Introduction

The history of research in Northern krill mirrors a dichotomy apparent in the wider field of research in biological oceanography that has been apparent since the 1880s (Kunzig, 2000). Back then, Victor Hensen thought that the oceans could be described in purely numeric terms and spent decades counting plankton. Ernst Haeckel was meanwhile scathing, saying “Mathematical treatment of these does more harm than good, because it gives a deceptive semblance of accuracy, which in fact is not attainable” (Mills, 1989). Because of the difficulties encountered in trying to observe plankton in situ (Hamner & Hamner, 2000), modern day oceanographers have been drawn towards Hensen’s approach, something made obvious by the complete absence of behavioural observation as a technique in the zooplankton biologists’ handbook by Harris et al (2005). Kawaguchi et al (2010) suggest that many studies view krill as passive particles drifting at the behest of physical processes, yet an understanding of the behaviour of individual animals is fundamental to oceanic ecology. Here we review the work that has been attempted on *Meganyctiphanes norvegica* and similar organisms in aquaria housed on land or at sea. Our hope is that we encourage a re-appraisal of the utility of observational science and promote the individual plankter to a position more than that of a mere integer.

Potentially, *M. norvegica* is an ideal model organism for laboratory based behavioural studies of plankton – it is large, robust, locally abundant and widely distributed, and it is well-studied with an extensive literature base (Mauchline & Fisher, 1969; Mauchline, 1980; Johnson & Tarling, 2011). However, the major drawback to its routine use in the laboratory is the difficulty in keeping stocks alive for an extended period. *M. norvegica* rarely survives for more than 4 weeks in aquaria, probably due to injury during capture and bacterial infections of the integument (Buchholz, 2003). However, they have been kept alive in outside tanks during winter for up to nine weeks, although only when maintained in virtual darkness; where the tanks were exposed to daylight, none of the krill survived beyond 15 days (Macdonald, 1927). In contrast to the long-term survival rates now achieved in Antarctic krill, *Euphausia superba* (Kawaguchi et al, 2010), it appears that even flow-through systems are insufficient for lengthy experiments using *M. norvegica*.

A further problem with the use of *M. norvegica* as an experimental animal in a laboratory setting is that, like all krill, they are social animals invariably found in swarms (Aitken, 1960; Nicol, 1984). Isolation of krill in aquaria has undoubtedly led to data being produced that are
difficult to interpret, for example in the study of swimming energetics (Ritz, 2000) and circadian migratory rhythms (Gaten et al., 2008).

**Pelagic animals in aquaria**

A major drawback to making observations of the behaviour of any pelagic animals in the laboratory is that they would rarely, if ever, encounter the limitations imposed by the walls of an aquarium. To an animal that spends its entire life in the ocean without ever necessarily reaching either the surface or the sea-bed, the confines of an aquarium usually result in a modification of their behaviour (Strand & Hamner, 1990). After working with mesopelagic hyperiid amphipods, Land (personal communication) suggested that, based on observations of pelagic crustaceans in aquaria, one could come to the naive conclusion that they lie at the bottom of the tank on their sides swimming in circles repeatedly or are attracted to solid surfaces. Over the years there have been steady developments in techniques used to house krill in laboratory-based and ship-based aquaria that permit experiments on larval and adult growth, food preferences and assimilation, all carried out under varying environmental conditions (Ross & Quetin, 2003).

Feeding behaviour of krill is one area where researchers have attempted to use aquaria in order to develop an understanding of growth, consumption rates, selectivity and the role of krill as grazers and/or predators. Early qualitative observations suggested that krill were able to generate a feeding current with their pleopods that drew food towards the basket formed by the periopods (Macdonald, 1927; Berkes, 1975). More quantitative work carried out on *M. norvegica* in aquaria suggested that there was an ontogenetic shift from herbivory to carnivory with increasing size and that it selectively predated on larger copepod species (McClatchie, 1985). Torgersen (2001) carried out some work on feeding by *M. norvegica* in aquaria at different light intensities. He noted that predation rates on *Calanus* spp. and *Metridia longa* were approximately three times higher under light levels comparable to that found at the depths they inhabit during the day than they were in the dark. A difference in the relative proportion of the two species consumed under light and dark conditions was noted – under illuminated conditions more *Calanus* were consumed than in the dark, despite there being little apparent difference in the swimming speeds of the prey under either condition. Although Weissburg & Browman (2005) suggested that it would be inaccurate to characterise *M. norvegica* as a visual predator on the basis of this work, they later acknowledged that light intensity does seem to affect predation efficiency of this species (Abrahamsen et al., 2010).

