

Influence of individual state on swimming capacity and behaviour of Antarctic krill *Euphausia superba*

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ABSTRACT: Swarms of Antarctic krill are frequently biased towards certain body sizes, sexes, maturities and physiological states. However, the mechanisms causing such biases remain unclear, with some speculating on differential responses to chemical or visual cues. This study examines the influence of individual state on swimming behaviour and performance, which, in turn, may cause biases in swarm composition. The frequency and strength of pleopod beats as well as the overall swimming capacity of Antarctic krill *Euphausia superba* were measured using a tethering technique. Individual state was considered in terms of body size, sex, maturity, moult stage and stomach fullness. Significant differences in the rate and strength of pleopod strokes as well as in the overall swimming capacity were found between individuals in different states. Body size had one of the strongest influences, with beat rate decreasing and beat strength increasing with increasing size. The sex and maturity of an individual altered this pattern, with adult females having faster but weaker pleopod beats than males of equivalent size. The difference in swimming style of females may impose a different lifestyle to that of males. Moulting stage also had a significant influence on swimming, with moulting/post-moult individuals having significantly lower swimming capacities, making them unlikely to keep pace with intermoult counterparts. Both the rate and the strength of the pleopod beat were significantly lower in satiated compared to unsatiated individuals, which may contribute to the dispersion of swarms during feeding.

KEY WORDS: Sex-dependent · Moulting · Maturity · Swarm · Satiation · Pleopods · Antarctic · Southern Ocean

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INTRODUCTION

Antarctic krill *Euphausia superba* are among the most successful taxa in the Southern Ocean, comprising up to 50% of the biomass of the zooplankton community (Godlewski 1996). They are negatively buoyant and must swim continuously in order to retain position in the water column (Kils 1981). For this reason, these organisms are among the most active of any oceanic crustaceans (Childress & Nygaard 1974), using up to 73% of their total daily metabolic expenditure on swimming (Kils 1981, Swadling et al. 2005). Such a large energetic investment means that any factors affecting swimming performance are fundamental to the ecology of this species.

Krill swim through the beating of 5 pairs of pleopods on the first to fifth abdominal segments. The pleopods

are stroked forwards and backwards in a metachronal rhythm, with the posterior-most pair striking first, followed by each of the others in quick succession. The complete wave of strokes (termed a beat) can take up to 400 ms (2.5 beats s^{-1}) in Antarctic krill swimming at ~ 3 cm s^{-1} (Kils 1981). This rate increases to beyond 7 beats s^{-1} as krill swim at speeds above 15 cm s^{-1} (Kils 1981, Swadling et al. 2005).

Particularly notable in many krill species is the tendency to swarm. Most swarms have a thickness of a few centimetres to many metres and a length that may extend for many kilometres (Mauchline 1980). Acoustic investigations have demonstrated that swarm structure can be maintained both day and night (Everson & Bone 1986), and that individual krill within the swarm space themselves around 10 to 20 cm apart (Hamner 1984). SCUBA observations have indicated that krill

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organise themselves in a similar fashion to fish schools with respect to orientation matching and collective response to stimuli (Hamner & Hamner 2000).

A number of studies have found that the composition of individuals within krill swarms is biased towards certain types or physiological states. Marr (1962), for instance, determined that many swarms have similarly sized individuals, which he believed resulted from cohorts remaining as integral units over their entire lifespans. Other studies, such as those of Watkins et al. (1992) and Nicol (1984), have shown biases with regards to sex and maturity, which were attributed to mating and reproductive activity. Buchholz et al. (1996) found that the distribution of moult stages in numerous swarms were non-random and posited the existence of a hormonal synchrony between individuals.

Although state-based biases in the composition of swarms are well established, the mechanisms by which krill sort themselves between swarms remain unclear. Fregin & Wiese (2002) demonstrated that krill are capable of signalling through photophores. Hamner et al. (1983) proposed that krill use rheotactic cues to maintain swarm integrity. The use of sex pheromones to attract mates has also been speculated on (F. Buchholz pers. comm.), although little direct evidence presently exists. In this study, we investigate whether the different swimming capabilities of krill may be a reason for such biases, with the state of an individual having a measurable influence on swimming behaviour and performance. According to this hypothesis, individuals in different states will lose touch with each other, while individuals in similar states remain intact, resulting in swarms developing state-based biases. This explanation is simpler than the inference of complex communication between conspecifics. However, it relies on the premise that state has a significant influence on swimming behaviour and performance, which has yet to be fully investigated.

Most studies on the swimming performance of krill have been carried out on individuals that are allowed to swim freely within tanks. Such an approach has obtained information on various aspects of krill physiology and behaviour, such as circadian rhythms (Velsch & Champalbert 1994), respiratory rates (Swadling et al. 2005), feeding (Torgersen 2001), and reactions to UV and photosynthetically active radiation (Newman et al. 2003). However, minimising experimental stress in these types of studies can be difficult because such active animals regularly collide with container-walls.

The use of tethers is one means of minimising collision-induced stress as well as allowing close observation of individual behaviours. In our study, the organism is attached to a counter-weighted rotational arm so that it swims only on a single plane, well away from the container edges. Investigators have used similar

techniques to examine the responses of pelagic crustaceans to light intensity (Frank & Widder 1994) and hydrodynamic stimuli (Hartline et al. 1999, Patria & Wiese 2004). The force and duration of swimming has also been calculated through measuring the angle of rotation of the counterweighted arm (Thomasson et al. 2003). Another advantage of the technique for the present work is the ability to keep track of individuals such that simultaneous measurements of state variables (size, sex/sexual maturity, moult stage and stomach fullness) can be made.

In this study, we will address the hypothesis that the state of an individual in terms of its biological and physiological characteristics has a significant effect on its swimming performance and behaviour. In so doing, we will examine different components of krill swimming performance, specifically, beat rate, power per stroke, and overall swimming capacity. This is the first study on *Euphausia superba* to relate swimming performance to individual state. Our results also have relevance to the wider study of swarming in pelagic Crustacea.

