al. (1999) demonstrated that FABP was higher in wild than captive gosling and adult barnacle geese, suggesting that some element of activity in the wild might be important in muscle hypertrophy, aerobic capacity, and fueling. The advent of biologging technology (Kays et al. 2015) has enabled researchers to record movement and aspects of animal biology and that can be used to address the question of training for migration in birds. Using implanted heart rate loggers, Portugal et al. (2011) showed that wild barnacle geese at their summer breeding area in Spitsbergen did not appear to increase the daily amount of time (22 min) spent flying in the weeks leading up to migration and therefore were likely not training for migration. Their work remains the only study to date on wild migratory birds.

In the present study, we set out to investigate whether there was any evidence of training for migration in a population of bar-headed geese deployed from nesting grounds (elevation 2050 m) in Mongolia. Bar-headed geese make one of the most demanding migrations on earth, crossing the Himalayan mountains and the Tibetan Plateau (mean elevation 4800 m) as they make their migration between India, China, and Mongolia (Hawkes et al. 2011, 2012; Bishop et al. 2015). We used implanted loggers to look for patterns of changes in heart rate that might suggest increases in physiological fitness and, for the first time, acceleration loggers to look for patterns in activity per day leading up to migration.

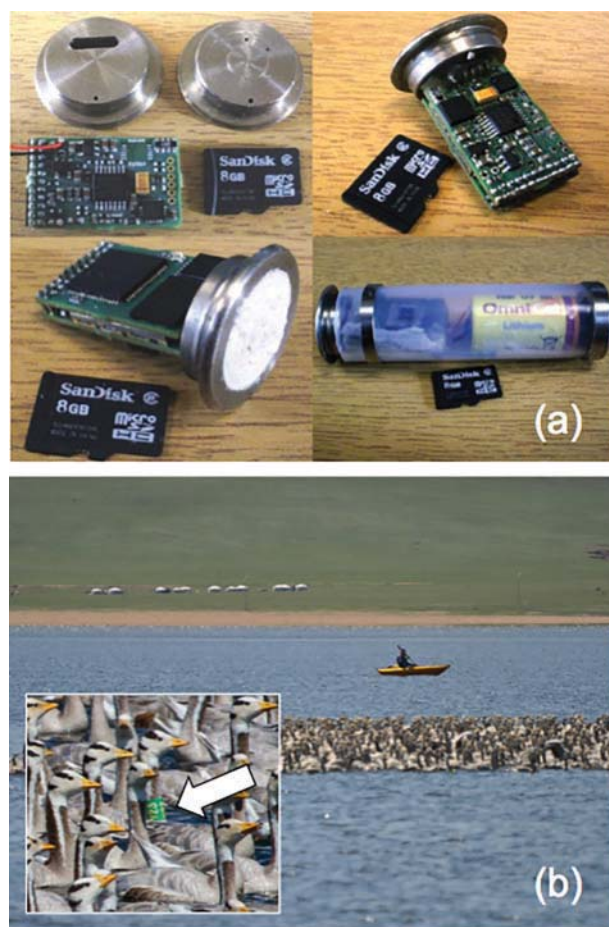
### Methods

Bar-headed geese ( $n=4$ , 2.4 kg mean body mass, range 2.1–2.6 kg) were captured on a breeding lake in western Mongolia (Terkhiin Tsagaan lake, 48.148°N, 99.577°E) where they annually moult their primary flight feathers and are unable to fly for 2–3 weeks of the summer. Geese were captured in July 2011 during moult using “drive trapping”,

herding the geese with inflatable kayaks on their breeding lakes into shoreline corral nets. In order to investigate whether the geese trained prior to their autumn (southward) migration, we implanted the geese with loggers to record heart rate, acceleration, temperature, and pressure (Fig. 1; Spivey and Bishop 2014). Geese were always captured, implanted, and released on the same day, and the loggers were surgically implanted into the abdominal cavity of the geese by a veterinary surgeon, under isoflurane anesthesia and using sterile instruments. Each logger was sterilized using ethylene oxide in the UK prior to the fieldwork. All work was carried out with ethical permission from Bangor University and following ethical regulations set out by the UK Home Office Animal in Scientific Procedures Act (ASPA 1986). Birds implanted with loggers were released with green neck collars bearing alpha-numeric identifying codes (P35, P37, P41, and P43; Fig. 1B). The following year, geese were re-sighted during surveys of the same breeding lakes and recaptured in order to remove the loggers and download the data.

