

Long Lag Times and High Velocities in the Motility of Natural Assemblages of Marine Bacteria

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The motility characteristics of natural assemblages of coastal marine bacteria were examined. Initially, less than 10% of the bacteria were motile. A single addition of tryptic soy broth caused an increase in the motile fraction of cells but only after 7 to 12 h. Motility peaked at 15 to 30 h, when more than 80% of cells were motile. These results support the proposal that energy limits motility in the marine environment. Cell speeds changed more than an order of magnitude on timescales of milliseconds and hours. The maximum community speed was $144 \mu\text{m s}^{-1}$, and the maximum individual burst velocity was $407 \mu\text{m s}^{-1}$. In uniform medium, speed was an inverse function of tryptic soy broth concentration, declining linearly over 0.001 to 1.0%. In media where concentration gradients existed, the mean speed was a function of position in a spatial gradient, changing from 69 to $144 \mu\text{m s}^{-1}$ over as little as 15 to 30 μm . The results suggest that marine bacteria are capable of previously undescribed quick shifts in speed that may permit the bacteria to rapidly detect and keep up with positional changes in small nutrient sources. These high speeds and quick shifts may reflect the requirements for useful motility in a turbulent ocean.

Motility is found among bacterial isolates from many environments, where it allows a chemotactic response to chemical gradients of attractants and repellents. These compounds represent signals for nearby nutrient sources and new habitats. Nutrient sources in the marine environment may be suspended particles or eukaryotic organisms to which bacteria are attracted because of the comparatively low nutrient concentrations dissolved in seawater (8, 9, 14, 21). Finding regions of increased nutrient concentration is important for maintaining motility as well as growth. Macnab (17) has pointed out the high genetic and biosynthetic expense of producing and maintaining the 60 or more proteins necessary for taxis in the single chemosensory system of *Escherichia coli*. The expense would appear to be even greater for some marine bacteria (*Vibrio* spp.) that possess two separate propulsion systems, one driven by protons and one driven by sodium ions (2). Additionally, Malmcrona-Friberg et al. (18) have suggested that two chemosensory systems exist in *Vibrio* spp., one each for starving and growing cells. Whether the dual-ion propulsion system and the dual-growth-state chemosensory system are the same remains to be determined, but sensory and motility costs are probably high in the marine environment.

The energetic expense of maintaining chemotactic movement in *E. coli* cultures has been calculated to be a small fraction of the total energy available to a bacterium (24). This allows *E. coli*, and presumably other bacteria, to move continuously in random walks biased toward an attractant. For this to produce chemotaxis, a 1- or 2- μm bacterium must produce enough power to swim at least $30 \mu\text{m s}^{-1}$ to compensate for rotation from Brownian motion and diffusion of the attractant (5, 24). For marine bacteria, commonly 0.2 to 0.6 μm long (16) and living at low ambient nutrient concentrations, rotation from Brownian motion and diffusion require these bacteria to move in excess of $100 \mu\text{m s}^{-1}$ (19). As the power requirement

increases with the square of the speed, the corresponding metabolic demand can consume all of the energy available to the cell. In some cases, it may be an energetic advantage to cease movement for some period. From this it follows that not all marine bacteria move all the time, despite models assuming the contrary (6, 14, 15).

Although models of marine bacterial motility exist (3, 15, 21) and there are studies of single isolates in culture (4, 18), work on the fraction of motile bacteria in natural samples is lacking. To determine the fraction of marine bacteria that are motile at a given time and how this community is influenced by changes in nutrients, experiments were conducted on natural communities of coastal marine bacteria.

MATERIALS AND METHODS

Samples were collected in sterile 125-ml glass bottles from the end of the Brighton Jetty in Gulf St. Vincent, South Australia. The gulf is approximately 7,000 km², with a mean depth of 20 m. The depth at the end of the jetty was about 3 m. Samples were returned to the laboratory within 30 min of sampling, and the experiments were begun immediately.

Tryptic soy broth (TSB) was added from sterilized concentrated stock to make final concentrations of 1, 0.1, 0.01, and 0.001%. TSB was chosen as a nutrient source because it did not appear to inhibit or limit motility. Seawater without nutrient addition was used as a control. Most experimental treatments were unshaken. To test the effect of shaking, however, one experiment was performed in which one set of bottles was shaken in a shaking water bath at 20°C while unshaken bottles were kept at the same temperature. Subsamples were withdrawn at intervals for optical density measurements (600 nm), acridine orange direct counts (AODC), and microscopic examination of motility. Poretics 0.2- μm -pore-size black polycarbonate membrane filters were used to prepare 2-ml aliquots for epifluorescence microscopy by the method of Hobbie et al. (12).