Price (1989) was able to observe feeding behaviour of krill (*Thysanoessa rachii*) in a mesocosm and reported changes in behaviour when the krill encountered food. When they came across phytoplankton patches that had been introduced into their large aquarium, the krill that encountered the patch swam faster and exhibited less turning than they had in the absence of food. Individual krill were also reported to turn back when they reached the edge of the algal patch. More recently Kawaguchi et al (2010) have managed to develop an aquarium set up that promotes schooling in *Euphausia superba* and can thus be used to develop a better understanding of complex social interactions amongst these animals. They
report that schooling krill swim faster than non-schooling and that schools appear to disperse under conditions of low light.

One interesting problem with interpreting the behaviour of krill in aquaria is whether it is the same as would be found in situ. Macdonald (1927) and Mauchline (1989) both observed *M. norvegica* apparently feeding at the bottom of an aquarium by either suspending sediment through the action of their pleopods before swimming through the suspended material, or by ploughing through the surface layers with their antennae and transferring resuspended material to their filtering basket with their periopods. These important reports appear to have been little commented upon until recent observations of *E. superba* actively feeding at abyssal depths by Clarke & Tyler (2008). Stomach content and stable isotope analyses also indicate that benthic feeding is a common trait in both Northern krill and Antarctic krill (Schmidt, this volume).

‘Tethered’ shrimps

One way of overcoming the problems presented by pelagic animals in aquaria is through the use of a ‘tethered shrimp’ approach. As with all tethering methods, this has the advantage of allowing the observer to keep the individual organism in view whilst still allowing it to react to various stimuli. It has the rather obvious disadvantage that it restricts free and natural movement preventing any interaction with other individuals. Attempts to assess stress levels of tethered krill singly or in close proximity suggested that association with conspecifics reduced heart rate to some extent (Ritz et al, 2003). Kils (1982) investigated the flow field of the water produced by *M. norvegica* during swimming by gluing the animal to thin cord and measuring the flow of drifting bodies resulting from swimming activity. Hamner (1988) used tethered animals to allow close investigation of the feeding mechanism of *E. superba* and was able to dismiss previous suggestions that food entered the feeding basket from behind or laterally, rather than from the anterior.

Frank & Widder (1994a), who had noted the profound disorientation experienced by deep-sea shrimps when encountering any surface, developed a form of tethering that allowed observation and a degree of quantification of pelagic crustacean behaviour. Their solution was to attach a small stainless steel swivel to the dorsal carapace of the animal with the other end attached to a rigid plastic rod. Using this arrangement, the shrimp could rotate freely in both horizontal and vertical planes without reaching the walls of the aquarium. Monitoring the tilt or flexion of the body and changes in swimming activity (determined by counting beats of the pleopods) enabled the authors to find a threshold sensitivity to test flashes of light. The sensitivities of various species of decapods shrimps to near-UV and blue-green light were established using this approach followed by video analysis (Frank & Widder, 1994a; 1994b). Abrahamsen et al (2010) used tethered *M. norvegica* to examine their feeding behaviour. They found that the krill generally detected copepods that came within 7-23 mm in a hemisphere centred on the head of the animal such that the main detection volume is ventral and lateral. They also noted that the perceptual distance of the krill was higher in light than dark conditions. This, together with other work on feeding from aquarium-based
studies, reinforces the concept of the importance of multi-sensory approaches to understanding pelagic plankton responses to stimuli (Weissburg & Browman, 2005).

Others have gone a step further and attached crustaceans to force-transducers (e.g Lenz & Hartline, 1999) or recorded the hydrodynamic signal generated by tethered krill using a microphone (Wiese & Ebina, 1995). One major advantage to this type of method is that swimming activity and behavioural responses can be recorded in such a way as to diminish the degree of subjectivity that may creep into analyses of video recordings. Another is that swimming activity and responses can easily be measured on several scales from instantaneous events such as escape responses (Lenz & Hartline 1999) to changes in rate of response to longer term diurnal changes. One attempt to monitor long term activity of krill using a combination of video and a carousel successfully recorded a single specimen of *M. norvegica* swimming 5.7 Km in 24 hours, despite the hindrance of a 10 cm wire attached to a simple swivel (Johnson & Thomasson, unpublished data). The krill was swimming at about 23 cm/s which is towards the upper end of swimming speed estimates from other sources (Kils, 1982).