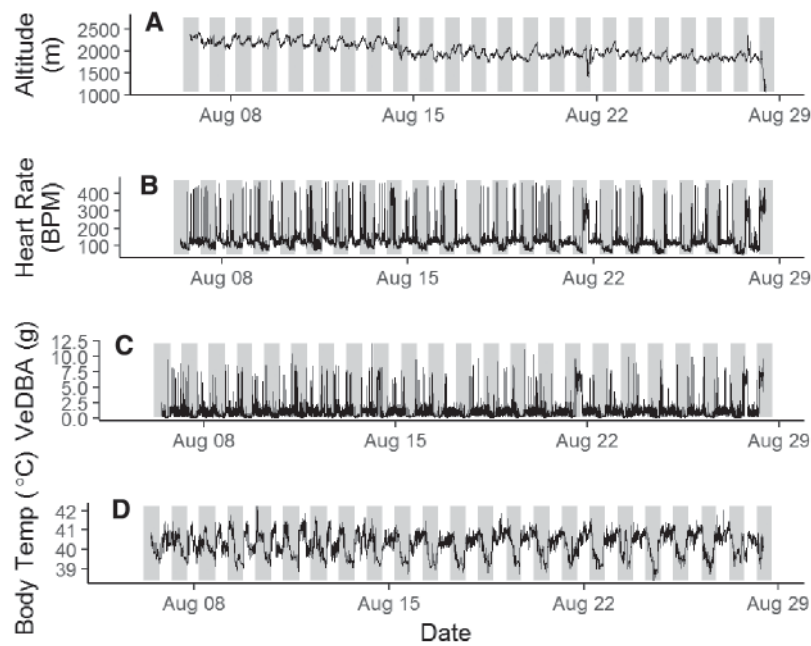
The logger recorded internal body temperature and pressure every 30 s, acceleration at 180 Hz in 18-s bursts every 2 min and electrocardiogram (ECG) via titanium end cap electrodes in the same 18-s burst every 2 min (Spivey and Bishop 2014). The logger was 7-cm long and 2-cm diameter, weighing a total of 32 g. Acceleration data were used to calculate vectoral dynamic body acceleration (VeDBA; Qasem et al. 2012) which describes dynamic movement by the geese in three dimensions (heave, surge, and sway). Heart rate was derived from the raw ECG data using a custom beat detection script in R. Because data were collected at intervals (to maximize logger battery life), data were summarized into 2-min averaged bins for analysis. The onset of migration, defined here as the first southerly movement away from the breeding grounds, was derived using plots of daily mean VeDBA and altitude (Supplementary Fig. S1).

In order to investigate whether geese trained for migration, we looked for significant changes in five key variables leading up to the onset of migration for each bird. (i) Whether the total amount of exercise undertaken increased leading up to migration. We used summed VeDBA per day to estimate total exercise because it encompassed all dynamic movement by the bird while the logger was recording. (ii) Whether the magnitude of the maximal activity undertaken each day increased prior to migration. We hypothesized that maximal activity could encompass strenuous take-off manoeuvres, climbing flights, or other demanding activities. We used the



**Fig. 1** (A) Custom designed implantable logger to record body temperature, pressure, three axis acceleration, and ECG in bar-headed geese (figure from Spivey and Bishop 2014); and (B) a flock of barheaded geese on Terkhiin Tsagaan lake, Mongolia being captured via drive trapping with inflatable kayaks (seen in background). Geese implanted with loggers were identified by their neck collars (inset photo in (B), goose wearing neck collar indicated with arrow).

maximum VeDBA recorded in any 2-min block per day to estimate this. (iii) Whether intense, probable “ground based wing flapping events” increased in frequency prior to migration. We inspected 1-h sections of data per goose to develop a thresholding approach: First we considered any section where VeDBA was four G or more for over 6 min as “flight”, and therefore excluded it from the analysis. For the remaining data, we extracted all instances where VeDBA exceeded 6 G, assigned it as probable ground based wing flapping, or a putative take-off event and counted the number of these “spikes” happening per day. There are no other activities undertaken by these birds that should lead to such a high dynamic acceleration value. (iv) Whether maximal heart rates increased leading up to migration. We hypothesized that maximal heart rate could