For microscopic examination of motility, a chamber was created on a glass slide from 100- μm -thick sections of coverslip, overlain with a whole coverslip. The sample was added at one edge, and the chamber was filled with sample by capillary action. Chamber edges were sealed with a small amount of petrolatum to prevent evaporation. The optics in this preparation were superior to standard depression slides. The coverslip props were necessary to keep the top and bottom glass surfaces apart and hence minimize wall effects in the center of the chamber. To test for confinement effects, two sets of slides were counted. The original slides were sealed at the beginning of the experiment and recounted at every time point. The new slides were made from the appropriate sample bottle at each time point and counted within 10 min.

Video taping was done midchamber, immediately after preparation of the slides, by using Zeiss Universal or Olympus BHT dark-field microscopy at max-

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imum light intensity. A Sony dxc 107p CCD or a JVC TK-1280E video camera set for maximum contrast was used. Resolution was estimated by using latex beads (Sigma). A bead radius of 0.23 μm was easily visible, but a radius of 0.05 μm was not visible. Except where noted, recordings were made of two microscope fields in the center and two microscope fields within 50 μm of the edge. Each slide was prepared in duplicate. For studies with cultures more than 50 h old, air bubbles were introduced into the chamber and prop walls were omitted. The air bubbles stimulated high-speed motility, and omission of the prop walls reduced the swarm thickness, enabling individual cells to be observed. Analysis for all treatment of the taped cells was done by the tracing and timing method described by Mitchell et al. (20), with the modification that for observing bacterial paths, particularly fast cells, measurements were made frame by frame. Recordings were made at 24 frames s^{-1} . Throughout this paper, the term "velocity" is used when the directional component of individual cell movement is important. The term "speed" is used in all other cases, such as those referring to average movement or to a change in the rate of movement without a change in the direction.

The isolation medium was based on that of Malmcrona-Friberg et al. (18) with the modification that the salts solution was replaced with artificial seawater. Isolates were identified by the carbon utilization method with commercially available plates and database (Biolog, Hayward, Calif.). Their protocol was modified by increasing the suspension solution to a salinity of 35‰, and a pH of 8.

RESULTS

Motile cells in all treatments made up less than or equal to 10% of the community of visible bacteria during the first 7 h of the experiment, and 7 of 15 samples had no motile cells (Fig. 1). During this time, the motile-cell fraction in 1% TSB was indistinguishable from that in the control seawater (Fig. 1b). Beyond 7 h, the motile fraction increased to a maximum of 84% (Fig. 1).

For all samples, cell numbers obtained from AODC and video tape began at 2.7×10^6 and 3.45×10^6 cells ml^{-1} at time zero. Total cell numbers peaked at 2.3×10^7 and 5.4×10^7 cells ml^{-1} for 0.01% TSB. Other TSB concentrations followed similar patterns, with maximum values of 3.8×10^8 (1%), 1.7×10^8 (0.1% TSB), and 9.7×10^6 cells ml^{-1} (0.001%). Seawater controls ranged between 1.9×10^6 and 2.5×10^6 cells ml^{-1} by AODC and 1.1×10^6 and 6.0×10^6 cells ml^{-1} by visible counts. The motile fraction of cells for the original slides showed a maximum for all treatments of 9% motile cells (0.01%) over the first 7 h, with a rise to 93% at 28 h. This paralleled the development of the motile fraction in newly prepared slides. The cells in the original slides had a speed range of 0 to 91 $\mu\text{m s}^{-1}$.

Mean speed during the 50-h experiments ranged from 0 to 112 $\mu\text{m s}^{-1}$. Figure 2a shows the changes in speed for new slides at concentrations of 0.001, 0.01, and 0.1% TSB. At 0.01% TSB, the speed stabilized at 94 $\mu\text{m s}^{-1}$ (95% confidence interval [CI], 22 $\mu\text{m s}^{-1}$). At 0.1% TSB, speed stabilized at 36 $\mu\text{m s}^{-1}$ (95% CI, 4.3 $\mu\text{m s}^{-1}$). Speed was monitored on a daily basis over a period of 9 days, until it returned to background for two consecutive days (Fig. 2b).

