The turbulent life of copepods: effects of water flow over a coral reef on their ability to detect and evade predators

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ABSTRACT: The heterogeneous distribution of water flow over structurally complex environments, such as coral reefs, may play an important role in the interactions between copepods and planktivorous fish by interfering with the copepods' ability to detect and evade predators. The escape response and capture rates of the copepod *Acartia tonsa* were examined in laboratory flumes that created both unidirectional and oscillatory flow conditions similar to those found near coral reefs. Two turbulent regimes were produced in each flume: 'smooth' flow was formed using a grid collimator and 'rough' flow was generated by placing a branched coral skeleton upstream of the flume's working section. A predator was simulated by a fixed siphon to generate a stimulatory flow field. Copepod detection of the siphon was measured as the distance from the siphon tip to where an escape response was initiated. This reactive distance remained the same in low-flow conditions as in still water, but was reduced by 25% at higher flow speeds, indicating a decline in the copepods' ability to detect velocity gradients formed by the siphon. Rough turbulence regimes intensified the effect of current speeds, resulting in an even shorter copepod reactive distance. Capture rates of copepods by the siphon increased with current speed, wave motion, and in rough flow, while the capture rates of non-evasive prey, *Artemia* nauplii, did not vary with flume conditions. The differences in capture rates between evasive and non-evasive prey suggest that behavioral shifts in copepod escape thresholds may account for increases in predation by reef-dwelling fishes observed in hydrodynamically complex coral environments.

KEY WORDS: Turbulence · Copepods · Escape behavior · Planktivores · Coral reefs

INTRODUCTION

The importance of turbulence in planktonic predator–prey interactions has been explored both theoretically (Rothschild & Osborn 1988, Mackenzie et al. 1994) and experimentally (Clarke et al. 2005, Gilbert & Buskey 2005). Turbulence, defined as the time-varying component of water flow, can be formed in the ocean by wind, waves, tides, coastal upwelling, and contact with rough surfaces. Coral reefs are coastal areas with particularly complex water movements. As currents and waves move over the reef, the rough structure of the seafloor disrupts flow to form a range of hydrodynamic microhabitats (Hearn et al. 2001). This variation in the fluid environment can alter rates of planktivory via effects on prey availability, capture success by planktivores, or the escape mechanisms of prey.

Calanoid copepods are abundant over coral reefs (Heidelberg et al. 2004, Holzman et al. 2005) and are an important part of the diet of many coral reef fishes (Hobson 1991, Clarke 1999). To avoid predation, many copepods are capable of performing vigorous escape responses (Singarajah 1969). Hydrodynamic stimuli generated by feeding fish are detected by copepods

using their first antennae (Hartline et al. 1996, Kiørboe et al. 1999), which initiates thrusts of the copepods' powerful swimming legs to dart away from the predator stimulus. This effective antipredator behavior allows copepods to reach speeds up to 500 body lengths s^{-1} (Strickler 1975). Although they are physically capable of detecting the slightest stimuli, their response is dependent upon a flexible behavioral threshold. The critical value of stimulus at which copepods elicit an escape response represents a balance between the cost of performing a jump and the risk of predation (Fields & Yen 1997, Kiørboe et al. 1999). Previous studies have suggested that calanoid copepods in turbulent waters become habituated to continual hydrodynamic stimuli and are not able to detect predator attacks with the same degree of sensitivity as in calm waters (Singarajah 1975, Hwang et al. 1994, Gilbert & Buskey 2005). The plasticity of this threshold prevents an expenditure of energy performing unnecessary escape jumps, but results in an increase of prey capture rates in turbulent flows (Mackenzie & Leggett 1991, Clarke et al. 2005).

A recent study showed that the planktivorous coral reef blenny *Acanthemblemaria spinosa* was more successful at capturing the copepod *Acartia tonsa* in turbulent water compared to in still water (Clarke et al. 2005). Because the investigators also looked at blenny predation on the non-evasive prey *Artemia* sp. nauplii (henceforth referred to as *Artemia*), they were able to determine that the success in *A. tonsa* predation was due to some disadvantage to the copepods. This 'prey handicap' suggested that increases in hydrodynamic stimuli might cause copepods to become habituated to turbulence (Hwang et al. 1994), or it might reduce their ability to detect predator advances (Fields & Yen 1997). Waggett & Buskey (2007a) and Gilbert & Buskey (2005) examined copepod responsiveness in still-water and turbulent conditions and found stimuli sensitivity (measured in reactive distance) was reduced under turbulent regimes.

The effect of turbulence on prey behavior has typically been tested under isotropic conditions, where turbulence was generated by an oscillating grid (for review see Peters & Marrase 2000). In order to determine the role of turbulence in coral reef predator–prey interactions, appropriate simulations of water motion over a reef were necessary. Measurements of water velocity, turbulence, and wave period on a Caribbean coral reef were used to characterize water movements experienced by blennies and their zooplankton prey, which provided a guide to determine the range of environmental conditions utilized in this study (C. M. Finelli unpubl. data).

The purpose of the present study was to examine how changes in the hydrodynamic regime affected the ability of copepods to detect and respond to a predator. A fixed siphon was used to simulate planktivorous fish such as coral reef blennies, which live in small holes in the coral and are considered 'hemi-sessile' because they rarely leave their burrows (Clarke 1999). Blennies feed by extending into the water column, using brief attack lunges to capture passing zooplankton and rely on water motion to supply zooplankton prey. Suction emulates the feeding mechanism of these fish and also provides a constant fluid shear that reliably elicits escape responses from *Acartia tonsa* (Singarajah 1969, Drost et al. 1988, Kiørboe et al. 1999). Studies using siphons to generate suction flows are able to simulate predation, which generates the same escape response from copepods (Waggett & Buskey 2007a,b). Using a siphon eliminated predator variability so that the effects of water motion on copepods could be separated from the effects of predator abilities or selectivity (Vinyard 1980). To test prey behavior in conditions that more closely mimicked the natural reef environment, we examined (1) changes in the calanoid escape response to increases in current speed and in oscillatory flow formed by passing waves, and (2) how turbulence formed by coral structures affected copepod predator detection and capture rates when compared to non-evasive prey.