**Fig. 2** Plots showing (A) altitude, (B) heart rate, (C) VeDBA, and (D) body temperature for three weeks leading up to migration (which occurs immediately after the last data point in the plot) for one bar-headed goose (P37). Data are averaged into 2-min bins. Diurnal patterns in heart rate, VeDBA, and body temperature can be seen as regular increases during the day and decreases overnight. Gray vertical boxes show night-time for all four panels. Similar plots for the other three geese can be found in Supplementary Figs. S3–S5.

encompass strenuous take off manoeuvres, climbing flights, or other demanding activities that might encompass training. We used the maximum heart rate recorded in any 2-min block per day to estimate maximal heart rate. (v) Finally, in order to investigate whether there was any evidence of the geese becoming physiologically fitter (not implying any evolutionary sense), we looked to see if the overnight resting heart rate decreased, which may be evidence of an increase in cardiac stroke volume if all other variables were to remain the same. We used minimum heart rate recorded in any 2-min block per day to estimate this parameter, which always occurred overnight.

Because heart rate and acceleration data from the loggers are serially auto-correlated (i.e., within each animal's dataset, subsequent data points are not independent), data were analyzed using generalized least squares models (Zuur et al. 2009) using the *gls* function in the R package “nlme” (Pinheiro et al. 2011). We applied a first-order autoregression (AR1) autocorrelation structure, which allows the user to account for serial autocorrelation within each bird's dataset, and compared models with (*gls*) and without (*lm*) auto-correlation structure using corrected Akaike values (AICc) using the R package “MuMIn” (Barton 2014). For each statistic, we report the model that had the lower AIC value, indicating better fit. The model took the

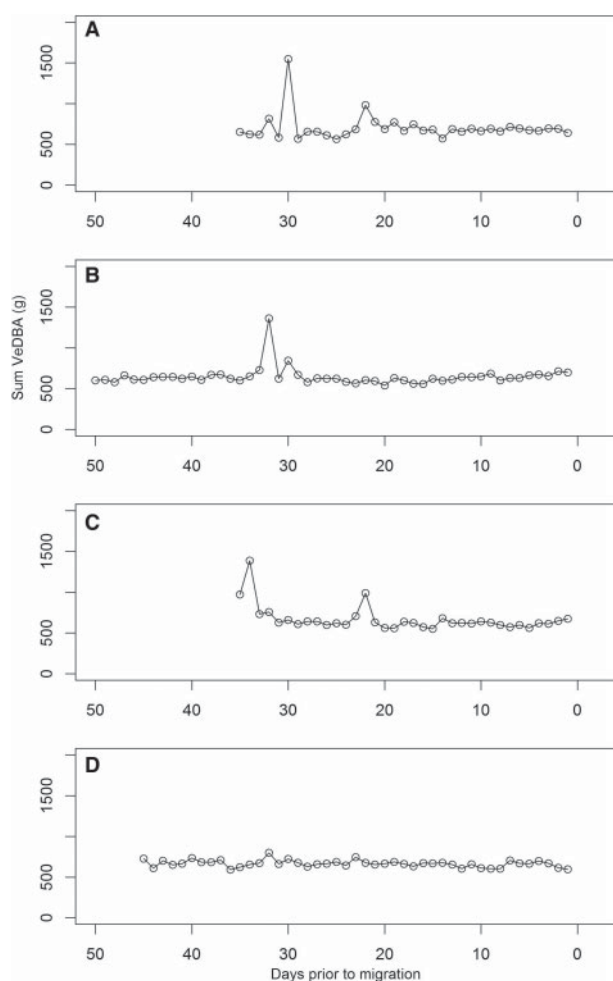
form  $gls(\text{parameter} \sim \text{days leading up to migration})$  where the parameters of interest were sum VeDBA, max VeDBA, maximum, minimum daily heart rate, and sum of daily spikes in VeDBA.

## Results

The four loggers recorded data for a median of 171 days in total (including the full migration and some time wintering, range 158–222 days; example in Fig. 2). The four geese departed for migration 35, 42, 46, and 51 days after logger deployment in late August and early September (Supplementary Fig. S1).

