Fallout From a Chemical Catastrophe

Methyl isocyanate, the deadly gas that last week leaked from a Union Carbide pesticide factory in Bhopal, India, killing more than 2,000 people, is one of the most dangerous chemicals regularly used in industry, according to the Occupational Safety and Health Administration (OSHA). At the same time, little is known about its long-term effects on human health and precisely how the substance causes its toxic effects.

The lack of data on methyl isocyanate reflects the fact that it's very potent and difficult to control, says Meryl H. Karol of the University of Pittsburgh's Graduate School of Public Health. Although nominally a liquid at room temperature, methyl isocyanate evaporates so quickly from an open container that it easily turns into a colorless, odorless, highly flammable and reactive gas. "I would hesitate having it in the laboratory," Karol says.

The OSHA standard for exposure to methyl isocyanate during an eight-hour day is 0.02 parts per million in air, far lower than what many Bhopal residents were subjected to. This standard is five times more stringent than that for phosgene gas, which was used as a chemical weapon during World War I and is still used in various industrial processes.

For the last 10 years, Karol and her colleague Yves Alarie have been studying how low doses of isocyanates, the family of compounds to which methyl isocyanate belongs, irritate the lungs and, over an extended time, cause allergies. Isocyanates are widely used not only, as in the case of methyl isocyanate, for making a series of compounds called carbamates (a popular class of pesticides that attack the nervous systems of insects) but also in other forms for making plastics like polyurethane foam. Thousands of workers potentially face occasional exposure to isocyanates.

"There are differences in potency among the isocyanates," says Karol, "but they generally have the same effects." At low levels, isocyanates cause eyes to water and damage to the cornea. At higher concentrations, muscles constrict, blocking the nasal and bronchial passages in something comparable to a severe asthma attack. This was responsible for most of the deaths in India. Isocyanates are easily absorbed through the skin and quickly enter the bloodstream. They react readily with proteins, disrupting membranes and killing cells. They may also affect the nervous system by inhibiting an enzyme called cholinesterase.

"One of our goals is to find the target molecules in the biological system that the isocyanate goes to [to produce] the physiological effects that we're seeing," says biochemist William E. Brown of Pittsburgh's Carnegie-Mellon University. The researchers are finding surprising evidence that despite their great reactivity, isocyanates that enter a biological system seem to have an affinity only for a few, specific proteins.

Ironically, the tragedy in India may allow the Pittsburgh group to check some of their results obtained from experiments performed on guinea pigs. "We would like to know if the protein that we're finding in guinea pigs is the same one that is affected in humans," says Brown. If this turns out to be the case, then a blood test — an "early-warning detection system" — can be developed for screening workers who may have been exposed to isocyanates. "A lot of the effects that you see are reversible," says Brown. "If you could see the danger level early enough, you could do

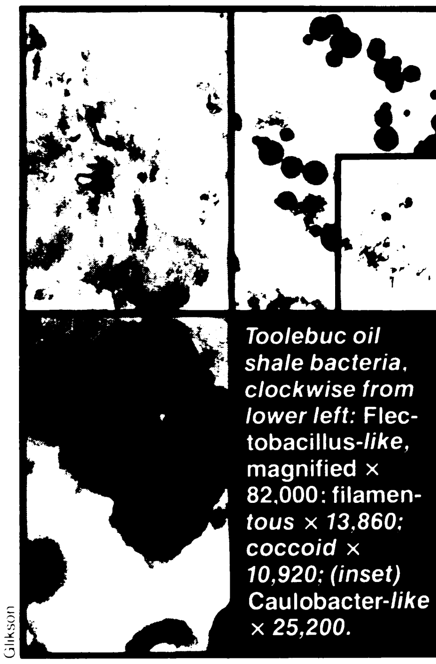
Viewing fossil fuels' bacterial sources

Conventional wisdom used to hold that fossil fuels originated mainly from marine plankton and higher plants, crushed and heated over the eons. But that ignored bacteria's role. Today, work on two fronts suggests that bacteria which dine on those plankton and plants are a dominant biochemical force in the creation of most petroleum and natural gas.

In Strasbourg, Pierre Albrecht of CNRS, the French national research organization, and co-workers recently established the presence of bacteria in fossil fuels. Although they never saw any bacteria, they uncovered a rich legacy of hopanoids — characteristic chemical fossils derived from the hopane molecule — that the bacteria left behind in petroleum. Photographic confirmation of bacteria's role was left to Australian geologist Miryam Glikson.

