
http://dx.doi.org/10.1016/j.jembe.2011.02.038
Boat-generated turbulence as a potential source of mortality among copepods

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ARTICLE INFO

Article history:
Received 25 November 2010
Received in revised form 18 February 2011
Accepted 19 February 2011

Keywords:
Acartia tonsa
Copepod carcasses
Copepod mortality
Motorized boats
Turbulence

ABSTRACT

Motorized boats present a wide array of stressors to aquatic organisms, but their impacts on zooplankton have not been studied in detail. This study investigated boat-generated turbulence as a potential source of mortality for copepods through a combination of field observations and laboratory experiments. Field sampling in the lower Chesapeake Bay showed that carcasses comprised 34% of the copepod population at a site with a high volume of boat traffic, whereas only 5.3–5.5% of the copepods were dead in the other two, less disturbed, nearby sites. Direct sampling behind passing vessels showed that the percentage of copepod carcasses increased from 7.7% outside the wakes to 14.3% inside the wakes. Laboratory experiments further showed that the fraction dead of the copepod population increased with increasing turbulence intensity, indicating that turbulence was causing mortality. In coastal waters with high volumes of boat traffic, boat-generated turbulence could be an important source of zooplankton mortality, altering trophic interactions among the plankton and shunting zooplankton biomass to the microbial loop.

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1. Introduction

Motorized boats present a wide array of stressors to aquatic organisms, such as chemical pollutants, noise, and direct physical harm. Their impacts on macrofauna, such as whales (Nowacek et al., 2004), manatees (Calleson and Frohlich, 2007), and turtles (Work et al., 2010) have been well documented, but their effects on zooplankton are often overlooked. As a boat is propelled through the water, a large amount of turbulence is generated in its wake. This episodic turbulence suddenly places zooplankton in a potentially stressful environment. In addition to influencing planktich trophic interactions (e.g. Marassé et al., 1990; MacKenzie and Leggett, 1991) and behaviors (e.g. Costello et al., 1990; Waggett and Buskey, 2007), turbulence can negatively impact zooplankton physiology such as excretion rates (Saiz and Alcaraz, 1992a), heart rates (Alcaraz and Saiz, 1991; Alcaraz et al., 1994), developmental rates (Saiz and Alcaraz, 1991), and growth efficiency (Saiz et al., 1992). Prior laboratory studies on turbulence effects exposed zooplankton to moderate levels of turbulence (energy dissipation rates \( \varepsilon = 0.05–0.15 \text{ cm}^2 \text{ s}^{-2} \)), which are within the range of turbulence naturally found in coastal zones and tidal fronts (Kiørboe and Saiz, 1995), but the impacts of higher turbulence levels have not been evaluated. Turbulence as encountered in boat wakes can be much higher than background turbulence, and zooplankton carcasses have been observed in many aquatic systems, including coastal waters with large volumes of boat traffic (Tang et al., 2006; Elliott and Tang, in press). Nevertheless, a link between boat-generated turbulence and in situ zooplankton mortality has not been considered.

In situ study of zooplankton mortality due to environmental or anthropogenic stresses has been hampered by the lack of convenient methods for identifying zooplankton carcasses in field samples. The recent refinement and rigorous evaluation of the Neutral Red staining method for the identification of A. tonsa carcasses has been evaluated exhaustively (Elliott and Tang, 2009); and Aniline Blue (Bickel et al., 2009) staining methods have helped alleviate this limitation, making it possible to directly assess the impacts of boat-generated turbulence on zooplankton mortality. In this study, we used the Neutral Red staining method to test the hypothesis that boat-generated turbulence can cause mortality among copepods in Chesapeake Bay. The calanoid copepod Acartia tonsa (Dana) is a dominant member of the mesozooplankton community in Chesapeake Bay, and the efficiency of the Neutral Red staining method for the identification of A. tonsa carcasses has been evaluated exhaustively (Elliott and Tang, 2009); hence, it was used as the representative species in this study. We conducted field sampling to quantify copepod carcasses at three sites with different levels of boat traffic. We also quantified copepod carcasses inside and outside of the wakes of passing vessels. Additionally, laboratory experiments were conducted to mimic boat-generated turbulence and measure its effect on copepod mortality.