Propulsion in krill is generated by highly efficient metachronal movements of the pleopods (Kils, 1982; Alben et al., 2010). The swimming movements of *Euphausia superba* were described by Kils (1982) who paid particular attention to the extension and unfolding of the biramous pleopods at the onset of the power-stroke and the folding down and contraction at the beginning of the return stroke. These movements have the effect of a slight antero-posterior jerk in the movement in the animal as it swims which has been recorded using pressure microphones for *E. superba* (Figure 1) and a pendulum for *M. norvegica* (Wiese & Ebina, 1995; Thomasson et al, 2003).

Figure 1: Example traces recorded from krill swimming while attached to a pendulum. As the animal swims the pendulum is displaced (increase on the Y axis). The top trace represents constant swimming, the middle cyclic swimming and the lower periodic swimming. Periodic swimming is found most often in satiated krill that, in situ, would be gliding downwards (Tarling & Johnson, 2006; reproduced with the permission of the publisher).

In order to record swimming behaviour and activity over long and short terms, Thomasson et al (2003) used a virtually friction free rotational transducer as a pendulum to which *M. norvegica* was attached along the dorsal mid-line of the carapace (Figure 2). This restricted the animal to movements along the main axis of their body. Swimming movements of the
krill resulted in a signal from the transducer that was recorded to a computer via an analogue/digital convertor. Any thrust generated by the animal swimming pushed the pendulum forward until the weight of the animal plus a calibration weight exactly compensated for that thrust.

Figure 2: Experimental set-up showing the principles of the pendulum technique. Krill were glued to the transducer arm. Movement of the transducer arm caused by the animal swimming was measured in mV and recorded on a PowerLab SP8, software Chart 4.0. The mV readings were translated into angular displacement ($\theta$), which was used to calculate swimming capacity in Newtons (Thomasson et al., 2003; reproduced with the permission of the publisher).

In the absence of any other weight, if the thrust generated by an animal was sufficient to allow it to hover then it would move a pendulum arm to 90 degrees from vertical. Half that amount of thrust would move it to 30 degrees. Thomasson et al (2003) connected transducers to an interface so that the exact angle that the animal was maintaining at any one time was known and the weight lifted by the animal (F in Newtons) could be calculated from:

$$F = [W_{krill} - (\rho H_2 O \times V_{krill}) + W_{arm}] \times g \times \sin \theta$$

where $W_{krill}$ = wet weight of krill in kg (total mass), $\rho H_2 O$ = specific density of seawater in kg $l^{-1}$ (generally taken as 1.03), $V_{krill}$ = volume of the krill, $W_{arm}$ = calibration weight of the transducer, $g$ = acceleration due to gravity in m s$^{-2}$ (9.82) and $\theta$ = angle through which the transducer arm is displaced. In this manner, the relative swimming capacity of *M. norvegica* of different sizes, sexes and moult stages was determined. The swimming capacity was shown to increase with the size of the animal with no differences between male and female krill. There was a significant difference in swimming capacity between moult stages, with newly-moulted animals being weaker swimmers (Thomasson et al., 2003). This apparatus was further modified for use at sea through developing control systems able to account for
external motion. This allowed researchers to work on recently captured animals (Tarling & Johnson, 2006; Johnson & Tarling, 2008).

Ideally it would be possible to calculate energy expended by an individual animal from the displacement of the arm. Because an animal swimming at a constant speed and attached to the pendulum is not moving, it is difficult to calculate ‘work done’ directly from Newton’s 2nd law so we have to use the fact that it is the krill that is moving the water. If we assume a constant flow of water:

\[ F = \frac{d(mv)}{d(t)} = r \times v \]

where \( m \) = mass (kg), \( v \) = velocity in (m/s), \( t \) = time (s) and \( r \) = mass of water moved per second.