### Does the total amount of exercise increase prior to migration?

There was no evidence that there was an increase in the total amount of exercise carried out each day leading up to migration for any of the four geese (sum VeDBA did not change significantly; P37  $gls$   $t=0.91$ ,  $AIC=451.6$ ,  $P>0.05$ ; P41  $gls$   $t=0.08$ ,  $AIC=612.9$ ,  $P>0.05$ ; P43  $gls$   $t=1.95$ ,  $AIC=437.3$ ,  $P>0.05$ ; P35  $gls$   $t=2.00$ ,  $AIC=463.5$ ,  $P=0.05$ ; Fig. 3), suggesting they did not undertake more physical activity as they got closer to migratory departure. Three of the four geese exhibited a spike in activity, approximately 33 days prior to migration (Fig. 3A–C). The geese were not being directly observed during this time, so it is not clear what the

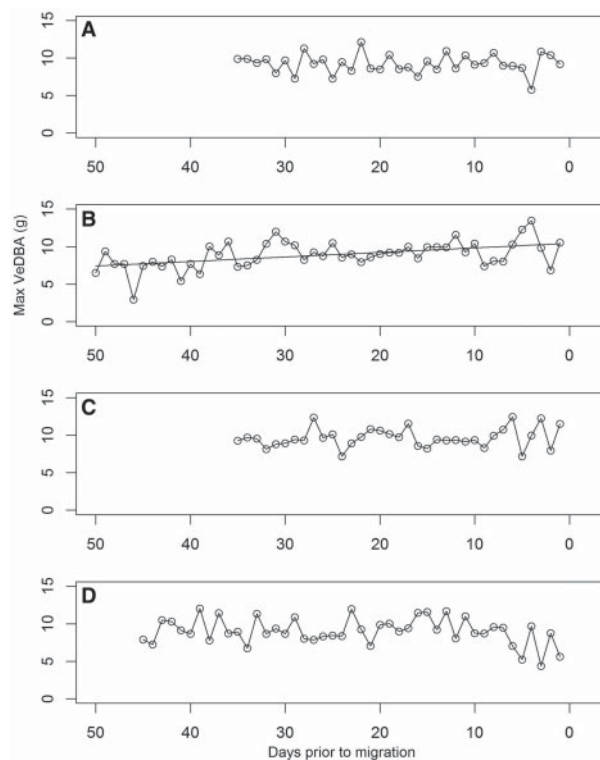


**Fig. 3** Plots showing daily sum of VeDBA for four bar-headed geese (A, B, C, D) over the period between device deployment and migratory departure, shown as days prior to departure, where 0 is the day of departure.

stressor might have been, although we suggest predator presence might be a likely explanation.

#### Does the magnitude of the maximal activity undertaken each day increase prior to migration?

The maximal amount of activity (as indicated by maximum VeDBA), which might have indicated particularly strenuous exercise bouts, did not change leading up to migratory departure for three of the geese (P35  $lm R^2=0.02$ ,  $F_{1,43}=1.94$ ,  $AIC=181.7$ ,  $P>0.05$ ; P37  $lm R^2=-0.03$ ,  $F_{1,33}=0.04$ ,  $AIC=121.5$ ,  $P>0.05$ ; P43  $lm R^2=0.00$ ,  $F_{1,33}=0.9$ ,  $AIC=123.2$ ,  $P>0.05$ ; Fig. 4A, C, D) but did increase significantly for the fourth (P41  $lm R^2=0.22$ ,  $F_{1,48}=14.46$ ,  $AIC=193.2$ ,  $P<0.01$ ; Fig. 4B). There was no correlation between maximum VeDBA and body temperature (Pearson's correlation P35,  $r=55.5$ ,  $P>0.05$ ; P37,  $r=38.7$ ,  $P>0.05$ ; P41,  $r=69.6$ ,  $P>0.05$ ; P43,  $r=52.9$ ,  $P>0.05$ ), which



**Fig. 4** Plots showing daily maximum VeDBA value for four bar-headed geese (A, B, C, D) over the period between device deployment and migratory departure, shown as days prior to departure, where 0 is the day of departure. Regression line shows significant linear relationship for goose B.