MATERIALS AND METHODS

Laboratory experiments using unidirectional and oscillatory flumes were conducted at the Marine Science Institute in Port Aransas, Texas. Copepods were collected from the nearby Aransas Ship Channel (27° 50.3' N, 97° 03.1' W) or Island Moorings canals (27° 48.40' N, 97° 05.53' W) by deploying a 0.25 m diameter, 153 µm mesh plankton net. The bulk zooplankton sample was diluted with whole seawater and aerated in the laboratory until used, within 18 h after collection. *Acartia tonsa* were isolated under a dissecting microscope using a large-bore Pasteur pipette. Only adult copepods with both first antennae intact were selected for experiments. The sorted *A. tonsa* were held in 100 ml beakers until they were added to a flume. All seawater used was filtered through a 20 µm mesh filter cup to remove debris.

Currents. Unidirectional water flow was generated in an oval 16.4 l paddle flume constructed of acrylic (Fig. 1). A 27.5 cm diameter paddle driven by a Dayton gear motor (Model 2H577A) with a Dart Micro-drive II controller was used to move water through a working channel with dimensions of $45 \times 10.1 \times 10.5$ cm (length $\times$ width $\times$ height). A 10 cm long working area was defined between 30 and 40 cm from the upstream end of the working section. A series of semi-circular vanes on
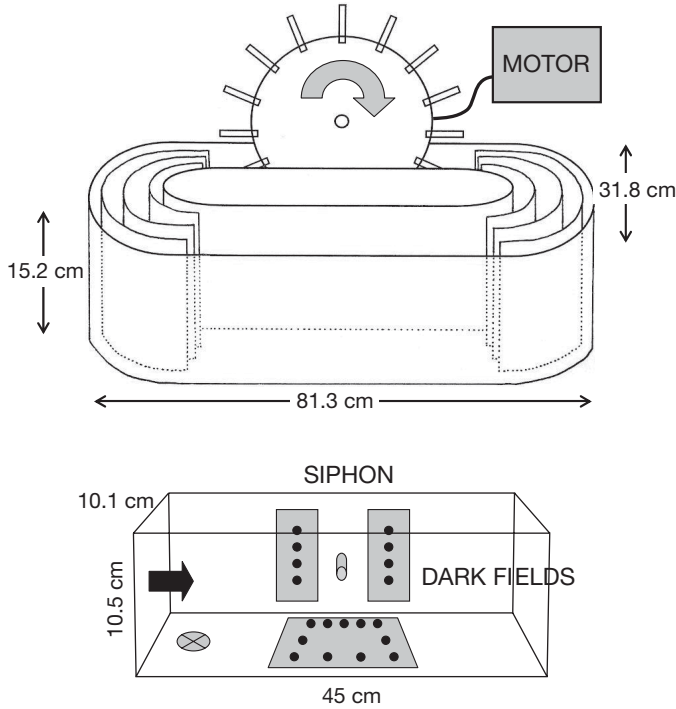
Fig. 1. Unidirectional flume that generated current velocities using a 27.5 cm diameter paddle wheel at 1, 3, and 9 rpm. Water was moved counter-clockwise through baffles on either end to straighten flow. The working area had a horizontally positioned siphon fixed 5 cm from the bottom of the flume. The arrow indicates direction of flow and the encircled X marks the location of an upstream branched piece of coral (rough flow treatment). For video data collection, dark field illumination surrounded the siphon for better image contrast

each end of the flume served to straighten flow through the semi-circular turns. A siphon made of glass tubing (1.8 mm inner diameter) was placed in the working area 5 cm from the bottom, and bent at a 90° angle in order to protrude horizontally 1 cm into the passing flow. Silicon tubing connected to the end of the siphon allowed water to flow continuously into a 500 ml beaker placed below the flume. Siphoned water was returned from the beaker to the flume by a peristaltic pump (Masterflex Model 7553-30) so that the water level and pressure head remained constant. The siphon suction was maintained at 2.2 ml s^{-1}, which produced a flow of 0.85 m s^{-1} at the tip of the siphon (empirical calculation based on flow velocity and the inner diameter of the siphon). This flow velocity is consistent with models of suction feeding in larval fishes (Drost et al. 1988). A 40 μm mesh filter cup encased the siphon outflow to prevent any captured copepods from returning to the flume.

Flow characterization: The flow of water through the unidirectional flume was set at mean velocities of 0, 3.6, 11.1, and 34.4 cm s^{-1} to correspond with the range of water velocities measured over the surface of a coral reef (Finelli unpubl. data; Table 1). Current speed and turbulence in the flume were measured with a SonTek 16 MHz Micro Acoustic Doppler Velocimeter (ADV). The ADV probe was positioned in the center of the working channel such that the volume of water sampled (0.09 cm^3 sample volume; default settings) was directly in front of the siphon tip. Measurements were made for 5 min at each flume setting at a sampling rate of 20 Hz. Measurements of turbulent water flow consist of mean and variance components such that:

$$u = \bar{u} + u'$$

To estimate turbulence parameters, we used a modified Reynolds decomposition to separate the mean and variance components of the 3 velocity signals (u, v, w). Each velocity measurement was subtracted from the mean for the entire 5 min record, thus leaving only the time-varying signals (Liiv 2001). The turbulent kinetic energy (TKE) was then calculated as:

$$\text{TKE} = 0.5(\overline{u'u'} + \overline{v'v'} + \overline{w'w'})$$

'Smooth' flow was created by placing a plastic grid $10 \times 3 \times 10$ cm (length × width × height, with 1 cm^2 openings) 30 cm upstream of the siphon. 'Rough' flow was formed by adding a branched coral structure 11.5 cm upstream of the siphon. The plastic grids remained in place for the rough flow treatments. These flow regimes were designed so that only turbulence varied between smooth and rough treatments, while water velocities remained similar (Table 1).