At Australian National University in Canberra, Glikson has used transmission electron microscopy to produce the first images of bacteria from fossil-fuel source rock. The technique requires first concentrating the organic matter by dissolving the rock. Then the organic matter is dried, embedded in resin and shaved into 0.05-micron slices with a handmade glass knife.

Though identification of the bacteria is difficult, Glikson says several types appear to resemble forms currently found in marine environments. Lacy, fan-shaped clumps suggest the living genus *Caulobacter*, she says, while a doughnut-shaped type appears similar to *Flectobacillus marinus*, a bacterium that degrades algae in the Florida Keys. In Australia's Toolebuc oil shales deriving from 120 million years ago, Glikson says "the bulk of the organic matter is made up of either filamentous bacteria or organic matter that has the same characteristics and consistency" but has lost its structure through compression. However, occasionally she identified in the shales a structure that she finds "reminiscent of the cyanobacteria" that



Glikson

Toolebuc oil shale bacteria, clockwise from lower left: Flectobacillus-*like*, magnified × 82,000; filamentous × 13,860; coccoid × 10,920; (inset) Caulobacter-*like* × 25,200.

form mats in the oxygen-poor zone at the seafloor (SN: 4/22/82, p. 284).

Shales formed from freshwater environments showed few if any bacterial remains. Glikson suspects this is due both to the type of bacteria-resistant planktons that likely served as a source of organic matter and to the highly alkaline — and therefore hostile to bacteria — nature of the initial environment.

Glikson's micrographs may generate more than academic interest, according to J. M. Moldowan of Chevron Oil Field Research Co. in Richmond, Calif. "One thing that all petroleums seem to have in common is bacterial reworking of organic matter," he says. Knowing more about the bacteria involved, he suggests, could help oil exploration teams gauge the potential yield of petroleum source rocks and the value of the oil they form.

Reports of Glikson's work are due out in the next issues of ORGANIC GEOCHEMISTRY and CHEMICAL GEOLOGY.

— *J. Raloff*

within the platform.

Karol, who is looking into ways of treating the allergies that result from low-level exposures to isocyanates, says, "Once you develop the allergy, it takes a long time to lose it. We are beginning to understand what type of exposure results in sensitivity to isocyanates." Blood samples from individuals who were exposed to methyl isocyanates and survived would provide clues about any antibodies that the human body may generate to battle the poison. The Pittsburgh group is trying to obtain blood samples from Bhopal, but they have not been successful yet.

Meanwhile, the researchers have come up with a simple technique for detecting low concentrations of isocyanates, based on their affinity for cholinesterase. The detector consists of a badge that, when treated, changes color after exposure to isocyanates. It could replace the large, cumbersome devices now worn by workers who handle isocyanates. Says Brown, "We're in the process of seeing if somebody would like to use it." — *I. Peterson*

'Planet' detected beyond the solar system

In 1983, three U.S. Naval Observatory astronomers reported finding a wobble in the motion of a star named Van Biesbroeck 8 (VB8), which they said indicated that it is being orbited by an object with no more than a few times the mass of the planet Jupiter (SN: 8/20/83, p. 116). About 21 light-years away, in the Milky Way constellation of Ophiuchus, VB8 was not the first star other than our sun for which such a "substellar companion" had been inferred. But now, says Donald McCarthy Jr. of the University of Arizona's Steward Observatory in Tucson, new observations have transformed that inference into "the first direct detection of an extra-solar planet."

That's strong talk. The term "planet" evokes images, if not of a rocky ball like earth or Mars, at least of a gassy one like Jupiter. And "VB 8B," as McCarthy and his colleagues have dubbed it, is considerably more massive than Jupiter (although it is only about nine-tenths the diameter). The point, however, is that it is not massive enough for its internal pressures to trigger its almost certain ball of hydrogen into the thermonuclear reactions that would make it a star. "And if you have a 'substar' revolving around a star," McCarthy says, "you have a planet."

McCarthy made the observations with colleagues Frank J. Low of Steward and Ronald G. Probst of the National Optical Astronomy Observatories, using a technique called speckle interferometry. "Speckle" is a way of compensating for the turbulence of earth's atmosphere, and is thus used for distinguishing between extremely close-together objects. The observations were made at infrared — thermal — wavelengths (1.6 and 2.2 microns), and part of the evidence that VB 8B is not a star is the fact that, by infrared, its temperature of 1,360 kelvins is about 100,000 times fainter than its "parent object."