Cell speed not only changed over time but also at any given time appeared to be nutrient concentration dependent. Figure 3 shows how speed decreased as a function of increasing concentration at the 12- and 27-h points. A linear regression of speed on concentration for these points gave an r^2 value of 0.997, with a y intercept of 100 $\mu\text{m s}^{-1}$ (standard error, 2.7 $\mu\text{m s}^{-1}$) at a minimum concentration of 0.001% TSB. At time points before 12 h, there was too small a fraction of motile cells to obtain reliable measurements.

Shaking the bottles delayed the onset of full-speed motility and motile fraction of the population by 5 h and delayed the loss or slowing of motility by at least 5 to 10 h for the average cell (Fig. 4). Individual cells showed large variations in velocity, causing the large confidence intervals in Fig. 4.

Investigation of variation in speed showed that not only were

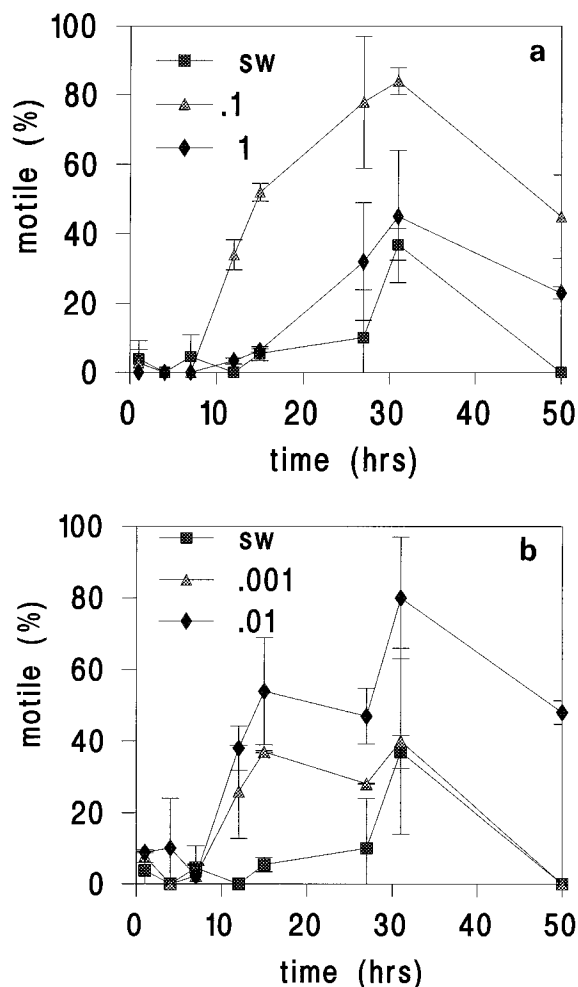


FIG. 1. Fraction of motile cells as a function of the time after sampling. (a) Comparisons of the two higher concentrations of TSB with the seawater control. (b) Comparison of the two lower concentrations of TSB with the seawater control. The highest nutrient concentration used (1%) was most similar in motile fraction to the seawater control when compared with other concentrations. Error bars are 95% CI. Symbols show concentrations of TSB (wt/vol). sw, seawater.

some cells consistently faster than others but also the speed of an individual cell could vary by up to an order of magnitude over 0.2 s (Fig. 5). These changes occurred during straight runs, midchamber, and were not associated with direction changes or stopping. The frequency and duration of speed changes varied from individual to individual. Figure 5 shows how speed changed for three cells over the period of up to 2 s, or 48 measurements. The speeds are presented as five-point running means to reduce speed spikes. While population speeds averaged up to 112 $\mu\text{m s}^{-1}$ and individual speeds averaged 238 $\mu\text{m s}^{-1}$, short bursts were up to 407 $\mu\text{m s}^{-1}$. Fast bacteria were difficult to monitor for periods longer than 1 to 2 s because they moved out of the field of focus or disappeared off the screen.