Copepod reaction sensitivity: Two Cohu solid state video cameras with Nikon Micro-Nikkor 55 mm lenses were positioned around the working area to focus on (1) the horizontal plane (10.5 cm^2) directly in front of the siphon tip and (2) the vertical plane (11.5 cm^2) surrounding the siphon tip. The 2 images were displayed

Table 1. Water velocities (cm s^{-1}) and turbulent kinetic energy (TKE in cm^2 s^{-2}; in parentheses) for conditions in unidirectional-flow- and oscillatory-flow-generating flumes

Flume Unidirectional	Flow	Current speeds		
		Low	Moderate	High
	Smooth	3.74 (0.07)	11.0 (0.31)	32.5 (1.17)
	Rough	3.56 (0.49)	11.2 (2.90)	36.3 (38.2)
		Wave amplitudes		
Oscillatory		10 cm	20 cm	30 cm
	Smooth	7.80 (0.09)	15.9 (0.41)	24.3 (1.30)
	Rough	8.63 (0.77)	22.3 (5.99)	33.5 (20.6)

on 1 monitor using a Pelco VSS200DT video splitter, which was then recorded on a Panasonic AG2560 video-cassette recorder. To produce high contrast images, dark field illumination was produced by infrared light-emitting diodes placed both beneath and behind the working area, but outside the video camera field of view (Fig. 1). Two fiber optic lights (Fostec EKE 150 W illuminator and Dolan-Jenner Fiber Lite 180 high-intensity illuminator) provided additional lighting from above.

Groups of 200 copepods (densities of 12 l^{-1}) were placed in the unidirectional flume to acclimate to the water flow for 10 min. The copepods were tested in smooth and rough flows at mean water velocities of 0 (control), 3.6, and 11.1 cm s^{-1}; the highest velocity treatment (34.4 cm s^{-1}) was not tested in this case because bubbles generated at high flow speeds made accurate image recognition of copepods difficult. Experiments lasted a maximum of 20 min and were timed based on the moment the siphon was turned on until it was turned off again. Limiting the duration of experiments and emptying and refilling the flume between each replicate so that a new group of cope-pods was used, prevented repeated stimulation of copepod escape responses and reduced escape perfor-mance due to copepod fatigue. Each treatment was repeated 4 to 5 times. Experiments were recorded at 30 frames s^{-1} on standard VHS video tapes. Video seg-ments of escape jumps were converted from analog to digital using Pinnacle Studio Plus (V. 9.4.3 Pinnacle Systems). Using Virtual Dub freeware (V. 1.6.14 Avery Lee), individual jumps were dubbed and saved as sequences of bitmap images. The frame showing the initial escape behavior was imported to Image J free-ware (V. 1.366 Wayne Rasband). Having both vertical and horizontal aspects on the same split screen pre-vented the use of 1 uniform calibration scale. By locat-ing the position of the copepod in both planes, reactive distance was measured from combining the distances between the tip of the siphon and the copepod using the Pythagorean theorem. All measurements were taken in pixels and later converted to millimeters using individual calibration scales in a Microsoft Office Excel (2003 V. 11.6113.5703) spreadsheet.

Capture rates: The rate of prey capture by the siphon predator was compared for evasive copepods (*Acartia tonsa*) and non-evasive *Artemia* nauplii by counting the number of organisms actually drawn into the siphon and retained within a filter cup. *A. tonsa* adults were collected using the same methods de-scribed above. *Artemia* nauplii were hatched by hy-drating cysts in aerated whole seawater for 36 h. To form natural prey densities of 12 l^{-1}, 200 individuals were selected under a dissecting microscope with a large-bore Pasteur pipette. Once the copepods or

Artemia were added to the unidirectional flume, water currents were generated at mean velocities of 3.6, 11.1, or 34.4 cm s^{-1} (low, moderate, and high) with smooth or rough conditions. Organisms were allowed to accli-mate for 10 min. Experiments ran for 10 to 20 min and were repeated 4 to 5 times with each replicate using new organisms. The duration of experiments was set to limit the possibility of reducing the effectiveness of escape responses due to copepod fatigue, while also using time frames comparable to the reaction distance experiments. A 40 µm mesh filter cup encased the siphon outflow and was removed at the end of the experiment to count all captured copepods or *Artemia* by rinsing the contents into a Petri dish. Swimming behavior of captured individuals was also observed after each experiment to determine any detrimental effects of being run through the flumes. Capture rates were calculated as the number of organisms caught relative to the duration of the experiment (min^{-1}).

Statistical analysis: We used a 2-way analysis of variance (ANOVA) with Tukey's pairwise comparisons (SPSS V. 13.0, SPSS) to test for the effects of flow speed, turbulence treatments, and the interaction be-tween flow and turbulence on copepod reactive dis-tances measured in the unidirectional flume. To ana-lyze capture rates, we first log-transformed all data, then performed a 3-way ANOVA to test for significant differences in capture rates between prey behaviors (evasive and non-evasive) in combination with flow speed and turbulence. To more closely analyze the effects of flow and turbulence on the capture rate of evasive and non-evasive prey, we performed a 2-way ANOVA with Tukey's pairwise comparisons. We used linear regression to examine the relationship between reaction distance and capture rate.