2. Materials and methods

2.1. Sampling at field sites

Three sites, separated by less than 200 m, were sampled within the lower Hampton River, a tributary of the Chesapeake Bay. Site 1 was a...
marina where multiple sail boats and small yachts were moored, and boat generated turbulence was minimal due to an imposed speed limit. Site 2 was next to the marina in the middle of a navigational channel. Boats frequently traveled through this area at high speeds and generated considerable turbulence in their wakes. Site 3 was a relatively shallow, rocky shoreline opposite from the marina. Boats were rarely found in this area and consequently the site experienced little to no boat-generated turbulence.

Samples were collected on three separate dates in May 2010. At each site, temperature, salinity, and dissolved oxygen concentration of surface water were measured with a YSI data sonde immediately before zooplankton collection. Zooplankton were collected by short, low speed (<1 m s$^{-1}$) horizontal tows just beneath the surface with a 0.5 m mouth diameter, 100 μm mesh net and non-filtering cod end. Zooplankton samples were gently concentrated down to approximately 200 ml, transferred to a separate jar, and stained with Neutral Red within 20 min. The entire stained samples were counted immediately, or refrigerated and counted within 24 h. The net and cod end were rinsed thoroughly between tows to avoid any carry-over of carcasses. The dominant copepod in the samples, A. tonsa, was enumerated and identified as live (stained) or dead (unstained) (Elliott and Tang, 2009).

2.2. Sampling of boat wakes

To determine if higher abundances of copepod carcasses were present within boat wakes, net tows were taken opportunistically behind passing vessels in the lower York River, a tributary of Chesapeake Bay. “Inside wake” samples were collected within the wakes of the passing vessels, and corresponding “outside wake” samples were collected a short distance outside the wakes. A maximum of 5 corresponding pairs of “inside wake” and “outside wake” samples were collected behind each vessel. Since samples were collected opportunistically, size and speed of the passing vessels varied; actual speed and type of motor were not known.

Zooplankton samples were taken with a 0.5 m mouth diameter, 200 μm mesh net with a 200 μm filtering cod end. The net was towed horizontally at low speed (<1 m s$^{-1}$) for one minute just beneath the surface. The net and cod end were rinsed thoroughly between tows to avoid any carryover of carcasses. The cod end content was concentrated down to 100–200 ml, and gently poured into a staining jar through a 2500 μm mesh sieve to remove any gelatinous zooplankton, which interfere with staining. The concentrated samples were stained with Neutral Red. For longer term storage, the stained samples were filtered onto 200 μm nylon mesh disks, and rinsed copiously with filtered seawater to remove any excess stain. The mesh disks containing the zooplankton were then transferred to small petri dishes and placed on ice. Upon return to the laboratory the samples were stored at −40 °C until analysis (within 3 months).

2.3. Laboratory turbulence experiments

Laboratory experiments were conducted to directly measure A. tonsa mortality as a function of turbulence intensity. An experimental chamber (61 cm × 42 cm × 30 cm) was filled with 40.4 L of unfiltered York River water. Turbulence was generated with a stirrer made from a mixing paddle attached to a dual-speed motor (Fig. 1). The actual turbulence intensity in the chamber, expressed as energy dissipation rate ($ε$; cm$^2$ s$^{-3}$), was calculated as:

$$ε = c_p^{3/4} k^{3/2} l^{-1} \tag{1}$$

where $c_p$ is a coefficient with a typical value of 0.09 (Libby, 1996), $k$ is the Turbulent Kinetic Energy (TKE) calculated from the equation:

$$k = \frac{1}{2} \left( \overline{u_1'^2} + \overline{u_2'^2} + \overline{u_3'^2} \right) \tag{2}$$

where $u_1'$, $u_2'$ and $u_3'$ were velocity deviations calculated from particle velocities in the x, y and z directions as measured by a Sontek Acoustic Doppler Velocimeter (ADV) sampling at 10 Hz (e.g. Fig. 2). Finally, $l$ in Eq. (1) is given by:

$$l = K z \tag{3}$$

where $K$ is von Karman’s constant (0.41) and $z$ is the average distance of the ADV sample volume from the wall, 18 cm below the sensor.

![Fig. 1. Schematic diagram of experimental setup for turbulence experiments. Experimental chamber depth = 30 cm. Acoustic Doppler Velocimeter (ADV) was used to quantify turbulence intensity, and was removed prior to experiments with copepods.](image1)

![Fig. 2. Example of data collected with the ADV. Top panel: velocity of particles in the x, y and z directions within the experimental chamber through time. Bottom panel: turbulent kinetic energy (TKE) produced within the experimental chamber. The bold horizontal line depicts the average TKE.](image2)
height. In our experimental chamber, \( z = 16 \text{ cm} \). Turbulence intensity was measured for different combinations of paddle size \( (\text{small and large}) \) and motor speed \( (\text{slow and fast}) \). For the experiments with copepods, the ADV was removed from the experimental chamber. All other aspects of the setup remained the same to obtain the pre-determined turbulence intensities.

The copepod \( A. \) tonsa was taken either from a laboratory culture or from the York River, gently concentrated on a 200 \( \mu \text{m} \) mesh sieve, and resuspended in approximately 400 ml of 0.2 \( \mu \text{m} \) filtered seawater. These concentrated copepod samples were then split up into four equal portions with a plankton splitter. Each portion was exposed to one of four turbulence intensities in the experimental chamber, including zero turbulence as the control; each turbulence intensity was tested 4–5 times (Table 1).

To begin the experiments, copepods were gently transferred to the experimental chamber containing 10 \( \mu \text{m} \) filtered York River water and allowed to acclimate for 10 min. Afterward, the stirrer was placed in the chamber and turned on for 30 s. For the control, the setup was the same but the motor was not turned on. The control therefore would account for any carcasses naturally present among the copepods, and any mortality associated with the experimental procedure not due to turbulence. After exposure to turbulence, the experimental chamber was drained and all copepods were gently concentrated onto a 70 \( \mu \text{m} \) mesh sieve, resuspended in filtered seawater, and stained with Neutral Red. After staining, the samples were either processed immediately, or stored at \(-40^\circ \text{C}\) for up to 2 days before processing.

2.4. Statistical analyses

The fraction dead of each sample was calculated by dividing the number of dead \( A. \) tonsa (as determined by the Neutral Red staining method) by the total number of \( A. \) tonsa. All values of fraction dead were normalized with an arcsine square root transformation, and tested for normality with the Komolgorov–Smirnoff test. For the field study (Section 2.1), a nested ANOVA (date nested within site) of the normalized data was first performed to test for an overall significant effect due to sampling date and location. Because sampling date did not have a significant effect, all dates were subsequently pooled for each site, and a one-way ANOVA was performed to test for effect of sampling location. The 95% confidence intervals were compared to determine differences among sites. For the boat wake sampling (Section 2.2), a one-tailed paired \( t \)-test was used to determine if there was a higher fraction dead copepods in the “inside wake” samples than in the “outside wake” samples. For the laboratory turbulence experiments (Section 2.3), Pearson’s correlation and linear regression analyses were performed between fraction dead and turbulence intensity. All statistical analyses were done with Minitab Statistical software.