MATERIALS AND METHODS

The study was carried out aboard the RRS 'James Clark Ross,' from December to January 2005 in the vicinity of South Georgia (54.5° S, 37° W). Krill were caught by locating krill swarms with multi-frequency acoustics (38 kHz, 120 kHz, 200 kHz) and sampling them with a rectangular midwater trawl (RMT8). A non-filtering cod end was fitted and hauls lasted no longer than 20 min to minimise the stress of capture. Once on-board, the cod end contents were diluted immediately into ambient surface seawater in 160 l circular containers held in a constant-temperature (CT) room maintained at ambient sea-surface temperature. The krill were transferred individually into separate 500 ml perforated polycarbonate jars before being placed into a flow-through tank (500 l capacity). Once seeded with a single krill, the jars were stacked within perforated tubes (9 cm diameter plastic drainage pipes) within this tank, to increase space efficiency and facilitate regular maintenance checks. The whole procedure from the point of the catch arriving on board to the completion of the transfer of krill into jars took a maximum of 1 h.

The perforations in the jars and tubes were sufficient to allow free exchange of water with the surrounding tank. The seawater was supplied at a rate of $\sim 60 \text{ l h}^{-1}$, a rate that exchanged the entire tank contents approximately 3 times daily. This resulted in water temperature in the tanks being close to the ambient temperature in the surface layers from where the water

was extracted (an inlet 6.5 m below the water's surface). Over the course of the experimental period, mean surface temperature was $2.72 \pm 0.54^\circ\text{C}$.

Krill were extracted from this apparatus over regular intervals until a maximum of 9 d after capture. During this time, they were exposed to ambient trophic conditions as a result of the continuous flow of only coarsely filtered surface water through the tanks. This meant that food availability changed depending on whether or not the ship had recently passed through any food patches. Accordingly, krill extracted from the apparatus at various times had stomachs that ranged from full to empty. Mortality rates during captivity were low (1 to 2% d^{-1}).

To measure swimming activity, individuals were extracted and attached to the arm of a TRO 15 virtually friction-free rotational displacement transducer (Pan-Lab) using cyanoacrylate glue. The krill could only move along its antero-posterior axis when attached to the transducer. The attached krill was lowered into an experimental aquarium containing seawater at ambient surface-water temperature (see above). The whole process from extraction to attachment and reimmersion took less than 30 s. Four transducers were prepared for simultaneous recording. Attached to 1 transducer was an average sized individual that was recently killed through the injection of alcohol to the cerebral ganglion, a procedure that minimised stress and any distortion to the body. This individual acted as a control and provided a base-line of the vibrations and movements caused by the ship over the course of the measurement period. Live krill were attached to the other 3 transducers. Displacement of the transducer arms were recorded on a PowerLab SP8 (AD Instruments) set at a sampling rate of 100 Hz.

Visual disturbance was minimised by housing the experimental apparatus in a black polythene tent within which light levels were $<1 \text{ W m}^{-2}$. Prepared krill were left undisturbed for 5 min before recording 10 min or more of swimming activity. After the measurements on each krill had been completed, the aquaria was emptied, rinsed and refilled with fresh seawater from the continuous surface-water supply.

Movement of the arm could be calculated through the linear relationship between mV output and degrees displaced relative to 0. The transducer arms each had a pre-calibrated counter-weight with a range from 0 to 5 g. This weight was set to give a maximum range of angle

for each individual while preventing the arm reaching its maximum possible displacement ($\sim 15^\circ$).

Pleopod beat rate and amplitude. The forward thrust generated by the pleopods has 2 main components: (1) the pleopod beat rate (PBR, Hz) and, (2) the pleopod beat amplitude (PBA, $^\circ$). We measured both of these components through analysing fine-scale vibrations of the rotational arm. The frequency of these vibrations was identified through discrete fast fourier transformation (FFT) using the spectrum function in Chart v.4.2.2 (PowerLab, AD Instruments). Although occasionally a number of spectral peaks were identified, there was usually only 1 major peak around the expected beat frequency of krill (3 to 8 Hz, Wiese & Ebina 1995). The centre of the peak was taken as the PBR and its height above baseline PBA (Fig. 1). PBA was considered in the log domain because its values were small and varied between individuals by orders of magnitude.

Swimming capacity. The large scale displacement of the rotational arm reflected the swimming capacity of the individual. The recordings allowed estimation of propulsion generated in N (1 N being the force required to give a mass of 1 kg an acceleration of 1 m s^{-2}). We refer to this parameter throughout as swimming capacity (F_p).

$$F_p = [w_{\text{krill}} + w_{\text{arm}}] \times g \times \text{Sin}\theta \quad (1)$$

where w_{krill} = underwater weight of krill in kg (total mass), w_{arm} = weight of the transducer arm in kg, g =

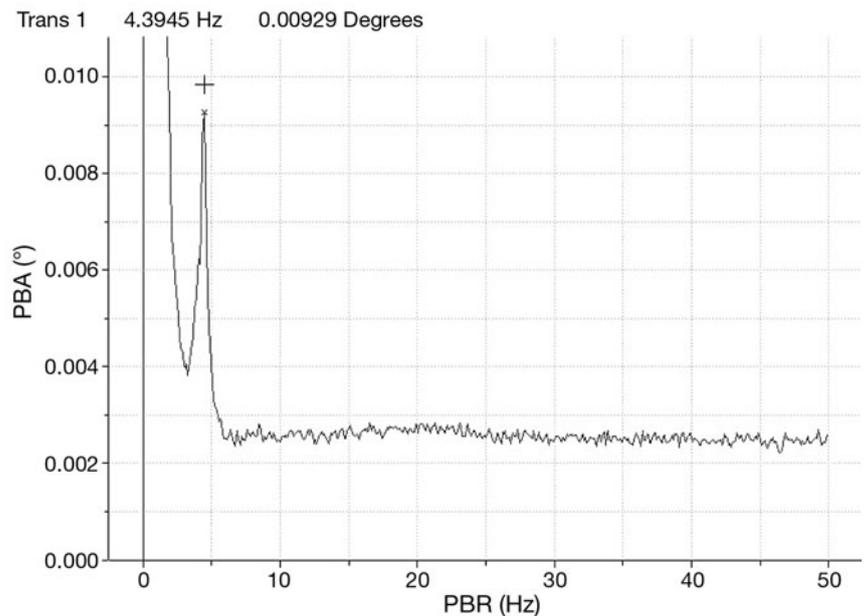


Fig. 1. *Euphausia superba*. Sample Fast Fourier Transform trace from a swimming krill attached to a rotational transducer. A clear peak is visible at 4.3945 Hz, the frequency at which the pleopods were beating (PBR), and the height of the peak (0.0929 $^\circ$) is indicative of the amplitude of degrees movement produced by each complete metachronal cycle (pleopod beat amplitude, PBA)

acceleration due to gravity in m s^{-2} (taken as 9.82) and θ = angle through which the arm was displaced.

