FERMENTOR DESIGN

CONTINUOUS CULTURES WITHOUT WASH-OUT

PHILADELPHIA—Plant and animal cells—in addition to microorganisms—grow to extremely high concentrations in the Bio Reactor, a novel fermentor that incorporates a spin-flow filter.

Developed at Arthur D. Little (Cambridge, MA), the Bio Reactor's configuration permits continuous feeding of a culture without loss of biomass. Hybridoma cells, for example, grow to densities of nearly 10^8 cells/ml, about 50 times higher than with normal culturing techniques. Scientists from DNA Plant Technology (Cinnaminson, NJ) reported similarly encouraging results with plant cell cultures.

Traditional continuous-culture bioreactors are based on the chemostat principle—steady-state growth in a continuous-flow culture. A chemostat provides a consistent environment for a culture by feeding it at a constant, controlled rate. High cell growth rates require high feeding rates. But since the overflow rate equals the feeding rate, a proportional fraction of the cells is lost from the culture.

Because the nutrient supply that determines the growth rate of chemostat cultures is always balanced by cell wash-out, high densities can be achieved only at the expense of rapid cell growth. The Bio Reactor is designed around an output filter that removes fermentation liquor but retains the cells within the culture vessel. "What we have done," explained Dennis Johnson, an A. D. Little chemical engineer speaking at a recent DNA Plant Technology meeting, "is to uncouple control of growth rate from control of cell density."

The design of the spin-flow filter is quite simple. A cylindrical filter rotates at the center of the culture vessel, immersed in the cell culture medium. The inner chamber of the filter is attached to an overflow outlet. As additional nutrient fluid is added to the vessel, the spin-flow filter mixes it in with the whole culture while removing an equal volume of cell-free culture fluid. "The idea is so simple," said Johnson, "that we were sure someone else must have tried it."

Apparently no one had, and A. D. Little now holds a patent on the design. The Virtis Company (Gardiner, NY) manufactured a spin-filter fermentor until recently, but suspended production pending a decision by A. D. Little on just how it wants to modify and commercialize the technology. "We originally developed the Bio Reactor to simulate large-scale process fermentations in the laboratory," explained Johnson. "We came up with the design well before people were very interested in large-scale cultures of mammalian or plant cells." Johnson hopes to use these new applications to extend patent protection for the Bio Reactor.

A. D. Little offers the technology as part of its technical services program for clients. Johnson says they have built spin-flow fermentors as large as 14 liters. He predicts that, thanks to the high cell densities achieved in Bio Reactors, 15–100 liter batches will be sufficient for manufacturing many animal or plant cell products.

A recent success with the Bio Reactor was reported by Chip Styer of DNA Plant Technology. Styer has been using a spin-filter fermentor to grow a number of different plant cell cultures. Although his studies are still preliminary, Styer expects to achieve the same increases in cell density with his plant cell cultures that others have obtained with animal cell cultures. Moreover, the system is ideal for multiple-phase cultures, such as those used to produce secondary metabolites.

"This design has real advantages over any other bioreactor," said Styer, "because you can completely exchange the culture media without cell wash-out." He suggested a scenario in which the cells are first grown to maximum density in a growth-promoting medium. Then the researcher turns off the continuous feed valve and removes the culture fluid through the spin filter. A new, production-enhancing medium can then be added to the cells without diluting them. If the product is secreted, it can even be harvested through the spin filter.

The spin filter design is a particular godsend for cell culturists contending with cells that grow too slowly to survive in traditional continuous processes. "This is really a whole new approach that has opened a lot of new possibilities," says Styer. "We know what to do to make bacteria and yeast commercially productive. Now we can see what we can do with plant cells."

—Tazewell Wilson