

# New light on an important microbe in the ocean

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Why is there such a fuss over the genome sequence of a marine bacterium, discussed by González *et al.* (1) in this issue of PNAS, when they are now a dime a dozen and number in the hundreds? It is well known that marine bacteria (and archaea) are extremely diverse and numerous and run the planet's biogeochemical cycles, including that of carbon. These microbes include CO<sub>2</sub>-fixing (photoautotrophic) cyanobacteria, the most numerous being *Prochlorococcus*, and CO<sub>2</sub>-evolving (heterotrophic) bacteria, a prominent example being *Pelagibacter*, the cultured representative of the SAR11 clade. We know the genomes of these microbes and many others, so what's so special about *Polaribacter* MED152? The answer is connected to one of MED152's two lifestyles described by González *et al.*

Despite intense effort, the number of sequenced marine bacterial genomes still pales in face of the ocean's diversity. *Polaribacter* MED152 is one of the first marine members of the *Bacteroidetes* to be sequenced and analyzed in depth, as more attention has been paid to marine *Proteobacteria* (e.g., *Pelagibacter* and *Silicibacter*) and *Cyanobacteria* (e.g., *Prochlorococcus*). *Bacteroidetes* is a complex division (or phylum) consisting of anaerobic genera, such as *Prevotella*, *Porphyromonas* (2), and *Bacteroides*, which have been extensively examined because of their role in human biology [*B. thetaiotaomicron* is the dominant bacterium in our intestines (3)], and of aerobic genera, such as *Cytophaga* and *Flavobacteria*, which have not been examined as extensively, but are known to be abundant in both freshwaters and the oceans. This division is still sometimes referred to as *Cytophaga-Flavobacteria-Bacteroides*. *Polaribacter* MED152 is a cultured representative of *Flavobacteria*.

González *et al.* (1) begin the argument for these bacteria by pointing to their abundance in the oceans and perhaps a specialized role in degrading organic material and thus in the carbon cycle. Estimates of *Flavobacteria* abundance range from 5% to 20%, depending on the oceanic regime and the detection method, but this bacterial group can be even more abundant during or after phytoplankton blooms, providing one of the first clues of its proclivities in organic material degrada-

**Table 1. Genomic features of selected representatives of some major bacterial groups in the oceans**

Feature	<i>Polaribacter</i>	<i>Pelagibacter</i>	<i>Silicibacter</i>	<i>Prochlorococcus</i>
Genome size, mB	2.97	1.31	4.60	2.41
Proteorhodopsin	1	1	0	0
Rubisco	0	0	0	1
PEP carboxylase	1	1	1	1
Pyruvate carboxylase	1	0	1	0
HCO <sub>3</sub> <sup>-</sup> transporters	2	0	0	2
Phytochrome	1	0	0	0
Photolyase	3	3	0	0
BLUF	1	0	0	0
Peptidases	93	16	37	19
DMSP use	–	+	+	+
Transporters	106	88	429	119

The strains examined here are *Polaribacter* sp. MED152 (*Flavobacteria*) (1), *Pelagibacter ubique* HTCC1062 (*Alphaproteobacteria*), *Silicibacter pomeroyi* DSS-3 (*Alphaproteobacteria*, in the *Roseobacter* group) (10), and *Prochlorococcus* MIT9313 (*Cyanobacteria*). Data are from González *et al.* (1) or <http://img.jgi.doe.gov> using the indicated features as search terms. Rubisco, ribulose 1–5 biphosphatase carboxylase oxygenase (CO<sub>2</sub> fixing enzyme); BLUF, blue light sensor using FAD.

\*DMSP use has not been demonstrated for MIT9313, but it has been demonstrated for natural *Prochlorococcus* communities (11).

tion. *Flavobacteria* may respond directly to dissolved organic material (DOM) produced by the high biomass levels of phytoplankton that define blooms, but they are also abundant on the particulate detritus often produced during blooms. Use of detritus is one of the lifestyles described by González *et al.*

*Polaribacter* MED152 appears to be well equipped for life on particulate detritus. González *et al.* (1) found several genes for attaching to and gliding on surfaces and for the production and export of extracellular polysaccharides, the slime used by attached microbes for adherence and protection. MED152 also has several enzymes to hydrolyze the polymers making up particulate detritus or that may occur in the dissolved phase. González *et al.* found that the largest protein families in MED152 were peptidases (93 genes) and glycosyl hydrolases (30 genes). This bacterium has more of these genes, it seems, than do other marine bacteria (Table 1), consistent with the reputation of *Bacteroidetes* using high molecular weight (HMW) polymers and carrying many hydrolytic enzymes. However, MED152 lacks ABC transporters for sugars, in contrast to the large number of these transporters in marine *Alphaproteobacteria* and *Gammaproteobacteria*, again consistent with some previous work. González *et al.* mention that *Proteobacteria* are less proficient than *Bacteroidetes* in using HMW DOM, a statement that may cause some

microbial ecologists to wince, given the huge diversity of marine *Bacteroidetes* and *Proteobacteria* and numerous counterexamples. But such generalizations can be useful guides and starting points for understanding complex interactions.