The pendulum with the dead krill attached provided the baseline ($F_{p \text{ base}}$), which was subtracted from readings made by the live krill ($F_{p \text{ gross}}$) to give a value for F_p that was a result of the net movement of the krill alone ($F_{p \text{ net}}$). A further factor we took into consideration was the potential offset caused by the process of attaching the krill to the pendulum using cryanoacrylate glue. Alcohol was injected into the cerebral ganglion of the 'live' krill once swimming recordings had been completed. A further 3 min of recordings were then made to identify any offsets relative to the baseline. Any measured offsets were factored into $F_{p \text{ base}}$ to give the final estimate of $F_{p \text{ net}}$.

The accurate estimation of stomach fullness (see below) required manipulation under a binocular microscope, a procedure that was found to cause considerable stress in individuals prior to experimentation. All stomach content examinations were therefore carried out after the measurement of swimming performance. Unfortunately, the injection of alcohol administered at the end of the procedure (see above) made the krill opaque and prevented the accurate assessment of stomach fullness. Therefore, around two-thirds of the krill (216 individuals) were injected after the initial recordings, while a further third (114 individuals) were not injected so that they were in a suitable condition for the assessment of stomach fullness. This meant that it was not possible to obtain both swimming capacity and stomach fullness measurements on the same individuals. However, PBR and PBA were measured for all 330 krill.

Measurements of individual state. The individual state of experimental animals was determined once the swimming measurement had been completed. All individuals were measured for total length (TL), moult stage and sexual stage. TL was measured from the front of the eye to the tip of the telson to the nearest mm. The moult stage was determined following the categorisation of Buchholz (1991), with a total of 6 stages being identified: A/B, C, D0, D1, D2 and D3. These were grouped for the purpose of statistical analyses according to broader phases of the moult cycle, namely: moulting/postmoult (D3, A/B), intermoult (C, D0) and premoult (D1, D2). Five sexual stages were identified: juvenile, sub-adult female, sub-adult male, adult female and adult male, following the guidelines of Makarov & Denys (1980).

Stomach fullness was assessed on a subset of the above individuals (114 out of 330). Three categories were distinguished: individuals with a stomach that was more than three-fourths full were classified as 'full'; between one-fourth and three-fourths full, as 'half-full' and less than one-fourth full, as 'empty.'

Statistical analysis. The significance of any relationships between dependent and independent variables were investigated with best-fit linear regression algorithms. Unequal variance in certain relationships necessitated the use of a weighted-mean fitting method. We employed 1-way ANOVA to test for any significant differences between groups. In many instances, the dataset was found to be non-normally distributed, which required analysis with non-parametric methods, such as Kruskal-Wallis 1-way ANOVA on ranks. The distribution of data in a group was mainly presented in medians and percentiles for the same reasons. Regressions were fitted using a weighted-mean method, which has the capacity to deal with unequal variance in the response variable, which we found in certain instances.

RESULTS

Influence of state on pleopod beat frequency and amplitude

PBR varied between 3 and 5 Hz, and showed a decreasing trend with increasing body length. This trend was not significant when all maturity stages were considered together. When taking maturity stage into account however, it was evident that adult females had distinctly higher PBR than all other stages. The negative relationship between PBR and body length was significant once adult females had been excluded ($F = 13.65$, $df = 256$, $r^2 = 0.05$, $p = 0.0003$):

$$\text{PBR} = 5.12 - (0.02 \times \text{TL}) \quad (2)$$

where TL is total body length in mm and PBR is in Hz.

Although not significant, females also exhibited a negative relationship between PBR and body length. Fig. 2 illustrates the similarity in slope but higher y-intercept of the female trend compared with the relationship for all other maturity stages.

The difference in PBR between males and females can be further seen in Fig. 3. Despite the fact that the TL of adult females, adult males and subadult males overlapped considerably, the average PBR for adult females (4.5 ± 0.7 Hz, mean \pm SD) was significantly higher than that of males (3.9 ± 0.7 Hz and 3.8 ± 0.9 Hz for subadult and adult males, respectively; 1-way ANOVA $F = 8.98$, $df = 325$, $p < 0.001$). Even the average PBR of juveniles (4.2 ± 0.7 Hz) was not as high as the female rate, despite the females being 13.5 mm smaller. This increased PBR was not evident in subadult females, however, which had an average rate of 3.9 ± 0.5 Hz. All other pairwise comparisons between maturity stages similarly did not show significant differences.

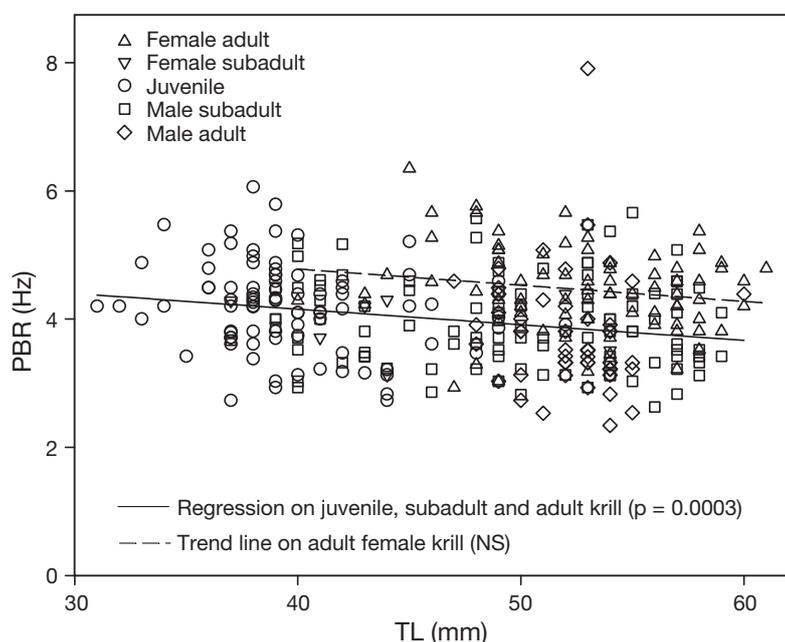


Fig. 2. *Euphausia superba*. Relationship of pleopod beat rate (PBR, Hz) with total length (TL, mm). Note that a significant negative linear relationship (solid line: $PBR = 5.12 - 0.002 TL$, $p = 0.0003$, $r^2 = 0.05$) was obtained for a combined data set of juveniles, sub-adult males, sub-adult females, and adult males. The relationship was not significant when adult females were additionally included. A trend line (dashed) for adult females only is added