Sample bottles still contained motile bacteria after 2 weeks. Examination of the 1% TSB treatment revealed a highly motile population of bacteria. Measuring of the motile-cell fraction was prevented by the propensity of the cells to swarm around air bubbles and at the coverslip edge. The speed of the bacteria depended on whether they were inside or outside a swarm (Fig. 6). The mean cell speed was 144 $\mu\text{m s}^{-1}$ (95% CI, 7.7 $\mu\text{m s}^{-1}$)

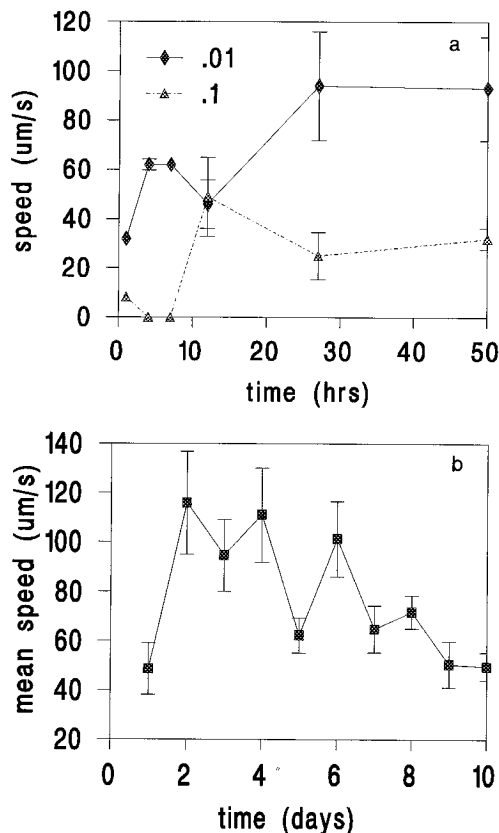


FIG. 2. Mean speed of motile cells as a function of time. (a) High-resolution sampling over a 50-h period. (b) Daily sampling over 9 days, with new bottles opened each day. Speed measurements were made in the center of the chamber away from surfaces. Zero values indicate that no motile cells were observed in these treatments. Sample sizes ranged between 10 and 25 cells for samples. Samples were assayed in triplicate. Error bars are 95% CI. Symbols in panel a show concentrations of TSB (wt/vol).

inside the swarm, $129 \mu\text{m s}^{-1}$ (95% CI, $9.7 \mu\text{m s}^{-1}$) at the swarm border, and $69 \mu\text{m s}^{-1}$ (95% CI, $3.1 \mu\text{m s}^{-1}$) outside the swarm. Turning at the edge of the swarm took place in a narrowly defined region (Fig. 7).

Under the given enrichment conditions, bacteria were in lag

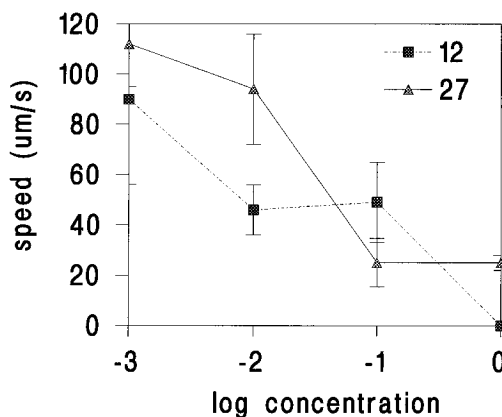


FIG. 3. Cell speed as a function of TSB concentration at 12 and 27 h. These two times were chosen because they had the highest mean speeds across the different concentrations (Fig. 2). Error bars are 95% CI.

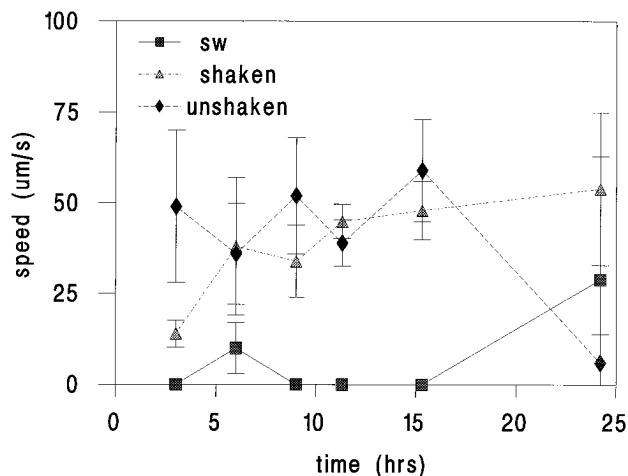


FIG. 4. Influence of shaking on cell speed at 0.1% TSB. No difference between shaken and unshaken treatments was found, except for the first and last points of the experiment. The seawater (sw) control was unshaken. Error bars are 95% CI.

phase for 12 h as measured by spectroscopy at 610 nm. No single conversion factor was found between optical density and cell number (Fig. 8). The maximum resolution of the method meant that one visible bacterium represented 1.1×10^5 bacteria ml^{-1} . The small size of marine bacteria meant that even with high-intensity dark-field microscopy, not all cells might be visible. For this reason, AODC counts were used, and it was found that the mean ratio of AODC to dark-field counts was 1.1 (95% CI, 0.25). The distribution was not consistent over time (Fig. 8).