The fate of particulate detritus is crucial for understanding the oceanic carbon cycle, but so far *Polaribacter* MED152 seems to be just another marine bacterium, probably important, still hardly noteworthy. It is the second lifestyle which perhaps enables MED152 to survive the organic-poor waters between stays on detritus, that really grabs our attention. *Polaribacter* MED152 has proteorhodopsin and, even more unusual, actually responds to light in experiments, in contrast to most other proteorhodopsin-bearing bacteria examined so far. Proteorhodopsin is a light-dependent H<sup>+</sup> pump that could drive ATP synthesis or in other ways help a cell meet its requirements for energy. Since its discovery in a gamma proteobacterium (4), proteorhodopsin has been found in many types of marine microbes and may be in roughly half of all bacteria in the surface oceans. Like the other proteorho-

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dopsin-bearing microbes examined so far, MED152 is heterotrophic and depends on organic compounds for carbon and presumably for most of its energy. So, this light effect is a radical departure from previous ideas about what controls heterotrophic microbes and about their role in the carbon cycle (5, 6).

In addition to the genome analyses, González *et al.* (1) present some data indicating how light actually affects MED152 metabolism. Despite intensive effort, only one of three published studies (not counting experiments with *Escherichia coli* bearing cloned proteorhodopsin) has been able to demonstrate any effect of light on a proteorhodopsin-bearing bacterium. Even *Pelagibacter ubique*, which has proteorhodopsin, grows the same with and without light in seawater with its naturally occurring organic compounds (7). The one success story is *Dokdonia* sp. MED134 (8), a close relative of *Polaribacter* MED152. However, unlike *Dokdonia* sp., *Polaribacter* MED152 does not grow any faster with light, at least in the growth media tested so far.

But MED152 does assimilate CO<sub>2</sub> faster in the light than in the dark, a very surprising and puzzling result. Because this bacterium does not have the main CO<sub>2</sub>-fixing enzyme (Rubisco) found in cyanobacteria (Table 1) or genes for any of the other three known autotrophic pathways, González *et al.* (1) hypothesize that the light-stimulated CO<sub>2</sub>-fixation is caused by anaplerotic reactions mediated by phosphoenolpyruvate (PEP) carboxylase or pyruvate carboxylase. The MED152 genome has

genes for both carboxylases, in addition to two bicarbonate transporters and carbonic anhydrase, all rather unusual for a

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heterotroph (Table 1). González *et al.* argue that the energy from proteorhodopsin allows more carbon to be diverted from oxidation to biomass synthesis, creating an imbalance in tricarboxylic acid cycle intermediates used for biosynthesis. Normally, however, the “filling in” by anaplerotic CO<sub>2</sub> fixation is only necessary when heterotrophic cells grow on two or three carbon compounds, such as glycolate, which is produced by phytoplankton. Other studies have examined light effects on amino acid uptake (9), but little of anything is known about the uptake of compounds like organic acids requiring a cell to carry out anaplerotic CO<sub>2</sub> fixation.

In addition to proteorhodopsin, the MED152 genome has several light-sensing genes, perhaps more than found even in other proteorhodopsin-bearing bacteria (Table 1), but why MED152 needs these genes is not quite clear. Presumably, a heterotrophic microbe armed with light-sensing mechanisms could key in more efficiently on the organic compounds produced by light-

driven phytoplankton, an important attribute in oligotrophic oceans where labile organic material is scarce. But MED152 does not have the genes necessary for using the compounds typically produced by phytoplankton, including glycolate and notably dimethylsulfoniopropionate (DMSP), which is the precursor to the climate-active gas, dimethylsulfide. Perhaps this presumed deficiency is a comment more on what is known about phytoplankton-derived organic compounds than about actual coupling between MED152 and phytoplankton. It is also possible that heterotrophic microbes need light-sensing mechanisms even if not directly tied to phytoplankton activity because of the many other impacts of light on the oceanic environment. No environmental signal is as strong and consistent as the transition from night to day in the oceans.

Undoubtedly the actual environment faced by MED152 and other marine microbes is more complex than the two sketched out by González *et al.* (1). Detrital particles are not very numerous in the oceans, and the “desert” between particles may be studded with colloids, gels, and various forms of HMW DOM, all potential sources of carbon and energy for MED152-like bacteria. Still, González *et al.* add an important chapter to our understanding of what controls marine microbes in the ocean, and their work should stimulate new directions in research for examining how light affects heterotrophic activity. The future is certain to bring more twists and turns to the lifestyles described here.

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