In addition, says McCarthy, the speckle observations and the Naval Observatory's original astrometric measurements together suggest that VB 8B may be only about 10 times as massive as Jupiter, while many researchers feel that about 80 Jupiter masses would be necessary to ignite the internal fires of a star. (The object is also being called a "brown dwarf," a term that connotes an object too low in mass for starhood but which avoids using the loaded term "planet." And various theoretical brown dwarf models, McCarthy notes, allow for objects from about 80 Jupiter-masses on down.)

McCarthy's group has applied the infrared speckle technique to a number of other objects for which there have been reasons to suspect companions (VB 10, Barnard's Star, CC 1228, G 2416, BD +43°4305, BD +68°946 and Stein 2051), and, he says, "there are certainly no very bright companions there." If the astrometric measurements are correct, he says, those companions are either very small, or very dim for other reasons.

McCarthy says he has also been looking at — but declines to identify until the observations are complete — another faint star that is "likely" to have a substellar companion, and adds that there are several other possible candidates. (In fact, according to McCarthy and other astronomers, there could well be many.)

Meanwhile, as for how to categorize VB 8B, McCarthy sees no problem. "If you put that object in our solar system," he asks, "would you call it a planet? It's a big Jupiter." —*J. Eberhart*

Gene splicing to protect crop roots

Bacteria that colonize the roots of corn plants have now been endowed with the ability to fight insect pests. This work initiates "a new approach to insect control," says Robert Kaufman of the Monsanto Company of St. Louis. Monsanto's plan to field-test genetically engineered microbes next spring will be submitted to the Environmental Protection Agency (EPA) later this month in accordance with that agency's interim policy on testing of genetically engineered microbes (SN: 10/13/84, p. 229), Kaufman announced at a press conference this week.

The strategy of putting pesticidal genes into microbes that naturally colonize crop plants, if successful, will "totally revolutionize pest control, making it a safer arena for both farmer and consumer," Kaufman says. He says the approach will be very selective, affecting only organisms that actually eat the plant tissue. It will also save energy because ultimately one application of the microbe should persist as long as the plant survives. He suggests that the microbial methods will be more feasible for developing countries than are pesticides based on petrochemicals.

The Monsanto work began almost four years ago with *Pseudomonas fluorescens*, a bacterium that normally lives on the roots of corn and some other plants. Using recombinant DNA techniques, the scientists transferred a gene from another bacterium, *Bacillus thuringiensis*, into chromosomes of *P. fluorescens*. That gene encodes a protein that kills insects. *B thuringiensis* and its insect-killing toxin have been used as an insecticide for more than 20 years, are components of almost 100 commercial products and have "excellent safety properties," according to Monsanto.

In laboratory and greenhouse tests, the Monsanto scientists coated corn seeds with the genetically engineered *P. fluorescens* and demonstrated that the bacteria can colonize the roots of the plants that grow from those seeds, produce the insecticidal protein and kill worms that nibble on the roots. Monsanto has also conducted field tests with the parent bacteria, not genetically engineered, to determine that the microbes survive on the corn roots for much of the growing season but do not persist longer in the soil.

"We know it works in the greenhouse; now we need to move to the field," Kaufman says. The information to be submitted to EPA includes data on the range and life cycle of the soil bacteria to be field-tested and results of toxicity and infectivity studies, which show that the bacteria do not harm test animals such as mice, fish and quail, Monsanto says. The company also plans to supply descriptions of methods its scientists have developed for tracking the microbe in the environment and for limiting spread of the microbe.

The black cutworm is the most important corn pest that this microbe now could be used to control. However, before putting the new pesticidal microbe on the market — perhaps in three to five years — Monsanto plans to add more insecticidal genes to increase the number of insects it could destroy. Currently about 35 percent of farmers in the Corn Belt spray their fields with a chemical insecticide during planting to prevent worm damage.

The microbial strategy could be applied to a wide variety of pests. The scientists have found high levels of naturally occurring bacteria on the above-ground regions of crop plants. "I could imagine going to the husk, isolating bacteria and transforming them," Kaufman says. "Chemical insecticides as we know them could be phased out in the next 25 years." —*J. A. Miller*