Wave motion. Waves passing over a coral reef push water back and forth below the surface. A 21.5 l U-shaped oscillatory flume (Fig. 2) reproduced this motion at a wave period of 5 s, which is similar to the wave period measured on a Caribbean coral reef (C. M. Finelli unpubl. data). Water flow in the flume was driven by a piston attached to a Scotch Yoke that trans-lated the rotational motion of the motor (Dayton Model 2H577A) to the linear motion in the piston. The motor was controlled with a Dart Micro-drive II controller. The working channel of the flume measured 36.8 × 10.1 × 10.5 cm (length × width × height). A glass siphon was positioned 5 cm from the bottom of the working area and protruded 1 cm horizontally into passing flow using identical materials and settings as the unidirec-tional flume. Wave amplitude (i.e. piston travel dis-tance) was set to 10, 20, and 30 cm via a moveable pin connecting the drive wheel to the Scotch Yoke. Again, the mean and variance components of the flow signal were separated using modified Reynolds decomposi-
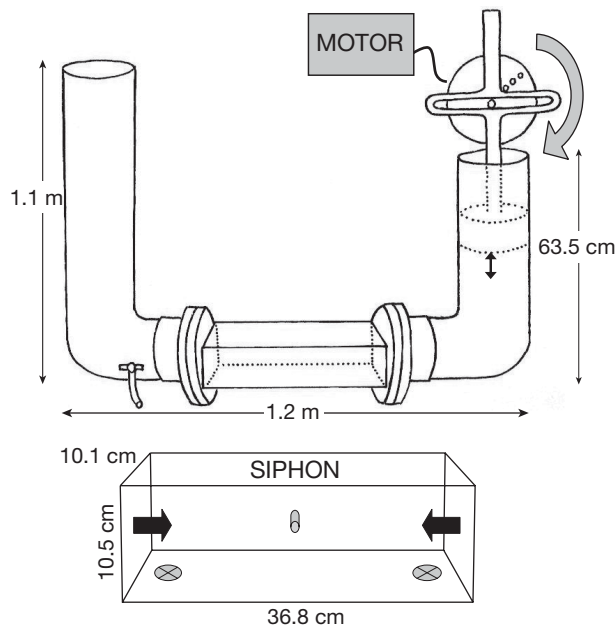
Fig. 2. Oscillating flume generating wave motion using a piston driven by a gear motor and Scotch Yoke. As the piston was lowered and raised, water moved back and forth through the working area (arrows indicate direction of flow). A siphon was positioned horizontally and located 5 cm from the bottom of the flume. To form smooth flow, plastic grids (not shown) were placed on either side of the siphon. The encircled Xs mark the locations where identical branched coral structures were placed to create rough flow

tion. As in the unidirectional flume experiments, the time-varying signal was generated by the difference from the mean. However, because the flow is oscillatory, an appropriate mean was estimated by subjecting the raw time series to a zero-phase displacement running average (*filtfilt* function, Matlab V.14, Mathworks; Table 1). To create smooth flow in both directions, 2 plastic grids $10 \times 4 \times 10$ cm (length × width × height, with 1 cm^2 openings) were placed in the working area, 18.4 cm on either side of the siphon. Rough flow was formed by adding uniform aquarium models of branched coral, 12 cm on either side of the siphon (Fig. 2). The plastic grids remained in place for the rough flow treatments.

Capture rates: Capture rates of evasive and non-evasive prey were calculated by counting the number of organisms drawn into the siphon and collected within a filter cup, as previously described. Using comparable prey densities ($12\,l^{-1}$), groups of 260 individuals of *Acartia tonsa* or *Artemia* nauplii were placed in the oscillatory flume and tested at wave amplitudes of 10, 20, or 30 cm, with both smooth and rough flow treatments. Experiments were run for 15 to 20 min in order to reduce the potential of copepod fatigue due to repeated escape responses and for consistency between

experiments in both flumes. Capture rates were measured by the number of individuals caught per minute.

Statistical analysis: Capture rates for both evasive and non-evasive prey were log transformed. A 3-way ANOVA was used to test the difference in capture rates between prey behaviors (evasive and non-evasive), in combination with the effects of wave amplitude and turbulence. The effects of wave amplitude and turbulence on copepod and *Artemia* capture rates were analyzed using a 2-way ANOVA, with Tukey's pairwise comparisons. In addition, the differences between capture rates in unidirectional and oscillatory flow generated by the 2 flumes were tested using an ANCOVA. To isolate the effect of flow direction on capture, an optimal model was selected from a full-factorial analysis. The initial model included a 3-way interaction, 2-way interactions, and all the effects of current, turbulence, and direction. Interaction terms that were not statistically significant were removed, leaving an optimal model examining the effect of unidirectional or oscillatory flow on capture rates.

RESULTS

Current speeds

Copepod reaction sensitivity

The distance from the siphon tip at which *Acartia tonsa* first reacted with an escape jump was compared between smooth and rough flow in still water and in low and moderate current regimes (Fig. 3). Coral was added in still water as a control and had no effect on
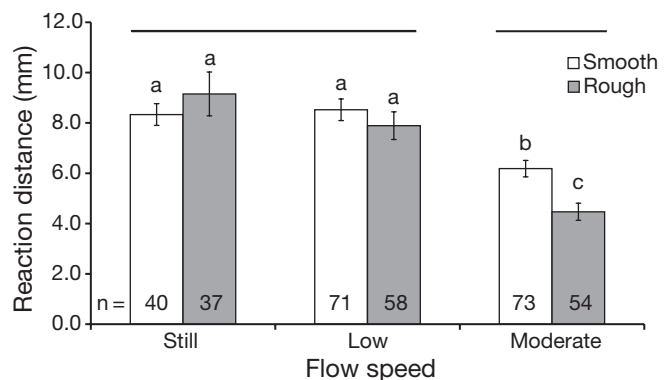

