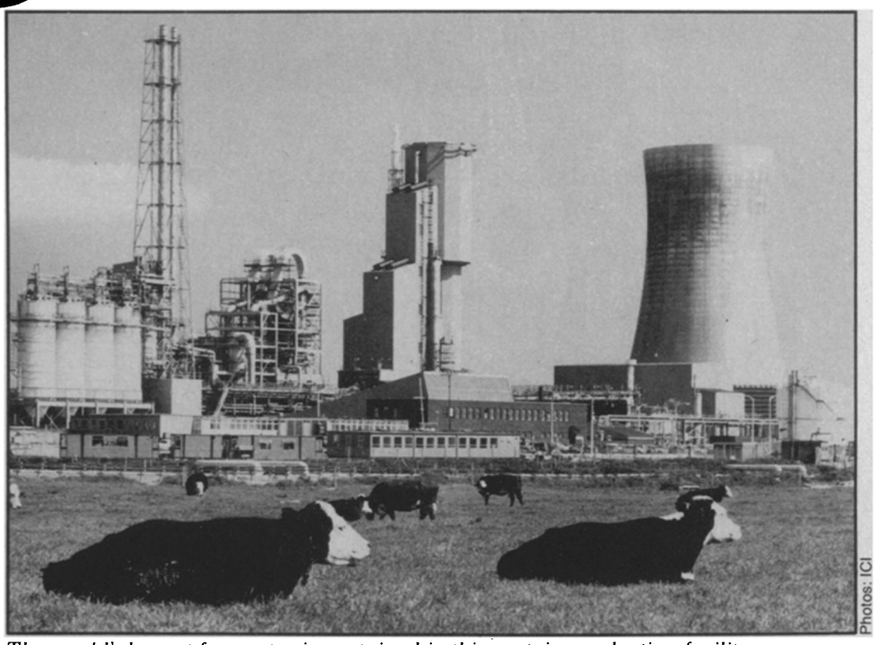
A Single-Cell Feast

Full-scale production of a microbial high-protein feed supplement is underway, at last, after almost 25 years of research

BY MARTIN SHERWOOD



Photos: ICI

The world's largest fermenter is contained in this protein production facility.

During 1981, about 50,000 tons of high-protein animal feed will be dispatched to European farmers from a 15-acre production site at Billingham in northern England. This "miracle" crop, which apparently yields more than 3,000 tons an acre — compared with about 2 tons for wheat — is made up of dried cells from a microorganism that lives on methanol. The key item on the 15-acre site, owned by the chemical company Imperial Chemical Industries Ltd., is the world's largest fermenter. The product of the fermenter is Pruteen — a result of nearly a quarter century of research on single-cell protein production and a good example of the potential of the growing field of biotechnology. And now that a fully operational plant exists, there could be rapid growth for this technology in countries that, unlike the United States, cannot produce sufficient animal feed by conventional agriculture.

In western Europe, for example, the protein in livestock diets comes mainly from soy and fishmeal. After oil, soy — which will not grow in most parts of Europe — is the second biggest European import in money terms. Ironically, the idea of producing large quantities of microbial cells as a feedstuff was first explored commercially because oil was cheap and supplies seemed more reliable than those of agricultural products.

In France, in the late 1950s, the British Petroleum Co. began to study the growth of yeasts with gas oil instead of carbohydrates as their carbon source. This crude oil fraction contains between 10 and 25 percent straight-chain waxy hydrocarbons that can be metabolized by yeasts. But, because the major part of the gas oil is not metabolized, another company in the BP group — this one based in Scotland — developed an alternative process that used a purified straight-chain hydrocarbon mixture as the yeasts' carbon source. Other companies, notably in Japan, developed similar processes.

At the same time, following major discoveries of natural gas in the North Sea, other companies — including ICI and Shell — became interested in feeding bacteria on methane. Shell experimented with this for several years, but then gave up; ICI, after less than a year, decided to switch from methane to methanol.