3. Results

3.1. Copepod carcasses at the field sites

There were no systematic differences in the measured physical parameters among the three sampling sites in the Hampton River (Table 2). Over the one month sampling period surface water temperatures ranged from 18.6 to 20 °C. On any given sampling day the temperatures among the three sites varied by no more than 0.4 °C. Salinity ranged from 18.7 to 19.7 psu over the month, and varied by less than 0.7 psu among the sites on each sampling day. During May, surface water dissolved oxygen content was between 6.31 and 6.91 mg L\(^{-1}\), and varied by no more than 0.5 mg L\(^{-1}\) on any sampling day. Within each site, the sampling date was not a significant source of variation in the fraction dead of the \( A. \) tonsa population (nested ANOVA, \( p = 0.639 \)). When samples from all sampling dates were pooled for each site, there was a significant difference in fraction dead of \( A. \) tonsa among sites (Fig. 3, one-way ANOVA, \( p = 0.012 \)). A comparison of the 95% confidence intervals showed that fraction dead of the copepod population was not significantly different between the marina and the shoreline, but was significantly higher in the channel.

3.2. Copepod carcasses in boat wakes

A total of 10 pairs of “inside wake” and “outside wake” samples were collected (Fig. 4). Wakes were generated by a variety of vessels, including small fishing boats, tugboats, tugboats pushing barges, yachts and a military landing craft. Samples with high zooplankton density were split, and at least 200 \( A. \) tonsa were counted within each sample and identified as live or dead based on staining patterns. \( A. \) tonsa accounted for 9–83% of the total zooplankton in the “inside wake” samples (average 38%), and 16–88% in the “outside wake” samples (average 39%). There was no significant difference between the two sample types in terms of the percent contribution of \( A. \) tonsa to the total zooplankton population \( (p = 0.786) \), or the total abundance of \( A. \) tonsa \( (9–7700 \text{ individuals m}^{-3}; p = 0.265) \). A higher fraction of \( A. \) tonsa was dead in the “inside wake” samples than in the corresponding “outside wake” samples, with one exception \( (p = 0.007) \). On average, 14.3% of \( A. \) tonsa were dead within the boat wakes, but only 7.7% were dead outside the wakes.

![Fig. 3. C弧carass prevalence at field sites. Average (± Standard Deviation) fraction dead of the Acartia tonsa population at each of the three sampling sites in the Hampton River.](image-url)
with an r² value of 0.34 (p=0.011). This equation indicates that a much higher boat-generated turbulence that could have caused copepod mortality.

Additional evidence for turbulence-induced mortality was provided by the field sampling in the York River, which showed that the fraction dead of copepods was twice as high within boat wakes as that outside boat wakes. The variable degree of increase in the fraction dead of A. tonsa among our “inside wake” samples (Fig. 4) could be attributed to different turbulence intensities generated by the different vessels. By sampling opportunistically, we could not control the type or speed of vessels producing the wakes, and we were not able to measure turbulence intensity within these wakes. The higher fraction dead within boat wakes could be explained by two possible mechanisms: 1) pre-existing copepod carcasses being concentrated from the surrounding water by the wakes coupled with active avoidance of the wakes by live copepods, or 2) mortality induced by a suddenly turbulent and stressful environment within the wakes.

Boat-generated turbulence is capable of resuspending sediments and increasing turbidity, especially in shallower systems (reviewed in Mosisch and Arthington, 1998). Also, wind-generated turbulence has been shown to effectively concentrate weak-swimming zooplankton on a vertical scale (Haury et al., 1990, 1992). It is possible that boat-generated turbulence may resuspend carcasses that have settled to the sediments or concentrate carcasses from the surrounding water column. However, if this occurred, an increase in the total A. tonsa abundance (live plus dead) would be expected among the “inside wake” samples in addition to an increase in carcass prevalence, but this was not observed in our study.