Fig. 3. *Acartia tonsa*. Mean reaction distance (mm) at which copepods responded to a siphon predator in still water and in low (mean velocity = 3.6 cm s^{-1}) and moderate (mean velocity = 11.1 cm s^{-1}) currents for smooth and rough flow treatments. Error bars represent standard error. Horizontal lines indicate similarities in reaction distance between current speeds (Tukey's test; $p > 0.05$), while different letters above bars indicate significant differences between flow treatments (1-way ANOVA; $p < 0.05$)

reaction distances, since the water was not moving (average ± standard error; smooth = 8.3 ± 0.35 mm, rough = 9.2 ± 0.58 mm). Copepods responded at similar distances in still water and in low flow (smooth = 8.5 ± 0.29 mm, rough = 7.9 ± 0.40 mm). At moderate flow speeds, copepod reactive distance decreased by ca. 25% in the smooth treatment (6.2 ± 0.22 mm) and by 27% with rough flow (4.5 ± 0.25 mm). When compared to still water, the combined effect of water current and increased turbulence in the moderate/rough treatment reduced the distance that copepods reacted to the siphon predator by more than half. Overall, current speed significantly affected copepod reactive distance (Table 2, $p < 0.001$). Although the singular effect of turbulence was nearly significant ($p = 0.072$), the interaction between current and turbulence did have a significant effect on reaction distances ($p = 0.002$).

Capture rates

Capture of non-evasive *Artemia* nauplii did not vary between flow treatments (Fig. 4, Table 3, $p > 0.05$) and was significantly different from copepod capture rates (behavior df = 1, $p < 0.001$, $R^2 = 0.903$; 3-way ANOVA). In comparison, the evasive *Acartia tonsa* showed a significant increase in capture rate with current speed (Fig. 4, Table 3, $p < 0.001$). At low current speed, there was no distinguishable difference between capture rates in smooth and rough flow (capture rate ± standard error; smooth = 0.10 ± 0.01 individuals min^{-1}, rough = 0.10 ± 0.04 individuals min^{-1}). Turbulence had the greatest effect at moderate current speed, more than doubling capture rates between smooth and rough flow (smooth = 0.20 ± 0.06 individuals min^{-1}, rough = 0.55 ± 0.07 individuals min^{-1}). At the maximum current speed tested, capture rates were highest, but the effect of rough flow conditions negatively affected capture (smooth = 0.91 ± 0.07 individuals min^{-1}, rough = 0.75 ± 0.12 individuals min^{-1}). The changing effect of enhanced turbulence in rough flow
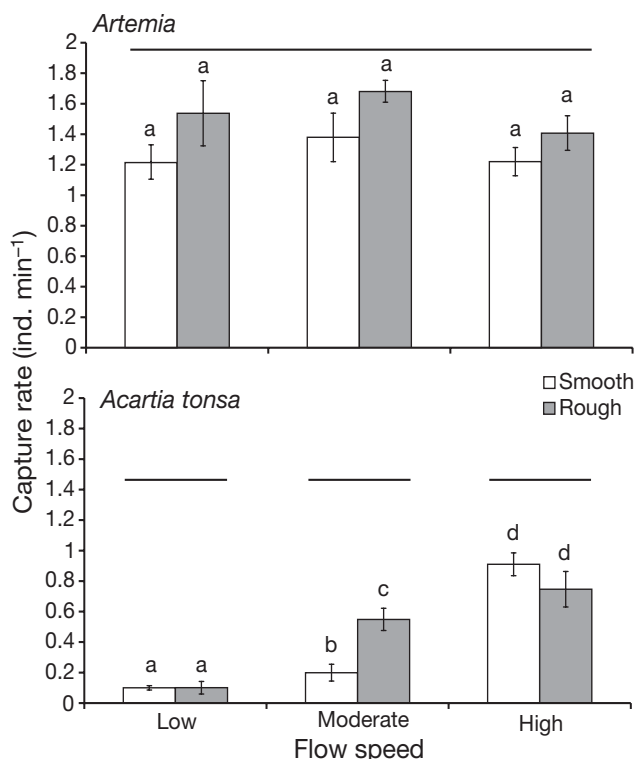

Fig. 4. *Artemia* sp. and *Acartia tonsa*. Mean capture rates (min^{-1}) for non-evasive *Artemia* nauplii and the evasive copepod *A. tonsa*, at low, moderate, and high current speeds (mean velocities = 3.6, 11.1, and 34.4 cm s^{-1}, respectively) in a unidirectional flume. Error bars represent standard error; n = 5 for each treatment. Horizontal lines indicate similarities in capture rates between current speeds (Tukey's test; $p > 0.05$), while different letters above bars indicate significant differences between flow treatments (1-way ANOVA; $p < 0.05$)

Table 2. Two-way ANOVA of copepod reactive distances in response to a siphon predator at 3 unidirectional current velocities in smooth and rough flow treatments. Significant values are shown in bold

Source of variation	df	MS	*F*	p
Current	2	373.945	60.475	**<0.001**
Turbulence	1	20.192	3.266	0.072
Current × Turbulence	2	38.288	6.192	**0.002**
Residual	327	6.183		

$R^2 = 0.289$; adjusted $R^2 = 0.278$

at different water velocities on copepod capture suggests a significant interaction between turbulence and flow regimes (flow × turbulence $p = 0.001$).