PBR was also significantly influenced by moult stage (Fig. 4). PBR was normalised to account for size effects through dividing the rate by TL (mm), following Thomasson et al. (2003). Normalised PBR was significantly higher in individuals that were moulting or recently moulted (A/B, D3) than in either intermoult (C, D0) or premoult (D1, D2) individuals (Kruskal-

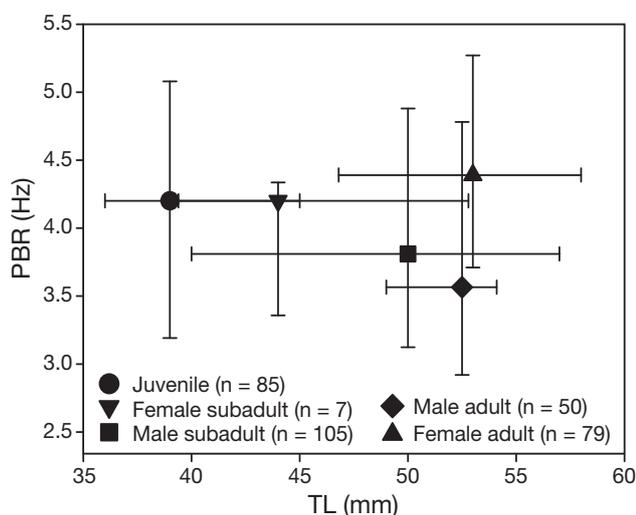


Fig. 3. *Euphausia superba*. Maturity and sex-specific variation in pleopod beat rate (PBR, Hz) with total length (TL, mm). Whiskers denote 10th and 90th percentiles. No attempt was made to fit a regression line as data points represent medians

Wallis 1-way ANOVA, $H = 6.777$, $df = 2$, $p = 0.034$). Intermoult and premoult individuals did not differ significantly from each other.

Log PBA showed the opposite trend to PBR, in that it increased significantly with increasing size ($F = 26.98$, $df = 323$, $r^2 = 0.08$, $p < 0.0001$).

$$\log PBA = -5.6850 - (0.0267 \times TL) \quad (3)$$

where TL is total body length in mm.

The fact that 70% of adult females fell below the regression line (Fig. 5) indicates that they had a lower than average log PBA for their size. This is illustrated further by a 1-way ANOVA. Adult female log PBA values were significantly lower than those of subadult and adult males ($H = 54.63$, $df = 4$, $p < 0.001$). Juveniles also had significantly lower log PBA values than the 2 male categories, but were not significantly different from those of adult females. Subadult and adult males did not differ significantly from each other. The lack of any difference between the 2 male categories is unsurprising given their overlapping body size ranges (Fig. 6). However, the similarity in the log PBA values of juveniles and adult females is less expected given that they occur at opposite ends of the body-size range.

Log PBA was significantly affected by moult stage (Fig. 7). The data were first normalised by dividing log PBA by TL, to account for the relationship with body length. A Kruskal-Wallis 1-way ANOVA on ranks found intermoult individuals (C, D0) had a significantly higher length-normalised log PBA than both moulting/postmoult individuals (A/B, D3) and premoult indi-

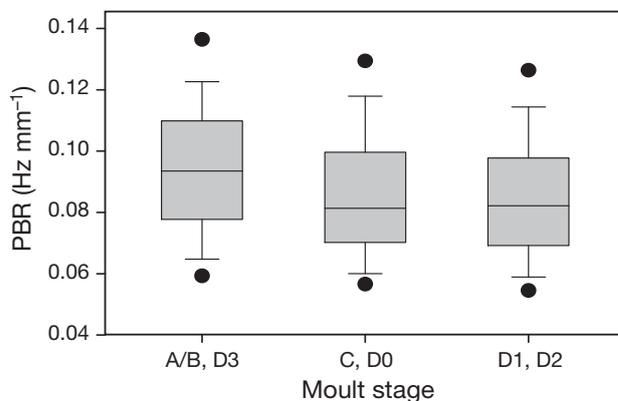


Fig. 4. *Euphausia superba*. Mean length-normalised pleopod beat rate (PBR, Hz mm^{-1}) for different moult stages; A/B, D3: moulting/postmoult; C, D0: intermoult; D1, D2: premoult. Boxes show the range of the 25th and 75th percentiles and the whiskers the 10th and 90th percentiles. Dots are 5th and 95th percentiles. Horizontal lines are medians

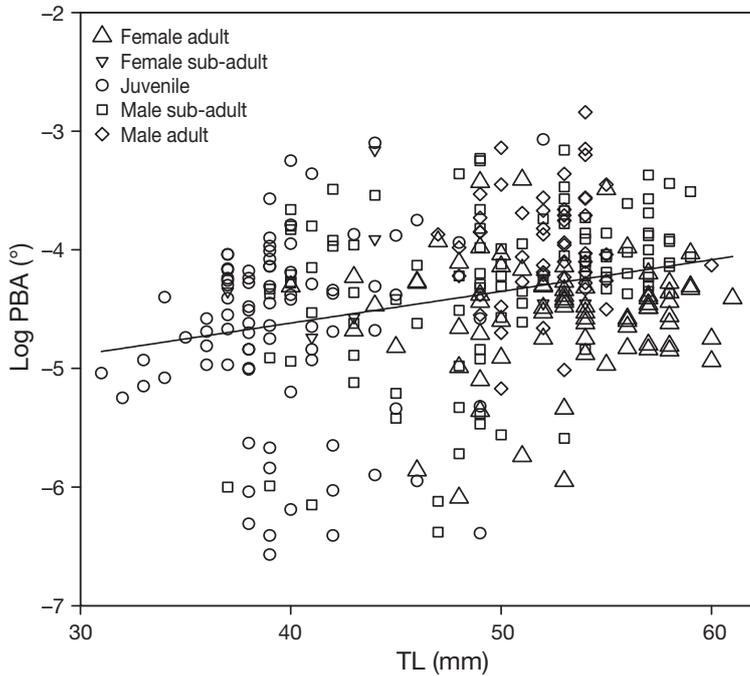


Fig. 5. *Euphausia superba*. Relationship of pleopod beat amplitude (log PBA, °) with total length (TL, mm) ($\log PBA^\circ = -0.0267 - 5.68 TL$; $p < 0.001$, $r^2 = 0.08$)

viduals (D1, D2; $H = 26.115$, $df = 2$, $p < 0.001$). This is the reverse of the PBR pattern, where intermoult individuals had the lowest values. Length-normalised log PBA was not significantly different between moulting/post-moult individuals and premoult individuals, which

