

## Deep thinking: Scientists sequence a coldloving marine microbe

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At home in the deep, dark Arctic Ocean, the marine bacterium Colwellia psychrerythraea 34H keeps very cool--typically below 5° degrees Celsius. How does the bacterium function in this frigid environment? To find out, scientists at The Institute for Genomic Research (TIGR) and collaborators have sequenced and analyzed C. psychrerythraea's genome.

That genome analysis, posted in the Proceedings of the National Academy of Sciences (PNAS) Online Early Edition July 25-29, reveals key biochemical tools that cold-adapted, or psychrophilic, bacteria can use to survive in subzero temperatures. In particular, some of C. psychrerythraea's estimated 4,937 genes apparently code for adaptive traits such as cell membranes packed with polyunsaturated fatty acids that resist freezing, polyester compounds that offer extra energy reserves, protective solutes inside cells, and ordinary enzymes altered to function in chilly seawater.

"Our goal was to compare C. psychrerythraea to a variety of bacteria that live at different optimal temperatures," remarks Barbara Methé, a molecular biologist at TIGR and lead author of the PNAS paper. "This organism must adapt to extreme cold--in fact, it cannot live above room temperature. How does it cope in such freezing conditions?"

Among cold-adapted bacteria, Colwellia species have a decidedly chilly comfort zone. The bacteria typically require temperatures less than  $20^{\circ}$  to grow. Scientists have found C. psychrerythraea--which prefers temperatures of  $-1^{\circ}$  to  $10^{\circ}$  --hugging sediments along the Arctic floor,



floating in the open ocean, and nestled inside Arctic and Antarctic sea ice.

To explain C. psychrerythraea's unique cold-adapted chemistry, TIGR researchers and collaborators first sequenced the bacterium's genome and analyzed its gene content for telltale proteins that might be cold-adaptive. Next, they predicted C. psychrerythraea's proteome--or complete inventory of protein sequences--and then compared it to 21 predicted proteomes from known bacterial genomes. These included both mesophilic bacteria, which live at room temperature, and thermophilic bacteria, which thrive in ultra-hot environments.

Finally, the scientists overlaid key C. psychrerythraea protein sequences onto known 3D structures from similar proteins among the diverse bacteria. Like matching up blueprints, this strategy allowed the team to spot any striking distinctions in C. psychrerythraea's amino acid composition, highlighting differences unique to the cold-loving organism.

Together, these analyses offer a picture of evolution in action, as C. psychrerythraea uses subtle tweaks in common bacterial biology to adapt to its chilly environs. For instance, the bacterium taps a group of four to five genes to generate polyunsaturated fatty acids and pack those acids into cell membranes, resulting in membranes that are fluid and functional--rather than a frozen chunk of biomass--below the freezing point. The genome also possesses a number of duplicated genes important to cell membrane biosynthesis. What's more, C. psychrerythraea dresses in layers, generating plenty of extracellular polysaccharides (sugars) that coat cell membranes.

Aside from its cellular outerwear, C. psychrerythraea generates a range of potential cold-protective compounds. One example is a family of polyesters, known as polyhydroxyalkanoate (PHA) compounds, that may



also boost reserves of nitrogen and carbon, which could be in short supply in the extreme cold. The organism also engineers cold-hardy versions of ordinary enzymes found in free-living bacteria, such as enzymes that break down organic matter. C. psychrerythraea possesses genes that may break down complex compounds, including pollutants, as well.

Collectively, Methé suggests, these subtle changes in biology allow C. psychrerythraea--and possibly similar bacteria--to thrive in the cold. More than just marvels of nature, cold-adapted enzymes hold industrial promise, as active ingredients in coldwater detergents, clean-up for industrial contaminants, and food treatments. Psychrophiles could hold clues to microbial life on other planets, as well, such as the frozen surface of Mars or one of Jupiter's moons, Europa. As yet, however, there's little chance that deciphering C. psychrerythraea's genome will yield insights into another cool character, its namesake. Deming, who led the establishment of the Colwellia genus, named the genus after her onetime advisor: noted marine biologist Rita Colwell.

In addition the TIGR team, led by Methé, study collaborators include: Jody Deming of the University of Washington at Seattle; Bahram Momen of the University of Maryland at College Park; Adrienne Huston of Pennsylvania State University at University Park; and a team from the University of Maryland's Center for Advanced Research in Biotechnology in Rockville.

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