One species, *Vibrio harveyi*, made up more than 99% of culturable cells isolated from the enrichments at the end of the experiments. Identification by Biolog was at the 0.919 similarity level (>0.5 similarity indicates positive species identification) for *V. harveyi*, with a modified octal identification number of **34.321-25.207-30.122-0.4050-01.40.20.1-01.677-00.114-3403.4**, where boldface numbers indicate intermediate responses in the Biolog identification system. Positive oxidase and catalase tests combined with yellow oxidation-fermentation tubes and growth on thiosulfate-citrate-bile salts-sucrose (TCBS; Oxoid) plates supported the genus identification. *V.*

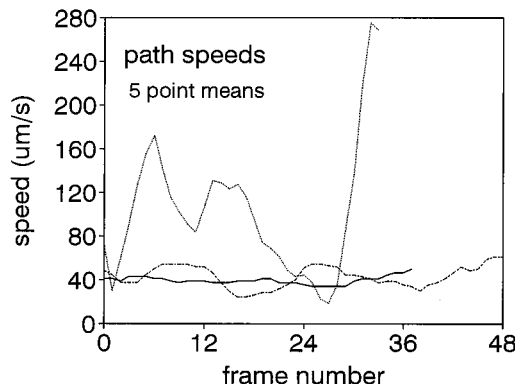


FIG. 5. Five-point running means of the speeds of three individual bacteria. The total elapsed time for the 48 frames corresponds to 2 s. The bacteria were chosen to illustrate three different modes of bacterial motility: constant, oscillating, and transient large-amplitude shifts. Bacteria were from the 31-h 0.01% TSB treatment.

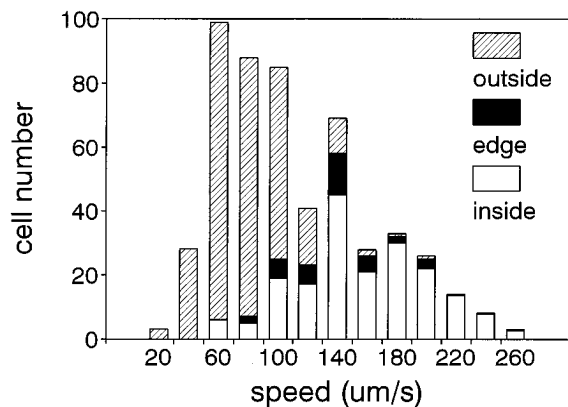


FIG. 6. Speeds for bacteria inside, outside, and at the edge of a swarm. At the lower two and upper three frequency bins, there is no overlap in speed between cells inside and outside a swarm.

*natrigen*s was run with the isolates as a control, and identifications were checked twice.

DISCUSSION

The usefulness and adaptive advantages of motility have been well documented, but models of marine bacterial motility assume that all cells are always motile (6, 14, 15). In most

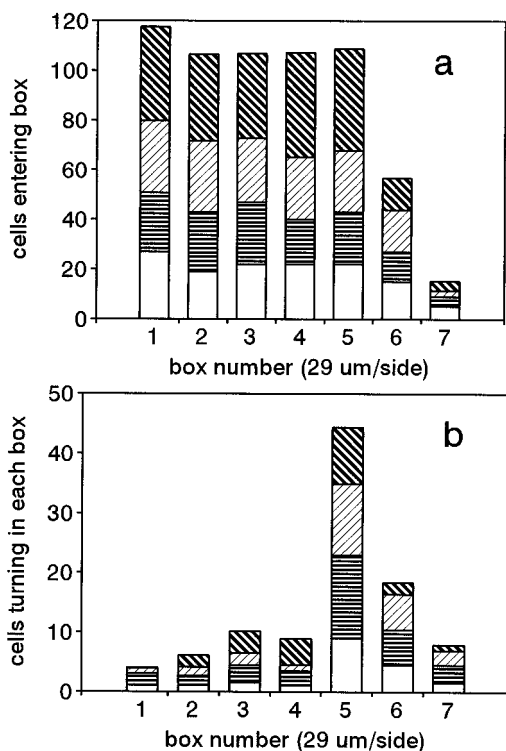


FIG. 7. Spatial distribution of swarming bacteria and their turns. To obtain these data, a transect across the screen was divided into seven boxes. Each box had a side equivalent to $29 \mu\text{m}$ on the microscope slide. The swarm was defined by high cell abundance. Box 1 was in the middle of the swarm, and box 6 was the swarm boundary. (a) Abundance of motile bacteria measured as the number of cells entering each box. (b) Number of bacteria turning in each box during 100 frames. For both panels, the procedure was repeated four times, with each replicate represented by a different fill pattern in the frequency distribution.