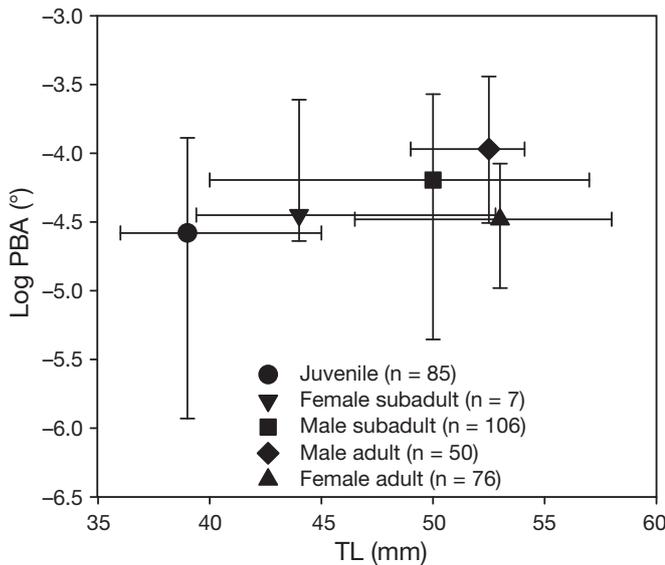


Fig. 6. *Euphausia superba*. Maturity and sex-specific variation in pleopod beat amplitude (log PBA, °) with total length (TL, mm). Whiskers denote 10th and 90th percentiles. No attempt was made to fit a regression line as data points represent medians

may be a product of their relatively large variances.

Regressing log PBA against PBR showed a significant negative relationship (Fig. 8, $F = 14.14$, $df = 326$, $r^2 = 0.04$, $p = 0.0002$):

$$\log PBA = -3.647 - 0.184PBR \quad (4)$$

Hence, individuals that have the fastest PBR also impart the lowest log PBA, while those individuals that beat slowly do so with greater PBA. However, what is more notable is the cluster of individuals with both low PBR and low log PBA, of which a high proportion had full stomachs (Fig. 8). To examine this further, we grouped the residuals from the PBR/log PBA relationship according to the 3 stomach fullness categories (i.e. full, half-full and empty). Individuals with full stomachs had a significantly higher residual difference than either those with half-full or empty stomachs (Fig. 9, 1-way ANOVA, $F = 5.787$, $df = 97$, $p = 0.004$). Individuals with half-full or empty stomachs were not significantly different from each other. Feeding state therefore has a significant effect on both the strength and the frequency of pleopod beats, with satiated individuals being more likely to exhibit lower PBR and log PBA than non-satiated individuals.

Influence of state on swimming capacity

Values for $F_{p\ net}$ were found to be negative in 3% of instances. This indicates that $F_{p\ base}$ could sometimes

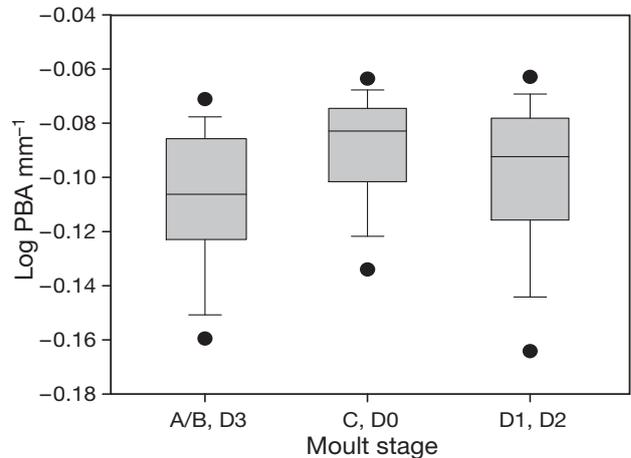


Fig. 7. *Euphausia superba*. Mean length-normalised pleopod beat amplitude (log PBA, ° mm^{-1}) for different moult stages; A/B, D3: moulting/postmoult; C, D0: intermoult; D1, D2: pre-moult. Boxes show the range of the 25th and 75th percentiles and the whiskers the 90th percentiles. Dots are 5th and 95th percentiles. Horizontal lines are medians

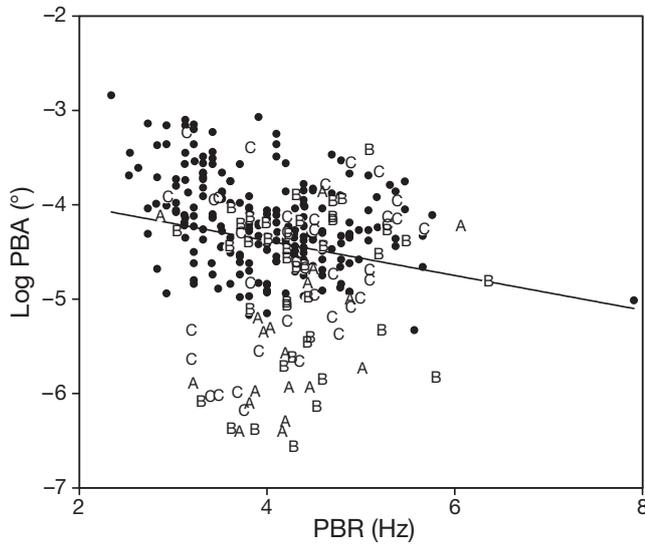


Fig. 8. *Euphausia superba*. Relationship between pleopod beat rate (PBR, Hz) and pleopod beat amplitude (log PBA, °). A significant negative regression ($\log \text{PBA}^\circ = -3.647 - 0.184 \text{PBR}$, $p = 0.0002$, $r^2 = 0.04$) was fitted to the entire date set. A: full stomach; B: half-full stomach; C: empty stomach. Black dots represent measurements where accompanying information on stomach fullness was not obtained (see 'Swimming capacity' in 'Materials and methods')

be larger than gross measurements of $F_{p \text{ gross}}$ as a result of measurement error. It is likely that the ship's movement sometimes affected total displacement of the 'live' and 'background' rotational arms differently, leading to some measurements being falsely low and negative, and the others being falsely high. Thus, the method is unsuited to the accurate estimation of the swimming capacity in any 1 individual krill. However, the method is valid for estimating the mean swimming capacity of krill of a given length, provided that the error introduced by the ship's movement is effectively