Current speeds and turbulence affected both copepod reaction distances from the siphon tip and capture rates by the siphon, suggesting that a reduction in their ability to detect and evade flow fields resulted in increased capture success. To examine this, copepod reactive distances and capture rates at low and moderate current velocities were first tested in an ANCOVA to determine which factor (current or turbulence) was a more suitable predictor. Turbulence had a significant effect ($p = 0.011$, $R^2 = 0.767$) and co-varied with copepod reactive distances ($p = 0.016$) so that in a regression analysis, smooth and rough flow were analyzed separately. A linear regression model showed that in rough conditions, capture rate increased as reactive distance was reduced (Fig. 5; $p = 0.035$, $R^2 = 0.447$). In smooth flow, there was no significant relationship between copepod reaction distance and capture rate ($p = 0.360$, $R^2 = 0.110$).

Table 3. Two-way ANOVA of non-evasive and evasive prey capture rates at 3 current velocities generated by a unidirectional flume in smooth and rough flow treatments. Significant values are shown in bold

Source of variation		Non-evasive *Artemia*				Evasive *Acartia tonsa*		
	df	MS	F	p	df	MS	F	p
Current	2	0.004	1.284	0.295	2	0.120	68.802	**< 0.001**
Turbulence	1	0.018	5.797	**0.024**	1	0.004	2.283	0.144
Current × Turbulence	2	0.000	0.119	0.889	2	0.016	8.876	**0.001**
Residual	24	0.003			24	0.002		
		$R^2 = 0.264$; adjusted $R^2 = 0.110$				$R^2 = 0.868$; adjusted $R^2 = 0.840$		

Wave motion

Capture rates of non-evasive prey remained relatively constant with increasing wave amplitude and corresponding water velocities (Fig. 6, Table 4, $p > 0.05$), and were significantly different from capture rates of evasive prey (behavior df = 1, $p < 0.001$, $R^2 =$ 0.639; 3-way ANOVA). In contrast, copepod capture rates increased with wave amplitude (Fig. 6, Table 4, $p = 0.017$). Copepods were captured at minimal rates for the smallest level of wave motion, while, in waves of 20 cm amplitude, rough conditions increased capture by 72 % (smooth = 0.65 ± 0.17 individuals min^{-1}, rough = 1.12 ± 0.07 individuals min^{-1}). Differences between prey capture in smooth and rough flows existed at wave amplitudes of 20 cm, but were less distinguishable in both lower and higher waves. In larger wave amplitudes of 30 cm, capture rates remained consistently high (Fig. 6). Yet at these levels of wave motion, copepods in smooth conditions were captured 45 % less often than non-evasive *Artemia* prey (evasive = 0.817 ± 0.09 individuals min^{-1}, non-evasive = 1.5 ± 0.10 individuals min^{-1}), while in rough conditions, copepod capture rate was similar to *Artemia* capture rate (evasive = 1.08 ± 0.31 individuals min^{-1}, non-

evasive = 1.2 ± 0.14 individuals min^{-1}). This suggests that the copepod escape behavior is less effective in rough turbulence and copepods more closely resemble non-evasive zooplankton.

An optimal ANCOVA model was constructed to test the effect flow direction (unidirectional or oscillatory) had on capture rates for non-evasive and evasive prey (Table 5). The initial model included 2-way and 3-way interactions between water velocity, turbulence, and
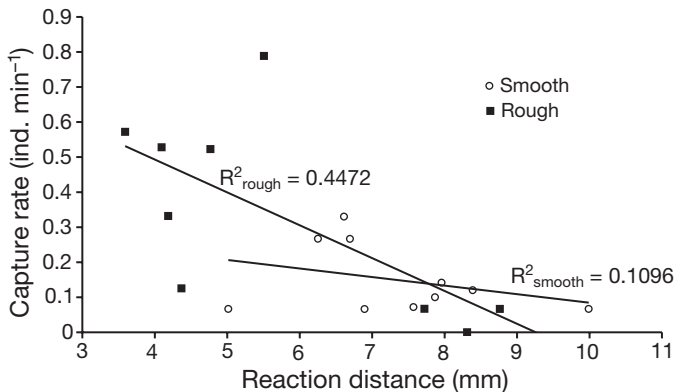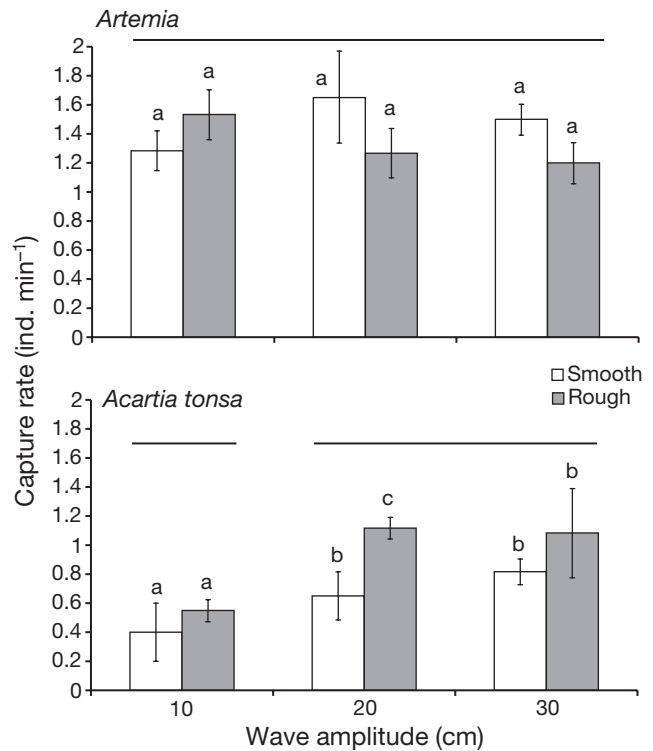