We know \( F \) from the angle through which the pendulum is displaced but in order to estimate the power output and energy consumed by a swimming krill it is necessary to know how much water it is displacing to produce thrust. Yen et al (2003) estimated the volume of fluid displaced by *Euphausia pacifica* as being about 18 times the volume of the animal. The volume of a single *M. norvegica* can be estimated from Kils (1981) as:

\[
Volume = 3.67 \times 10^{-6} L^{3.16}
\]

where volume is in ml and length (L) is in mm. The mass of the water displaced per second (\( r \)) can be estimated using:

\[
r = PBRp(Vol \times 18)
\]

where \( PBR \) = the pleopod beat rate, \( p \) = the specific gravity of water and \( Vol \times 18 \) is the estimated volume of water displaced during each pleopod beat cycle relative to the volume of the krill. Average pleopod beat frequency decreases with increasing length and can be estimated from Thomasson et al (2003):

\[
PBR = -0.107L + 10.18
\]

where \( PBR \) = pleopod beat rate in Hz and \( L \) = total length of a krill. It should be noted that the pleopod beat rate varies significantly during the moult cycle of krill (Thomasson et al 2003). From the data obtained from the pendulum system (\( F \) and \( PBR \)), power in theory could be estimated as:

\[
\frac{1}{2} F^2 (18PBRpVol)
\]

Nevertheless, further parameters using this approach still need refinement. We do not know, for example if there are differences in the volume of water moved at different swimming speeds or by different sizes animals or how Reynolds numbers affect different sizes/species
of krill in water bodies of different densities. Generally, the most efficient krill would move a large volume of water slowly. We also do not know how efficient pleopod based swimming is and it is possible that a significant amount of energy is wasted in displacing water laterally. However the theory suggests that there is an exponential increase in the energy required for swimming with increasing size, something that undoubtedly limits the maximum size of negatively buoyant animals such as krill. These factors, based on observations of individual animals, may shed some light on subtle, larger scale ecological impacts of changes in local water density or reduced calcification rates induced by climate change.

The pendulum arrangement allows researchers to ask a broad range of ethological and physiological questions of individual krill of known state with regard to sex, size, moult stage and stomach fullness, and in response to olfactory, visual and physical stimuli or environmental variables such as light intensity, time, food availability, temperature or salinity. On occasion, observations of tethered animals can lead to interesting links between behaviour and physiology or anatomy. Tarling et al (2009) linked swimming behaviour to egg release in *E. superba* and demonstrated that it was a slow and steady occurrence, where the swimming behaviour during spawning may explain some of the sorting evident in schools of this species. In mesopelagic shrimps, Shelton et al (1999) linked the eye-blink of mesopelagic decapods with the escape response and anatomy to demonstrate that previously unexplained tapetal distributions were linked to camouflage and bioluminescence. Gaten & Johnson (unpublished) found that brief flashes of dim blue light (c.10$^{13}$ photons.m$^{-2}$.s$^{-1}$) were sufficient to cause dramatic changes in pleopod beat rate. Because swimming patterns may be affected by individual state and the artificial situation of the animal, as with simpler observational techniques, careful interpretation of short term responses may still be required (Thomasson et al, 2003; Johnson & Tarling, 2008). Tarling & Johnson (2006) noted that there was a difference in swimming activity and swimming style depending on the stomach fullness of krill. Those that were satiated tended to be much less active than those that had empty stomachs. It was suggested that this behaviour could have significant implications for the carbon budget of the southern oceans as it was a mechanism that could carry biological material to depth more quickly than passive sinking of detritus. This work has implications for the standard tenet of biological oceanography that assumes that mesopelagic organisms migrate only once each day.

Using the pendulum system, comparison of swimming activities between species reveals some interesting differences between *M. norvegica* and *E. superba* (Thomasson et al 2003; Johnson & Tarling, 2006). Generally *M. norvegica* swam with a faster pleopod beat than equivalent sized *E. superba* and it was noted that in the latter species the males swam with a slower, stronger beat. Such a swimming style would be ideal as an honest signal (Zahavi & Zahavi, 1999) to female krill looking for a mate that is large (which is costly in the case of a negatively buoyant animal like krill), and would generate strong vortices that females could use to their advantage (see below). It is interesting that when we compared pleopod beat rates between moult stages of krill from the two species, a slightly different picture emerged. Soft, newly-moulted *M. norvegica* had a low pleopod beat rate, suggesting that their strategy
in this vulnerable condition was to sink quietly (Tarling et al., 1999). *E. superba* had the highest pleopod beat rates when their carapace was soft which suggests that they may be using an increased beat rate to compensate for a lower output per beat (Johnson & Tarling, 2006). This would suggest that their strategy mandates staying with the school – a very different approach compared to *M. norvegica*, which may be indicative of important differences in lifestyles between the two species.