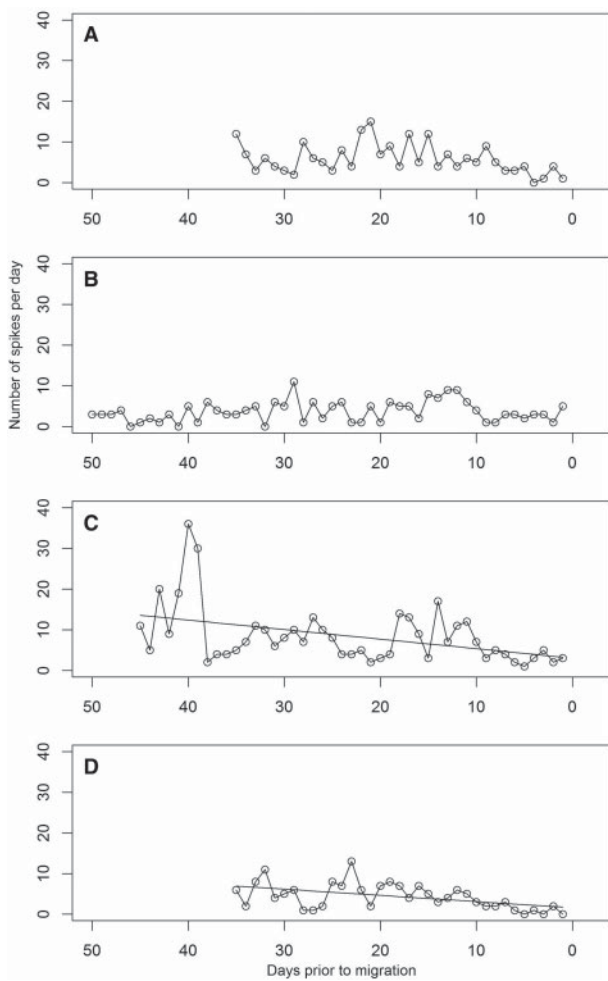
suggested that when maximal activity occurred, it was not necessarily sustained long enough to produce a hyperthermic response.

#### Do probable “ground based wing flapping events” increase in frequency prior to migration?

The frequency of probable “ground based wing flapping events” (number of spikes in VeDBA per day) did not increase prior to migration for two geese (P37  $lm R^2=0.07$ ,  $F_{1,33}=3.49$ ,  $AIC=192.6$ ,  $P>0.05$ ; P41  $lm R^2=0.01$ ,  $F_{1,48}=1.67$ ,  $AIC=238.7$ ,  $P>0.05$ ; Fig. 5A, B) but actually decreased prior to migration for the other two geese (P35  $lm R^2=0.22$ ,  $F_{1,33}=10.80$ ,  $AIC=175.8$ ,  $P<0.01$ ; P43  $lm R^2=0.17$ ,  $F_{1,33}=10.00$ ,  $AIC=300.2$ ,  $P<0.01$ ; Fig. 5C, D).

#### Does the magnitude of the maximal heart rate each day increase prior to migration?

For two of the geese, there was no change in maximum heart rate leading up to migration (P41  $gls t=-0.88$ ,  $AIC=466.7$ ,  $P>0.05$ ; P43  $gls t=0.80$ ,  $AIC=439.5$ ,  $P>0.05$ ; Fig. 6B, C) but two geese had a small but significant decrease in daily maximum heart rate leading up to migration