A major difficulty in trying to feed microorganisms on hydrocarbons is that the organisms need an aqueous environment and hydrocarbons — either as liquids or gases — are not water soluble. Methanol, on the other hand, dissolves easily in water. Consequently, ICI, which was already making methanol from natural gas on a large scale at Billingham, felt that the engineering problems would be much less severe with a methanol-based process.

As happened with the discovery of the major antibiotics, thousands of soil samples were screened to try to find an organism that would thrive with methanol as its carbon source. Thirteen years ago, ICI found *Methylophilus methylotrophus,* the organism it has used ever since. This bacterial species uses carbon from methanol, nitrogen from ammonia, oxygen from air and a variety of trace elements to grow. Given a balanced diet and a temperature of 35°C to 40°C, the cells will divide every two and a half hours, although division takes place at only about half this rate in ICI's full-scale plant.

It took the company several years and several million dollars — total R&D expenditure on the protein project has now probably exceeded $40 million and continues to rise — to move its chosen microorganism from the laboratory to the pilot plant. A 1,000-ton pilot plant was completed at Billingham in 1973 and provided much of the basic design data for the full-scale plant. Both plants use an air-lift fermenter in which there are no moving parts; the contents are circulated and stirred by air injected under pressure at the base of each fermenter.

The pilot plant also provided the hundreds of tons of Pruteen that were needed for animal feeding trials and toxicological testing. It was in the latter area that ICI's competitors came unstuck. Early in 1973, two Japanese companies—Dainippon and Kanegafuchi — had to abandon plans for single-cell protein plants because of public concern about minute traces of carcinogenic hydrocarbons, such as 3,4-benzopyrene, in the yeast produced with their technology.

Japanese technology was used, however, in a plant constructed in southern Italy by Liquichimica. At the same time, BP —in conjunction with the Italian company Anic — also built a full-scale plant in southern Italy for its pure hydrocarbon process. Neither of these plants has ever operated because, in 1976, the Italian government withdrew permission for marketing the products. It claimed that they contained unsatisfactory levels of unconverted hydrocarbons although, as BP pointed out, the amounts of hydrocarbon in the products were substantially lower than those found in commercially sold rice that had been polished with mineral oil.

Both BP and ICI have carried out very extensive toxicological testing of their

Martin Sherwood is a British science writer and editor of CHEMISTRY AND INDUSTRY.

products, including multi-generation trials, in a variety of animal and bird species. Neither product has ever given any indication that it is not completely safe as an animal feedstuff. However, following the intransigence of the Italian authorities, BP has now given up its interests in single-cell protein and ICI, which has approval to market Pruteen from regulatory authorities in about a dozen countries, has not considered it worthwhile to apply for approval in Italy.

On nutritional grounds, single-cell material offers a valuable high-protein supplement for animal diets. Pruteen, for example, contains more than 70 percent protein and has a good balance of essential amino acids. Given the inefficiency with which livestock converts its feed into meat for human consumption, why has ICI concentrated on marketing its new product as an animal feed rather than one for direct human consumption?

Part of the reason is that, because of its role as a major European fertilizer manufacturer, ICI already has a good knowledge of the agriculture business, while its involvement with the food industry is very limited. Second, the eating habits of animals are unaffected by social factors. There is no problem in persuading a pig to eat a diet containing microbial cells; most humans would need coaxing before they switched to such an unconventional food source. And there is an additional, health reason why single-cell protein may not be suitable for direct human consumption: its nucleic acid content.

In humans, the purine bases in the nucleic acids are metabolized to insoluble uric acid, which may cause kidney stones and other disorders. Consequently, the United Nations Protein Advisory Group has recommended a maximum daily intake of nucleic acids by humans of 2 grams. Dried bacterial cells may contain up to 20 percent nucleic acids. In most animals, on the other hand, the purines are metabolized to a soluble compound, allantoin, which is excreted, so the problem does not arise.

