New petroleum-degrading bacteria found at Rancho La Brea Tar Pits in Los Angeles

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A tar pit at Rancho La Brea. Pits like these have yielded thousands of animal fossils. Credit: D. E. Crowley, UCR

Environmental scientists at UC Riverside have discovered that the Rancho La Brea tar pits in downtown Los Angeles, Calif., house hundreds of new species of bacteria with unusual properties, allowing the bacteria to survive and grow in heavy oil and natural asphalt.

Trapped in soil that was mixed with heavy oil nearly 28,000 years ago, the bacteria are uniquely adapted to the pits' oil and natural asphalt, and contain three previously undiscovered classes of enzymes that can naturally break down petroleum products, the researchers report.

"We were surprised to find these bacteria because asphalt is an extreme and hostile environment for life to survive," said Jong-Shik Kim, a postdoctoral researcher in the Department of Environmental Sciences, who initiated the study. "It's clear, however, that these living organisms can survive in heavy oil mixtures containing many highly toxic chemicals. Moreover, these bacteria survive with no water and little or no oxygen."

The bacteria and their enzymes have potential application for bioremediation (cleaning oil spills), medical treatments (new medicines), alternative energy (biofuels), enhanced oil recovery, and industrial applications (biochemicals and biotechnology).

Study results appear online in the April 6 issue of *Applied and Environmental Microbiology*.

Kim and his advisor, David E. Crowley, a professor of environmental microbiology, used DNA-based methods to identify the new bacteria as well as the DNA encoding the three classes of petroleum-degrading enzymes.

"Previously, some bacteria had been cultured from the asphalt, but no one had been able to extract DNA from the asphalt to study the entire microbial community," said Kim, the first author of the paper.

Providing a natural observatory for the unusual bacteria, the Rancho La Brea tar pits, which formed in the last ice age, are located in Hancock Park, Los Angeles. Rancho La Brea, one of the world's fossil localities, is recognized for having the largest and most diverse assemblage of extinct ice age plants and animals in the world.

"The living bacteria contained in the asphalt are most likely the progeny of soil microorganisms that were trapped in the asphalt, although some

may also have been carried to the surface in the heavy oil that seeped upwards from deep underground oil reservoirs," said Crowley, the research paper's other author.

According to the researchers, most of the more than 200 species of microorganisms they identified represent entirely new branches in the tree of life, some being classified as new families of bacterial species.

While the bacteria remain to be grown in the laboratory, the researchers found that the closest relatives of many of the bacterial families are able to survive in high salt, toxic, and even radioactive environments.

"One family that was represented by many species is related to a group of bacteria that are the most radiation-resistant organisms on the planet," Crowley said. "Indeed, this family of bacteria has been previously investigated by the Department of Energy for cleanup of hydrocarbon contamination in radioactive environments."

It was the continual production of bubbles of methane gas that come up through heavy oil overlying the asphalts that clued the researchers to the presence of bacteria in the asphalt. "In the absence of oxygen, methane is produced by bacteria that use carbon dioxide for respiration instead of oxygen," Crowley explained.

He noted that the bacteria are not uniformly distributed in the tar pits. While one reason for their presence could be bacteria rising through the soil via a subterranean oil flow, other explanations are possible. "Probably there has also been genetic exchange and natural selection of new species over the thousands of years the bacteria have been living in the asphalt," Crowley said.

To identify the bacteria and their enzymes, Kim and Crowley analyzed the genetics of the bacteria extracted from the tar pits. To accomplish

this, they first froze the tar with liquid nitrogen and then pulverized it into a powdery mixture using a mortar and pestle. This process allowed the researchers to extract DNA from bacteria in the asphalt, after which it could be purified by other more standard methods used for environmental samples.

Next in their research Kim and Crowley plan to perform a thorough, quantitative and qualitative assessment of the bacteria in the tar pits to identify genes that may have application for petroleum processing, oil recovery, and biotechnology.

Source: University of California - Riverside