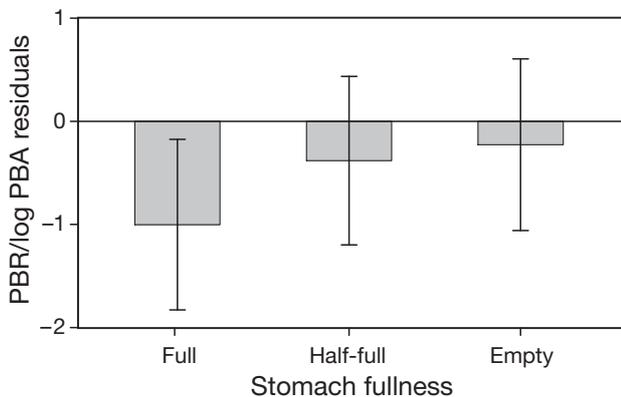


Fig. 9. *Euphausia superba*. A comparison of the residuals from the PBR/log PBA relationship (Fig. 8, Eq. 4) for different states of stomach fullness. The height of the column denotes the mean residual, and the error bars ± 1 SD. Number of krill in each state were: full: 19; half-full: 41; empty: 38

random. This assumption is made explicit by representing the estimated swimming capacity of an individual krill as follows

$$F_{p \text{ net},k} = f(\text{TL}_k) + S_{\text{live},k} - S_{\text{dead},k} + \varepsilon_{\text{live},k} \quad (5)$$

where $f(\text{TL})$ denotes the relationship between mean swimming capacity and TL, $S_{\text{live},k}$ and $S_{\text{dead},k}$ are effects for the motion of the ship on the live and dead krill, and $\varepsilon_{\text{live},k}$ represents variation in the actual swimming capacity of a live krill of a particular length. The error due to the ship's movement is a difference $S_{\text{live},k} - S_{\text{dead},k}$, between 2 comparable quantities. Thus, the assumption that such errors would tend to cancel in the long-run therefore seems reasonable. We tested this assumption below.

The relationship between mean swimming capacity and TL appears linear over the observed range of TL (33 to 60 mm, Fig. 10), with no evidence of any curvilinearity (linear: $R^2 = 21.4\%$; quadratic: $R^2 = 21.5\%$, $p_{\text{quadratic}} = 0.84$). However, there is a marked increase in variability with increased swimming capacity. A straight line was therefore fitted by weighted regression assuming a variance (V) related to the mean swimming capacity (M) as $V = a + bM^c$, using weights $W = 1/V$. The model was fitted iteratively using estimates of a , b and c derived from nonlinear regression of the squared residuals on the fitted values. This gave $a = 0.016$, $b = 0.48$, $c = 2$. The fitted line was:

$$F_{p \text{ net}} = -2.74 + 0.085\text{TL} \quad (6)$$

A plot of the residuals against the fitted values showed a random scatter consistent with the straight-line model and the above variance function.

Given the above findings, we went on to examine how differences in other body condition indices affected the mean/median swimming capacity. Maturity had a relatively minor influence on swimming capacity, with the major difference between maturity groups being a result of the maturity-specific differences body lengths. Fig. 11 shows that the average swimming capacities of each maturity group increased in a broadly linear fashion, according to its average TL. Particularly notable is the fact that adult females fitted within this linear trend, despite the differences in their PBR and log PBA relationships noted above. A Kruskal-Wallis 1-way ANOVA found that adult female swimming capacity did not differ significantly from that of subadult and adult males, whereas juveniles were significantly different from all other stages ($H = 60.15$, $df = 4$, $p < 0.001$).

Intermoult individuals (C, D0) had a significantly higher swimming capacity than moulting/postmoult individuals (D3, A/B; Kruskal-Wallis 1-way ANOVA on ranks, $H_2 = 28.374$, $df = 2$, $p < 0.001$). Premoult individuals did not differ significantly from the other 2 categories given that they exhibited intermediate values.

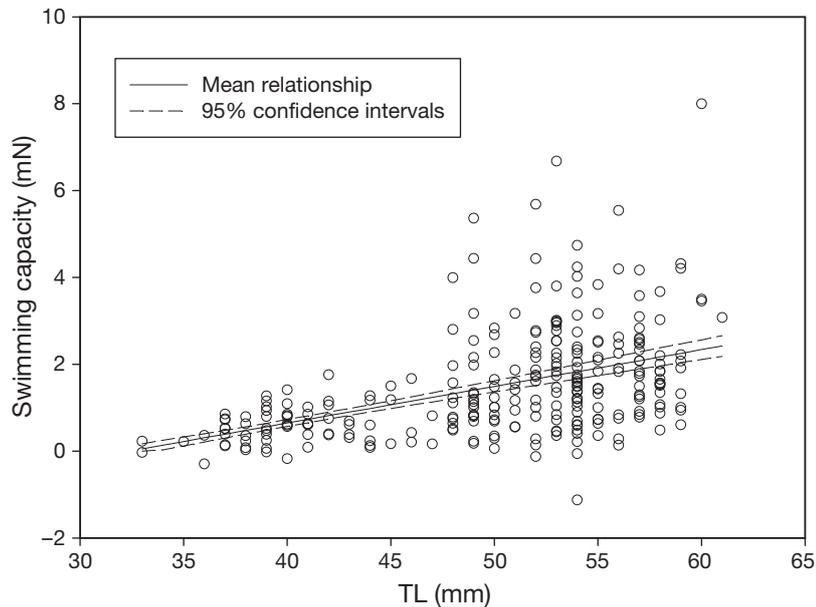


Fig. 10. *Euphausia superba*. Relationship between average swimming capacity (F_{pnet} , mN) and total length (TL, mm), showing 95% confidence intervals

DISCUSSION

General findings

We found that the PBR and PBA as well as the overall swimming capacity of krill was influenced by the state of the individual krill. Body size had one of the strongest influences, with PBR decreasing and log PBA increasing with increasing size. These trends were altered somewhat by sexual maturity and moult stage, with males and intermoult individuals having slower, stronger beats and females and moulting individuals