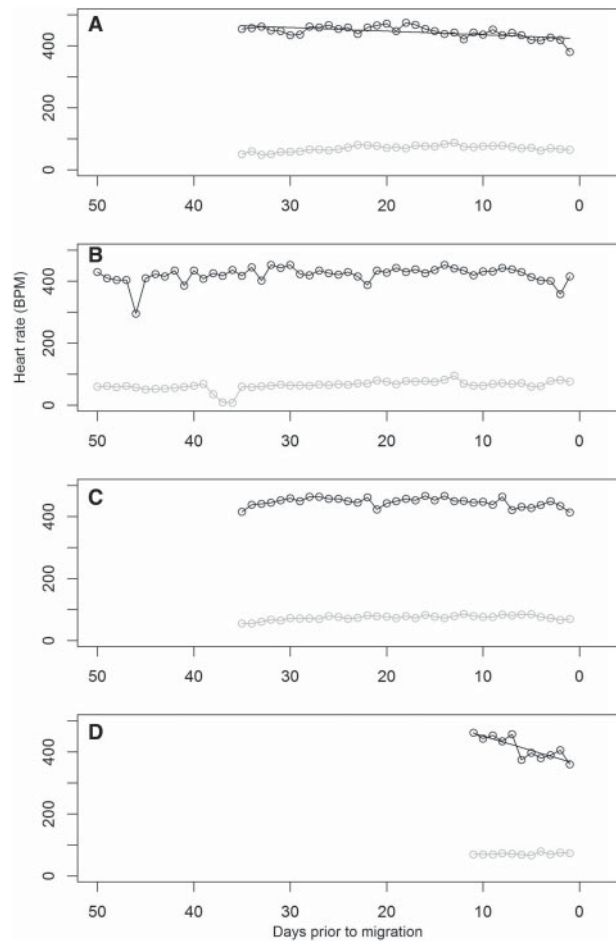


**Fig. 5** Plots showing number of spikes in VeDBA data per day for four bar-headed geese (**A, B, C, D**) over the period between device deployment and migratory departure, shown as days prior to departure, where 0 is the day of departure. Regression line shows significant linear relationship for geese C and D.

(P35 *gls*  $t=2.26$ ,  $AIC=402.2$ ,  $P<0.01$ ; P37 *gls*  $t=2.45$ ,  $AIC=283.8$ ,  $P<0.01$ ; **Fig. 6A, D**), contrary to the prediction of an increase in maximum heart rate.

### Is there any evidence of physiological improvements prior to migration?

There was no significant change in minimum heart rate for any of the geese over the period leading up to migration (P35 *gls*  $t=-0.93$ ,  $AIC=330.3$ ,  $P>0.05$ ; P37 *gls*  $t=-0.44$ ,  $AIC=221.8$ ,  $P>0.05$ ; P41 *gls*  $t=-1.91$ ,  $AIC=380.4$ ,  $P>0.05$ ; P43 *gls*  $t=-1.32$ ,  $AIC=217.7$ ,  $P>0.05$ ; **Fig. 6**), which might have been indicative of cardiac hypertrophy. There was also no significant change in the range of body temperatures experienced by the four geese (P37 *lm*  $R^2=0.04$ ,  $F_{1,33}=2.42$ ,  $AIC=21.5$ ,  $P>0.05$ ; P41 *lm*  $R^2=0.03$ ,  $F_{1,48}=2.48$ ,  $AIC=33.8$ ,  $P>0.05$ ; P43 *lm*  $R^2=0.00$ ,  $F_{1,33}=1.06$ ,  $AIC=9.3$ ,  $P>0.05$ ; P35 *lm*  $R^2=0.06$ ,  $F_{1,43}=3.94$ ,  $AIC=40.0$ ,



**Fig. 6** Plots showing daily minimum (gray points) and maximum (black points) heart rate for four bar-headed geese (**A, B, C, D**) over the period between device deployment and migratory departure, shown as days prior to departure, where 0 is the day of departure. Regression line shows significant linear relationship for goose in part (A). Heart rates for all but the last 11 days for the goose in part (D) were excluded from the analysis as they were erroneous.

$P<0.05$ ; Supplementary Fig. S2), which might have otherwise reflected the difference between intense periods of exercise and sleep at low metabolic rates.

## Discussion

Here, we provide evidence that bar-headed geese do not obviously engage in training for migration. Our study is the first to use accelerometry data to study this in wild birds, and suggests that overall (for three out of four birds) there was no increase in the sum or the maximal amount of activity each day leading up to migration. Interestingly, while there is notable variation in the maximal activity undertaken by each bird (doubling or halving from one day to another), the sum of activity is remarkably consistent both within and between individuals at around

600–700 g (defined as 9.806 metres per second squared) per day. The accelerometry data were also used to identify ground based flapping events, which might have represented “power training.” These do not appear to increase leading up to migration, and indeed actually decreased for two birds. Likewise, the results showed that maximum daily heart rate decreased for two of the four birds leading up to migration. Perhaps, therefore, study birds might have been conserving energy to lay down greater fat deposits for migration, rather than training, but the cause of this, or prevalence, is not clear and requires further investigation. There may be several reasons for the present observations, which we discuss in turn:

