industry is not as well developed as that of the U.S. The price of fluid milk has been controlled at low levels, while the input costs, largely feed, have recently been allowed to seek world levels. Thus, Mexican dairy farmers want the cost savings and productivity gains afforded by BST. Another example involves Du Pont's (Wilmington, DE) aggressive marketing of its Optimum high-oil corn, which increases feed efficiency, to Mexican poultry integrators. Although the current version is a product of conventional plant breeding, biotech is being applied to develop future products.

Obviously, Mexico is not the American market. But, as a future commercial equal, Mexico will demand increased attention from genetics companies as NAFTA is resolved. ///

I thank David Wheat of the Bowditch Group (Boston) for his review and input.

Can microbes economically remove sulfur?

Worldwide capacity for high-temperature, high-pressure, desulfurization represents a $25 billion capital investment that costs $6 billion a year to operate.

ATLANTA—Although refiners now rely on costly physical-chemical procedures to remove sulfur from crude oil, an alternative microbial-enzyme-based approach to this industrial challenge is "technically feasible," according to several researchers who reported findings during the American Society for Microbiology's (Washington, DC) recent meeting here. However, a "considerable number of chemical engineering" details will need to be worked out during the next few years before this alternative approach is ready for large-scale use, these researchers caution.

Sulfur removal from crude oil is a vital step in the high-volume petrochemical industry, an industry that processes some 20 million barrels of high-sulfur crude oil per day, which is roughly one-third of the total crude oil processed daily worldwide. During the next decade, moreover, increased demand and a depletion of low-sulfur crude-oil supplies will increase the daily volume of high-sulfur crude being processed by another 10 million barrels.

Although sulfur removal is only one component of the multistep oil-refining process, huge amounts of money are involved. For example, Chevron (San Francisco, CA) recently invested $150 million to build a new desulfurization plant. The current worldwide capacity for this high-temperature, high-pressure, desulfurization process represents a $25 billion capital investment that costs an additional $6 billion to operate each year.

Hence, an efficient, lower-cost, biotechnology-based alternative process might well attract some of the money now being spent on the traditional chemical process. One of the barriers has been to find microorganisms that can reliably metabolize sulfur-containing components in crude oil without simultaneously breaking down carbon components that need to be retained in the refined products.

Dibenzothiophene (DBT), which contains a single sulfur atom at the center of a three-ringed organic molecule, is the accepted model organic compound for studying desulfurization. Although a number of microorganisms are known to remove sulfur from DBT, many of them are better suited for bioremediation—which typically involves extensive metabolism of the carbon structure—than for the selective desulfurization required in the oil-refinery business.

However, a bacterium designated *Rhodococcus rhodocrous* and isolated by John Kilbane of the Institute of Gas Technology (IGT, Chicago, IL) is now "getting lots of attention" for its desulfurizing potential, Kilbane says. Although it can remove sulfur from DBT without degrading the carbon ring structure, the metabolic process requires air, which is considered a drawback in a petrochemical setting where air-liquid mixtures are likely to be highly flammable, if not explosive.

One way to overcome some of these difficulties would be to identify the specific genes needed for the critical desulfurization steps and then to move them into an anerobic organism or an organism that would simply produce the enzymes, says John Rambosek of Panlabs (Bothell, WA). Since last year when Rambosek and his colleagues began studying the genes of *R. rhodocrous*, they have cloned a DNA segment that appears to contain the genes encoding the enzymes responsible for desulfurizing DBT. They are now characterizing several of those genes, as well as the proteins produced by them.

According to Kilbane, at least two additional microorganism-based desulfurization processes were identified during the past few years. One of the microorganisms, an obligate anaerobe, is being characterized by investigators in Korea. It appears to require hydrogen gas as a reducing agent and to release hydrogen sulfide gas from DBT without degrading the organic ring structure.

The other microorganism, being studied by William Finnerty of Finnerty Enterprises (Athens, GA), also produces hydrogen sulfide from DBT and leaves its organic ring system intact. Moreover, because the reactions can be carried out in organic solvents, which are compatible with other oil-refining steps, Finnerty's studies represent an "exciting" development, Kilbane says.

The microorganism Finnerty is studying is a gram-positive bacterium that "requires specialized conditions to grow," Finnerty says. Unlike the Korean group's bacterium, Finnerty's microorganism does not require anaerobic conditions for growth, although the desulfurization reaction is carried out in an anerobic hydrogen atmosphere at ambient temperatures. First, though, the bacterial cells are treated with solvents so that they are no longer viable. They are then washed before being suspended in dimethylformamide, an organic solvent.

Besides the model compound DBT, Finnerty's biocatalyst can also remove in excess of 80 percent of the organic sulfur from several samples of crude oil. Although these reactions have been carried out on a small scale in a batch reactor, Finnerty is developing a continuous-flow, immobilized-bed system and scale-up efforts are under way.

—Jeffrey L. Fox