**Swarming and swimming**

The understanding of swimming and swarming behaviour (and the underlying mechanisms) was limited in krill by the difficulty in maintaining these animals under experimental conditions that are conducive to these activities. However, the seminal experiments of Kils (1982) provided real insight into the behaviour and physiology of Antarctic krill, *E. superba*, particularly with respect to their swimming behaviour. The reasons suggested for swarming behaviour in krill and other organisms include reduction in energy expenditure, protection from predators, increased possibility of locating either food or a mate and improved decision making (Ritz, 1994; Parrish & Hamner, 1997; Ritz et al, 2001; List 2004). Strand & Hamner (1991) showed that swarming would occur in relatively large aquaria, provided they were free from any contrasting visual stimuli or external disturbance. Swarm formation can now be readily induced in *E. superba* at the Australian Antarctic Division research aquaria in Tasmania (Kawaguchi et al., 2010). As Watson (2000) points out, the degree to which krill swarm can be of great ecological and commercial importance through knock-on effects on predator species.

There have been many in situ studies of swarm behaviour of *M. norvegica* that have revealed something of their spatial and temporal occurrence (Aitken, 1960; Nicol, 1986; Tarling et al, 1998, 1999). Aspects that have been of interest to behavioural physiologists looking at the level of the individual have included the mechanisms that may promote swarm formation and maintenance and the possibility of advantages to individuals in terms of reduced costs of swimming (Wiese & Ebina, 1995; Swadling et al, 2005).

The propulsion jet flow of *M. norvegica* and *E. superba* may qualify as a communication signal. To maintain a constant position in the krill formation and for fast re-assemblage even in darkness or after predator invasions, individuals have to be able to (A) perceive and analyse the flow field produced by the locomotion of their forerunners and (B) to synchronically beat their own pleopods. Three conditions are important for this task: the pleopods must generate a flow signal with specificity, the antennular sensor system must analyse the three-dimensional flow field, and a sensory-motor reflex from antennules to pleopods must exist (Yen et al, 2003; Patia & Wiese 2004; Abrahamsen et al., 2010).

In the flow jets produced by euphausiids the specificity rests in the spatial and temporal properties. The spatial properties determine the most favourable position energetically for an individual to swim within the formation, which is most certainly the contact point of the turbulent fringes of two adjacent propulsion jets (Wiese 1996). The temporal component is characterised by the frequency spectrum of flow turbulence. Metachronal beats of the
pleopods modulate the propulsion jet flow at 6 Hz in *M. norvegica* (Patria & Wiese 2004) and 3 Hz in *E. superba* (Kils 1982), and thus provide a rhythmic mechanosensory input to the antennules of the following krill. The specificity of modulation of the flow signal renders it suitable for the purpose of communication (Markl 1983).

The jet flow, which powers propulsion, may be as fast as 10 cm s$^{-1}$ (Kils 1983, Patria & Wiese 2004). The flow of water constitutes the carrier; it is directional and has a substantial range, considering the relatively small size of the krill (max. 50 mm). This range is determined first by the attenuation of flow velocity with distance (x) from the source, which is proportional to $1 \times \frac{1}{x}$ in jet flows of circular cross-section (Schlichting, 1982). Secondly, the range depends on the sensitivity of the receiving sensory organs, which is assumed to be 1 mm s$^{-1}$ in the antennular flow receptors of tethered swimming euphausiids (Wiese & Marschall 1990, Patria & Wiese 2004). Under these conditions an assumed velocity of flow of 10 cm s$^{-1}$ at a position 1 cm to the rear of the pleopods (Kils 1982) would attenuate to 1 mm s$^{-1}$ in a distance of 10 cm. Even in less densely organized schools of krill the nearest neighbour distance is estimated to be not larger than 50 cm and hence well within range of the flow signals from forerunners.

A modulated flow signal is produced by the propulsion jet pump of *M. norvegica* and *E. superba*. Kils (1982) has described in detail the action of the pleopods of *E. superba* (considered roughly equivalent to that of *M. norvegica*), especially the extension and unfolding of the biramous appendage at the onset of power-stroke and the folding down and contraction at the beginning of the return stroke. This rectifier effect in the periodic oscillation of pleopods leads to the occurrence of pressure jerks (pressure because a pressure sensitive microphone has been used in recording) during build up and decay of pressure peaks (Fig. 3; Wiese & Ebina 1995).