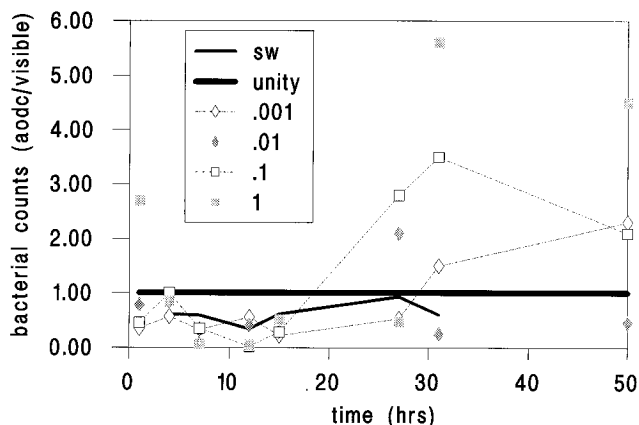


FIG. 8. AODC expressed as a fraction of dark-field video microscopy counts for four concentrations of TSB (percent [wt/vol]) and a seawater control (sw). Symbols are connected by lines at alternating concentrations for clarity. Unity (thick line) indicates AODC and dark-field counts equal to each other.

marine environments, however, motility is energetically expensive because nutrient concentrations are low and cells are small (19). Under these conditions, it is equally adaptive for cells to turn motility on and off. The results presented here support this hypothesis and make the testable prediction that for marine bacteria, induction should be closely coupled to changes in ambient nutrient concentration.

Measurement errors. Measurements of speed and the fraction of motile cells both contained known systematic bias. Determination of the motile-cell fraction took place over approximately 1 min. During this time, cells came onto the screen from the edges and from above and below the plane of focus. The result was that motile cells just beyond the edge of the screen and just out of the focal plane had a 50% probability of being counted while nonmotile cells in the same positions would not be counted. Given speeds here on the order of $100 \mu\text{m s}^{-1}$ and turning times of about once per second, the distance from which motile bacteria immigrated can be estimated by using the bacterial diffusivity:

$$D = v^2\tau/3 \quad (1)$$

where v is velocity, τ is turning frequency, tumbling is assumed to be random (5), and the length travelled in a given time can be expressed as a function of the diffusivity as follows:

$$x = (2D/t)^{1/2}, \quad (2)$$

where x is length, D is diffusivity, and t is the time of the taping. We calculate the maximum overestimate to be 27% when all cells are moving. This tends to shift visible-cell counts (Fig. 8) downward, bringing the two methods close to agreement (unity) for the first 15 to 30 h of the experiment. This correction was not applied to the data presented in the figures, because it varies with velocity and turning frequency. As presented in Results and discussed below, speed varied within each treatment, population, and individual. The velocity measurement itself also contained a systematic bias, in that the method underestimates the speed. This occurs because the tracing method flattens a three-dimensional path onto a two-dimensional surface, with the subsequent loss of the velocity component perpendicular to the screen. If the mean angle incident to the plane of focus is 45° , the velocity is underestimated by about 40%. The aspect ratio of the screen to the depth of focus was about 10:1, so that the average bacterium inclined to the

plane of focus remained in focus for only 100 to 200 ms. To minimize this error, only long runs of 1 s or more were considered for analysis of speed change (Fig. 5). For the motile-fraction measurements, all paths were used. The sealed slides indicate that volume and time on the slide had little effect on the cells, compared with those kept in 100-ml enrichments.

Initial motility response. Figure 1 indicates that fewer than 10% of the marine bacteria in Gulf St. Vincent are motile at any given time but that a much larger fraction of the cells are potentially motile and that shaking has minimal effect on motility (Fig. 4). The time required for motility to begin was about 7 h after the addition of nutrients. For unshaken seawater with no added nutrients, this time was 25 to 30 h. From the motile-fraction increase in the absence of nutrient addition, we infer that weak gradients form in the bottles because of reduced turbulence. From the long lag before the onset of motility and the subsequent rapid rise in the motile fraction of cells, we propose that existing cells became motile, rather than motile subpopulations gradually overgrowing the microbial community. That existing cells became motile is similarly indicated by the depression of motility at high nutrient concentrations (1% TSB), at which growth was highest. Further experiments are needed to confirm this proposal.