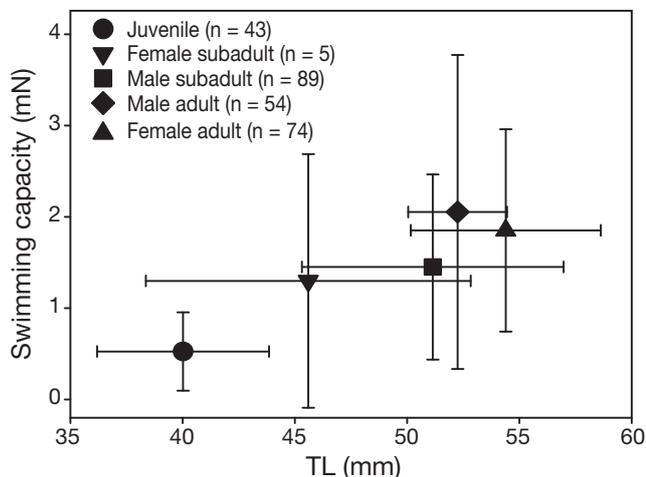


Fig. 11. *Euphausia superba*. Maturity- and sex-specific variation in swimming capacity (F_{pnet} , mN) with total length (TL, mm). Error bars denote 10th and 90th percentiles. No attempt was made to fit a regression line as data points represent medians

having faster, weaker beats. Weight specific swimming capacities were relatively similar in all maturity stages, indicating much of the variation in PBR and log PBA cancelled itself out in terms of overall swimming performance. This was not true of recently moulted individuals, however, which had significantly lower swimming capacities. Similarly, the swimming capacity of satiated individuals was probably reduced given that they exhibited both low PBR and low log PBA.

Measurement uncertainties

As a ship-board study, this work had the advantage of having access to a ready supply of animals in fresh condition, thus reducing the influence of prolonged captivity on behavioural patterns and performance. However, the unsteady platform that a ship provides also makes accounting for measurement error a more difficult task. We took measures to insulate the instruments from ship vibration but it is unlikely that we were able to prevent all such error getting through to our traces. We believe our FFT analysis successfully filtered out the vast majority of this type of error, given that PBR was within expected levels (see below) and varied between individuals in a coherent way. Log PBA measurements were a relative rather than absolute measure of power imparted per pleopod beat. Being unable to calibrate this particular feature is another constraint imposed by carrying out our experiments on an unstable platform. Nevertheless, even relative measurements of this parameter give unique insights into the swimming performance of krill.

The measurement of swimming capacity was most influenced by the pitch and roll of the ship, the majority of which we accounted for by obtaining concurrent baseline measurements from a rotational arm attached to a dead krill. However, the fact that some swimming capacity measurements turned out to be negative is evidence that we were not successful at accounting for all background motion in all instances. We presume that certain combinations of pitch and roll resulted in one rotational arm being affected more than another, even within the small distances that separated them. Over a large number of measurements, these will go both ways, causing some measurements to be falsely high, others falsely low. This will create scatter but, assuming this scatter is random, the mean value will be unbiased. Our analysis showed that scatter around the mean was indeed consistent with random errors.

Therefore, even though our study was not suited to obtaining swimming capacity values for any 1 individual krill, it was robust in determining the mean swimming capacity of an experimental population.

Comparability of measurements

We found that PBR for krill 30 to 60 mm TL varied between 2 and 7 Hz. This is comparable with measurements on *Euphausia superba* made by Swadling et al. (2005), who found rates of between 1.5 and 6.5 Hz. Kils (1981) measured rates of between 2 and 10 Hz. These average rates are somewhat lower than those observed in *Meganyctiphanes norvegica*, a euphausiid of equivalent size found in the Northern Hemisphere (Fig. 12) (Thomasson et al. 2003). For instance, at around 40 mm TL, our measurements of *E. superba* indicate a maximum PBR of 5 Hz, whereas the maximum PBR of *M. norvegica* of the same size was up to 9 kHz. PBR is set by the metachronal rhythm of the pleopod action (Kils 1981) and operating temperature is instrumental in setting an upper limit to this rhythm. *E. superba* characteristically inhabits temperatures of between 0 and 4°C, whereas *M. norvegica* may be found in temperatures of between 6 and 24°C. The cold-water lifestyle of *E. superba* may therefore limit its potential to increase PBR. Another factor that may contribute to the difference is the morphology of the swimming appendages, such as their relative length and width, as well

as their muscular arrangements and the thickness of the cuticle. Further studies focussed on such comparative morphology would be highly beneficial to the understanding of krill swimming performance.

The other means of increasing swimming capacity is to increase the power imparted during each pleopod beat. Our approach was to determine the amplitude of the degree of movement produced by each complete metachronal cycle (log PBA). Although it was a relative rather than absolute measure of power imparted per beat, our results give one of the first measurements of this parameter yet obtained. Comparing patterns in this parameter to those in PBR is instructive in identifying some of the tradeoffs made by krill in optimising their swimming performance.

Effect of moulting

Moulting had a significant effect on PBR, log PBA, and swimming capacity. Intermoult individuals beat their pleopods at a slower rate and with larger amplitudes than moulting, post-moult or pre-moult individuals. The thickness and rigidity of the exoskeleton changes throughout the moult cycle and reaches its peak during the intermoult period (Buchholz & Buchholz 1989). As an individual enters premoult, the cuticle will gradually become resorbed and the new cuticle developed underneath. This will decrease the rigidity of the exoskeleton and reduce the capacity of the individual to impart power to the water, thereby decreasing PBA. As PBA decreases, PBR must increase in order to maintain swimming capacity. This compensation effect was observed in our results, particularly in the moulting/post-moult phase, where PBA was the lowest of all 3 major moult stages and PBR, correspondingly, the highest. Nevertheless, this compensation cannot completely maintain swimming capacity at a constant level, since both moulting/post-moult and pre-moult individuals have significantly lower capacities than intermoult individuals.