Active avoidance of boat-generated turbulence by live copepods is much more questionable. Various species and developmental stages of copepods can actively avoid turbulent surface waters through vertical migration (Mackas et al., 1993; Lagadeuc et al., 1997; Visser et al., 2001; Maar et al., 2006), and returned to shallower depths once the turbulent event has passed (Incze et al., 2001). These observations, however, are more relevant to moderate, sustained turbulence. While copepods may increase their jumping frequency in response to slightly elevated turbulence (ε = 0.054 cm² s⁻¹⁻¹; Saiz and Alcaraz, 1992b), it is uncertain whether this behavioral response could allow the copepods to escape much higher and sporadic turbulence in boat wakes. The reaction times of A. tonsa to hydrodynamic stimuli are on the order of milliseconds, and the copepod can move approximately 4.5 mm with one escape jump (Buskey et al., 2002). If we consider a copepod located along the mid-line of a 2 m-wide wake, 222 consecutive escape jumps perpendicular to the direction of wake formation would be necessary to remove the copepod from turbulent conditions. This rough calculation likely underestimates the required escape time and distance, as the copepod would also have to overcome random physical transport due to the turbulence (Yen et al., 2008). Considering these factors, concentration of copepod carcasses by boat wakes is possible, but avoidance of boat wakes by live copepods is unlikely, if not impossible.

As copepods were essentially trapped within the boat-generated wakes, the question then becomes whether or not turbulence levels were strong enough to cause mortality. Fields and Yen (1997) estimated that A. tonsa typically reside in estuaries with turbulence levels
that produce a Kolmogoroff scale of 6.5 mm, which is well above the body length of the copepod (ca. 1 mm). This means *A. tonsa* is normally not adversely affected by background turbulence in the environment. The intermittent, intense turbulence generated by boats could place the copepod in a suddenly stressful environment to which it is not accustomed and may result in death. It is possible that boat-generated turbulence created a suddenly stressful environment to which the copepods were unable to adapt, resulting in death. This hypothesis was addressed in our laboratory experiments.

In coastal zones and tidal fronts, the energy dissipation rates are on the order of 0.001–1 cm² s⁻³ and 0.1 cm² s⁻³, respectively (Kiørboe and Saiz, 1995). In our experiments, the medium (1.31 cm² s⁻³) and high (2.24 cm² s⁻³) energy dissipation rates were on the high end of the natural turbulence range in coastal environments. Nonetheless, these experimental turbulence levels were already strong enough to cause copepod mortality. Therefore, our results suggest that high background turbulence in coastal waters likely creates a stressful environment for copepods, and elevated turbulence, such as during a storm event, may directly result in copepod mortality. Turbulence created by motorized boats is likely orders of magnitude higher than what we were able to generate in our experiments. Loberto (2007) observed a dissipation rate as high as 310 cm² s⁻³ at a distance of 50 propeller diameters behind a 20 mm diameter, scale-model boat propeller running at 3000 rpm. This turbulence intensity is 2 orders of magnitude higher than the highest turbulence intensity we tested. A full size propeller running at comparable rpm is expected to produce even stronger turbulence in its wake. At these high turbulence levels, our Eq. (4) predicts that all of the copepods trapped within the wakes would be killed. Direct physical trauma incurred during contact with propellers or other solid surfaces may have also contributed to the copepod mortality observed in our studies. Microscopic examination of the samples, however, did not reveal any excessive mutilation of the copepods. Hence, the observed mortality was likely caused by stress induced by the high turbulence.

If boat-generated turbulence directly causes zooplankton mortality, as our data suggest, this mortality source is expected to be particularly important during summer months when recreational boat traffic increases, and in ports and harbors with heavy boat traffic. Non-predatory mortality such as this is rarely considered in the literature, yet it could be important for proper understanding of zooplankton ecology especially in coastal and estuarine waters (e.g. Elliott and Tang, accepted for publication). The fate of zooplankton carcasses is also of particular interest. If not immediately ingested, carcasses may also be subject to microbial decomposition within the water column, shunting zooplankton high quality organic material to depth, or be subject to other stress induced by the high turbulence.