As a consequence, the power spectrum of such a signal comprises a base frequency and 3 harmonics in fixed multiples of the base frequency (Wiese & Ebina 1995). This means in the case of *M. norvegica* 6 Hz and about 12, 18 and 24 Hz; in the case of *E. superba* 3 Hz and about 6, 9 and 12 Hz. The three harmonics derive from the pressure jerks seen in the oscillating time course of pressure (Figure 3), probably due to the rectifier effect in the pleopod action. The higher the order of the harmonic, the earlier this harmonic disappears from the signal with distance from the source. Provided that this rule applies to low frequency turbulent flow as well, and provided that the receiver system is able to encode them, harmonics may be used to indicate roughly the distance of the detector from the signal source (Wiese 1996). Bleckmann et al. (1991) proposed that oscillations of water between 1 and 40 Hz are monitored very thoroughly by crustaceans as potential signals indicating the presence of prey or of conspecifics, whereas events comprising oscillation frequencies 40 to 100 Hz often result in escape responses.
Flow in euphausiids is sensed by one-sidedly hinged flagella of the antennules, with the proprioceptors of the hinge serving as transducers. The antennular flagella often break in captivity. Levers, in contrast to membranes, are suitable devices to work under various pressure conditions and therefore various water depths. The threshold curve of sensitivity of antennular proprioceptors has been measured both in *E. superba* (Wiese & Marschall 1990) and in *M. norvegica* (Figure 5; Patria & Wiese 2004) by recording sensory cell activity in the basipodite of the antennule in response to quantitatively controlled vibrations of water applied to the vertically moving flagellum (Wiese et al. 1980). A sensitivity threshold of around 0.5 mm sec\(^{-1}\) was noticed (frequency range 1-40 Hz), with higher sensitivity in *E. superba* than *M. norvegica*. The reason for the encountered difference in sensitivity is not known although it may be explained in part by the differences in swarming and schooling activity between the two species. *M. norvegica* females appear unable to swim when newly moulted and hence tend to sink (Thomasson et al., 2003), whereas female *E. superba* retain...
greater rigidity in their pleopods which enables them to maintain their position in the swarm (Johnson & Tarling, 2008).

A sample of an actual recording of sensory nerve activity (Figure 4) shows two different action potentials (different amplitude in the extracellular recording) within one cycle of the water oscillation. Both bending and stretching in the antennular flagellar hinge produce a signal in the proprioceptive sensory cells.

Figure 4: Recordings of evoked sensory activity from the antennular nerve of *M. norvegica* at a site 5 mm proximal to the flagellar hinge (upper traces) in response to water vibration stimuli (lower traces)(see Patria & Wiese, 2004 for details of the recording method). The extracellular recording shows several nerve axons responding (deduced by the different amplitudes of the action potentials). Two different axons often respond per sine cycle of the stimulus. This fact is interpreted as a sign of sensitivity to both flexion and extension of the flagellar hinge.

Systems of communication very much depend on the range through which the generated signals extend from the source to the prospective receiver. In this context, flow velocities generated by the pleopod jet pump have been measured for *E. superba* using kinematography (Kils 1982) and for *M. norvegica* using computer assisted video analysis (Patria & Wiese 2004). According to these investigations, the range of generated flow velocities extends from 5-10 cm s\(^{-1}\) near the pleopods. Using the sensitivity threshold of 0.5 mm s\(^{-1}\) in velocity of flow (Figure 5), the rule of Schlichting (1982, see above) predicts a range of signals of modulated flow as produced by *M. norvegica* and *E. superba* of roughly 100 cm. That is, the dispersedly swimming krill more or less depend on accidental encounters with conspecifics to recruit new individuals to a potential formation.

Divers swimming close to formations of *E. superba* (Hamner 1984) reported that, in formation, individuals swim at the maximum force available and are apparently sorted by size. Tarling (personal communication) has measured inter-individual distances in krill formations and reported typically 20-25 cm between forerunner and follower. Measurements using tethered krill suggest that the position of the follower shrimp in the formation is linked to the ring-vortex established around the jetflow of circular cross-section produced by krill swimming ahead (Wiese, unpublished observations). In tethered *M. norvegica* this ring vortex is detected about 10-12 cm behind the pleopod-pump (Patria & Wiese 2004).
Light and daily rhythms

Light is a key factor in experiments with deep water crustaceans, although this is often overlooked with the result that unreliable data may unwittingly be collected. The problems of retinal breakdown in response to exposure to unnaturally high light levels have been demonstrated in a range of crustaceans, from the Norway lobster in Scottish sea lochs (Shelton et al., 1985) to hydrothermal vent shrimps (Herring et al., 1999). Working with *E. superba*, Newman et al (2003) noted that krill reacted to UVA and photosynthetically active radiation (PAR) by moving away from it. They suggested that this was a mechanism to protect them from over-exposure to damaging UVB light.