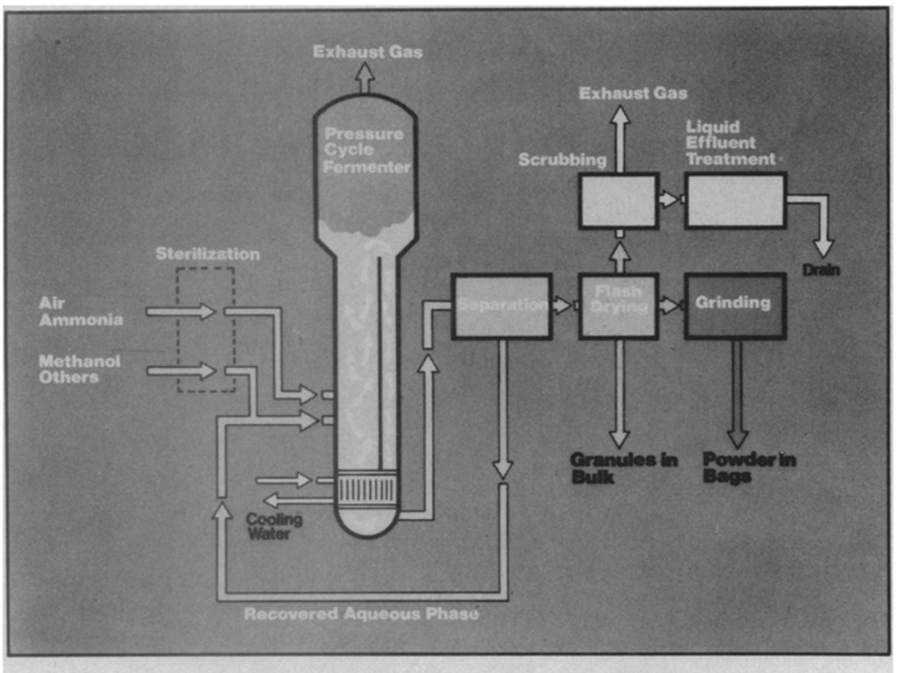
Having shown that its product could provide a useful addition to the diets of pigs, poultry and calves, ICI decided in 1976 to build its first full-scale plant. Completed at the end of 1979, at a cost of about $95 million, it took nearly another year to get it working satisfactorily.

A major problem was to achieve and maintain sterility. If the quality of the product is to be maintained, there must always be only one strain of microorganism in the 1,300-m^3 fermenter. This means that everything going into the fermenter must be sterilized and that all the instrumentation required to monitor conditions inside must be able to communicate readings to the outside without bleaching the sterile containment. Much of 1980 was spent in test runs to see how long the plant could be kept sterile. ICI now believes that continuous operation for periods in excess of a year will be possible.

In the Pruteen process, air, methanol and other raw materials are fed continuously to the fermenter. Carbon dioxide, produced by cell metabolism, is released at the top and a portion of the liquid content is removed continuously at the bottom. The liquid removed is steam-treated to kill and coagulate the microbial cells, which are then separated from the water by centrifugation. The water is recycled to the fermenter and the cell mass is dried and either sold directly as granules or ground to a powder.

Although the full-scale plant is now completely operational, the pilot plant is still being used for further research. The process may be improved in a number of ways and ICI is keen to make it more efficient before a second plant is built. Although there are no definite plans yet, the company is already talking in terms of an output five or six times greater than that of the first plant with a fermenter of about the same size.

One possibility is that, in addition to improving the engineering for the hardware, the organism itself could be reengineered. In the Oct. 2 NATURE, a team of ICI researchers published a paper describing genetic modification of *M. methylotrophus*'s nitrogen assimilation system. If the modified organism is stable, it could need a few percent less energy for metabolism than the original organism needs. And that means a few percent more methanol being incorporated into cell mass rather than going to waste as carbon dioxide. If these and other experiments prove successful, the future could look bright for Pruteen. □



Bacteria growing in fermenter are the key to Pruteen production (diagram above). Calves at an ICI research station willingly drink a milk substitute based on single-cell protein.