The lag time for the appearance of motility after nutrient addition was 6 to 12 h. Marine bacterial chemotaxis models consider cells fully motile when they first encounter a nutrient source, implying little or no lag time before the onset of motility (3, 4, 14, 15). While short lag times have the clear advantage of allowing a rapid response to environmental changes, more-rapid initiation of motility may not be possible given the 60 (17) or more genes involved, the structural regulation of expression (13), and ambient temperatures.

The long lag time and low initial fraction of motile cells make it appear that marine bacteria have to remain in or near the nutrient source for hours before motility is stimulated. There are two situations that may satisfy the time requirement: (i) when bacteria become associated with the zooplankton fecal pellets, and (ii) when bacteria become associated with aggregated particulate matter (marine snow). Gowing and Silver (11) found that bacteria begin breaking down fecal pellets from the inside. Cho and Azam (7) found that bacteria also begin breaking down pellets from the outside. As the pellet breaks apart, it may be highly advantageous for bacteria in the pellet to rapidly locate a pellet fragment before they are separated from it by turbulence and sinking. Marine snow, some of which is derived from fecal pellets, has lifetimes similar to or longer than those of fecal pellets, and bacterial concentrations on these particles are up to 10^{11} cells ml^{-1} (1), 4 to 5 orders of magnitude higher than bacterioplankton concentrations. Again, high speed may help bacteria remain on or near marine snow surfaces.

Energy limitation. The energy expenditure per unit time required to move a cell increases as the square of the speed (5). For this reason, fast cells expend more energy per unit time when moving than do slow cells. If energy is limiting for motility, then among separate treatments of different concentrations we might expect to see an increase in speed with an increase in nutrient concentration, the opposite of what is observed here. The crucial factor, however, is the absolute concentration of nutrients. Energy limitation is predicted to become important at about $1 \mu\text{M}$ or $10^{-5}\%$ (19), so there was abundant energy available for all treatment concentrations (0.001 to 1%). The prediction then is that the decrease in speed because of energy limitation should be seen between 10^{-5} and $10^{-3}\%$ (wt/vol) TSB. This range is three orders of

magnitude within the threshold concentration at which bacteria can perform chemotaxis (17).

Tens of hours after reaching peak speed, cells slowed (Fig. 1). Under the experimental conditions, return to background speed required more than a week (Fig. 2b). This decline may have been prolonged by organic carbon available from lysis and the existence of microgradients with the bottles. Additionally, it appears that after even longer times (2 weeks), cells were still capable of moving at $144 \mu\text{m s}^{-1}$ when stimulated. This high speed may be fuelled by storage product or concentrations of organic carbon from lysis. Both of these are absent in bulk seawater but may be present on particle surfaces, suggesting that energy limitation for high-speed motility could be spatially heterogeneous in seawater.

Speed and nutrient concentration. Figure 3 shows that speed is a function of bulk nutrient concentration for the maximum velocities the cells achieve, with low concentrations resulting in high velocities. There must, however, be a threshold below which motility is not induced, since the seawater controls showed zero or low speed in all experiments. Figure 3 indicates that it is below 0.001% for our experimental conditions with Gulf St. Vincent bacteria. Whether induction depends on the concentration of a specific compound or the overall energy state of the cells remains to be determined for these bacteria. The effect could indicate a chemokinetic mechanism that slows the cell in order to increase the likelihood of remaining near the nutrient source. The potential effectiveness of this strategy is illustrated by comparing translational diffusion for motile and immotile cells. Reducing just the velocity in equation 1 from 100 to $10 \mu\text{m s}^{-1}$ (Fig. 3) decreases the diffusivity (equation 2) from approximately 10^{-5} to 10^{-7} $\text{cm}^2 \text{s}^{-1}$. The corresponding time to diffuse $100 \mu\text{m}$ away from a particle increases from 1.5 to 150 s. Taking this strategy to its extreme, i.e., stopping, the time spent within $100 \mu\text{m}$ of the source is further increased by less than an order of magnitude over the slow value. The advantage of not stopping is increased sensitivity to environmental changes, as shown below. The slowing strategy is probably only effective over short distances (<1 mm). At centimeter scales, the effect is the same, but the relevance of staying near a source for days to months is less clear.