The daylight levels normally experienced by *M. norvegica* have been estimated to lie between $2.8 \times 10^{12}$ and $1.4 \times 10^{14}$ photons.m$^{-2}$s$^{-1}$ in the Oslofjord, Norway (Onsrud & Kaartvedt, 1998) and between $9.9 \times 10^{12}$ and $4.5 \times 10^{13}$ photons.m$^{-2}$s$^{-1}$ in the Gullmarsfjord, Sweden (Gaten & Johnson, unpublished) where the krill are normally found in the deepest part of the fjord during the day. Preliminary experiments have shown that following 3 h exposure to light at $1.8 \times 10^{19}$ photons.m$^{-2}$s$^{-1}$ the eyes of *M. norvegica* showed minor damage to the rhabdons when compared to animals fixed in the dark. However, krill fixed after being caught at midday and exposed briefly to sunlight during capture ($8.1 \times 10^{20}$ photons.m$^{-2}$s$^{-1}$) showed extensive rhabdom degeneration (Figure 6; Gaten & Johnson, unpublished). This shows that the eyes of this species (and probably all euphausiids) are vulnerable to light-induced damage that, although reversible following brief exposure, usually proved permanent in decapod crustaceans from various depths exposed to similar levels of daylight (Gaten et al., 1990).
There is little doubt that their behaviour is also affected by brief flashes of light as they have, in common with almost all euphausiids, the ability to produce light (Herring and Locket, 1978). Bioluminescence is produced in *M. norvegica* by 10 photophores, one on each eyestalk, four on the ventral thorax and four on the ventral abdomen (Herring and Locket, 1978). They have been shown to respond to various photic stimuli with flashes of light from the photophores (Mauchline, 1960). Although photophores, such as the ocular peduncle, have been proposed to illuminate food in the dark (Macdonald, 1927), the photophores are mainly thought to provide adaptive benefits through camouflage involving counter shading that matches the downwelling radiation (Clarke, 1963). Recent laboratory-based work on tethered krill has suggested that they may form the basis of a system of communication using light flashes (Fregin and Wiese, 2002).

Euphausiids lack statocysts so it is probable that their only way of orientating themselves to the vertical is via the direction of light. Land (1980) showed that this is probably correct in experiments on a mesopelagic euphausiid, *Nematoscelis atlantica*. He observed specimens that were restrained, but that could freely rotate, and monitored their reaction to a small, moving light source. It was clear that the tethered animals rotated their eyes through 180° to track the light and that the ventral photophores rotated in synchrony with the movement of the eyes. This strongly suggests that the eyes tracked the downwelling light, allowing the animal to orientate itself vertically, whilst the photophores simultaneously rotated to remain pointing downwards (Land, 1980). The latter point lends support to the suggestion that the photophores are concerned primarily with counter-illumination, at least in this species. *M. norvegica* similarly has thoracic and abdominal photophores that can rotate through 180° (Hardy, 1962).
In addition to the reliance on light for vision and orientation, *M. norvegica* almost certainly uses the daily variation in light levels to control its circadian clock. Circadian rhythms have been found to be present in all organisms so far examined and they underlie the ability to predict daily variations in light and other factors. *M. norvegica* undertakes an extensive vertical migration, rising to the surface each night to feed and moving into deeper water during the daytime, probably to avoid visually guided predators (but see Tarling & Johnson, 2006). The ability to predict the onset of day and night enables the animals to initiate their diel migrations at the appropriate time, irrespective of the ambient light conditions. Fine tuning of the migration depth then occurs in response to changes in the light climate, for instance at times of solar (Strömberg et al., 2002) or lunar (Tarling et al., 1999) eclipses.

The influence of a circadian rhythm on swimming activity of individual *M. norvegica* was clearly demonstrated in the laboratory by Velsch and Champalbert (1994) using both actographic analysis and infra-red video observations. After establishing that an activity rhythm was present under a light:dark cycle, with maximum activity during the night, they showed that an endogenous rhythm was present in total darkness with a period of less than 23 h. They concluded that light variations and the endogenous rhythm both had a role in vertical migration in *M. norvegica* (Velsch and Champalbert, 1994).