Fig. 6. *Artemia* sp. and *Acartia tonsa*. Mean capture rates (min^{-1}) for non-evasive *Artemia* nauplii and the evasive copepod *A. tonsa*, in 10, 20 and 30 cm amplitude waves (period = 5 s; mean velocities = 8.2, 19.1, and 28.9 cm s^{-1}, respectively) in an oscillating flume. Error bars represent standard error; n = 4 for each treatment. Horizontal lines indicate similarities in capture rates between current speeds (Tukey's test; $p > 0.05$), while different letters above bars indicate significant differences between flow treatments (1-way ANOVA; $p < 0.05$)

Fig. 5. *Acartia tonsa*. The relationship between reaction distance and capture rates in smooth ($y_{smooth} = -0.0244x + 0.3285$) and rough ($y_{rough} = -0.0938x + 0.8682$) flow treatments in a unidirectional flume

Table 4. Two-way ANOVA of non-evasive and evasive prey capture rates in wave motion (wave period = 5 s) at 3 wave amplitudes generated by an oscillating flume with smooth and rough flow treatments. Significant values are shown in bold

Source of variation	Non-evasive *Artemia*				Evasive *Acartia tonsa*			
	df	MS	F	p	df	MS	F	p
Amplitude	2	0.001	0.123	0.885	2	0.034	5.164	**0.017**
Turbulence	1	0.003	0.626	0.439	1	0.032	4.738	**0.043**
Amplitude × Turbulence	2	0.007	1.594	0.230	2	0.003	0.411	0.669
Residual	24	0.005			18	0.007		
	$R^2 = 0.184$; adjusted $R^2 = 0.043$				$R^2 = 0.469$; adjusted $R^2 = 0.321$			

direction. The 3-way interaction (velocity × turbulence × direction) and a 2-way interaction (turbulence × direction) were removed from the model to isolate the effect of flow direction. R^2 values for the evasive prey model were not altered by the omission of unnecessary interaction terms ($R^2_{initial} - R^2_{optimal} \leq 0.003$). The copepod capture rate was greater in oscillatory flow than in unidirectional flow (Figs. 4 & 6, Table 5, p = 0.023). The non-evasive capture rate was not significantly affected by the difference in flow direction between the 2 flumes (p = 0.667).

DISCUSSION

The effects of water flow and turbulence on planktonic predator–prey interactions were tested in the present study using flumes to mimic water movements over a coral reef. This provided the opportunity to observe changes in prey behavior in response to conditions that are more comparable to the environment zooplankton experience in nature. The range of water velocities, wave amplitudes, and turbulence generated by the unidirectional and oscillatory flumes corresponded to measurements taken over holes in coral occupied by blennies (C. M. Finelli unpubl. data). These values also fall within velocity ranges from previous field observations (Hamner et al. 1988, Leichter

et al. 1998, Sebens et al. 1998), flume studies (Helmuth & Sebens 1993, Falter et al. 2006), and hydrodynamic model estimates (Carleton et al. 2001). Using a siphon to simulate blenny feeding eliminated predator variability so that the effects of speeds, wave motion, and coral-generated turbulence on copepod escape behavior could be separated from the effects of predator behavior. Testing non-evasive prey under the same experimental conditions provided a standard to which the effectiveness of copepod escape behavior was compared. *Artemia* are widely considered to be representative zooplanktonic prey that lack any ability to evade predation, so their rate of capture can be interpreted as maximum predator success (Trager et al. 1994, Clarke et al. 2005). The difference between copepod and *Artemia* capture rates represents the efficiency of prey escape behavior.

The flumes used in this study generated flow to simulate currents and waves moving over a coral reef. Both processes of water movement play important roles in planktonic predator–prey interactions on reefs. Currents are responsible for delivering pelagic plankton, such as calanoid copepods, to the numerous and densely gathered reef planktivores (Hamner et al. 1988, Hobson 1991), while waves provide mixing and mass-transfer between surface waters and the benthos (Helmuth & Sebens 1993, Hearn et al. 2001). Both these physical processes can alter the biology of preda-

Table 5. Optimal ANCOVA model of non-evasive and evasive prey capture rates in 2 flow directions (unidirectional and oscillatory), in smooth and rough flow treatments. The full-factorial model included all interaction terms ($R^2_{Non-evasive} = 0.173$, $R^2_{Evasive} = 0.682$), while in the optimal model an insignificant 3-way interaction term (Velocity × Turbulence × Direction) and 2-way interaction term (Direction × Turbulence) were removed. Significant values are shown in bold

Source of variation	Non-evasive *Artemia*				Evasive *Acartia tonsa*			
	df	MS	F	p	df	MS	F	p
Velocity	1	0.005	1.189	0.281	1	0.226	51.472	**<0.001**
Turbulence	1	0.011	2.758	0.103	1	0.021	4.888	**0.032**
Direction	1	0.001	0.187	0.667	1	0.024	5.494	**0.023**
Velocity × Turbulence	1	0.006	1.483	0.229	1	0.011	2.409	0.127
Velocity × Direction	1	0.001	0.151	0.699	1	0.000	0.043	0.836
Residual	48	0.004			48	0.004		
	$R^2 = 0.080$; adjusted $R^2 = -0.015$				$R^2 = 0.679$; adjusted $R^2 = 0.646$			

tor–prey interactions based on the effect that small-scale water motions have on prey behavior. As current speeds increased, the ability of copepods to detect and react to a predator was reduced. At the low flow speed, copepod reactive distance remained relatively high (Fig. 3) and capture rates were minimal (Fig. 4). This suggests that at this current speed, the mechano-sensory capabilities of copepods were not diminished, or that the threshold for eliciting an escape response remained highly sensitive. In moderate flow conditions, reaction distances decreased significantly and the addition of coral-generated turbulence had an even greater effect in reducing copepod detection abilities (Fig. 3, Table 2). The reduction in prey sensitivity to predator-generated flow corresponded to increased capture rates, or predation success (Figs. 4 & 5). This negative relationship is evident; as copepods become less responsive they are more likely to become captured (Fields & Yen 1997). Interestingly, the relationship was different between smooth and rough flows. In smooth flow, variation in reaction distance did not have any effect on capture rates, which suggests that no matter how far away copepods react to the hydrodynamic stimulus, they are still able to effectively evade predation. In rough turbulent treatments, capture rates were more steeply negatively correlated with reaction distance so that even a small reduction in the distance at which copepods reacted to predators resulted in an increase in capture rates.