#### Limited data coverage

To prolong battery life, the archival loggers were duty-cycled to record for just 18 s of every 2 min throughout the day. Although this represents some of the highest resolution, long-term physiological data ever collected for a migratory bird, data were not collected for 85% of the time. It was not possible to collect complete recordings (covering 100% of the time) to compare with the duty-cycled data. It is possible that episodes of training did not occur during the 15% window when we did collect data. However, it would seem highly unlikely that all training occurred in bouts of <102 s, all of which we missed for the entire period of several weeks leading up to migration. Therefore while we recognize this shortcoming, we do not believe it is the best explanation for the apparent lack of training. Perhaps also longer recording prior to migration might reveal greater insight into preparation for migration. Increases in maximum VeDBA were observed for the one goose that remained for the longest period prior to departure (see below), and highlight that longer recordings in the future might shed light on whether this was an artifact.

#### Training at surprisingly small scales

Alternatively, training for migration may occur at a far smaller scale than previously appreciated. Many birds including waterfowl exhibit ground-based wing-flapping behavior, typically as a preening or social activity. These bouts of flapping usually only last a few seconds but may be metabolically extremely intense (Norberg 1996) and thus, it might be that these bouts act as short, intense explosive strength-training sessions. Muscle building in humans is known to respond strongly to such short but high intensity training sessions (Mitchell et al. 2013), so these events could contribute meaningfully to training in birds. In the present study we attempted to identify these from accelerometry data

and find that they do not increase prior to migration, suggesting that either these explosive training bouts do not contribute to migratory preparation in bar-headed geese, or that such preparation is continuous from the time of moult.

#### Endogenous development of flight musculature

It also remains highly possible that increases in flight muscle mass and performance necessary for migratory flight are endogenous, that is, not brought about as a result of exercise. This has been suggested previously for migratory birds (Bishop et al. 1998; Dietz et al. 1999; Guglielmo et al. 2002; Bauchinger and Biebach 2006; Vézina et al. 2007; McFarlan et al. 2009; Portugal et al. 2009; Price et al. 2011), although other studies suggest that regular flight can be augmented with “power training” (increasing load to the working muscles) and may produce muscle hypertrophy (Marsh and Storer 1981; Marsh 1984; Lindstrom et al. 2000). It remains to be empirically demonstrated if bar-headed geese can endogenously build flight muscle without power training because we did not measure flight muscle mass in the present study, but barnacle geese in captivity (i.e., deprived of the ability to fly around freely, and therefore exercise their flight muscles) increase flight muscle mass by a third as they regrow their flight feathers after moult (Portugal et al. 2009). In addition, there is no evidence that wild birds “de-train” (i.e., exhibit a partial or complete anatomical, physiological, or performance loss as a consequence of reduction of activity) during long periods without flight, for example, over the breeding or wintering seasons, although this can be seen in captive birds, such as pigeons (*Columba livia*; Chaplin et al. 1997). Certainly most wild birds probably experience large fluctuations in flight muscle mass over the course of a year (Jehl 1997)—consuming flight muscle during migratory journeys, rebuilding it on arrival to breeding and wintering grounds, and must regulate this via some process. It is also unclear to what extent power training might be beneficial for endurance exercise.

The mechanism for such endogenous change appears likely to be hormonally based, brought about by changing photo-period, leading to up-regulation of specific genetic pathways for muscle development or fuel mobilization. For example, McFarlan et al. (2009) showed that white-throated sparrows (*Zonotrichia albicollis*) increase gene expression for heart-type FABP and plasma-membrane FABP up to 1000% relative to pre-migratory resting levels which would facilitate transport of essential fatty acids by up to 110% into the flight muscle to fuel migration. In barnacle geese, FABP in the pectoralis muscle can be twice as high in pre-migratory wild goslings as that of captive goslings and

50% higher in wild than captive adults (Pelters et al. 1999). In another study, Price et al. (2011) demonstrated that mRNA expression of insulin-like growth factor 1 (IGF1) was increased in captive white-throated sparrows in migratory condition relative to sparrows in wintering condition. At the same time, it also is clear that training produces muscle hypertrophy and increases in specific gene expression—Price et al. (2011) trained starlings (*Sturnus vulgaris*) in a wind tunnel and demonstrated muscle hypertrophy as well as up-regulation of IGF1 and myostatin immediately following an acute bout of flight. It remains to be demonstrated what proportion of flight muscle hypertrophy may be exercise-induced versus endogenously generated across bird species, but it seems likely that there may be variation in the relative contributions across species and migratory pathways.