Effect of sex and maturity

It was apparent that adult males and females had different swimming styles, in that males beat their pleopods slowly and strongly, and adult females, quickly and weakly. Nevertheless, both sexes attained approximately the same average swimming capacity. Direct measurements on the

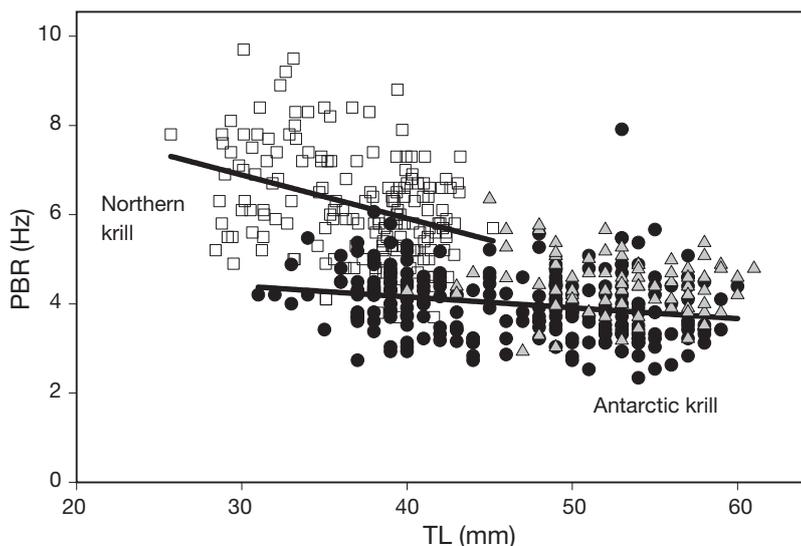


Fig. 12. *Euphausia superba*. Comparison of the relationship between pleopod beat rate (PBR, Hz) and total length (TL, mm) between Antarctic krill and Northern krill *Meganyctiphanes norvegica*. Data for Antarctic krill is as presented in Fig. 2, where the fitted regression was $\text{PBR (Hz)} = 5.12 - 0.02 \text{ TL (mm)}$. Data for Northern krill was extracted from Thomasson et al. (2003): $\text{PBR (Hz)} = 10.18 - 0.107 \text{ TL (mm)}$. (□) Northern krill, (Δ) adult female Antarctic krill, (●) all other maturity stages of Antarctic krill

relative costs, in terms of respiration, of these different swimming styles were not made. However, Swadling et al. (2005) found a linear relationship between PBR and respiration. Applying this relationship to the present study, the predicted respiration rate of adult females would be 50% higher than that of adult males (respective average PBR of 4.5 Hz and 3.8 Hz; equivalent respiration rates of ~ 6.0 and ~ 4.2 mg O₂ g(dry weight)⁻¹ h⁻¹). Although the per beat energetic cost is likely to be influenced by the flexibility of the pleopods (which will be higher in females) maintaining swimming capacity through increasing PBR could have an associated cost in terms of respiration. Given that females must also invest a great deal of energy into the provision of the ovary during the summer months, which can make up over 40% of the body mass (Cuzin-Roudy 2000), it is probable that adult females have a greater energetic requirement than males. This is likely to result in a sexual differentiation in feeding behaviours and risk-taking strategies, as posited for *Meganycitophanes norvegica* by Tarling (2003).

This change in pleopod beat style only became apparent in the adult stages; sub-adult males, sub-adult females and juveniles showed little comparative difference in these parameters, relative to body size. The divergence of pleopod beat styles on reaching adulthood suggests that they are a direct or indirect product of secondary sexual development. Tarling et al. (2006) found that adult males and females had distinctly different intermoult periods, with females having an average period between moults of 20 d, and males 30 d. This would mean that females would spend 50% more time in the moulting, postmoult and pre-moult stages compared to males. As already discussed, the thickness and rigidity of the cuticle during these periods is reduced, meaning that PBA would decrease and PBR must increase to compensate.

The ultimate benefits to either sex of maintaining such different moult rates remain unclear. However, Cuzin-Roudy & Buchholz (1999) have proposed that moulting and spawning are interlinked, so a fast moult cycle would result in an increased number of spawning episodes per season. Moulting is nevertheless a costly process, both in terms of energy required to generate a new cuticle and vulnerability to predators and conspecifics (Tarling et al. 1999). Males may maintain a lower moult rate in order to reduce this cost, resulting in the cuticle remaining rigid for a comparatively greater length of time.

Swimming speed and respiration

Swadling et al. (2005) developed an algorithm for determining swimming speed as a function of PBR.

Relating this to the present study, the observed PBR would result in krill swimming at speeds between 9.8 and 12.3 cm s⁻¹. This is towards the upper end of the current speeds to which Swadling et al. (2005) exposed their animals but still well within the maximum speeds of 40 cm s⁻¹ for these animals. Kils (1981) posited that the pleopod stroke is optimally developed towards 15 cm s⁻¹. In our calculations above, we predicted that the relatively higher PBR and hence equivalent swimming speed of krill in the present study would mean that their respiration rates were also likely to be high, between ~ 4.2 and ~ 6.0 mg O₂ g(dry weight)⁻¹ h⁻¹. Other measures of krill respiration in chambers have determined typical values to be much lower, around 1 mg O₂ g(dry weight)⁻¹ h⁻¹ (Kils 1981, Saborowski et al. 2000). One thing to consider in this instance is that the pendulum apparatus prevents the individual from building up any momentum and overcoming inertia. This is equivalent to a sprinter leaving their blocks at the start of a race, time after time, when cadence must be fast and energy expenditure high. Therefore, although instructive in terms of measuring relative performance, the present study is unlikely to provide representative measures of transport costs, as carried out by, for example, Swadling et al. (2005) and Torres (1984).

Effects of stomach fullness

We found that individuals with full stomachs had a lower swimming performance in terms of both PBR and log PBA compared to individuals with half-full to empty stomachs. Simultaneous observations on swimming pattern, reported in Tarling & Johnson (2006), found that satiated individuals were the most likely to exhibit parachuting behaviour, where the pleopods stop beating altogether and are fanned out in order to control the subsequent descent (Kils & Marschall 1995). Combined, it appears that having a full stomach decreases the capacity to swim and increases the probability of sinking.

It is known that the processes of ingestion and digestion have a significant energetic cost because respiration rates have been found to decrease in starved animals (Ikeda & Dixon 1982, Atkinson et al. 2002). However, whether this energetic cost is sufficient to cause a change in swimming behaviour is unclear. Another possibility is that krill enter a different behavioural mode when satiated, as a means of rapidly sinking out of the dangerous food-rich surface waters. The phenomenon of satiation sinking has been widely reported in a number of planktonic species, including euphausiids (Pearre 2003).

CONCLUSIONS

In this study, we provide some explanation of the means by which biases in the composition of swarms occur. Previous studies (Hamner et al. 1983, Nicol 1984, Watkins et al. 1992) have speculated that signals, cues or hormonal rhythms may be the main mechanisms by which krill sort themselves. Here we demonstrate that the effect of state on swimming performance and behaviour may be enough to create biases. In our results, sex, maturity and moult-stage, and feeding-state had a significant influence on swimming performance and behaviour. Such differences could determine whether an individual maintains contact with or falls behind a swarm, a process that will filter the composition of swarms and generate the biases that have been commonly observed.

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