Copepod capture rates also significantly increased with wave motion. Compared to non-evasive prey, copepods were captured at lower rates in 10 and 20 cm amplitude waves (Fig. 6), particularly in smooth conditions. Capture of both copepods and non-evasive *Artemia* were at approximately the same rate in rough conditions with 30 cm waves, suggesting that at these levels copepod evasive behavior was ineffective and they became non-evasive zooplankton. Copepods were either not able to detect the siphon-generated velocity gradient at such rapid speeds or they were physically incapable of performing escape jumps against the water flow. The difference existing between smooth and rough conditions, even in oscillating water movements, demonstrates that copepods are still capable of detecting predators as they are carried over the reef by waves, while the addition of coral-generated turbulence inhibits their escape response.

In the present study, capture rates of copepods were significantly different between unidirectional and oscillatory flow. While the velocity and turbulence levels associated with wave motion and currents overlapped (Table 1), copepods were captured at higher rates in the oscillatory flume, suggesting that the flow formed by waves more greatly reduced copepod escape behavior (Figs. 4 & 6). The constant change in

flow direction during wave oscillations may heighten the effect of current speed and turbulence on copepod escape behavior (Heidelberg et al. 1997). Helmuth & Sebens (1993) found that oscillatory flow reduced benthic boundary layer effects by increasing water velocities closer to the reef surface. Therefore, wave motion can raise encounter rates between copepods and tube-dwelling blennies, while an inhibition of copepod escape responses due to flow can result in increased predation or improved predator success. This apparent advantage to the blenny may account for the increase in copepod zooplanktivory near the benthos (Motro et al. 2005) and the depletion of copepods <1 m above the reef (Clarke 1999, Holzman et al. 2005).

Turbulent microhabitats leave copepods vulnerable to predation, yet they might not benefit from turbulence-avoidance behavior (Franks 2001). In coral reefs, turbulence can be formed at the surface by wind and waves and at the bottom by interactions with the rough coral structure (Carleton et al. 2001). Zooplankton that demonstrate turbulence-avoidance behavior are unable to reach the relative safety of deeper and less turbulent waters in the shallow reef environment and must opt instead to seek shelter amongst the coral structures. However, this secure environment may restrict flow and result in a lack of sufficient water movement. Copepods can benefit from small levels of turbulence, which may provide increased food concentration and improve feeding rates (Saiz et al. 1992, Saiz & Kiørboe 1995). While turbulence-avoidance behavior is not an efficient strategy for copepods, vertical distribution is one way to alleviate predation pressure (Alldredge & King 1985). Copepods may migrate short distances above the reef to avoid the limitations of the benthic boundary layer and reduce predation by sessile organisms. Migrating too far above the reef exposes plankton to fish that are commonly found just above the benthos, restricted to this region in order to avoid their own piscivorous predators (Motro et al. 2005). Predators like the tube blenny benefit from this biological–physical relationship. *Acanthemblemaria spinosa* preferentially occupy burrows higher up on the reef structure (Clarke 1999). This can elevate blennies above the depleted benthic boundary layer and expose them to more energetic microhabitats. Increased water movements can produce enhanced encounter rates with zooplankton prey (Sundby 1997, Mackenzie & Kiørboe 2000), but this does not always result in increased ingestion or capture rates (Marrase et al. 1990, Mackenzie & Leggett 1991). Therefore, it is the effect of turbulence in reducing prey escape behavior that plays an important role in predation success. For reef blennies, turbulence can result in more predator–prey interactions and greater capture

success of evasive copepods (Clarke et al. 2005, Waggett & Buskey 2007b).

The present study has examined the behavioral shift in copepod escape responses to multiple levels of turbulence. Turbulence over coral reefs is several orders of magnitude larger than turbulence found in other coastal, shelf, and even oceanic regions (Marrase et al. 1990, Hearn et al. 2001). Rough flow formed by water passing over coral structures can increase the spatial variation in water movements (Roberts et al. 1975), which produces more ambient hydrodynamic stimuli, thereby raising the threshold for copepods to perform escape jumps and potentially masking predator attacks (Singarajah 1975, Fields & Yen 1997). In such a variable environment, the dome-shaped effect of increasing turbulence on the predation success of planktivorous fish has been well described (Mackenzie et al. 1994), while changes in prey behavior over an increasing range of turbulence has thusfar been less well studied. The present study shows how copepod escape capabilities are efficient at low levels of flow and how turbulence begins to inhibit their escape response at intermediate levels.

This study elucidates the changes in evasive prey behavior in response to varying hydrodynamic conditions. In replicating water movements that more closely resemble the natural environment, the effect of currents, wave motion, and coral-generated turbulence on copepod escape behavior can be applied to further our understanding of planktonic predator–prey interactions on coral reefs. In addition, the use of hemisessile blennies as model predators makes this study relevant to other benthic planktivores, such as corals, which extend tentacles into passing water to capture prey, and particularly to siphon-feeders, such as bivalves.

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