### Expedition style

Waterfowl also generally do not migrate for very long periods at a time, with episodes of flight up to a day long, interspersed with stopovers (Arzel et al. 2006). Bar-headed geese migrate up to 5140 km in as much as 92 days in the spring (Hawkes et al. 2012) which would require them to fly as little as 1 h each day (although in reality they make flights of up to 23 h, interspersed with stopovers of several weeks). Thus, during migration, bar-headed geese have ample time to rest and recover between flights, possibly for days. They may therefore effectively train as they migrate, becoming steadily fitter as they travel and this remains to be investigated. Clearly, the differences in strategies used between human and avian high altitude specialists come about because wild birds do not have access to the facilities, resources, and equipment that expedition style human mountaineers do, but also likely come about as a consequence of their differing physiology and evolutionary pressures. Birds, of course, might also make use of assisting wind conditions to reduce flight costs, as has been shown previously (Sherub et al. 2016).

### The superior avian cardiorespiratory system

The avian respiratory system has evolved to allow birds to take up greater amounts of oxygen per unit of body mass than other vertebrates owing in large part to the huge metabolic demands for oxygen during flight (Maina 2000; Ward et al. 2002; Cieri et al. 2014), although see (Speakman and Thomas 2005; McGuire and Guglielmo 2009) for discussion of flight metabolic rate of bats. It might not be surprising if the *metabolic* costs of flight could be easily met by a migrating bird without much need for training, assuming the flight muscles have sufficient

fuel stores. In bar-headed geese, oxygen uptake is thought to be considerably enhanced compared with that of all other birds studied to date, with adaptations for enhanced oxygen uptake throughout the oxygen transport cascade (Scott and Milsom 2006, 2007; Butler 2010; Scott 2011; Scott et al. 2011; Meir and Milsom 2013). For example, Hawkes et al. (2014) tested the performance of bar-headed geese in severe hypoxia (7% oxygen) and showed that their capacity for exercise is enhanced compared with that of barnacle geese, which are otherwise very similar morphologically, but migrate at low altitudes (Butler et al. 1998; Prop et al. 2003; Eichhorn et al. 2009). It is also worth noting that the majority of work demonstrating the range of adaptations by bar-headed geese to enhance oxygen delivery at multiple steps of the oxygen transport cascade (Butler 2010; Scott 2011; Scott et al. 2015) have been carried out on captive bar-headed geese that had been pinioned and, therefore had never flown, i.e., wild birds might be expected to do better (but see Lague et al. 2016). Bar-headed geese have also been shown to use behavioral adaptations to reduce flight costs, such as staying close to the underlying terrain (Bishop et al. 2015), or flying at night in colder conditions where air density (and therefore oxygen availability) should be higher (Hawkes et al. 2011, 2012).

### Future directions

Given physiological and behavioral adaptations to reduce the challenge of flight at high altitudes, perhaps the major challenge for migratory bar-headed geese is not in supplying adequate oxygen to the flight muscles or in endogenously gaining flight muscle mass, but in maintaining an adequate supply of fuel to the flight muscles and in preventing dehydration and muscle damage (Butler 2016; Jenni-Eiermann 2017). First, fuel supply and water balance can probably be maintained by migrating bar-headed geese by regular “stop-overs” to rest and refuel (Hawkes et al. 2012). However, muscle damage might be expected during migratory flights in bar-headed geese, particularly during steep climbing flights from Indian wintering grounds onto the Tibetan Plateau in just 7–8 h (Hawkes et al. 2011). Both hypoxia and exercise are known to generate high levels of free radical species, which are thought to cause damage in proteins, lipids, and muscle myocytes (Powers and Jackson 2008; Zhang et al. 2015). Thus it might be expected that bar-headed geese, or many other high altitude migrants, would have a significant free radical production during migration,

yet we lack any understanding of rates of ROS production or their ability for dealing with it, for example through anti-oxidant loading (Powers and Jackson 2008). This is therefore a research priority remaining to be investigated.

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## Supplementary data

Supplementary data available at *ICB* online.

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