The effect of nutrient concentration on speed that was measured here may not have been reported from culture experiments because cultures are grown at high concentrations, with carbon sources usually added at greater than 1 g/liter (0.1%). In contrast, natural seawater concentrations for amino acids and sugars are at concentrations equivalent to 10^{-7} to $10^{-5}\%$ (wt/vol) (8, 9).

Absolute speed. The aggregate community speeds of 112 and $144 \mu\text{m s}^{-1}$ (Fig. 2), along with individual bursts of up to $407 \mu\text{m s}^{-1}$, are among the highest recorded (10, 23). These speeds are consistent with predictions that marine bacteria must be fast in order to be chemotactic, because of their small size (19). This prediction was based on the point that Brownian motion rotates cells as an inverse cube function of their size, so that the smaller marine bacteria rotate in excess of 10^3 degrees in 1 s. High velocities permit cells to swim for a short time in the straight lines necessary for chemotactic migration.

Speed may decrease with increasing nutrient concentration because size increases with increasing nutrient concentration. There is ample evidence that marine bacteria are capable of size changes of an order of magnitude in linear dimension in response to nutrient changes (22), and so we would expect that the bacteria in these experiments changed size. Size change measurements were not possible with the dark-field scattered-light microscopy used. Determining whether speed and size

changes are linked is the next step in finding what controls absolute velocity. If a link is found, then the question arises of what the connecting mechanism might be. Hughes et al. (13) show that repressor proteins pumped out via the motors stimulate flagellum production by reducing repressor concentration. An increase in cell volume might similarly reduce repressor concentration, determining the type and number of motors produced. High speed may be an advantage in this environment, because the modelling by Bowen et al. (6) suggests that slow bacteria have difficulty migrating and staying near small nutrient sources.

Speed fluctuations and bursts. Rapid speed changes are possible because bacteria have no inertia, coasting to a stop in less than 1 Å (0.1 nm) (24), but it is worth considering that the fluctuations may have been physiological artifacts. We believe that this is unlikely because of the magnitude of the fluctuation, the rapid recovery, the coordinated swarm, and its definite boundary. The variability of speed within an individual is unusual (Fig. 5). There is currently no chemotactic mechanism in which millisecond speed shifts are used. Fluctuations are not explained by standard chemokinesis, in which cells slow and accumulate in a region. Here, the cells sped up and accumulated in a region, maintaining position by turning.

The rapid shift may be a valuable adaptation to a flowing environment. High-speed bursts may help cells cross streamlines and take them away from regions of low nutrients, and the slowing may prevent overshoot of small nutrient sources. This reasoning at first sight suggests that bacteria should slow to a stop, since immotile bacteria have low translational Brownian diffusivities (10^{-9} to 10^{-8} cm² s⁻¹). However, the ubiquitous turbulence of the ocean creates shear at this scale, which over time separates a stopped or slowed cell and its source. The lower speed of about 70 μm s⁻¹ is, according to the model of Bowen et al. (6), sufficient to allow bacteria to remain near a nutrient source, presumably by enabling them to quickly detect and react to changes in the source position. Overall, high speeds, rapid speed changes, and narrow turning region (Fig. 6 and 7) would permit bacteria to temporarily remain near point sources while minimizing the large energy expenditure associated with very high speeds (19).

Swarming and signal-processing time. Segall et al. (25) found that the signal-processing time for *E. coli* was about 0.2 s. They pointed out that this is not a lower limit and that a processing time of a few milliseconds might be realized in the presence of selective advantage. The rapid speed changes open the possibility that processing times in marine bacteria are faster than 0.2 s. The experiments here do not allow for direct calculation of the processing time, but an upper-limit estimate can be obtained for comparison with the 0.2-s value.

Purcell (24) pointed out that the minimum distance, l_m , a bacterium must go to detect a change in the environmental concentration of an attractant is

$$l_m = D/v \quad (3)$$

The time, t , to go this distance (and hence the available signal-processing time) is $t = l_m/v$, or, substituting equation 3,

$$t = D/v^2 \quad (4)$$

where all variables for equations 3 and 4 are as previously defined. For the average bacteria outside the swarm moving at 69 μm s⁻¹, the detection time is about equal to 0.21 s. In the swarm, at an average speed of 144 μm s⁻¹, the time is about 0.048 s. The maximum available processing time for the highest observed burst speed (407 μm s⁻¹) is 0.006 s. From this, we

propose that signal-processing times are less than 0.1 s for some marine bacteria.

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