An adapted version of this actograph, using 12 experimental chambers, has been used with Antarctic krill in a ship-board experiment (Gaten et al., 2008). The activity monitor comprised 12 vertical tubes, each containing 5 l of seawater, retained upright in a light-tight box (Figure 7). Each tube had infra-red barriers 5 cm from the top and bottom of the tube and the output from these barriers was recorded continuously. The lighting within the apparatus
was adjusted to that experienced by the krill at normal daytime depths and the temperature in the cool-room kept at the level of the seawater supply. The krill were not fed during the experiment. Activity levels of the 12 individuals were recorded for five days under a light:dark cycle and then for a further five days in total darkness. The locomotor activity patterns showed a degree of complexity that suggested that the light:dark cycle was not the only Zeitgeber (external cue) in this species (Figure 8). The presence of a secondary 12 h rhythm in 68% of the rhythmic animals is consistent with field observations of a 12 h component in vertical migration that becomes predominant when food is scarce (Godlewska, 1996). In addition, ultradian components in the activity rhythm are thought to reflect the stress caused by the isolation of these extremely social animals (Gaten et al., 2008). Together these findings highlight the necessity of taking into account the effects of lack of food and of social cues when undertaking experiments on krill.

![Graph](image)

**Figure 8:** The presence of a 12 h activity rhythm in *Euphausia superba* is demonstrated by the filtered output from the activity monitor. The animal was in total darkness throughout, so the trace is evidence of an endogenous bimodal circadian rhythm. The data are plotted as relative activity as a function of time, with a superimposed sinusoid derived from the computed period of the oscillation (12.6 h). For further details see Gaten et al., 2008.

The eyes of *M. norvegica* have been well described, both in terms of their morphology (Hallberg and Nilsson, 1983) and their optics (Land et al., 1979). The eyes are of the refracting superposition type and, in contrast to the ‘double eyes’ found in many euphausiids, are spherically symmetrical. Arguably the most important property of an eye is the minimum resolvable angle as this can be used to define the resolution of the eye under ideal conditions. Land et al (1979) used the inter-receptor angle ($\Delta \varphi$) to estimate the resolution of the eye of *M. norvegica*:

$$\Delta \varphi = 57.3 \, d/f$$

where $d =$ maximum diameter of the receptor and $f =$ focal length (or posterior nodal distance). This gives a value of around 2.9° which is much lower than is usually found in the upper eyes.
of euphausiids (1.2° - Land et al., 1979) or mysids (1.5° - Gaten et al., 2002) from deeper water. This is assuming ideal optics and adequate lighting – in reality the resolution would be much poorer due to imperfections in the optics. What this means in practical terms is that *M. norvegica*, given the low light levels at which these animals normally operate, is unlikely to be predominantly a visual predator. The higher predation rates on *Metridia longa* by *M. norvegica* noted by Torgersen (2001) is thus much more likely to be due to the fact that this prey species swims faster and further than the other copepod used in this experiment and is hence more likely to encounter the predator (Browman, 2005).

**Conclusion**

Observations of *Meganyctiphanes norvegica* and other species of krill in aquaria or on tethers will never be as satisfactory in principle as in-situ observations of individual animals would be. There are some techniques developing that allow individual animals to be tracked in the medium term (e.g. McGehee & Jaffe, 1996) but they will probably never allow a full understanding of the state and circumstance of individual organisms. Aquarium based studies have huge potential to answer fundamental questions about krill biology, ecology and behaviour, as long as the limitations are recognised and care is taken over the conclusions drawn (Kawaguchi et al 2010).

One aspect that must be considered is that krill are highly social animals that do not function in a normal fashion when isolated in an aquarium (Strand & Hamner, 1990; Gaten et al., 2008). In addition, it is of paramount importance in designing experiments in aquaria to consider the light environment carefully. Light is a key determinant of behaviour of mesopelagic organisms and they tend to be specifically adapted to cope with a particular light intensity, in addition to defined spectral, spatial and temporal distributions of light (Gaten et al., 1990, 1992, 2002; Johnson et al, 2000a, 2000b, 2002). Failure to consider these important factors is likely to lead to anomalous behaviour, physiological damage and erroneous conclusions.

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